Mycotoxins and Health

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Professor

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Wheat Quality Laboratory

NDSU Wheat Quality Lab

Project Leader

Dr. Senay Simsek

- Survey: Regional Quality Report
- Export Cargo Survey
- Nursery Samples: Breeding Programs
  - Including Field Plots
- Research Projects

Established 1907

My Research Team

Graduate Students:

Marie Maneka Rubera (PhD)
Natsuki Fujiwara (PhD)
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Bethany Stebbings (MS)
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Md Mahfuzur Rahman (MS)
Ana Maria Lopez (MS)
Abdulrahman Alahmed (MS)
Daniel Dumas (MS)

Time Slips and Visiting Scientists

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Angela Ostenson
Tuba Turkmen
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Kaitlyn Peterson

Research Specialists and Technicians

Kaitlin Beck
Chris Cosette
Karen Dickey
Kelly McMonagle
DeLane Olsen
Kristin Whitney
Kathy Christianson

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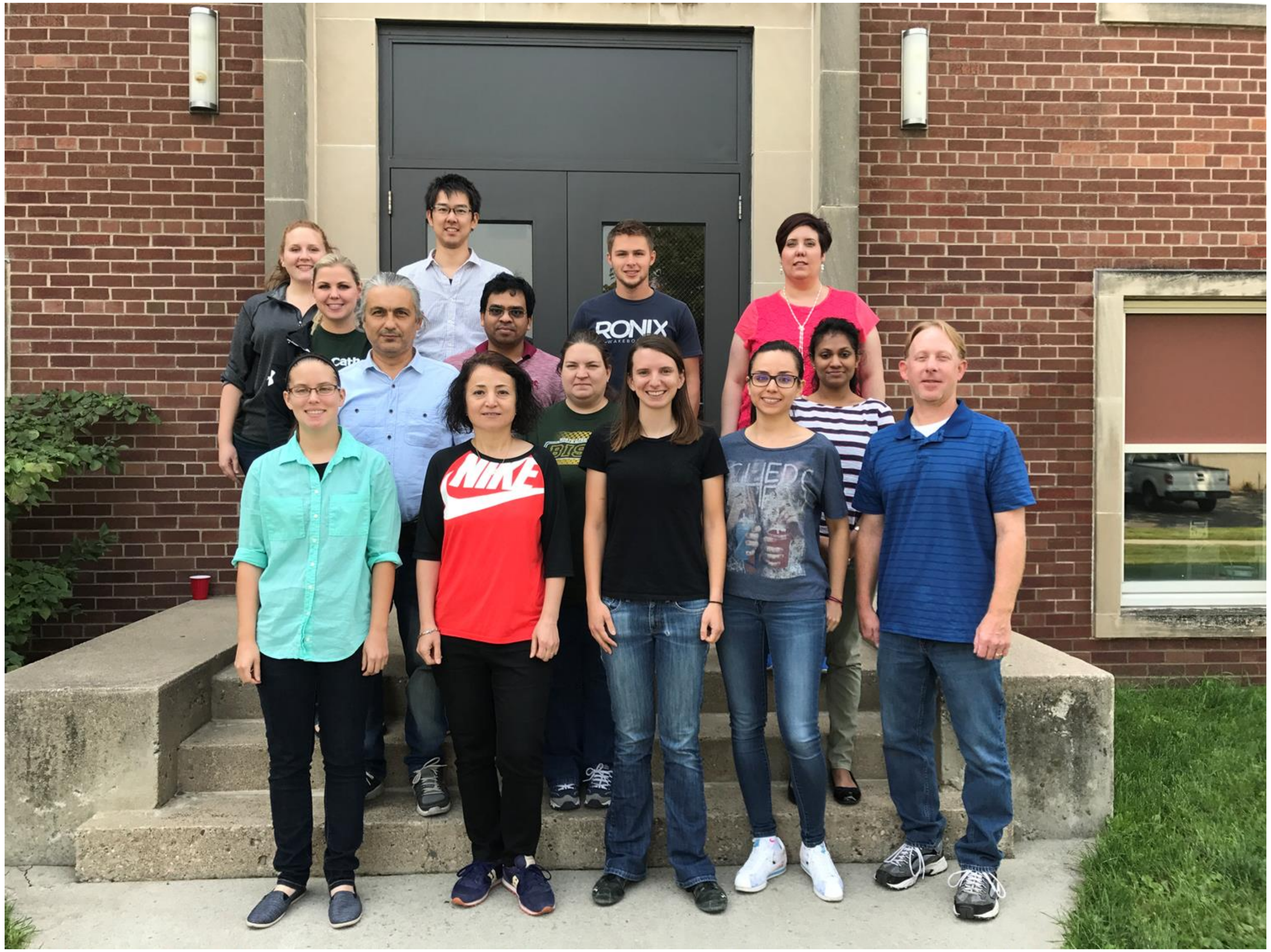
Total: 21 members + Dr. Simsek (Project Leader)


RONIX
NIKE

FUSARIUM HEAD BLIGHT

Hard Red Spring Wheat

Specialty wheat grown in the Northern Plains of United States

- Highest protein content
- Hard endosperm
- Red bran
- Strong gluten
- High water absorption

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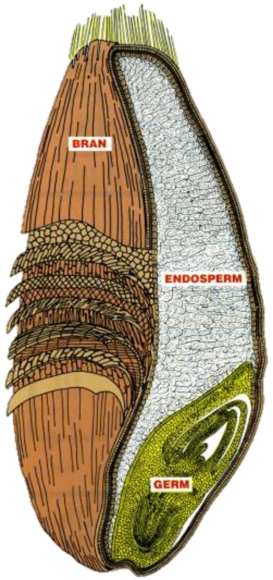
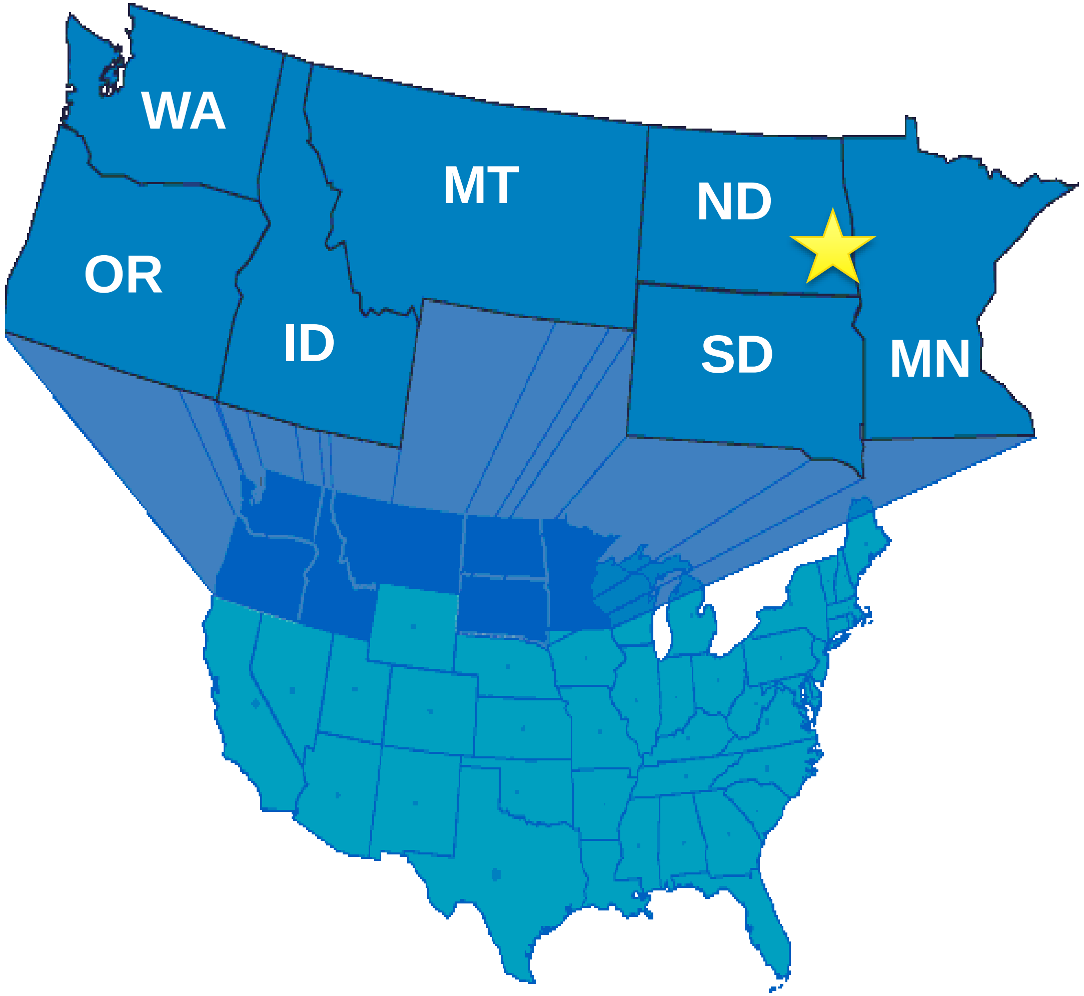




Mold

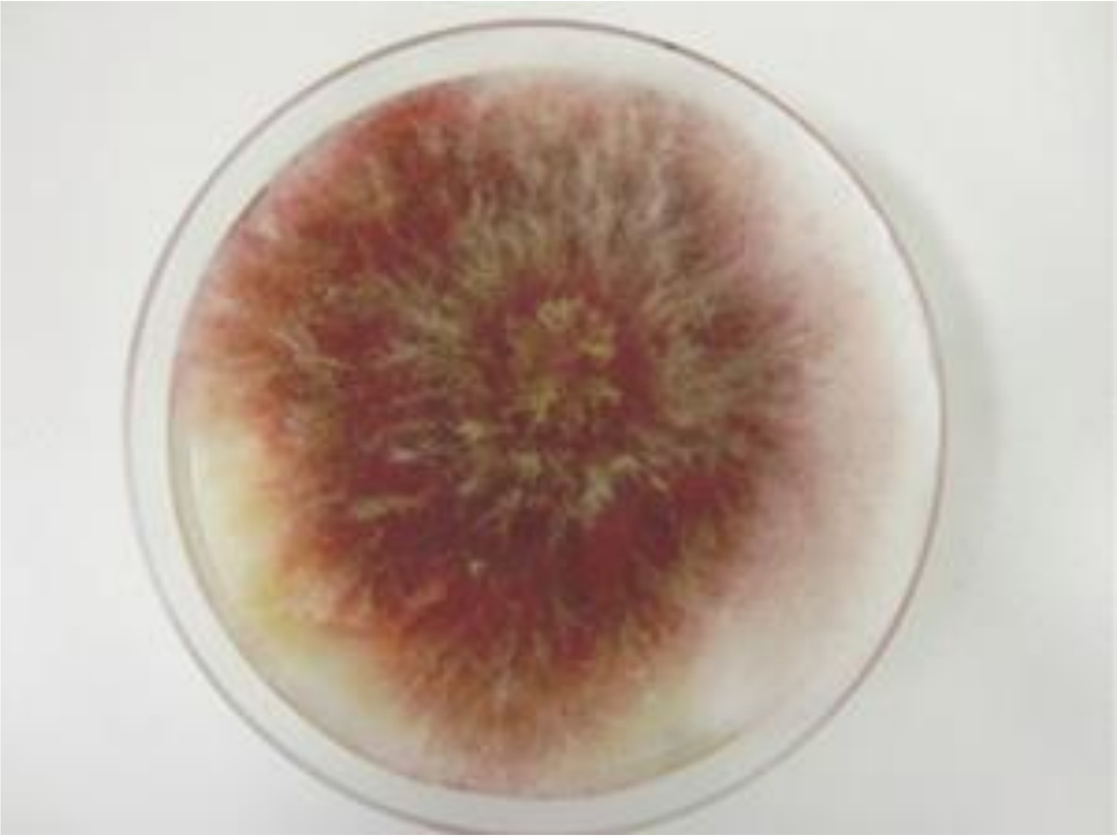


Figure 1. *Fusarium graminearum* mycelium growth on artificial growth media.

Nebraska Extension. 2012. Major *Fusarium* diseases on corn, wheat, and soybeans in Nebraska. http://extensionpublications.unl.edu/assets/html/g2181/build/g2181.htm (accessed 2017 December 17).

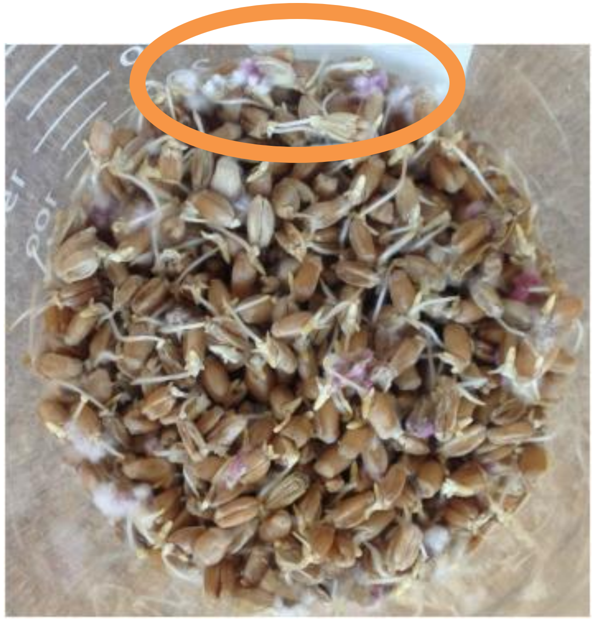
Important Definitions

- Toxin
- Toxicology
- Infection vs intoxication

FUSARIUM GRAMINEARUM INFECTIONS IN GRAIN

Wheat infected with *Fusarium graminearum.*

Wisconsin Field Crops Pathology. *Fusarium* head blight (scab) of wheat. https://fyi.uwex.edu/fieldcroppathology/fusarium-head-blight-scab-of-wheat/ (accessed 2017 December 17).

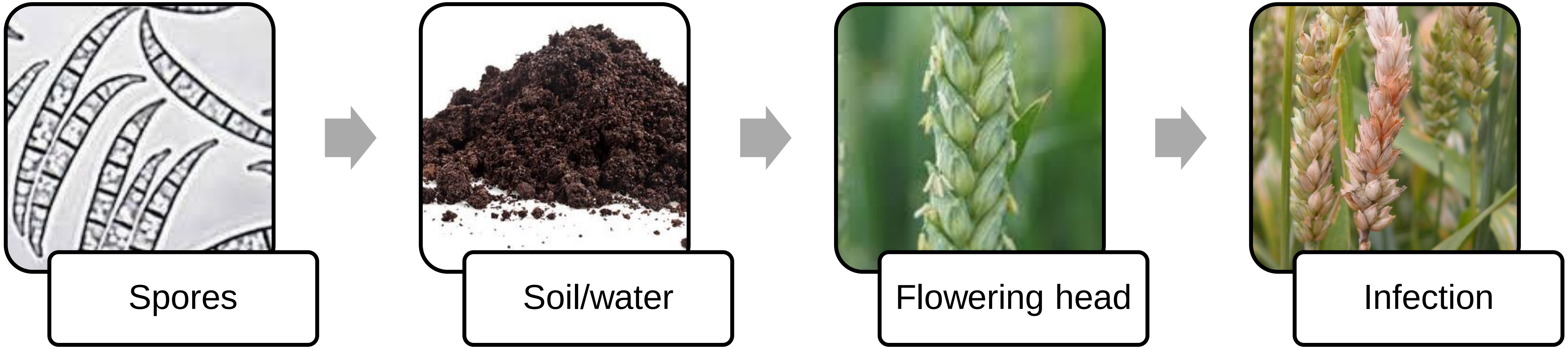


Fusarium graminearum mycelium growing on germinating wheat.

Jin, Z., Zhou, B., Gillespie, J., Gross, T., Barr, J., Simsek, S., Brueggeman, R., and Schwarz, P. 2018. Production of deoxynivalenol (DON) and DON-3-glucoside during the malting of *Fusarium* infected hard red spring wheat. Food Control. 85:6-10.

Wheat infected with *Fusarium graminearum*

Fusarium Head Blight (FHB) Cycle



Disease cycle of Fusarium head blight.

Ohio State University Extension. 2017. *Fusarium* head blight or head scab or wheat, barley, and other small grains. https://ohioline.osu.edu/factsheet/plpath-cer-06 (accessed 2017 December 17).

Fusarium graminearum Growth

Crops

- Wheat
- Barley
- Corn
- Rye
- Oat

Weather

- Warm
- Moist
- Humid

Farming style

- Tilling
- Crop rotation
- Fungicide

Production of DON by *F. graminearum*

Highly variable

Temperature, water activity, pH, nutrient availability, etc.

Optimum conditions

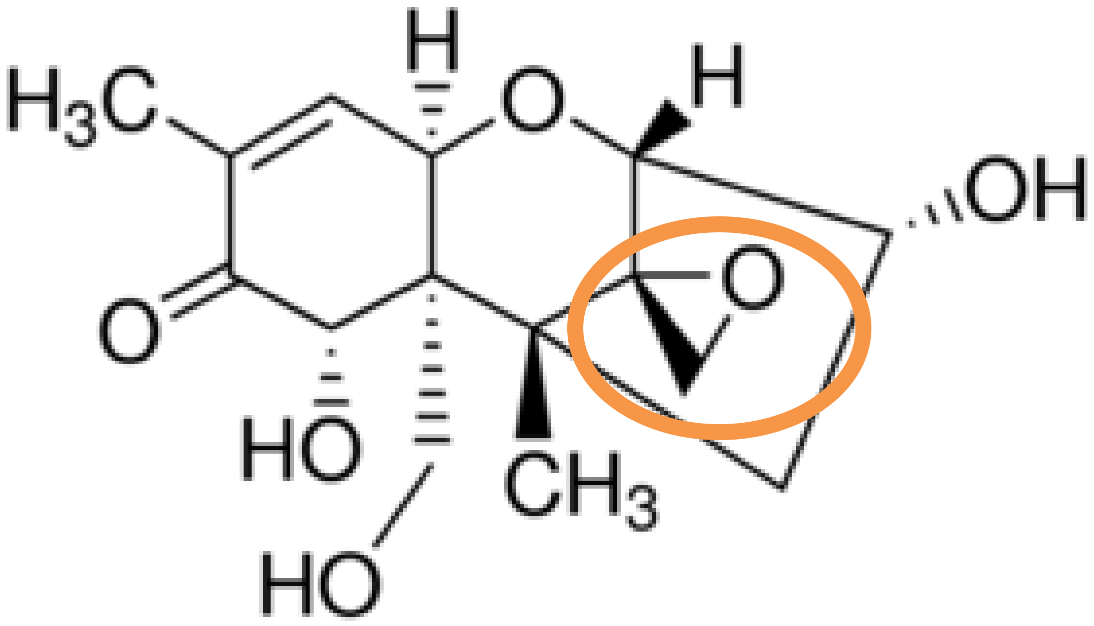
pH 4.91 and 23.75 °C

Inducer of DON production

Expression of the *Tri5* gene

Deoxynivalenol (DON)

- Mycotoxin produced by *Fusarium granimearium*
- Vomitoxin
- 12,13-epoxy trichothecene
- Water soluble toxin



Chemical structure of deoxynivalenol.

Sigma Aldrich. 2017. Deoxynivalenol. https://www.sigmaaldrich.com/catalog/product/sigma/d0156?lang=en&region=US (accessed 2017 December 17).

FDA Limits on DON in Food and Feed

Table 3. FDA limits on DON in food and feed.

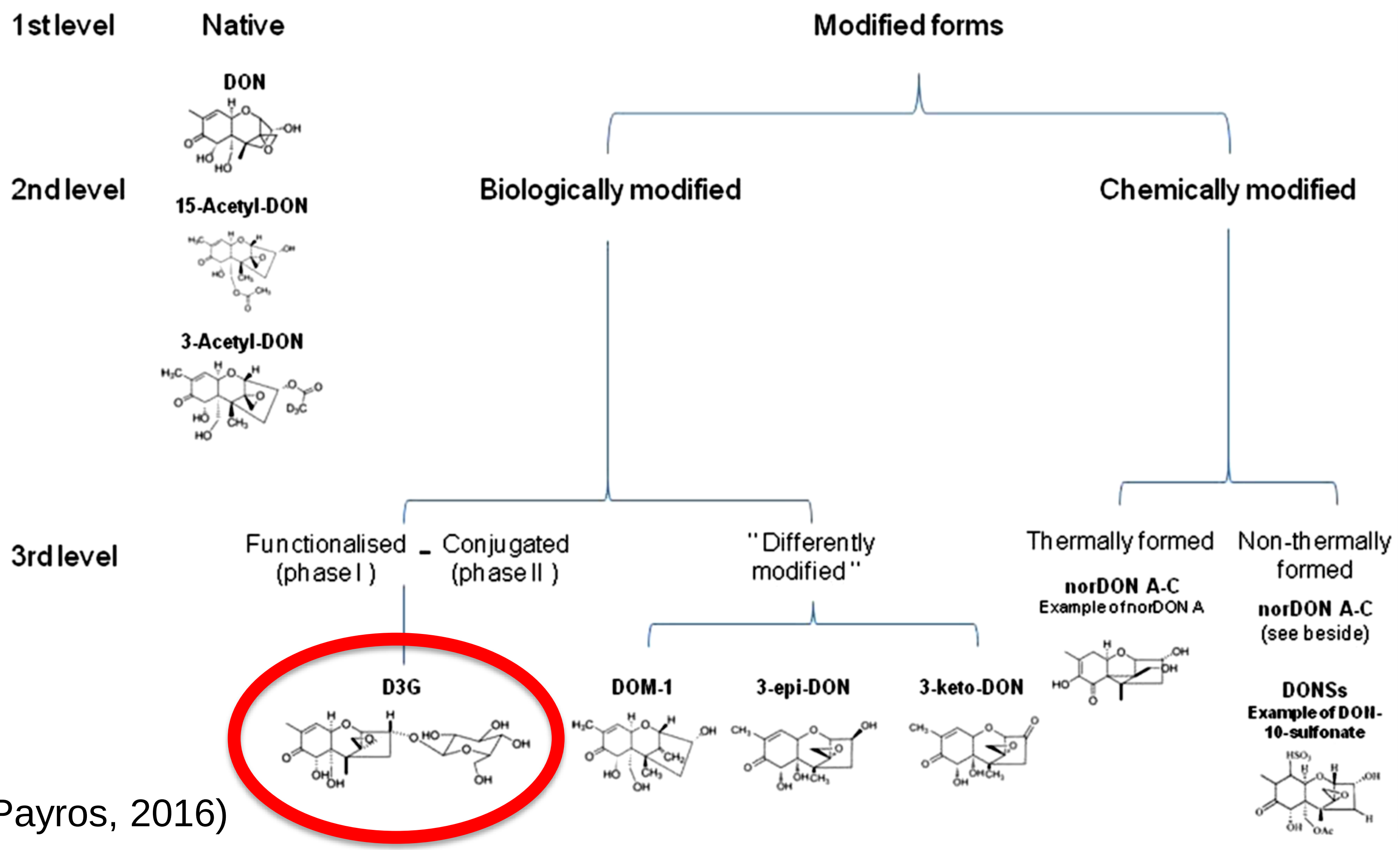
Product	DON limit (ppm)
Wheat products for humans	1
Feed for pigs (<20 % of diet)	5
Grain for chicken (<50 % of diet)	10
Grain for cattle (<50 % of diet)	10
Distillers grain and gluten feed (Cattle >4 months old)	30
Grain for all other animals (<40 % of diet)	5
Raw grain intended for processing	No limit

Modification of Mycotoxins

- Biological and chemical changes can modify mycotoxin structure
- “Modified mycotoxins”
  - Different forms of mycotoxins
- “Masked mycotoxins”
  - Only toxins conjugated by plants

(Payros, 2016)

Modification Mechanism



NDSU (Payros, 2016)

Bound Mycotoxins

Deoxynivalenol-3-glucoside (D3G)

- 3-β-ᴅ-glucopyranosil-4-deoxynivalenol

Produced by the plant as a detoxification method

Samples with DON may have DON-3-glucoside

D3G/DON ≈ 10-30%.

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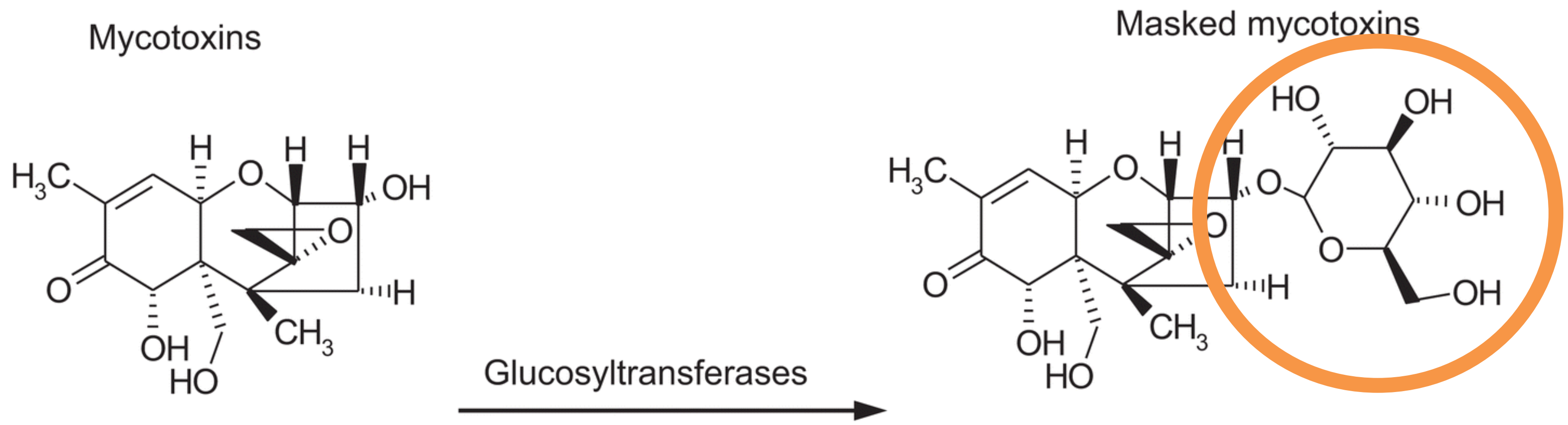
DON-3-glucoside (D3G)

- Phase II metabolism product
- Stored in plant vacuoles
- Less toxic than DON
- Masked mycotoxin



Reaction mechanism of the formation of deoxynivalenol-3-glucoside from deoxynivalenol.

Berthiller, F., Maragos, C. M., and Dall'Asta, C. 2015. Introduction to masked mycotoxins. Pages 1-13 in: Masked Mycotoxins in Food: Formation, Occurrence and Toxicological Relevance.

Agricultural Costs of *Fusarium graminearum* and DON

Decreases

- Yield, quality, and overall grade

Financial losses

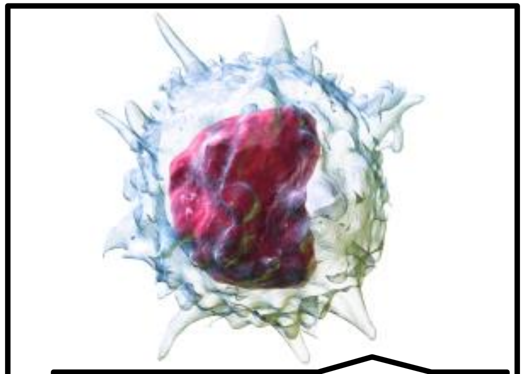
- Value of yield lost: $1 billion
- Testing at elevators: $21 million
- Added costs at mills: $8 million

DON, D3G AND HEALTH

Illness from Deoxynivalenol

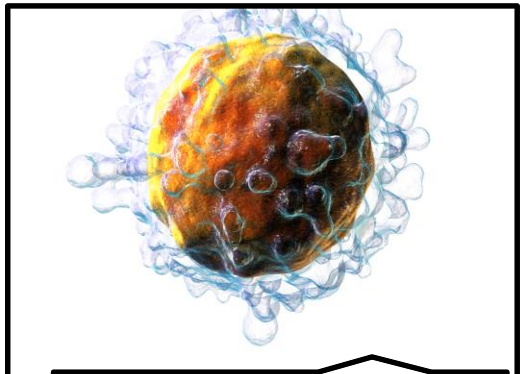
Acute exposure	Diarrhea	Vomiting	
High doses	Circulatory shock	Reduced cardiac output	Death
Chronic exposure	Anorexia	Reduced weight gain	Altered nutritional efficiency

Cellular Targets Of DON

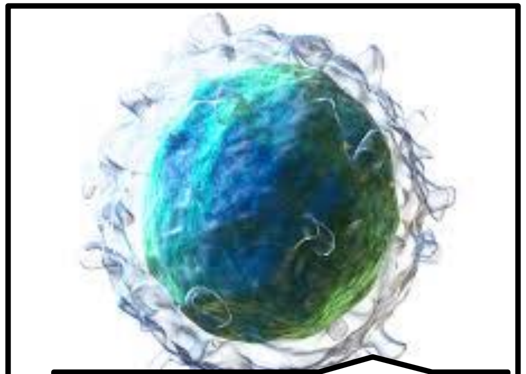


Monocyte

Macrophage



T-lymphocyte

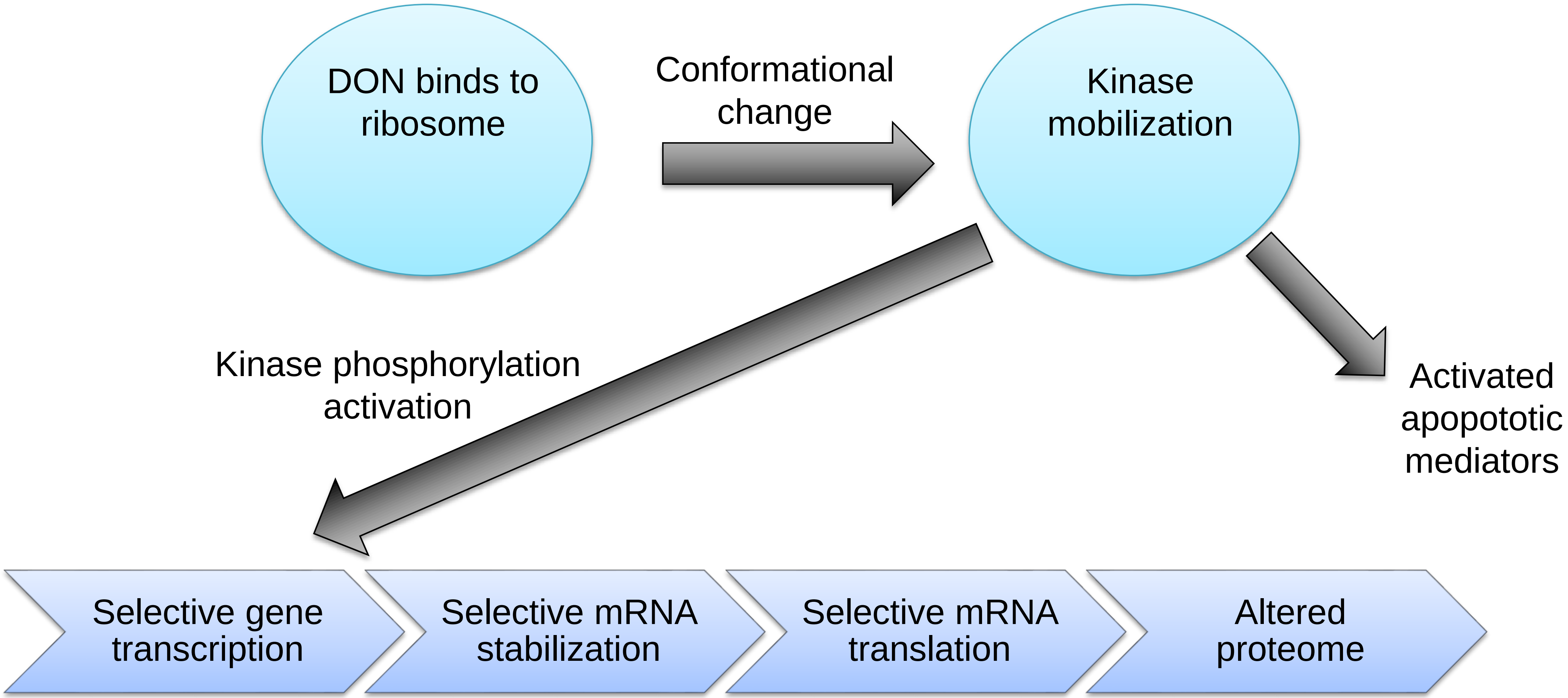


B-lymphocyte

Cellular targets of DON.

Selfhacked 2018. Danger of high or low monocytes. https://selfhacked.com/blog/monocytes/ (accessed 2018 January 18).

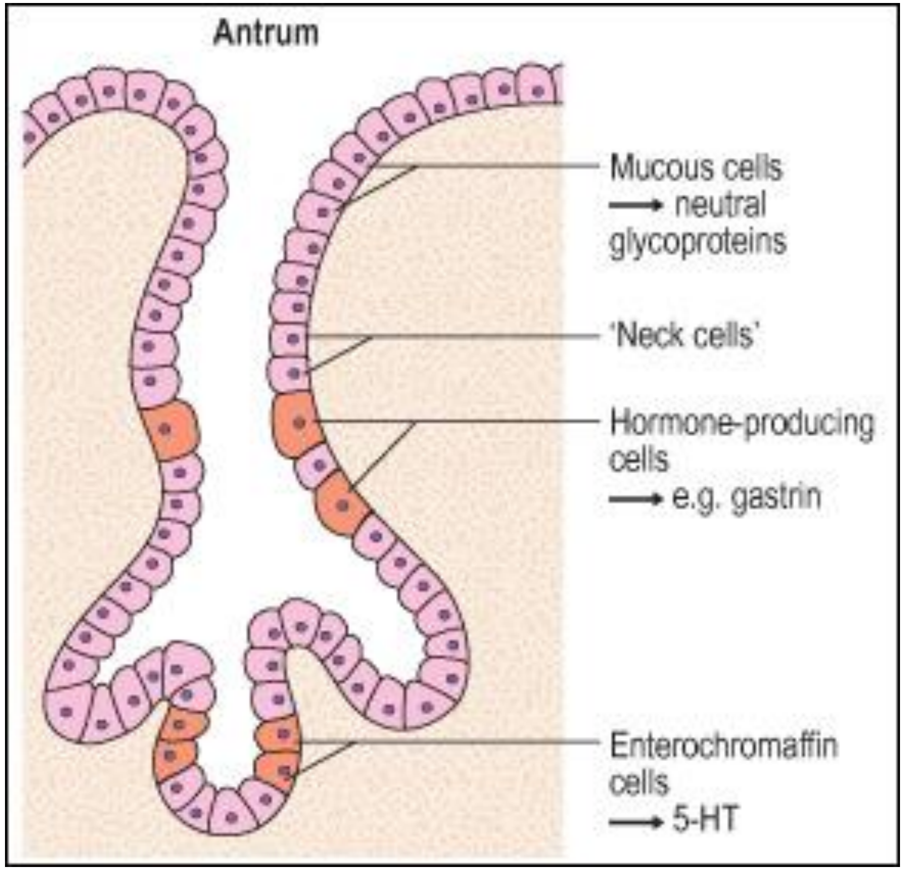
Ribotoxic Stress Response



Ribotoxic stress response overview.

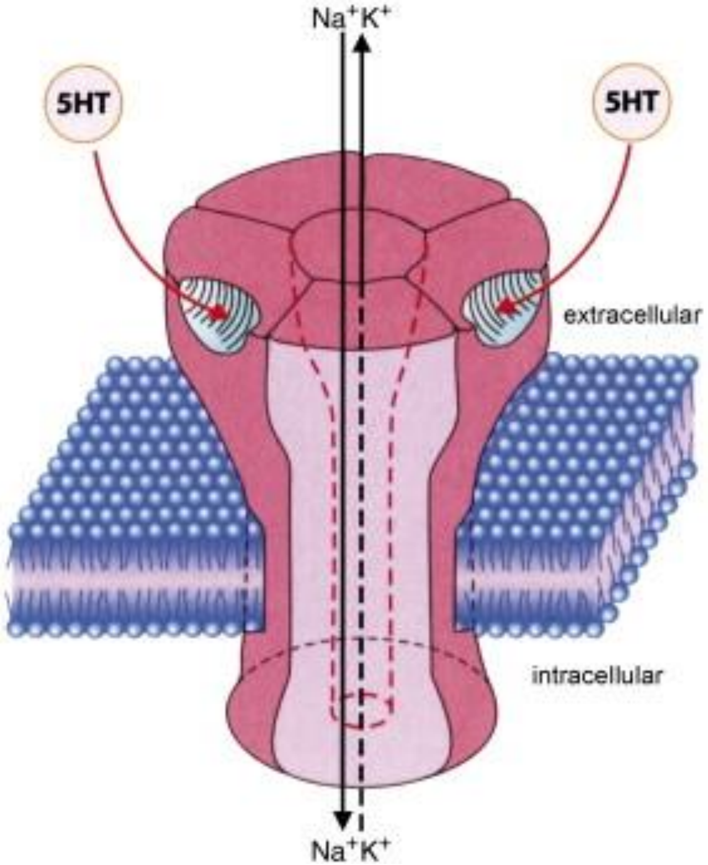
Pestka, J. J. 2010. Deoxynivalenol: Mechanisms of action, human exposure, and toxicological relevance. Arch. Toxicol. 84:663-679.

Reduced Food Intake



Enterochromaffin cells.

The Rise of Caliphate. 2010. L5-Patho peptic ulcer disease. https://ak47boyz90.wordpress.com/2010/09/02/l5-patho-peptic-ulcer-disease/ (accessed 2017 December 21).



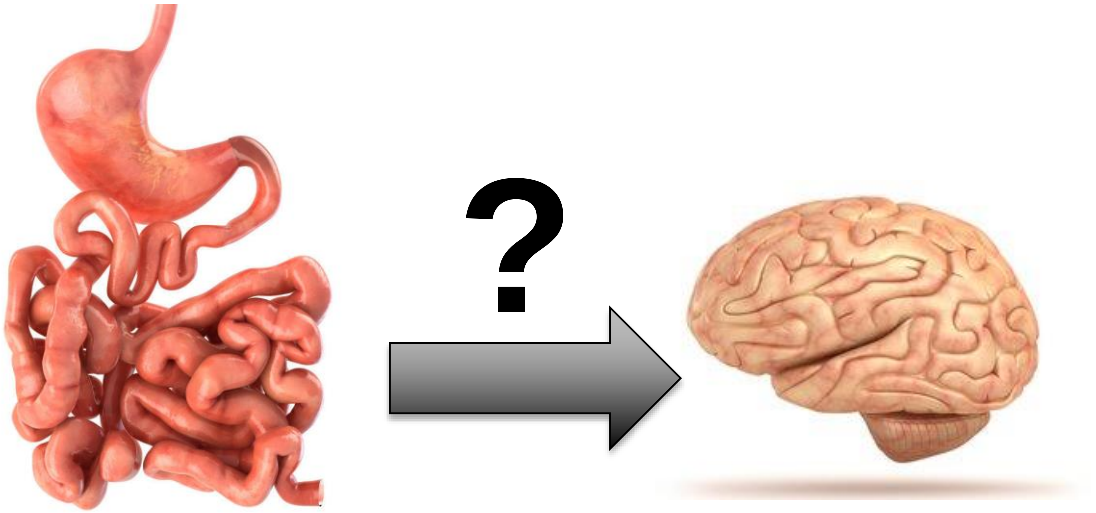
Serotonin receptor.

Faerber, L., Drechsler, S., Ladenburger, S., Gschaidmeirer, H., and Fischer, W. 2007. The neuronal 5-HT3 receptor network after 20 years of research – Evolving concepts in management of pain and inflammation. Eur. J. Pharmacology. 560(1):1-8.

Altered Nutrient Uptake

- Shortened villi
- Enterocyte lysis
- Glucose/galactose transporter inhibition
- Inhibition of fructose, serine, and glucose transporters

Vomiting



Norepinephrine ↑

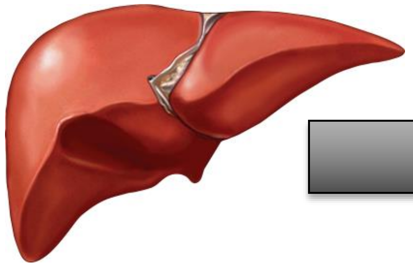
Dopamine ↓

Serotonin ↑

Vomiting due to ingestion of DON.

Chen, L., Peng, Z., Nussler, A. K., Liu, L., and Yang, W. 2017. Current and prospective sights in mechanism of deoxynivalenol-induced emesis for future scientific study and clinical treatment. J. Appl. Toxicol. 37:784-791.

Detoxification of DON



Phase I metabolism

Oxidized DON

Glutathione
S-transferase

Glutathione-DON
conjugate

Phase II metabolism

Deoxynivalenol detoxification.

Sobrova, P., Adam, V., Vasatkova, A., Beklova, M., Zeman, L., and Kizek, R. 2010. Deoxynivalenol and its toxicity. Interdiscip. Toxicol. 3:94-99.

Species Susceptibility to DON

Most: Pigs → Rodents → Dogs → Cats → Poultry → Least: Ruminants

Table 2. Species susceptibility to deoxynivalenol.

Animal	LD_{50} (mg kg^{-1} body weight)	Administration method
Mice	46-78	Oral
1-day old chicken	140	Oral

Digestion of D3G

- Limited studies on
  - Fate of D3G and other masked mycotoxins in the gut
  - Contribution to toxicity

Digestion and Absorption

	In vitro	*In Vivo*
Stomach	Stable	Stable
Small Intestine	Stable	Stable
Large Intestine	Hydrolyzed	Hydrolyzed (Indirect evidence)
Absorption	< 1%	< DON
Toxicity	< DON	< DON

Gratz 2017

Toxicity of D3G

- Thought to have limited bioavailability in the gut
  - Due to covalent binding

- D3G cannot sterically bind to ribosomal 60S subunit A-site pocket
  - Target for DON-induced ribosomal toxicity

Gratz 2017

Toxicity of D3G

- D3G
  - Does not induce ribotic stress
  - No activation of JNK and p38 MAP kinasis
  - Does not alter viability and barrier function of intestinal cells
  - Does not induce morphological lesions or pro-inflammatory gene expression in explants

(Payros, 2016)

D3G Hydrolysis by Fecal Microbiota

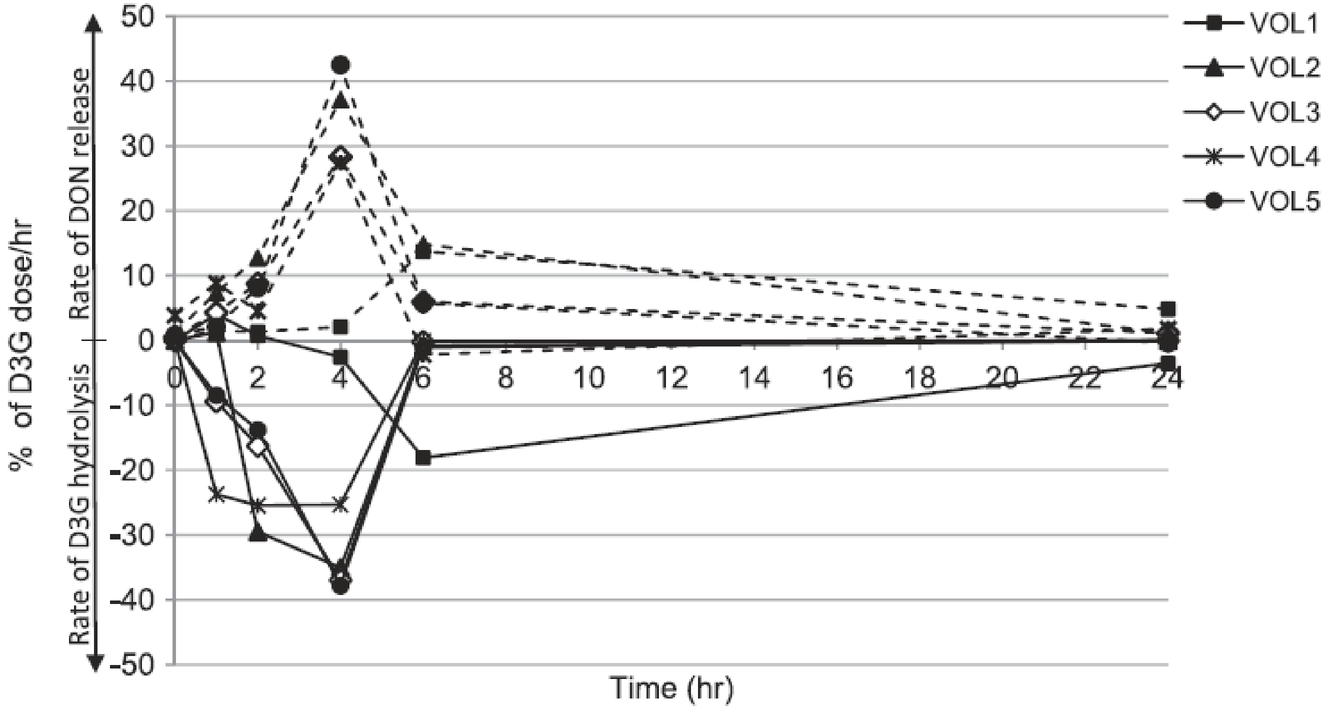


FIG 1 Rate of hydrolysis of D3G and release of DON by the fecal microbiota of 5 volunteers (VOL). Data are presented as D3G hydrolyzed per hour (% of D3G dose [solid lines]) and DON released per hour (% of D3G dose [dashed lines]).

Gratz et al., 2013

D3G Hydrolysis by Fecal Microbiota

- Fecal microbiota cleave D3G and release DON
- Hydrolysis rates range from 25-38% per hour after 4 hour incubation
- 100% D3G is degraded after 6 hours

Gratz et al., 2013

D3G Metabolism by Human Microbiota

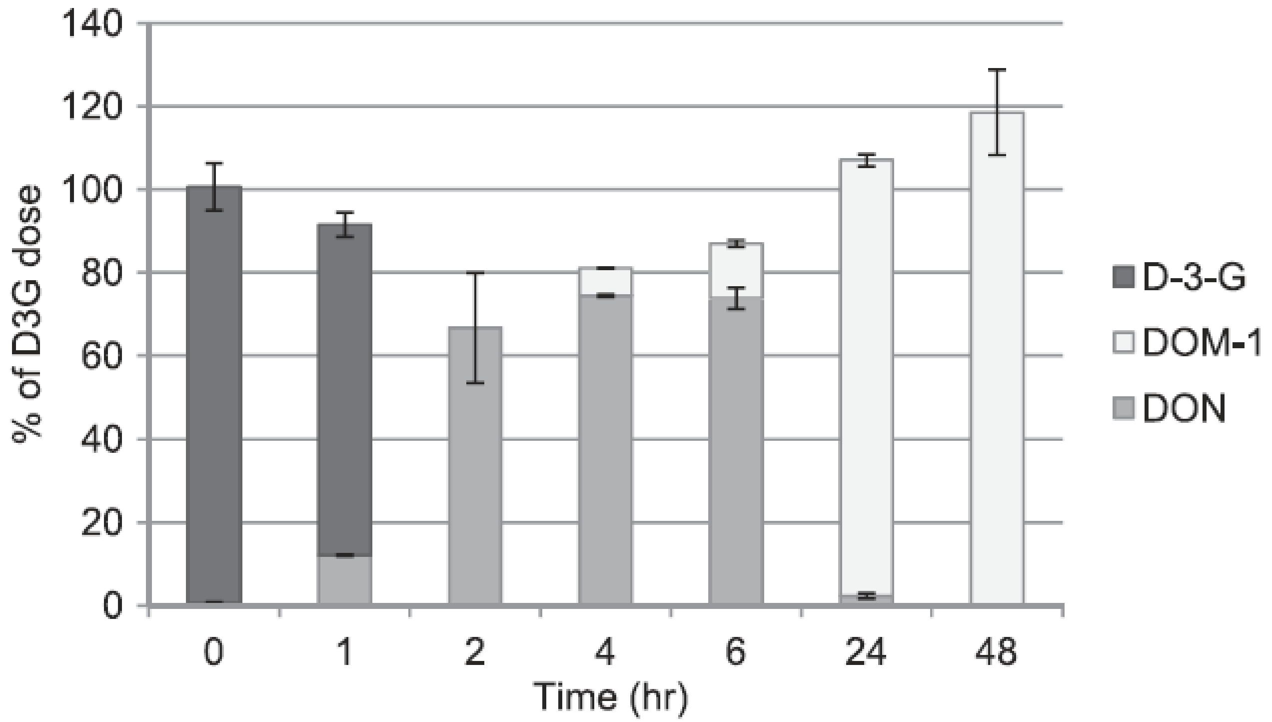


FIG 2 Metabolism of D3G by the human microbiota of volunteer 5. Results are presented as means of duplicate incubations for each time point.

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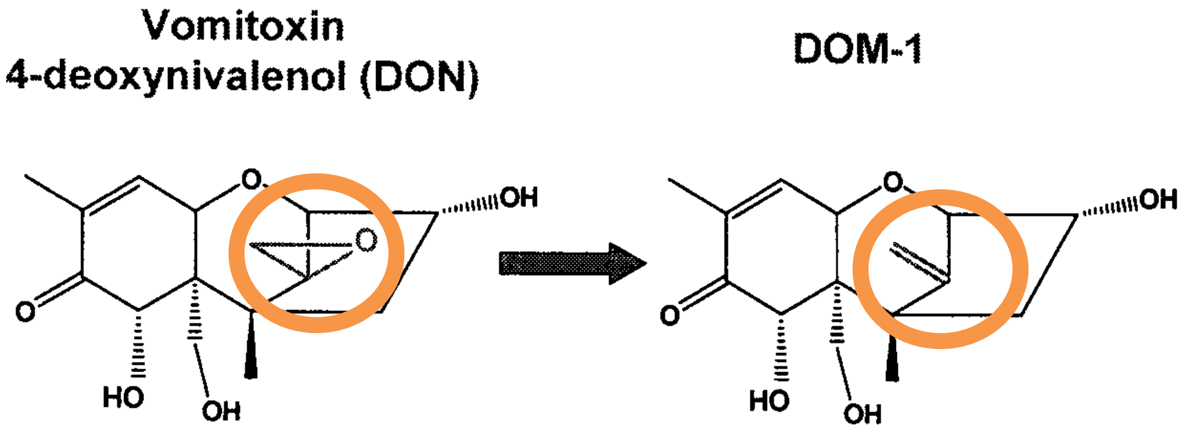
Gratz et al., 2013

De-epoxy deoxynivalenol (DOM-1)

- Bacterial conversion
- Large intestine
- Epoxide reductase
- Less toxic than DON



Reaction mechanism of the formation of deepoxy-deoxynivalenol from deoxynivalenol.

Zhou, T., Gong, J., Yu, H., and Li, X.-Z. 2008. Bacterial isolate and methods for detoxification of trichothecene mycotoxins. https://www.google.com/patents/US20100239537 (accessed 2018 January 18).

D3G Metabolism by Human Microbiota

During Incubation

- D3G is degraded
- DON is released
- DON is detoxified to deepoxy-deoxynivalenol (DOM-1)

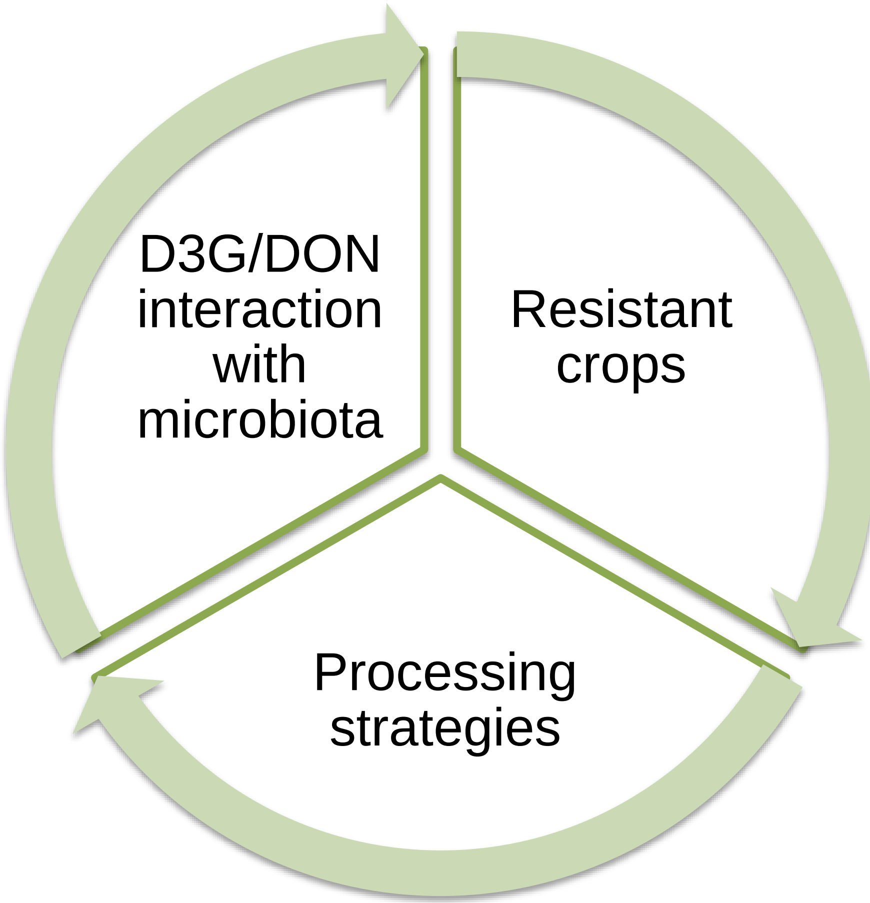
Gratz et al., 2013

D3G and Gut Microbiota

Bacterial hydrolysis of deoxynivalenol-3-glucoside.

Bacterial strain	Hydrolysis after 4 h (%)	Hydrolysis after 8 h (%)
Lactobacillus plantarum	34	62
Enterococcus mundtii	18	38
Enterobacter cloacae	17	25
Bifidobacterium adolescentis	5-17	17-25

Future Research



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- Sobrova, P., Adam, V., Vasatkova, A., Beklova, M., Zeman, L., and Kizek, R. 2010. Deoxynivalenol and its toxicity. Interdiscip. Toxicol. 3:94-99.
- U.S. Food & Drug Administration. 2017. Guidance for industry and FDA: Advisory levels for deoxynivalenol (DON) in finished wheat products for human consumption and grains and grain by-products used for animal feed. https://www.fda.gov/Food/GuidanceRegulation/GuidanceDocumentsRegulatoryInformation/ChemicalContaminantsMetalsNaturalToxinsPesticides/ucm120184.htm (accessed 2017 December 17).
- Wu, L., Qiu, L., Zhang, H., Sun, J., Hu, X., and Wang, B. 2017. Optimization for the production of deoxynivalenol and zearalenone by *Fusarium graminearum* using response surface methodology. Toxins. 9:57.

muchos gracias!

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