

# Crossing borders to mitigate mycotoxins

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### Myco-What?!

Mycotoxins are toxic secondary metabolites produced by fungi that contaminate crops and stored grains. The more than 400 chemically different mycotoxins display an array of toxic effects, ranging from gastro-intestinal complaints to infertility and liver cancer. Due to the severity of the problems, strict

Food matrix

Max. value

Cereals for direct human 75 μg/kg
consumption

Bread, pastries, biscuits,... 20 μg/kg

Table 1: Example of EU-regulations for ZEN. Non-compliant harvests are removed from the food chain.

**EU-regulations and guidelines** apply to food and feed applications.

There is no magic bullet when it comes to mitigating mycotoxins. An **integrated crop management system** is needed to prevent, intervene and remediate. **Microbial and enzyme-based detoxification** of contaminated food and feed is a promising bioremediation technique.

We study the use of **Actinobacteria** as detoxifiers of the non-steriodal estrogenic mycotoxin **zearalenone**. We use a poly-omics approach to unravel the degradation pathway. Screening is based on both degradation and loss of estrogenic properties.

### Fact sheet

25%

of the world's food crops estimated to be contaminated with mycotoxins (WHO)

28

mycotoxins and metabolites per sample in Biomin's latest global survey

of all samples in Biomin's survey contained 10 or more mycotoxins

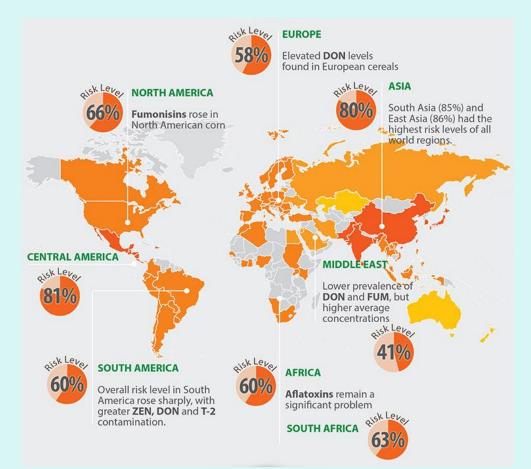
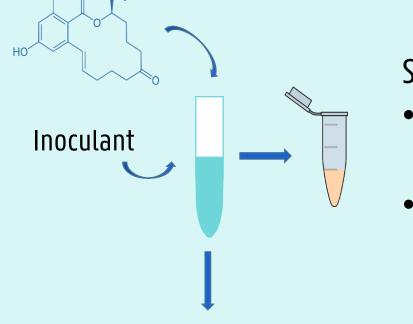


Figure 1: Global mycotoxin risk levels (Biomin survey, 2017). The more orange, the higher the risk.

### Workflow

# Screening of the Actinobacteria strain collection



#### Supernatant

- Detoxification? BLYES/BLYR assay
- Degradation? HPLC-FLD and LC-MS/MS
- Pellet
- Adsorption?Extraction andHPLC-FLD

#### Poly-omics approach



Genomic analysis of selected strains (PacBio SMRT)



RNAseq analysis (Illumina)



Differentially expressed genes and important enzymes



Degradation products

## Application in pre-and post-harvest remediation

Field fungi



- Storage conditions
- Degradation of toxins?
- Application in animal feed?

Storage fungi



Field & pot trialsApplication while

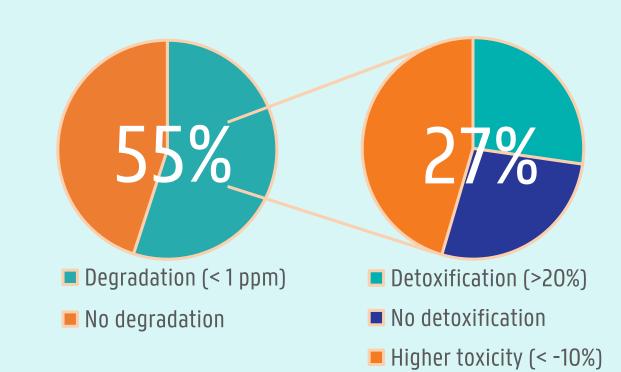
the plant is growing?

Biocontrol activity?

→ Food & feed

### Results

### Detoxification is not a given



R. globerulus R58 -42,4%

S. cacaoi subsp. asoensis -33,15%
S. rimosus K145 -29,22%
S. humiferus B5 -29,04%
S. humiferus B6 -26,6%
S. turgidiscabies 90 -26,36%
R. fascians SR13 -8,5%
R. fascians D188 -3%
R. fascians D188 -3%
R. pyridinivorans 0,38%
R. defluvii 2,3 %
R. erythropolis SQ14,75%
R. ruber N3615,97%
C. glutamicum 12,44%
C. glutamicum 12,44%
R. jostti RHA121,83%
R. rhodnii 29,6%
S. rimosus K189 97,96%

Figure 2: Overview of the screening thus far. Fifty-three different Actinobacteria have been screened for the transformation of ZEN in rich growth medium. Pie charts indicate percentages of strains that degrade and detoxify ZEN. The heat map shows an overview of degrading strains and the residual toxicity after 3 days

### Streptomyces rimosus among most performant strains

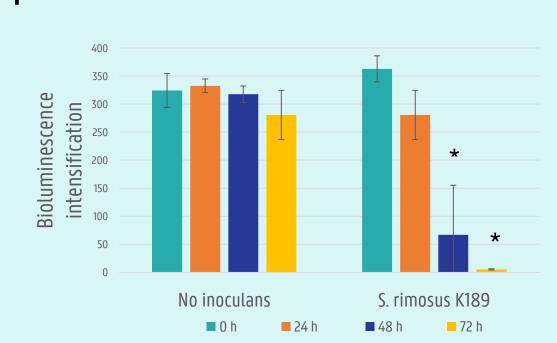


Figure 3: Detoxification of 5 mg/kg ZEN by *S. rimosus* K189; bioluminescene (RLU) based on BLYES/BLYR assay (490 BioTech). \* p < 0,001

### Application on stubble or on the plant? under investigation

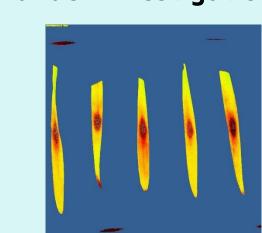




Figure 4: Biocontrol assays on: left: detached leaves; right: straw.

### Conclusion

- **Degradation of mycotoxins by microorganisms** is a promising tool to be implemented in integrated crop management systems
- Degradation of zearalenone does not always entail detoxification
- Rhodococcus and Streptomyces strains show divergent ZEN metabolism
- **Application potential** on leaves and on stubble currently being investigated
- The poly-omics approach will allow to identify biodegradation genes and enzymes, important for application in pre- and post-harvest remediation of grains