Molecular Phytopathology and Mycotoxin Research Unit
Department of Crop Sciences
University of Goettingen

www.gwdg.de/~instphyt/karlovsky

Fusarium head blight: From epidemiology to biological function of mycotoxins

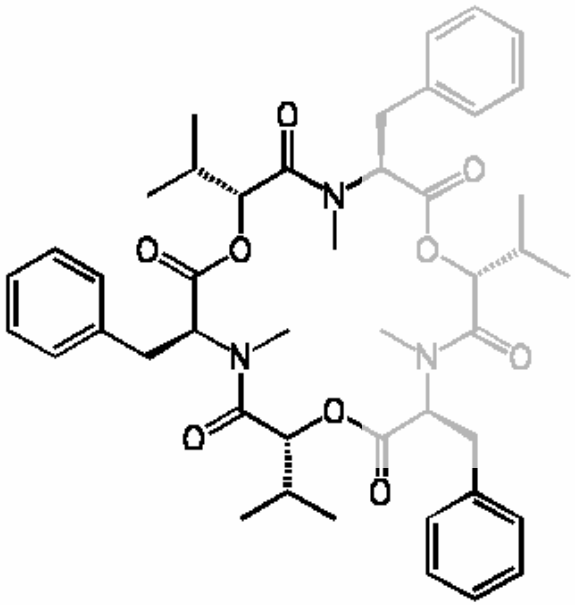
Petr Karlovsky

IFA Tulln, 7 December 2006

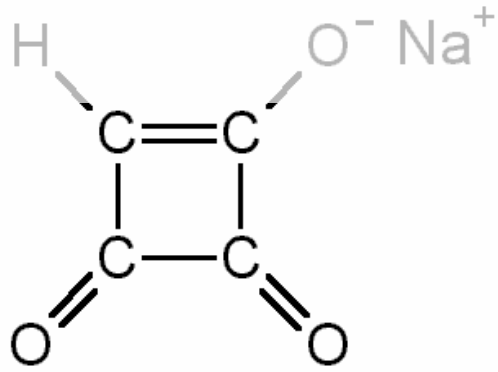
Georg-August-Universität Göttingen
Fakultät für Agrarwissenschaften

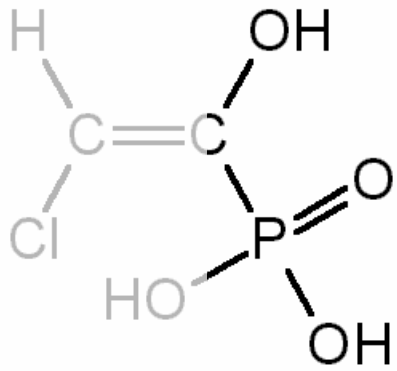
Department für Nutzpflanzenwissenschaften
Abteilung Molekulare Phytopathologie & Mykotoxinforschung

Selected secondary metabolites of *Fusarium* spp.



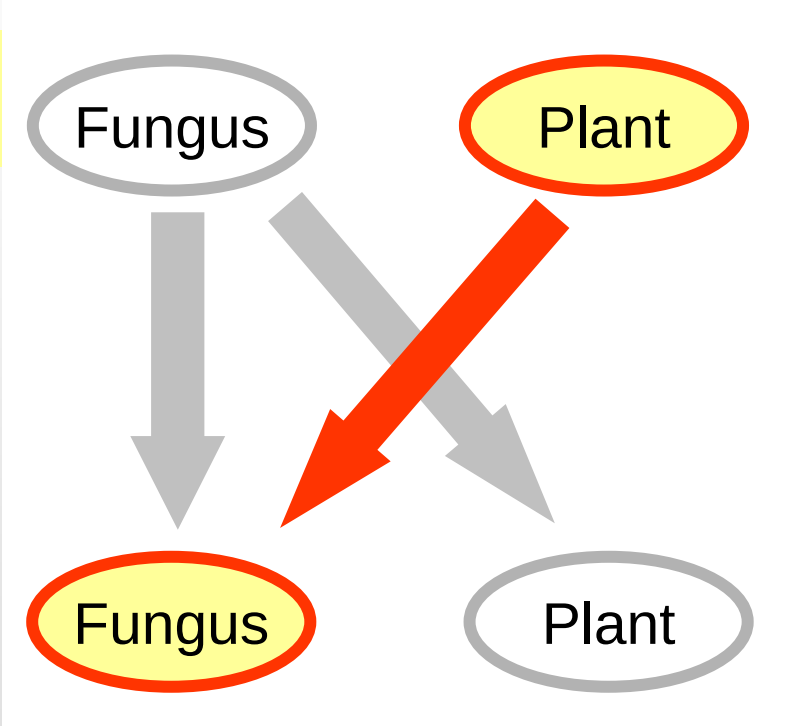
Function ?





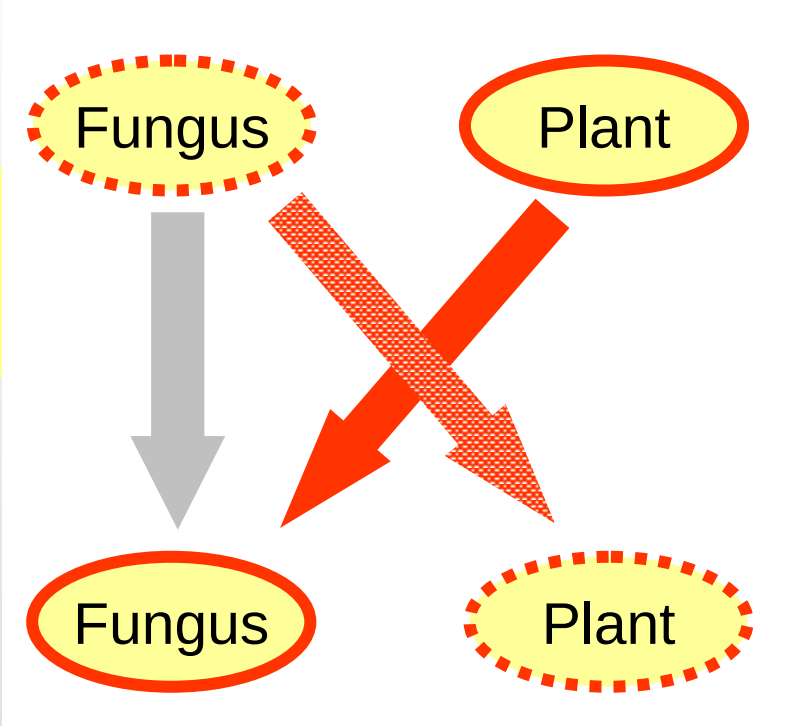
Secondary metabolites in biotic interactions

- Lignans of *Sesamum indicum*
- Metabolites in *Verticillium Brassica* interaction
- Deoxynivalenol:
  - Search for the source
  - Microbial detoxification
- Zearalenone: Search for biological function



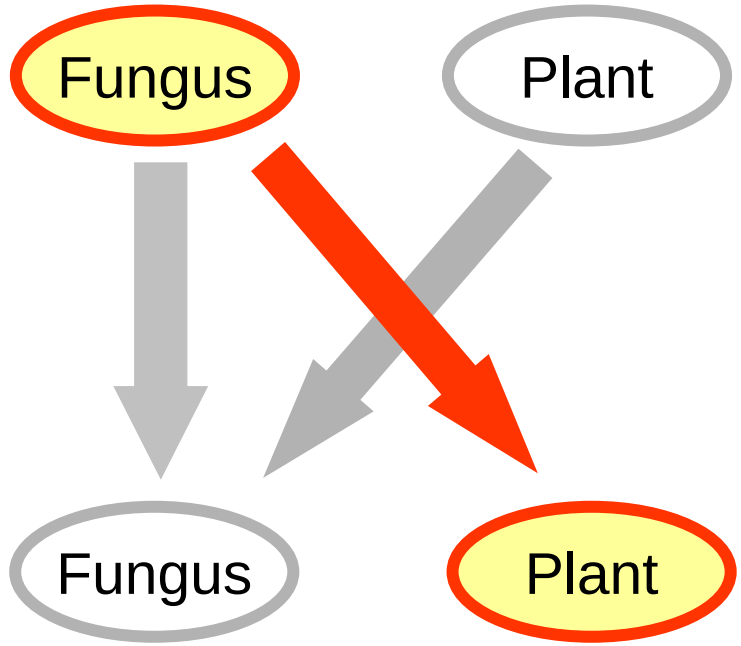
Secondary metabolites in biotic interactions

- Lignans of *Sesamum indicum*
- Metabolites in *Verticillium Brassica* interaction
- Deoxynivalenol:
  - Search for the source
  - Microbial detoxification
- Zearalenone: Search for biological function



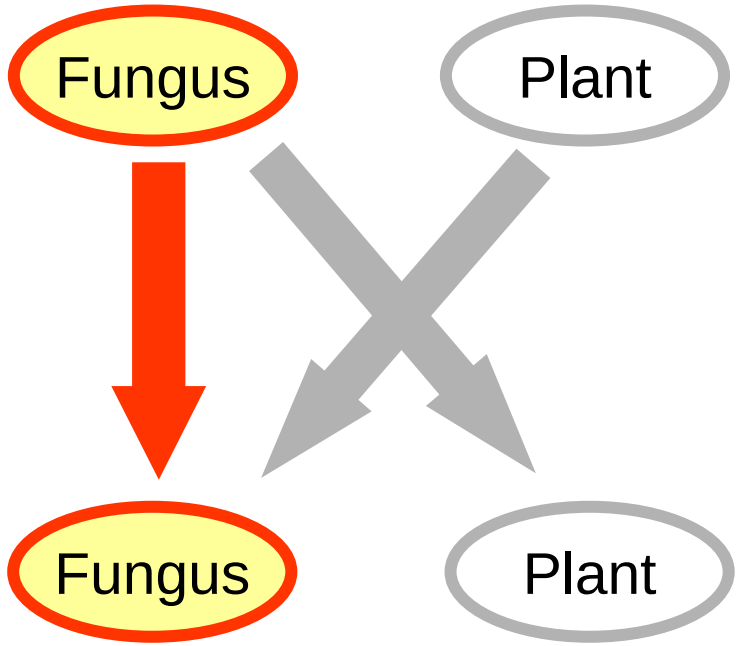
Secondary metabolites in biotic interactions

- Lignans of *Sesamum indicum*
- Metabolites in *Verticillium Brassica* interaction
- **Deoxynivalenol:**
  - **Search for the source**
  - **Microbial detoxification**
- Zearalenone: Search for biological function



Secondary metabolites in biotic interactions

- Lignans of *Sesamum indicum*
- Metabolites in *Verticillium Brassica* interaction
- Deoxynivalenol:
  - Search for the source
  - Microbial detoxification



- **Zearalenone: Search for biological function**

Heavily contaminated grains:

- Hazardous waste
- Fuel for thermal power stations

Epidemiology of Fusarium head blight

Fusarium species isolated from infected ears:

F. graminearum, F. culmorum,
F. acuminatum, F. avenaceum,
F. crookwellense, F. equiseti,
F. poae, F. solani...

Which species is relevant for FHB?

Textbook wisdom

1. Both *Fusarium culmorum* and *F. graminearum* contribute to DON contamination
2. The species form a North/South-gradient

Rationale

1. FG and FC produce DON in vitro and after artificial inoculation
2. FC has a lower temperature optimum

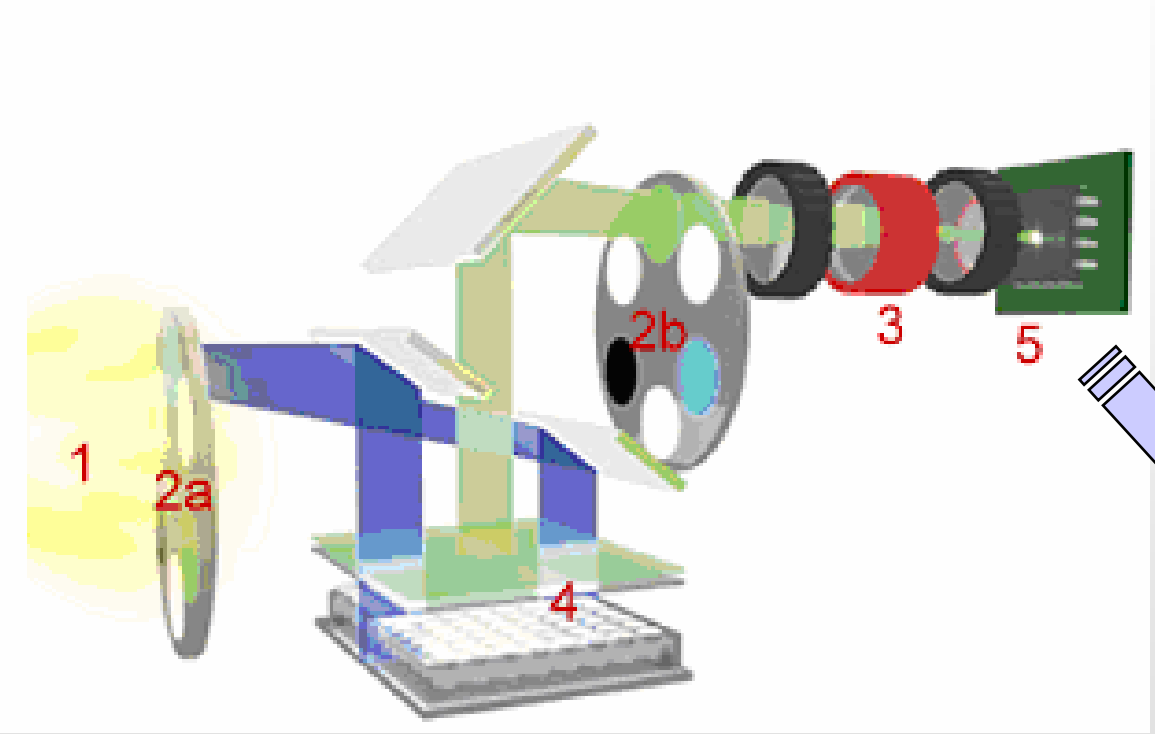
Sampling 2003

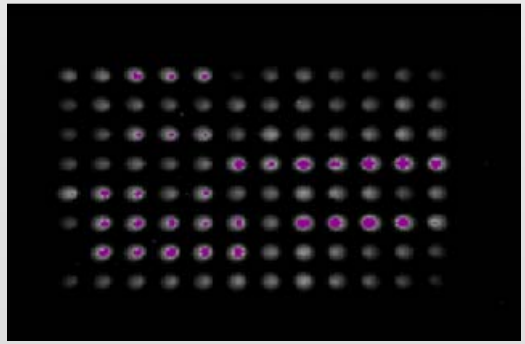
1200 ear with FHB symptoms

How to identify 2 fungal species in 1000 samples with the smallest effort?

C. Brandfass et al., unpublished

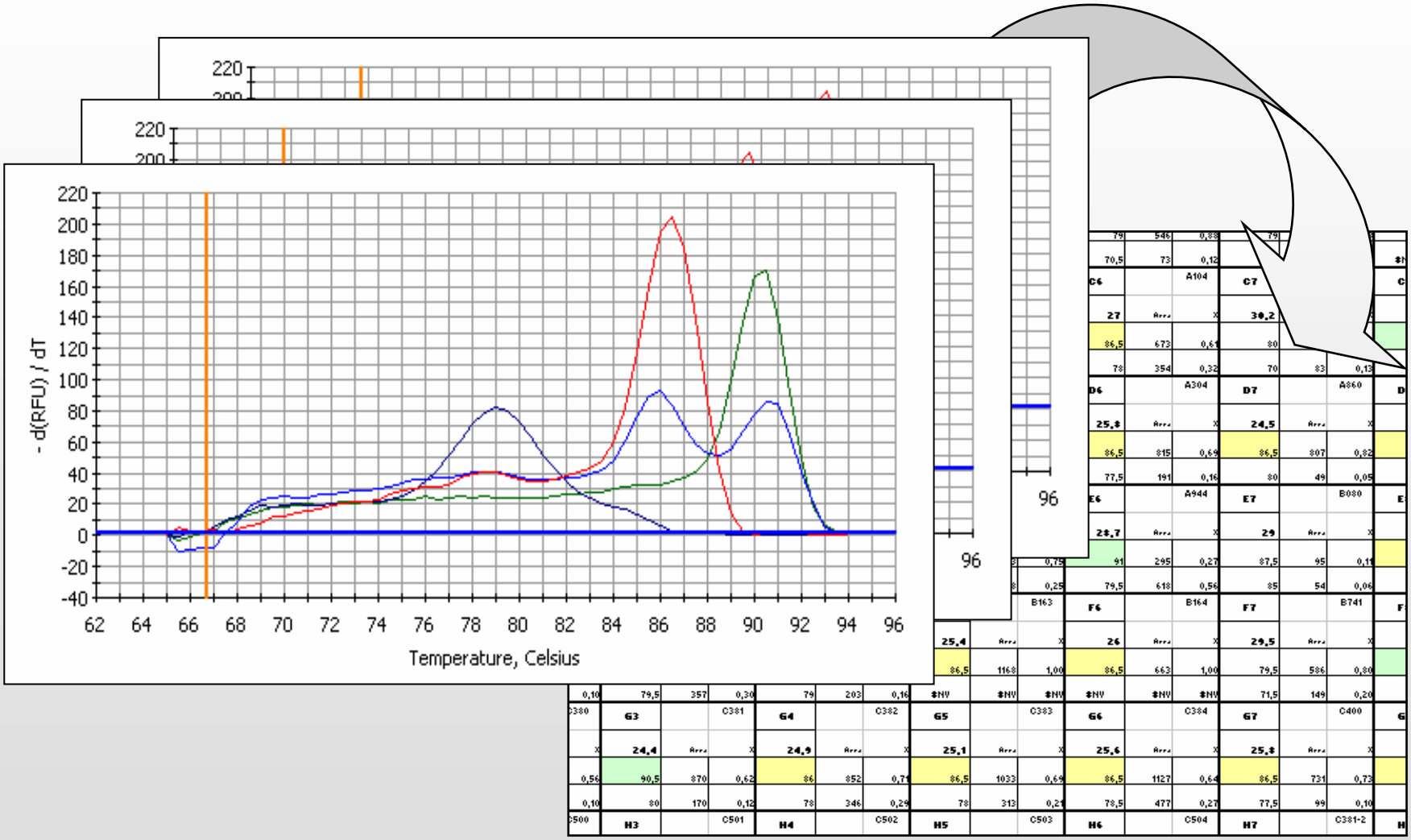
Real-time thermocycler





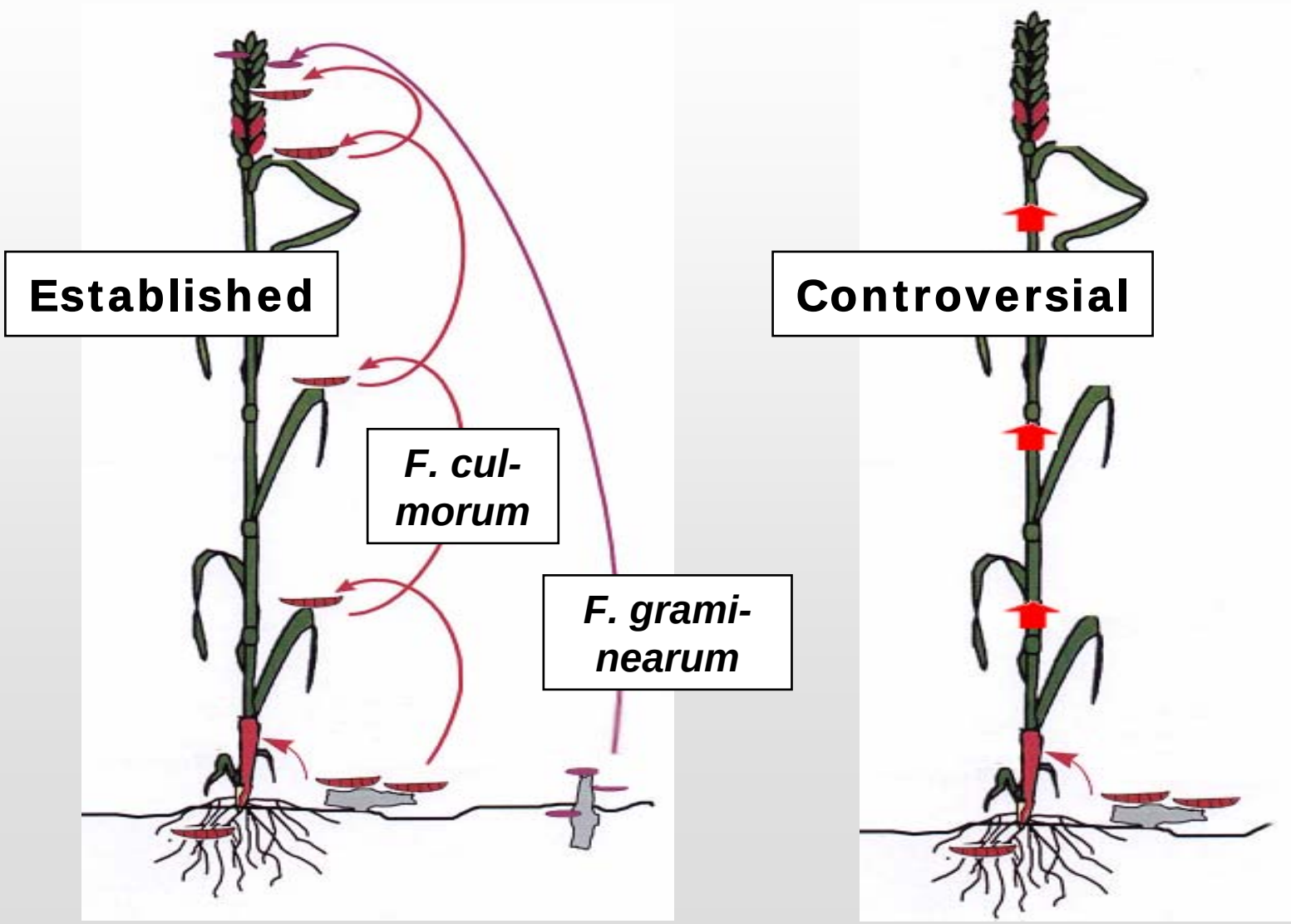
1: Light
2: Filter
3: Amplifier
4: Samples
5: CCD chip

Simultaneous identification of *F. culmorum* and *F. graminearum* by melting curve analysis

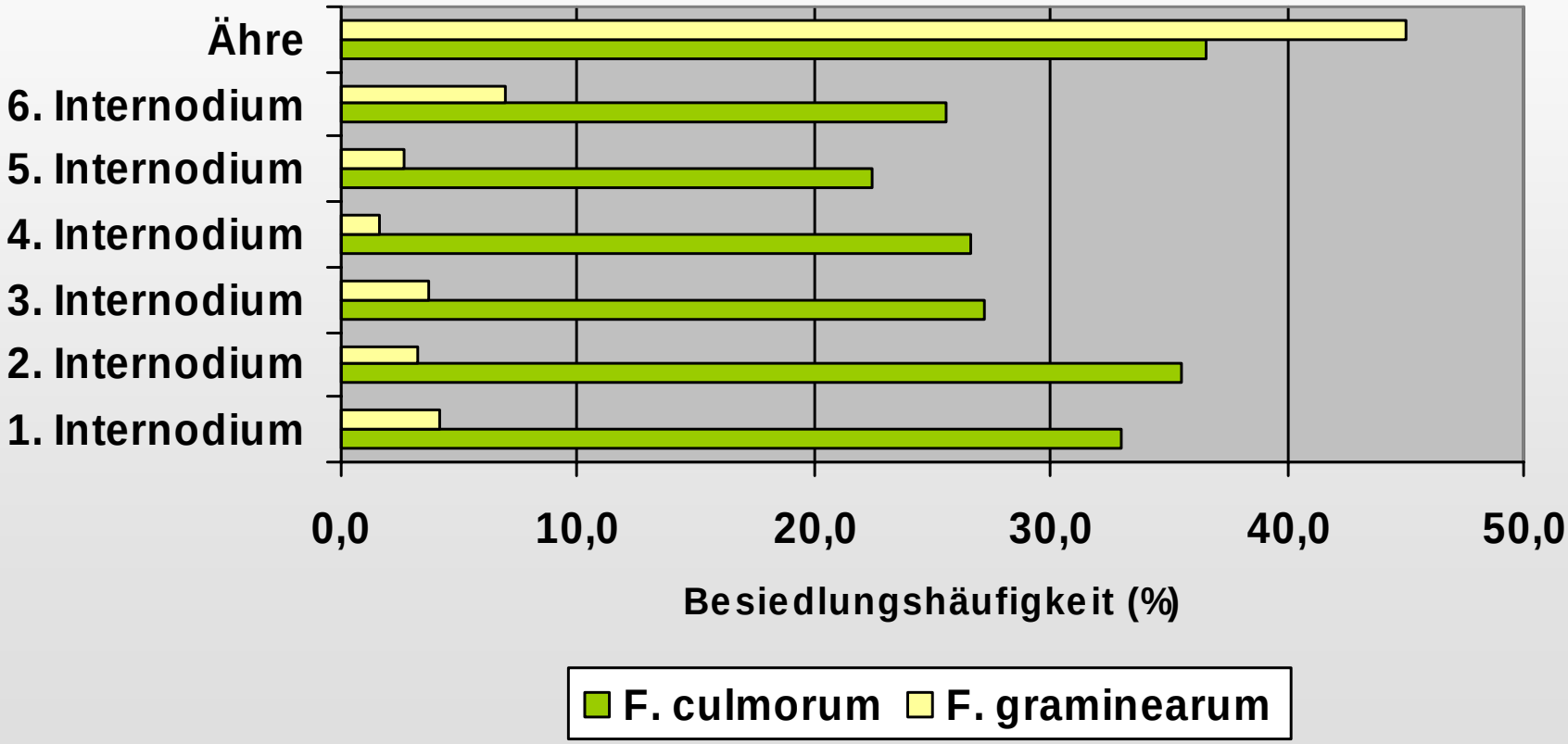


Brandfass et al. 2006: BMC Microbiol. 23:6

Does *F. culmorum* colonize wheat systemically?



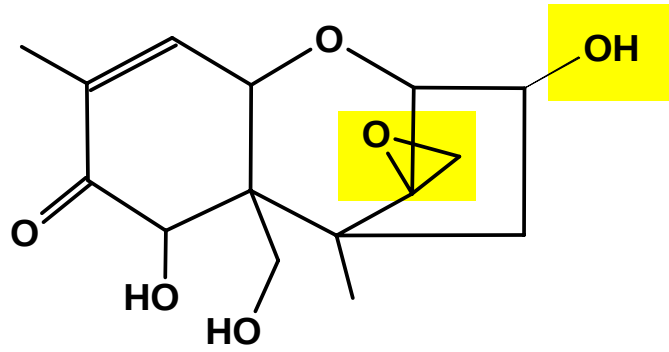
Colonization of wheat stem and ears by *F. culmorum* and *F. graminearum*



Schlüter et al. (2006): Gesunde Pflanze 58:107

Protection against deoxynivalenol

- Irreversible detoxification
  - Deepoxidation
  - Mineralization
  - **Epimerization**
- Reversible detoxification
  - Acetylation (*Tri101*)
  - Active export (*PDR*5, *Tri*12)
- Engineering resistant ribosomes (RL3)



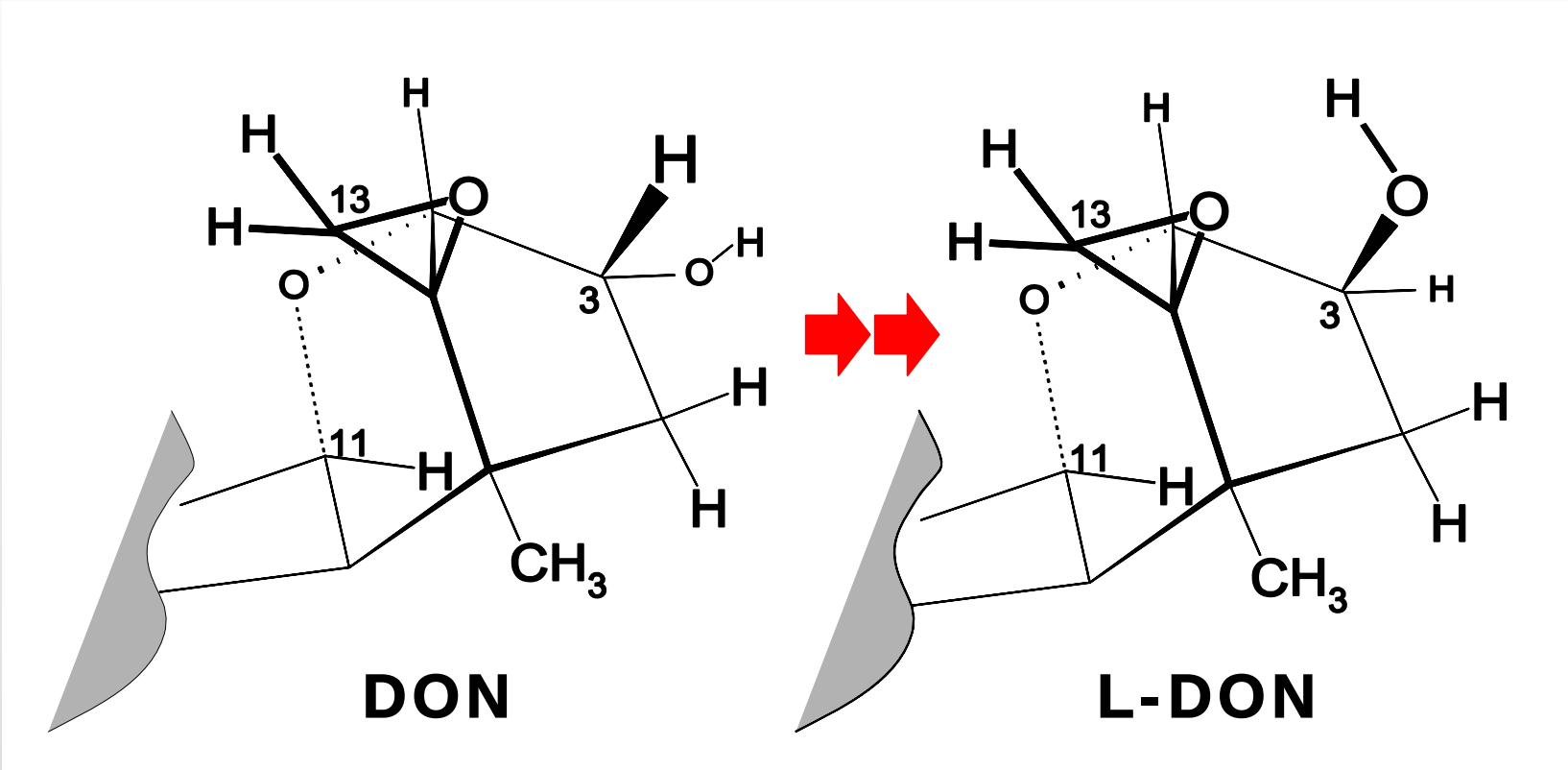
Search for the detoxification of DON

Source	Assays	Positive
Mixed cultures from grains	819	-
Pure cultures from grains	1434	-
Mixed soil cultures	200	-
Pure cultures from soil	975	-
Other sources	198	1

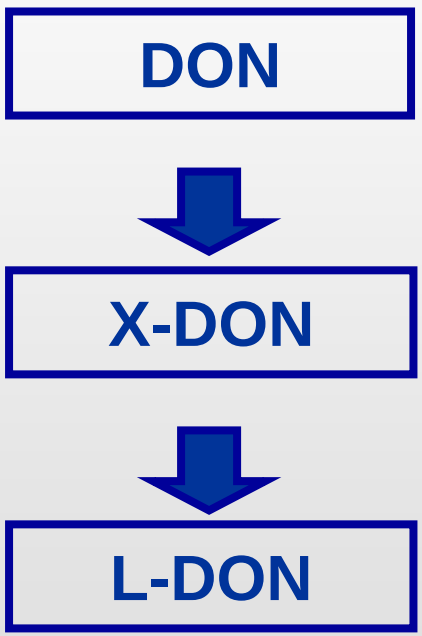
➡ **Active culture D107**

Völkl et al. (2004) J Bas Microbiol 44:147

L-DON: 3-epi-deoxynivalenol



Toxicity of transformation products



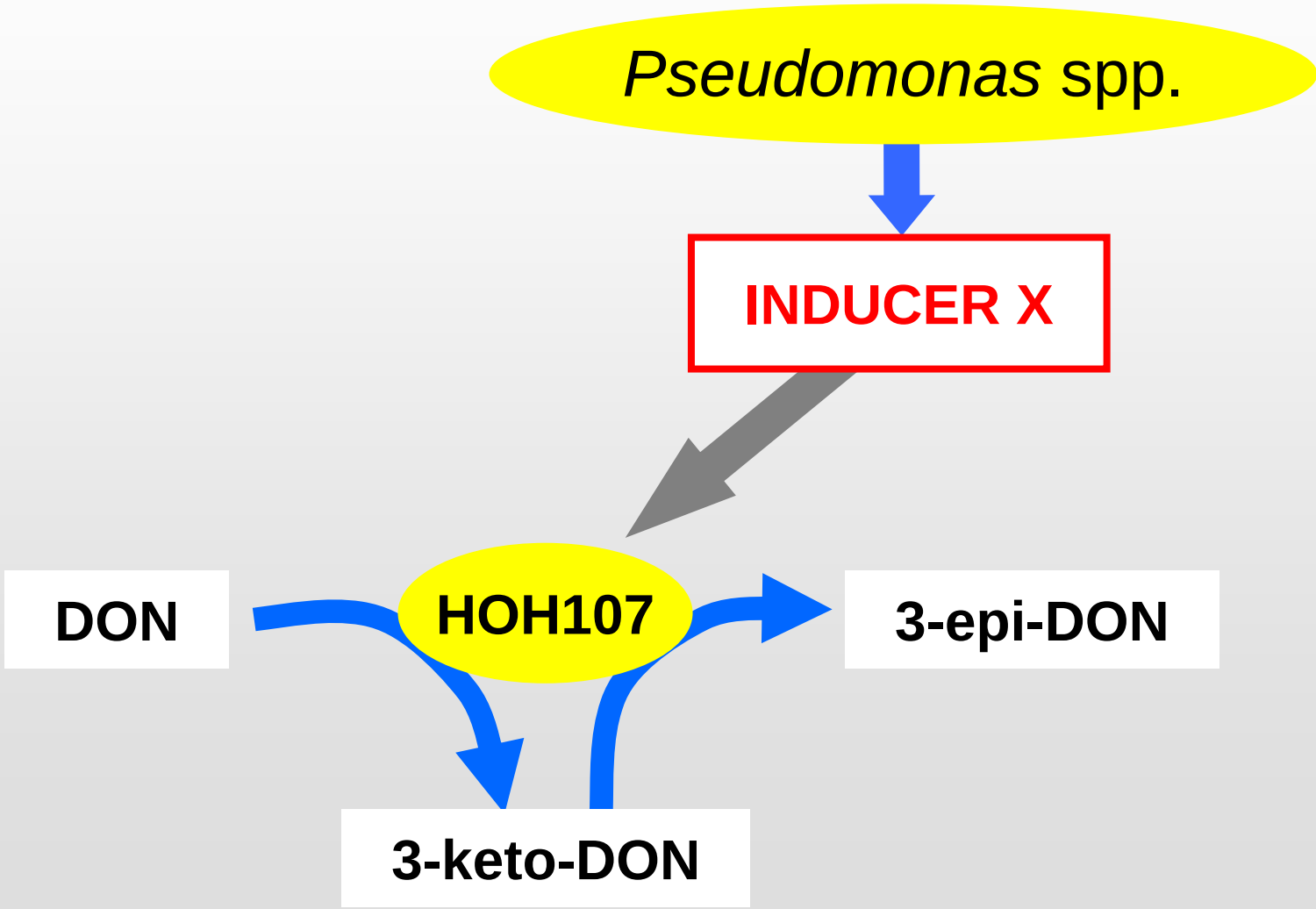
	DON [µg/ml]	X-DON [µg/ml]	L-DON [µg/ml]
A. salina [LC_{50}]	52	78	194
Immunos. [IC_{30}]	≈ 0.1	> 1.07	
MTT-assay [LC_{50}]	≈ 15	200	>> 200

Andreas Völkl & Manfred Garais, unpublished

Stability of pure culture HOH107

- Activity of HOH107 declined continuously
- Activity of mixed culture D107 was stable for over 5 months
- Culture supernatant of D107 reactivated HOH107

Inductibility of HOH107 activity



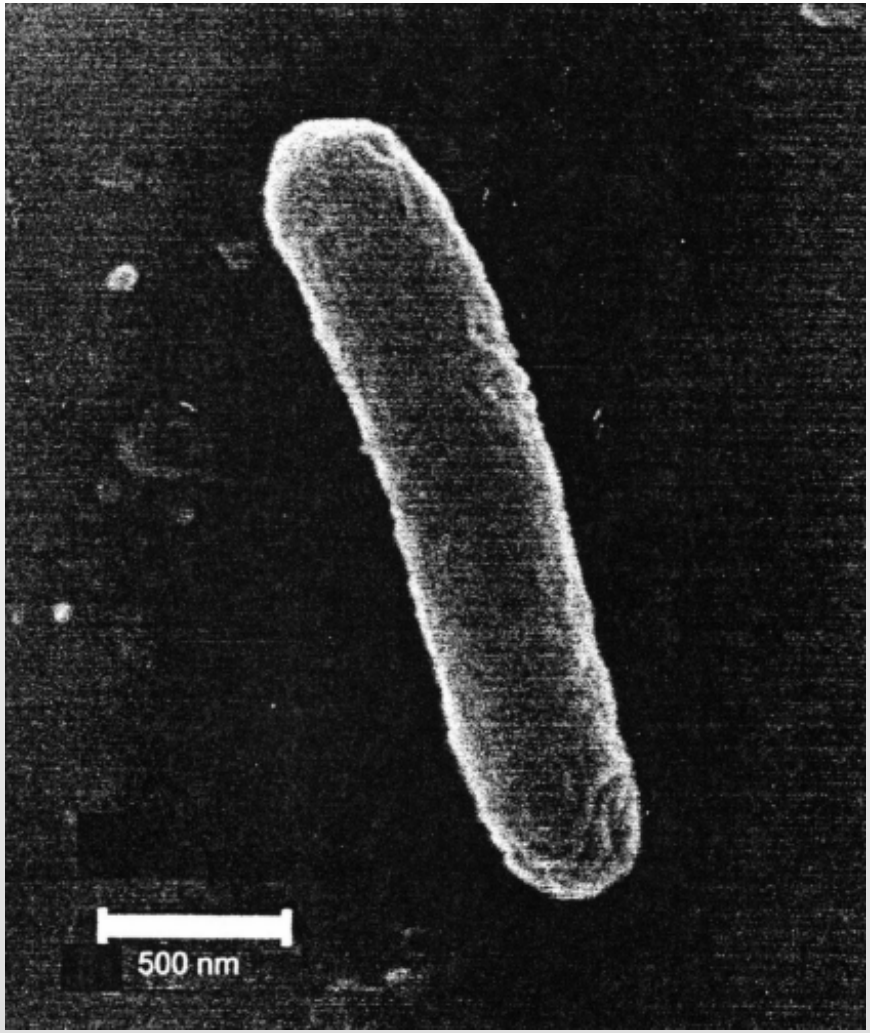
Arne Weiberg, unpublished

Inducibility of HOH107 activity

Species	Inducing activity
P. aeruginosa	**no**
P. fluorescens	**yes**
P. putida	**yes**
P. syringae	**yes**
***Tn5* mutants**	
P. putida JM213	**yes**
P. putida JM214	**yes**
P. putida JM218	**no**

Arne Weiberg, unpublished

HOH107

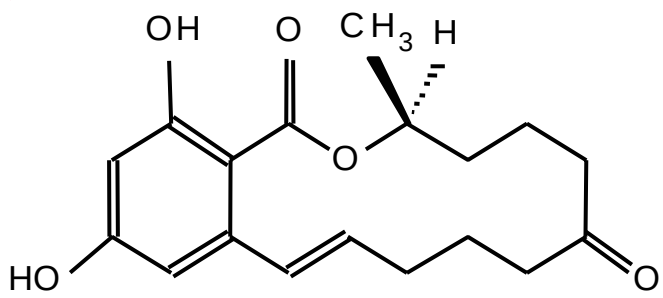


Anne Heller, unpublished

Zearalenone in headlines

Hog farms in trouble

EC embargo on US beef



Therapy of menopausal syndrome

Homosexuality in rabbits

Hypothesis about biological function

- Regulation of sexual reproduction

  ⇨ disproved

- Virulence factor

  ⇨ disproved for some plants

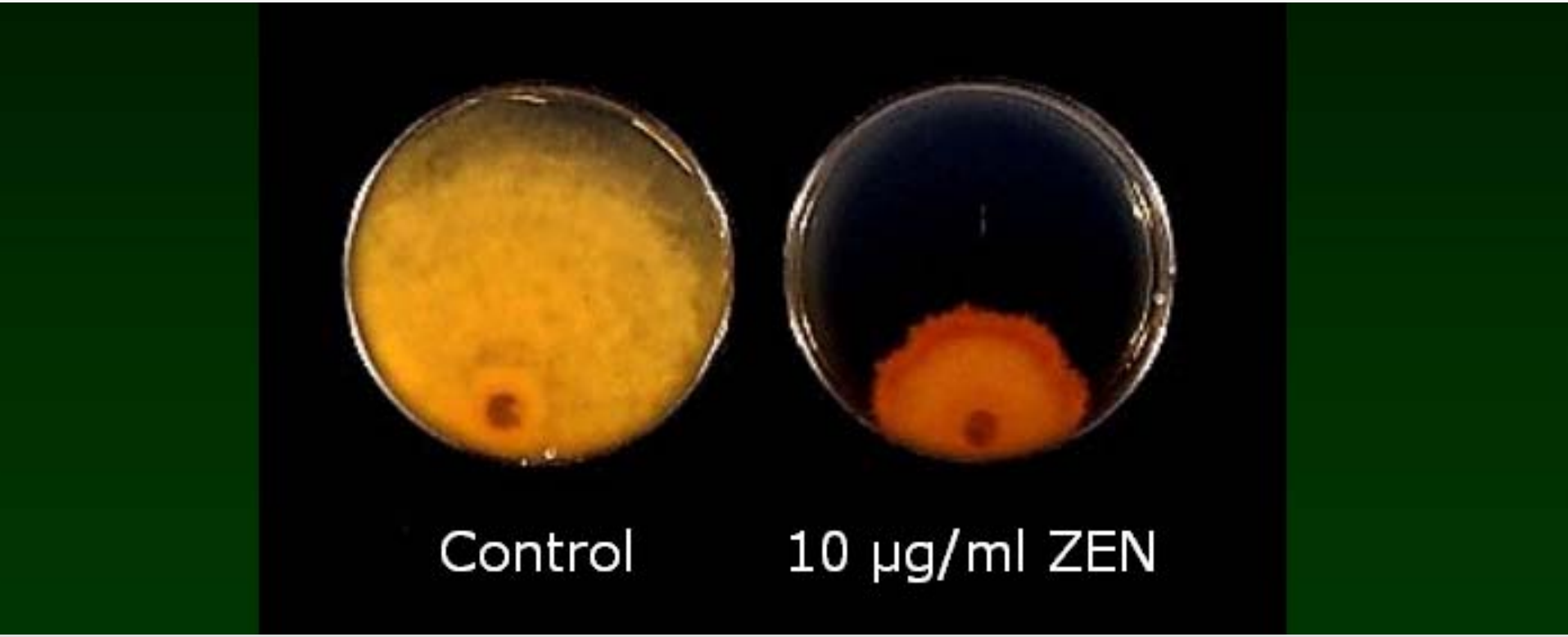
Zearalenone as fungicide

Alternaria alternata

Utermark & Karlovsky (2007): Appl Env Microbiol 73:637

Zearalenone as fungicide

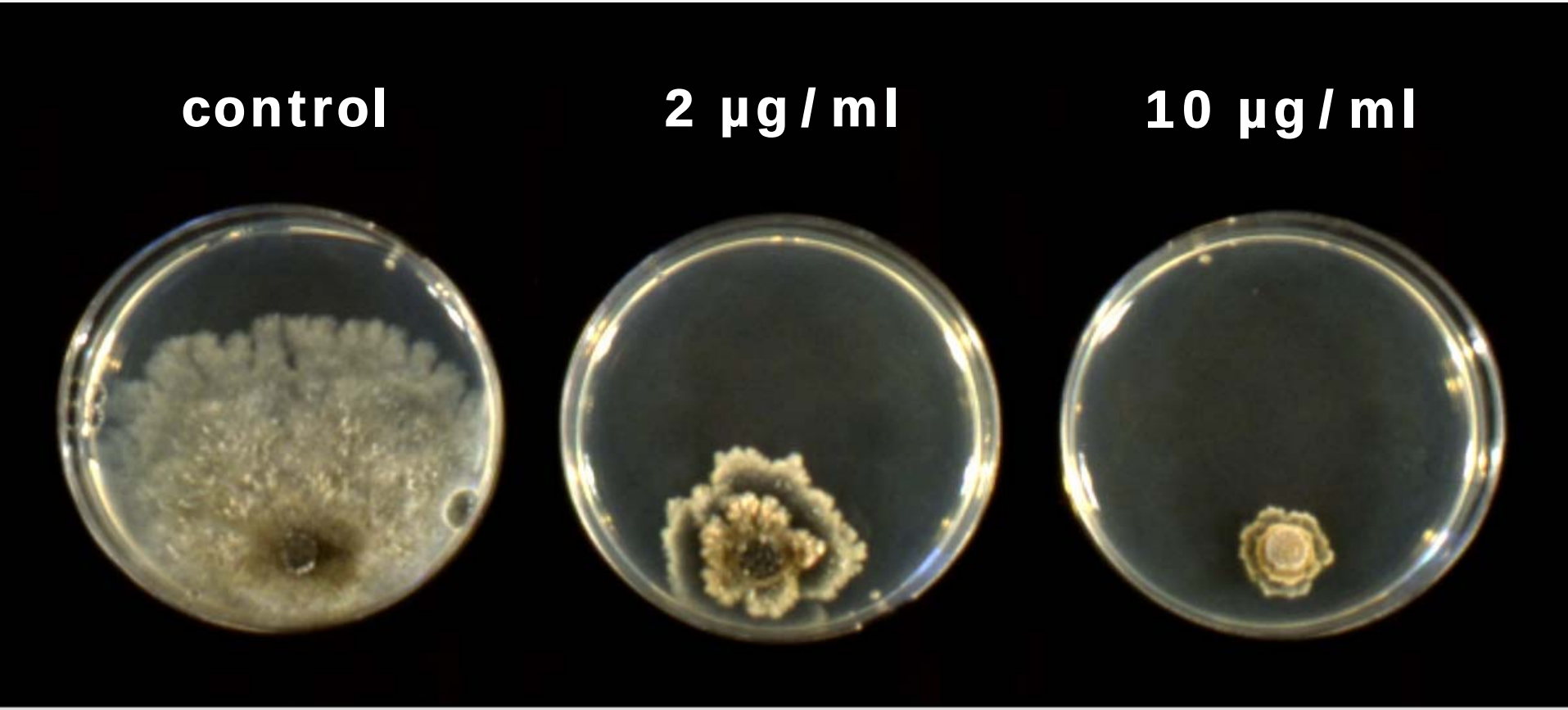
Epicoccum purpurascens



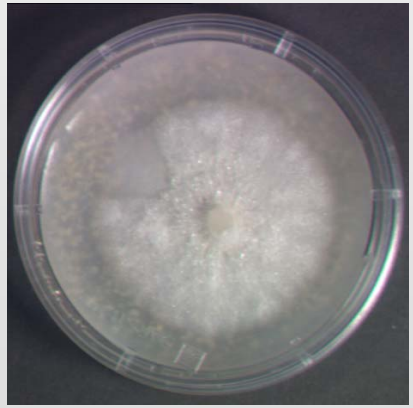
Utermark & Karlovsky (2007): Appl Env Microbiol 73:637

Zearalenone as fungicide

Sordaria fimicola



Gliocladium roseum is resistant to zearalenone

	Czapek-Dox	Oatmeal agar
Without zearalenone		
Zearalenone 20 µg/ ml		

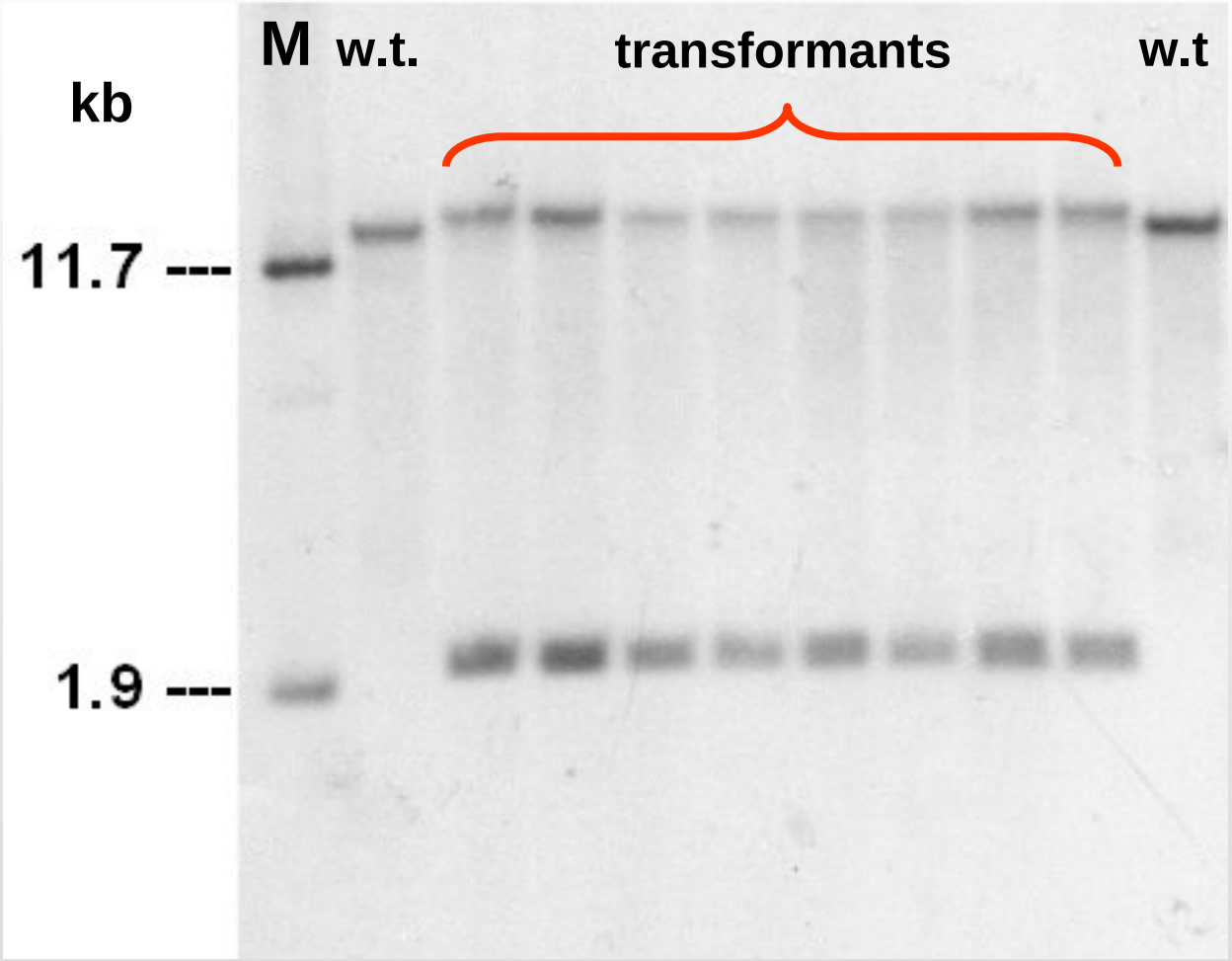
Interaction between *F. graminearum* and *G. roseum* on solid surface

3 d after inoculation

4 d after inoculation

Hanno Wolf, unpublished

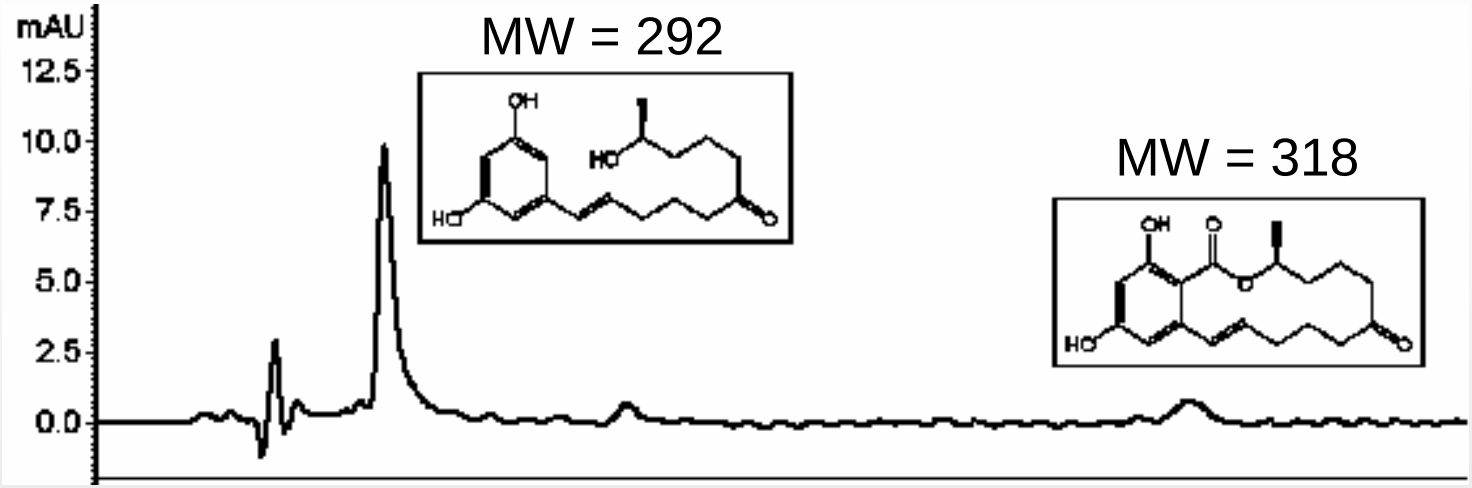
Knockout of *zes2* in *G. roseum*: Southern hybridization



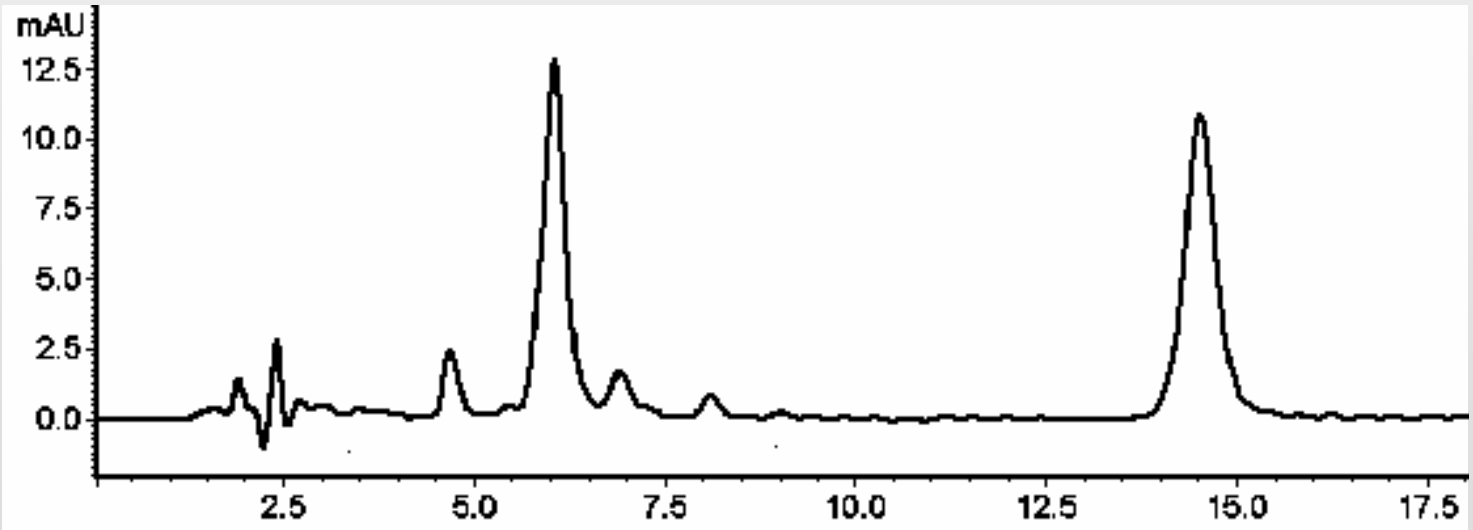
Utermark & Karlovsky (2007): Appl Env Microbiol 73:637

Transformation of zearalenone by *zes2* mutants of *G. roseum*

w.t. strain

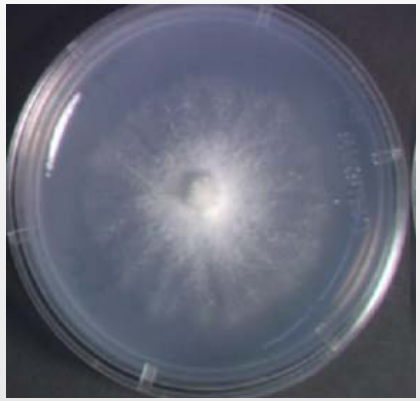
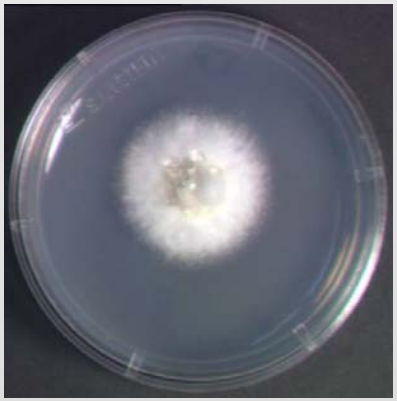
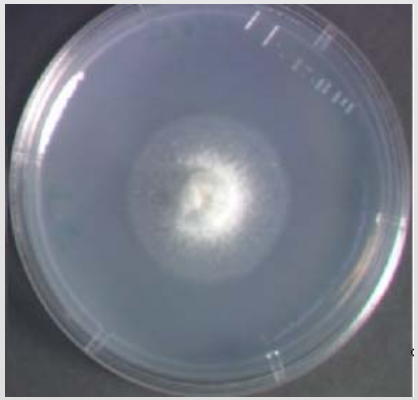


***zes2* mutant**



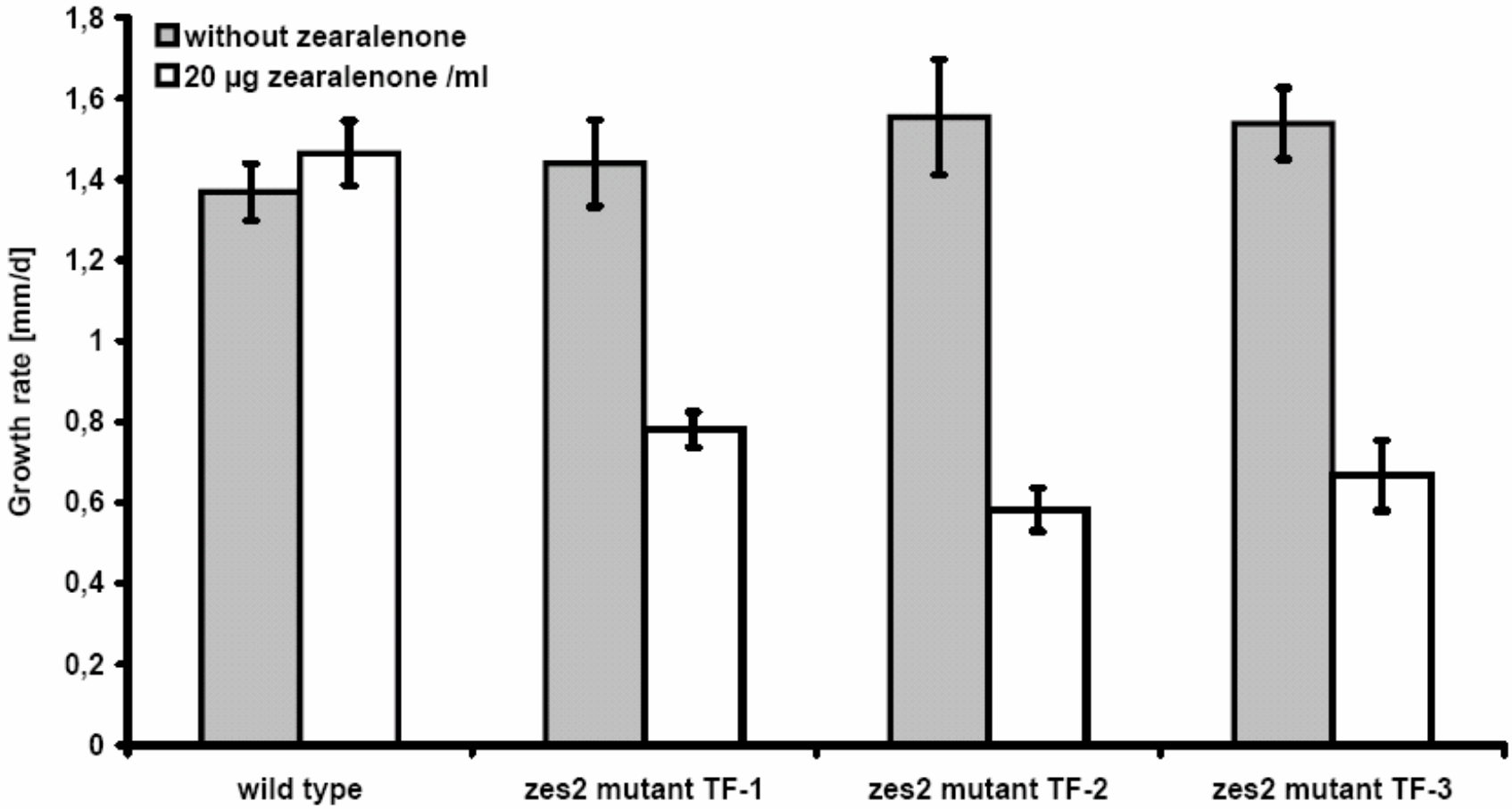
Utermark & Karlovsky (2007): Appl Env Microbiol 73:637

Growth inhibition of *zes2* mutants by zearalenone on solid medium

	Czapek-Dox	SNA
W.t.		
***zes2* mutant**		

All media with 20 µg/ml zearalenone

Growth inhibition of *zes2* mutants by zearalenone on solid medium



Acknowledgment

Dr. Christoph Brandfaß
Dr. Joachim Weinert

Prof. Dr. Klaus Schlüter
Dr. Ute Kropf

Dr. Andrea Völkl
Prof. Dr. Manfred Gareis
Dr. Anne Heller
Arne Weiberg

Dr. Gertrud Woerfel
Jan Utermark
Dr. Hanno Wolf
Dr. Uli Hettwer

Technical support

Heike Rollwage
Petra Romanczuk
Monika Boßmann
Patricia Bartoschek

Financial support

DFG
BMVEL
VW-Stiftung