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Agroscope

Cropping factors: The key for sustainable mycotoxin management in cereals

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T. Musa, T. Bucheli, F. Wettstein, H.R. Forrer

Agroscope, Ecological Plant Protection in Arable Crops
Luxembourg Institute of Science and Technology
University of Milan, Food, Environmental and Nutritional Sciences
Agroscope, Environmental Analytics

December 4, 2018
National FHB Forum, St. Louis (MO), USA





Outline

- Surveys on *Fusarium* species and mycotoxins in **wheat**, **barley** and **oats**
- Our resulting research to reduce the risk in cereals:
 - “Healthy & Safe” cereal varieties
 - “MycoKey”: Biocontrol, cover crops and biofumigation



+ Maximum / recommended limits for unprocessed cereals (ppm) – EU & CH

Toxin	Limit Cereals	Limit Maize	Rec. limit * Feed / indicative levels
DON	1.25	1.75	0.9 - 12
ZEA	0.1	0.35	0.1 - 3
T-2, HT-2	in prep.	in prep.	0.1 – 1 (wheat-oats)

No intention so far for NIV...

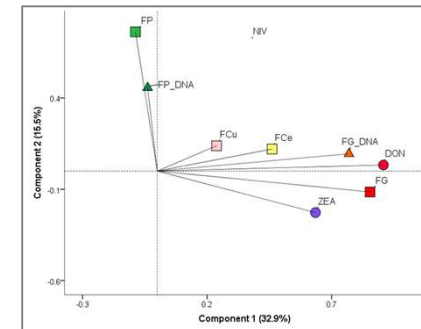
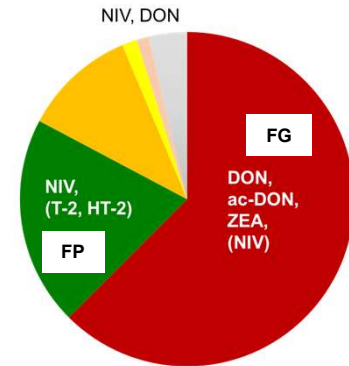
* complete feeding stuffs to all feed materials





Swiss *Fusarium* surveys Objectives

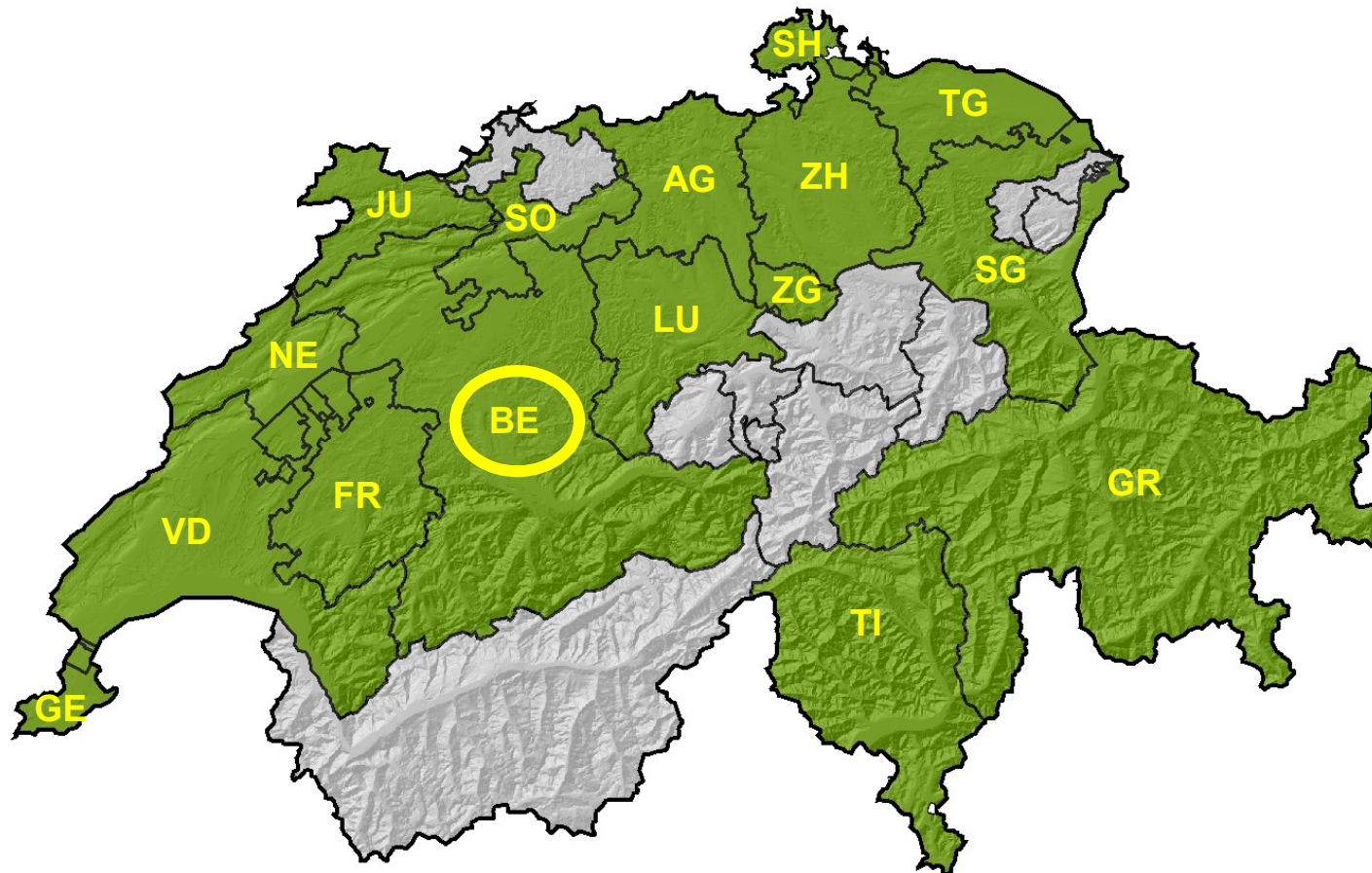
- Abundance & distribution of species
- Correlation between species abundance & toxin content
- Effect of cropping factors for the most prevalent species





Wheat samples from growers

- 527 grain samples from 17 cantons (2007-10)
- 159 grain samples from Berne (2011-14)





Questionnaire on cropping factors



Variety

Fragen zur Getreideprobe (Weizen) - Ernte 09

Angaben zum Schlag, aus dem die Probe stammt

Name und Adresse des Betriebsleiters
Millo, Renato De Giorgio, Mont-
2075 Thiele-Wavre
-756 90 09 E-Mail: Farm@doncamilo.ch

Konv.
ÖLN
IP-Label Suisse
Extensio
Bio

Production system

Ort des Schlags (falls nicht Wohnort):

Getreideart & Sorte: W1 Runal Vorfrucht (2008): keine Angabe Vor-Vorfrucht (2007):
Körnermais: KW

Previous crop

Haben Sie in ihrem Getreideschlag Fusarien beobachtet?
Ja Nein Bemerkung:

Mähdrescher mit Unterflurhäcksler (2008) ja nein unbekannt

Zusätzliches Häckseln/Mulchen von Ernteresten ja nein Wenn gemulcht, welchem Gerät:

Pflug ja nein

Einarbeiten von Ernteresten: Grubber Federzinkenegge Andere:
Scheibenegge Kreiselegge Rototiller

Direktsaat ja nein

Tillage & residue management



Sädatum: 20.10.2008 Blüh-Beginn (DC 61), falls bekannt: ✓

N-Düngung: kg N / ha
1 32 2 27 3 27
Handelsname, falls bekannt: Gelbor 4kale NA NA

Fungicide

Fungizid, falls verwendet: Produkt 3 Knoten dium

Wachstumsregulator, falls verwendet: Produkt



Bitte den ausgefüllten Fragebogen an:
Besten Dank!
Susanne Vogelgsang

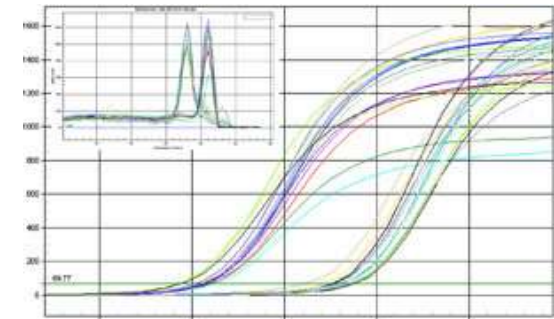
...cken.
riculture
re cantonal
-agronome
Aurore 1 • CH-2053 Cernier
T 032 889 36 82 • F 032 889 37 01
E Michel.Horner@ne.ch





Wheat survey 2007-2014

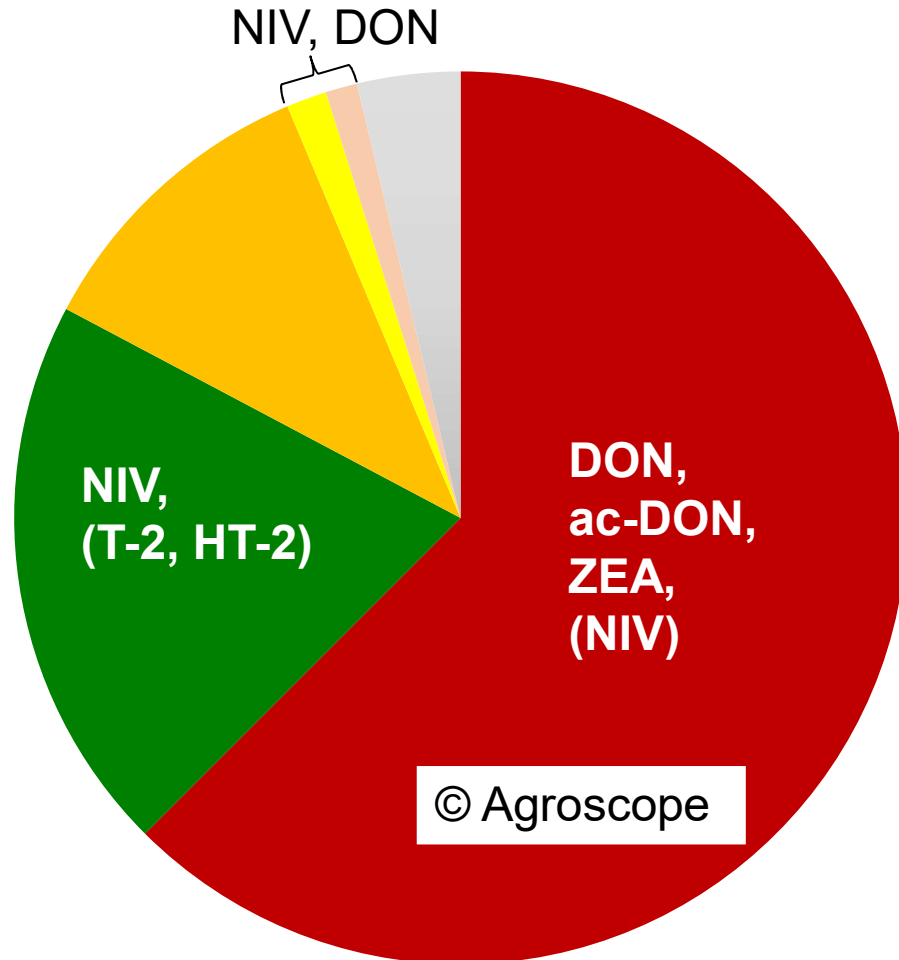
- **Harvest samples from growers**
686 wheat samples from 17 cantons
- **Questionnaire on production factors**
- **Seed health test**
Incidence of *Fusarium* species
- **Toxin content**
LC-MS/MS: DON, other trichothecenes & ZEA
- **qPCR** for top-two dominant species
- **Genetic chemotypes**
15-ADON, 3-ADON, NIV





Ratio of isolated *Fusarium* species

n=686



■ *F. graminearum*

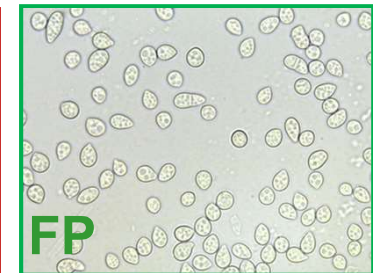
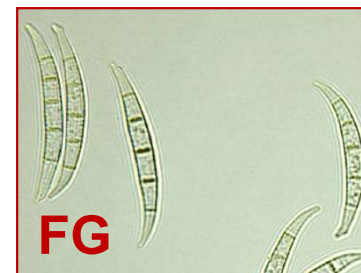
■ *F. poae*

■ *F. avenaceum*

■ *F. cerealis*

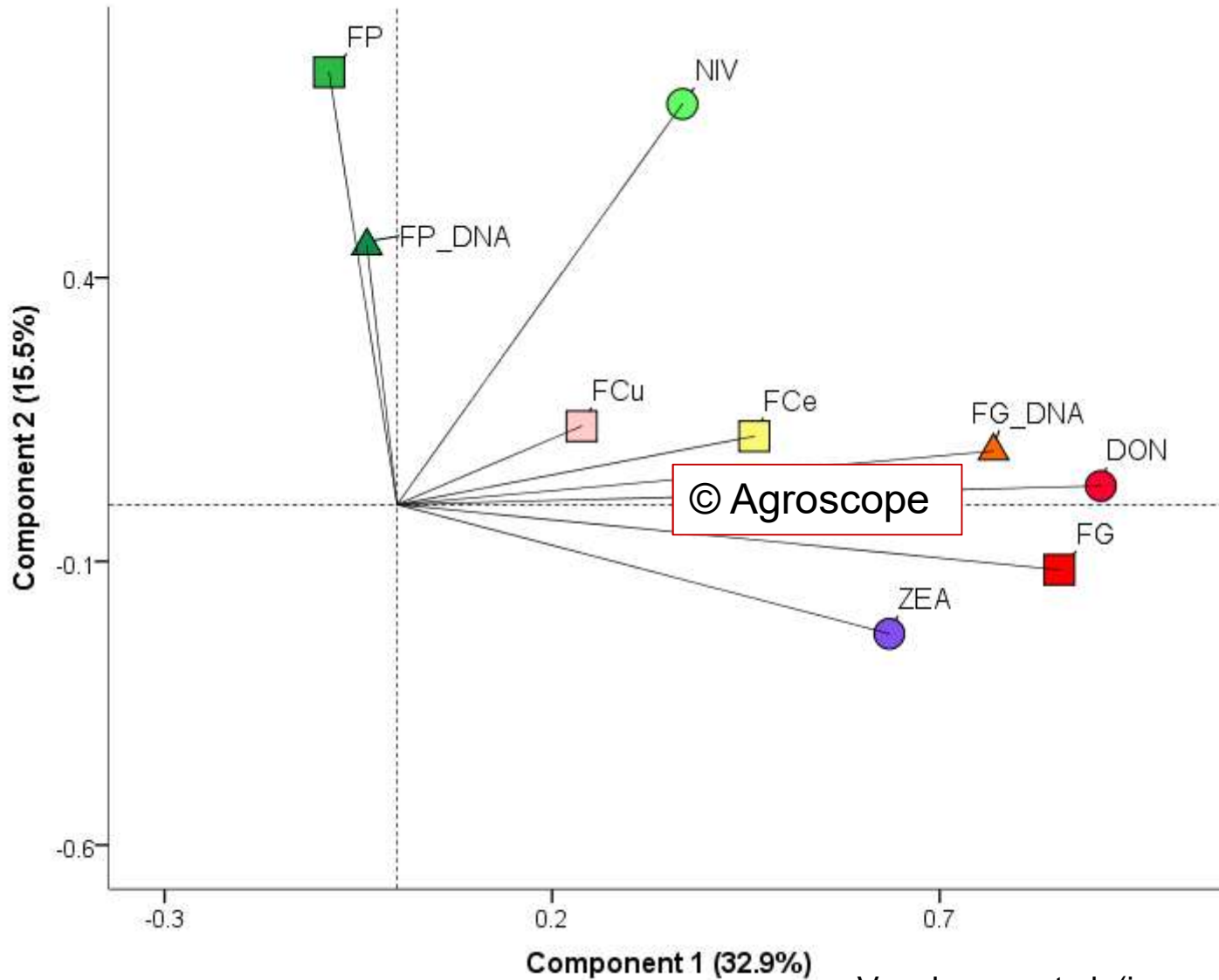
■ *F. culmorum*

■ *F. spp*




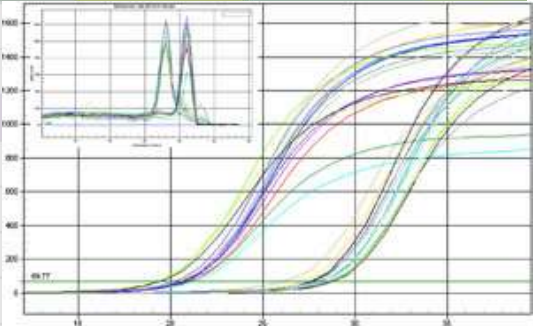



Correlations fungal abundance and mycotoxin contents - PCA





Three analysis methods to trace the origin of NIV

?	Incidences				qPCR		Chemotypes			
	NIV ppb	FG%	FP%	FC%	Fcrook%	DNA_FG	DNA_FP	15aDON_K	3aDON_K	NIV_K
470										
403										
210										
187										
187										
173										
170										
159	38	0	0		6	720.0	1.4	374.4	11.2	26.5
158	4	1	0		0	28.2	0.8	14.8	0.0	0.0
157	1	3	2		0	1.7	5.2	1.3	1.4	0.0
142	20	0	5		0	375.0	6.4	38.2	1.6	1.7
130	5	4	1		0	39.3	16.3	15.0	0.6	1.5
110	2	5	0		0	9.2	4.9	5.3	0.7	0.0
110	1	2	0		3	30.7	0.5	7.5	1.7	7.2
110	1	6	0		0	0.3	279.0	0.0	0.0	0.0
104	0	3	0		0	6.4	8.9	1.1	0.0	0.0
102	44	1	2		0	993.0	0.6	43.3	4.0	2.4
101	17	22	0		0	26.6	58.6	6.2	1.1	0.5
96	17	6	0		3	623	23.8	136.3	2.3	1.6

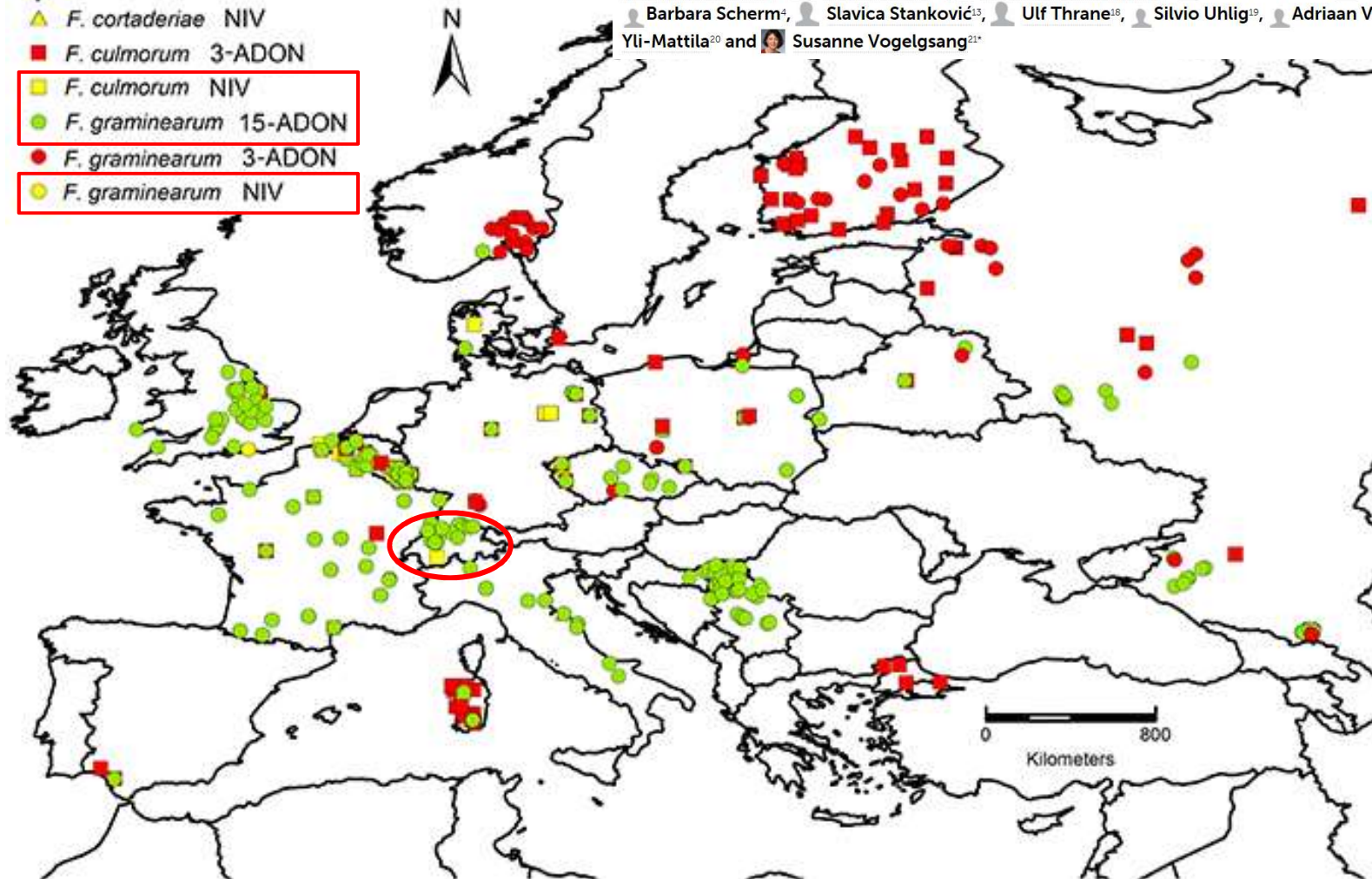


A European Database of *Fusarium graminearum* and *F. culmorum* Trichothecene Genotypes

Matias Pasquali^{1*}, Marco Beyer¹, Antonio Logrieco², Kris Audenaert³, Virgilio Balmas⁴, Ryan Basler⁵, Anne-Laure Boutigny⁶, Jana Chrpová⁷, Elżbieta Czembor⁸, Tatiana Gagkaeva⁹, María T. González-Jaén¹⁰, Ingerd S. Hofgaard¹¹, Nagehan D. Köycü¹², Lucien Hoffmann¹, Jelena Lević¹³, Patricia Marin¹⁰, Thomas Miedaner¹⁴, Quirico Migheli⁴, Antonio Moretti², Marina E. H. Müller¹⁵, Françoise Munaut¹⁶, Päivi Parikka¹⁷, Marine Pallez-Barthel¹, Jonathan Piec¹, Jonathan Scaufflaire¹⁶, Barbara Scherm⁴, Slavica Stanković¹³, Ulf Thrane¹⁸, Silvio Uhlig¹⁹, Adriaan Vanheule³, Tapani Yli-Mattila²⁰ and Susanne Vogelgsang^{21*}

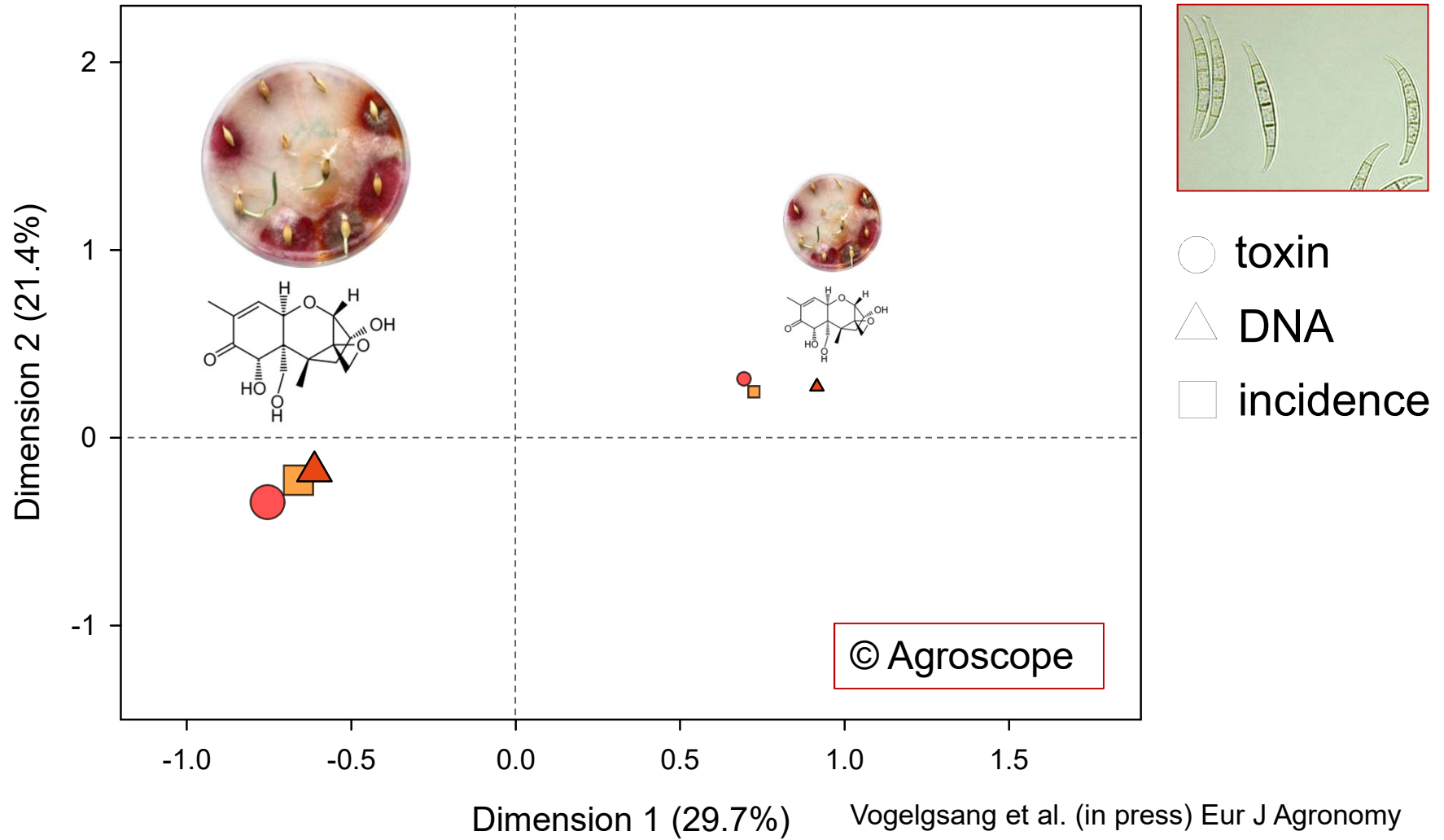
Species CHEMOTYPE

- ▲ *F. cortaderiae* NIV
- *F. culmorum* 3-ADON
- *F. culmorum* NIV
- *F. graminearum* 15-ADON
- *F. graminearum* 3-ADON
- *F. graminearum* NIV



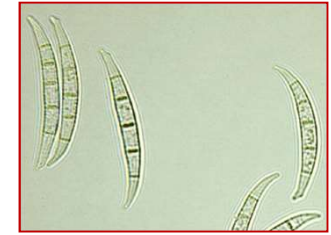
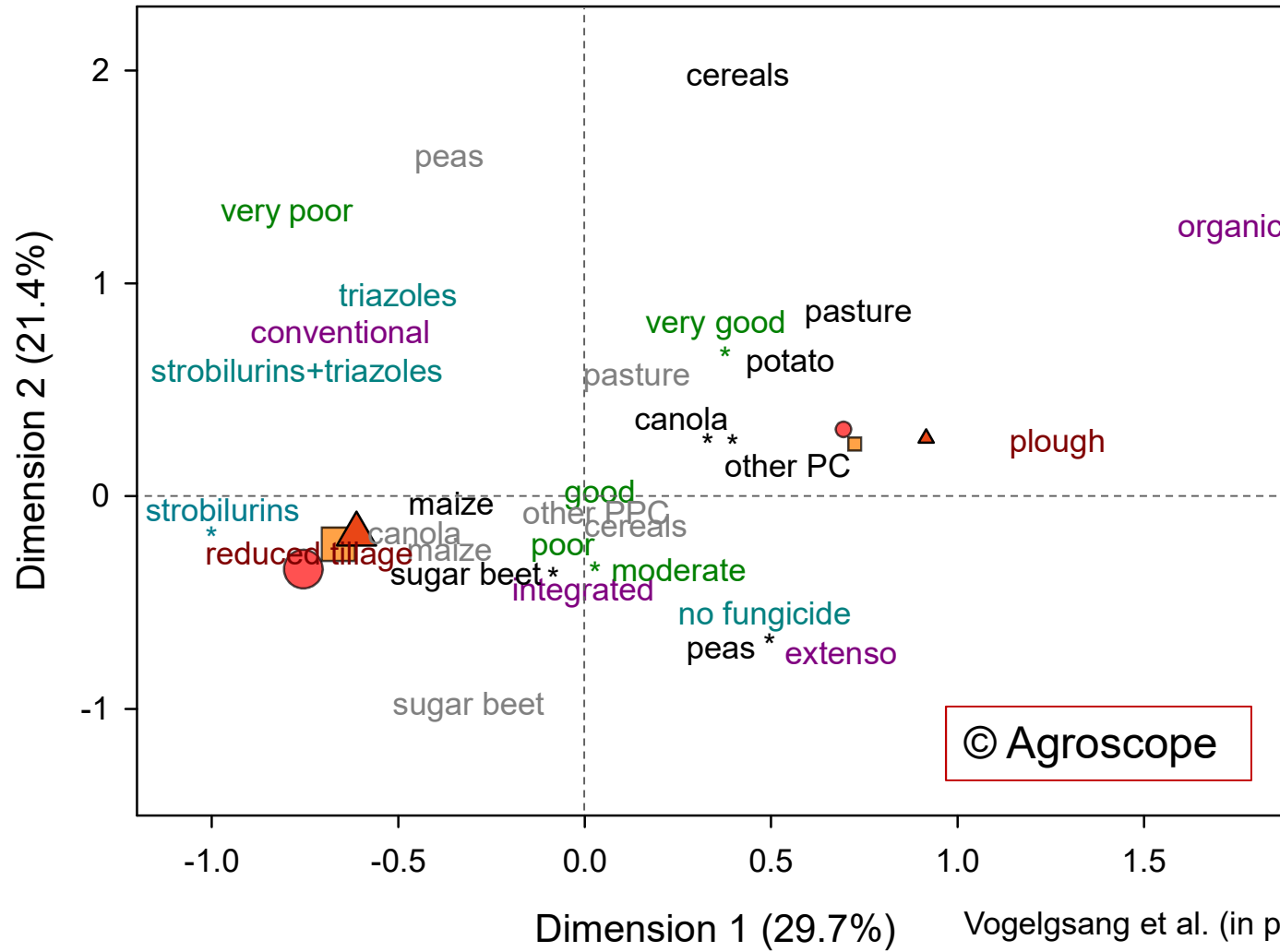


Multiple correspondence analysis **FG/DON**



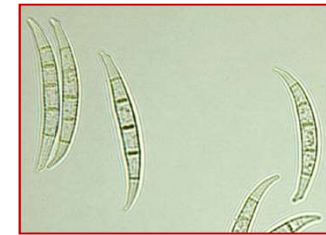
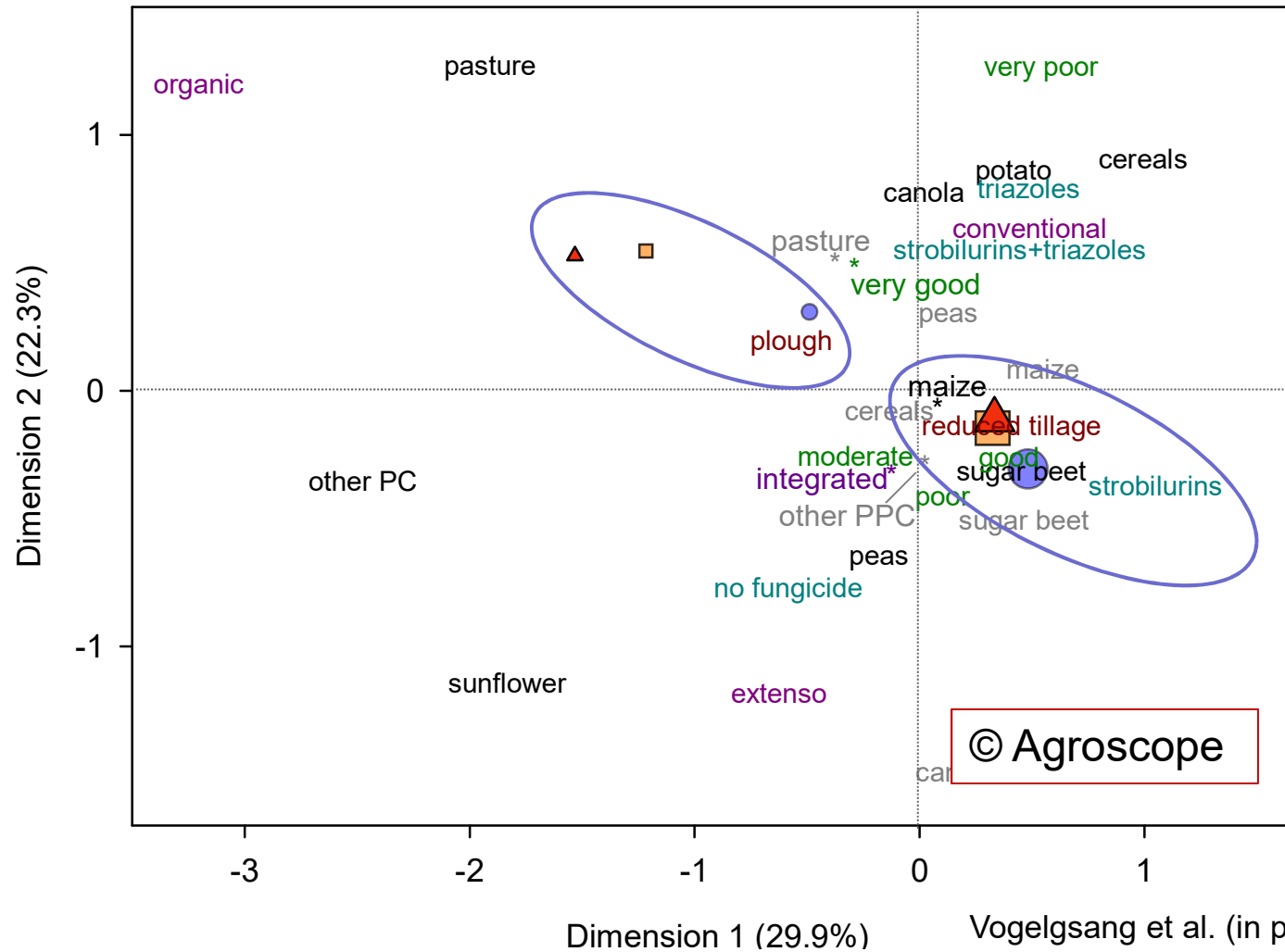


Multiple correspondence analysis **FG/DON**





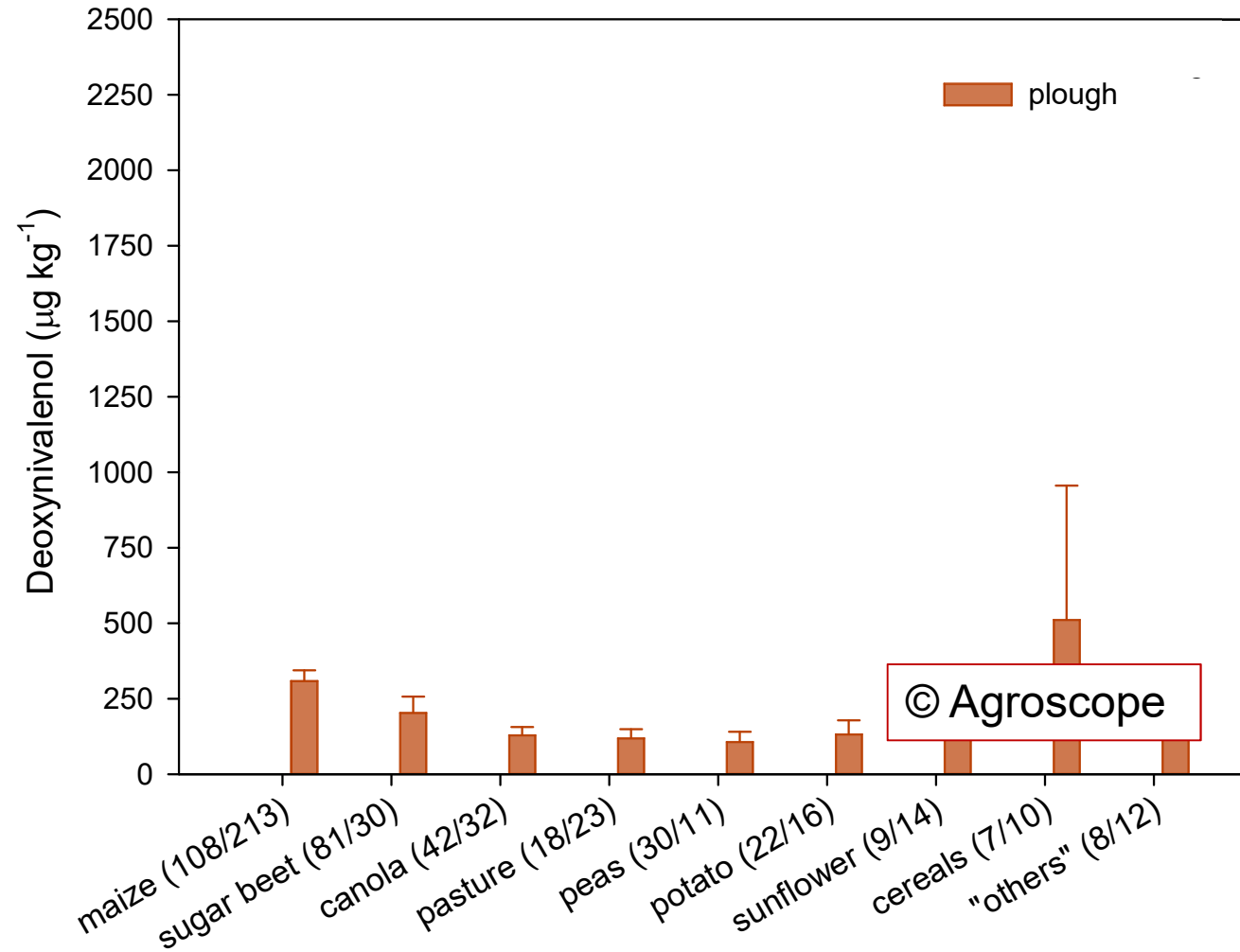
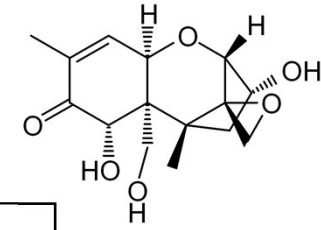
Multiple correspondence analysis **FG/ZEA**



- toxin
- △ DNA
- incidence

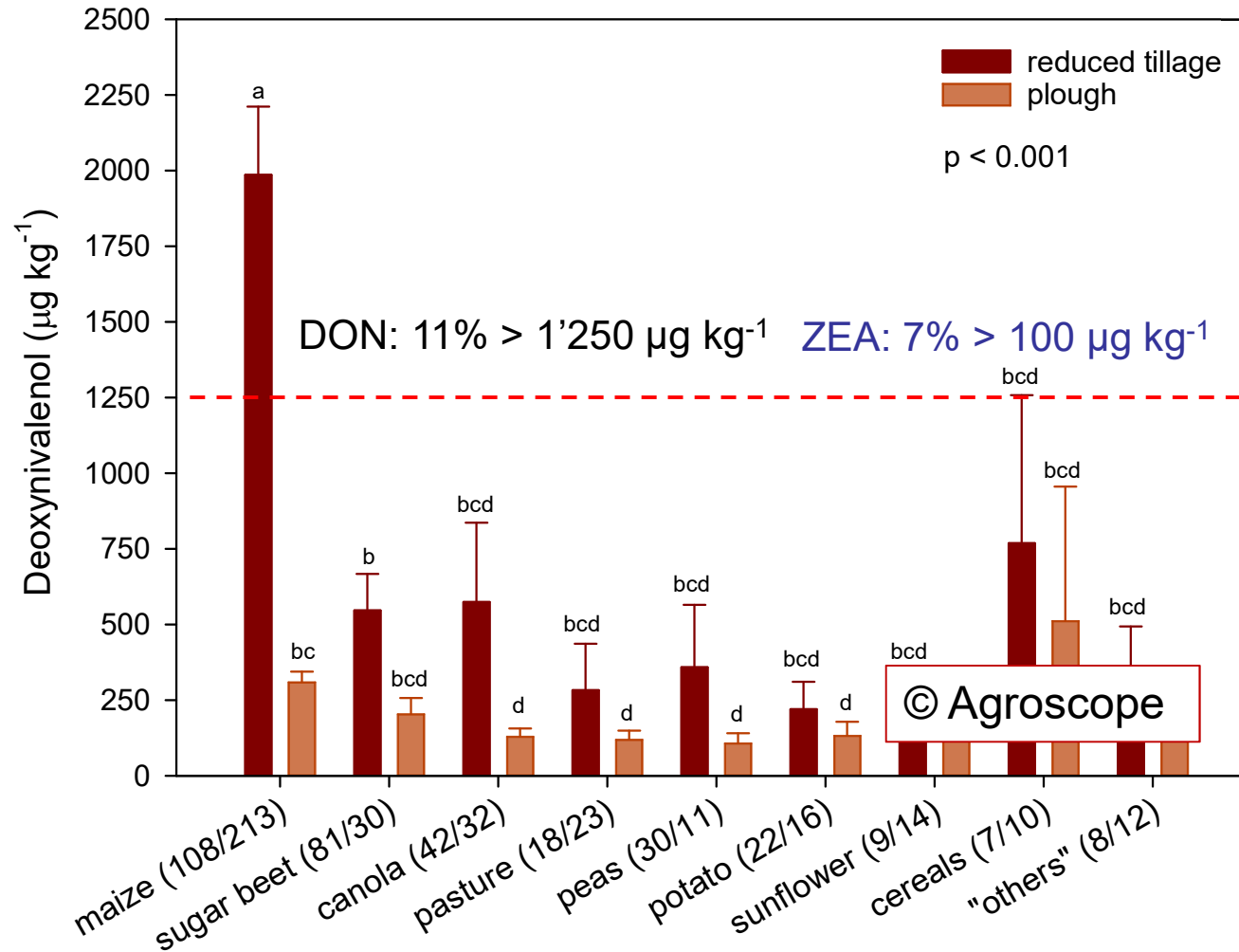
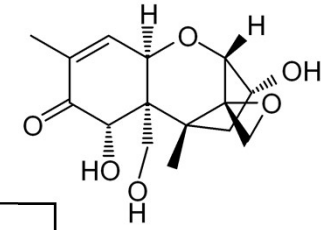


Exceeded maximum limits - DON





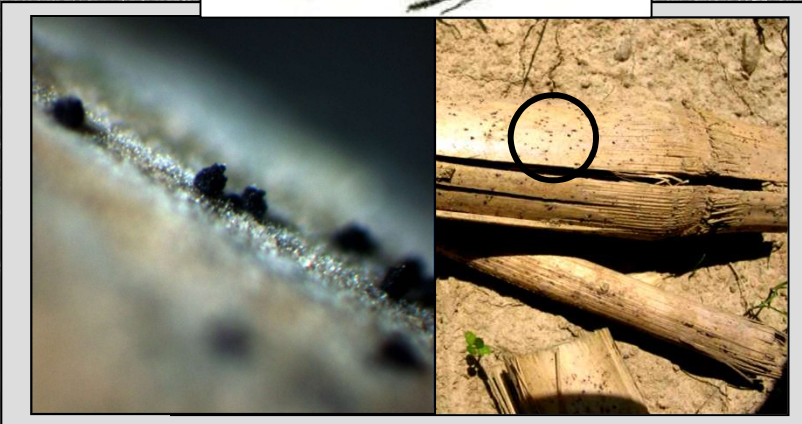
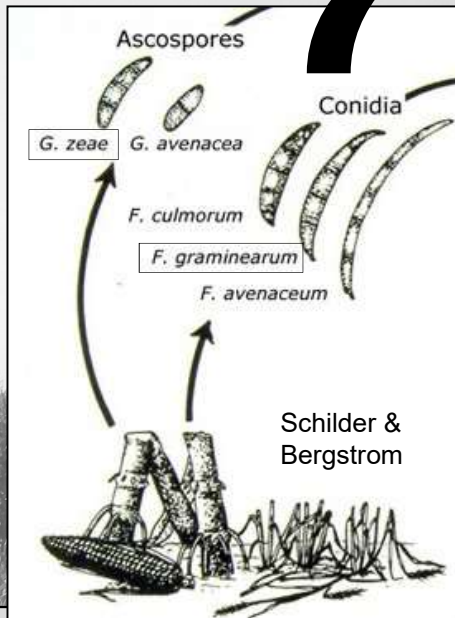
Exceeded maximum limits - DON



Life cycle *F. graminearum*



Life cycle *F. graminearum*



Determining ascospore deposition using „self-made“ and commercial spore traps



Trapped spores in wheat plot (RZ 30.05.07)

No precrop maize (plot 109)

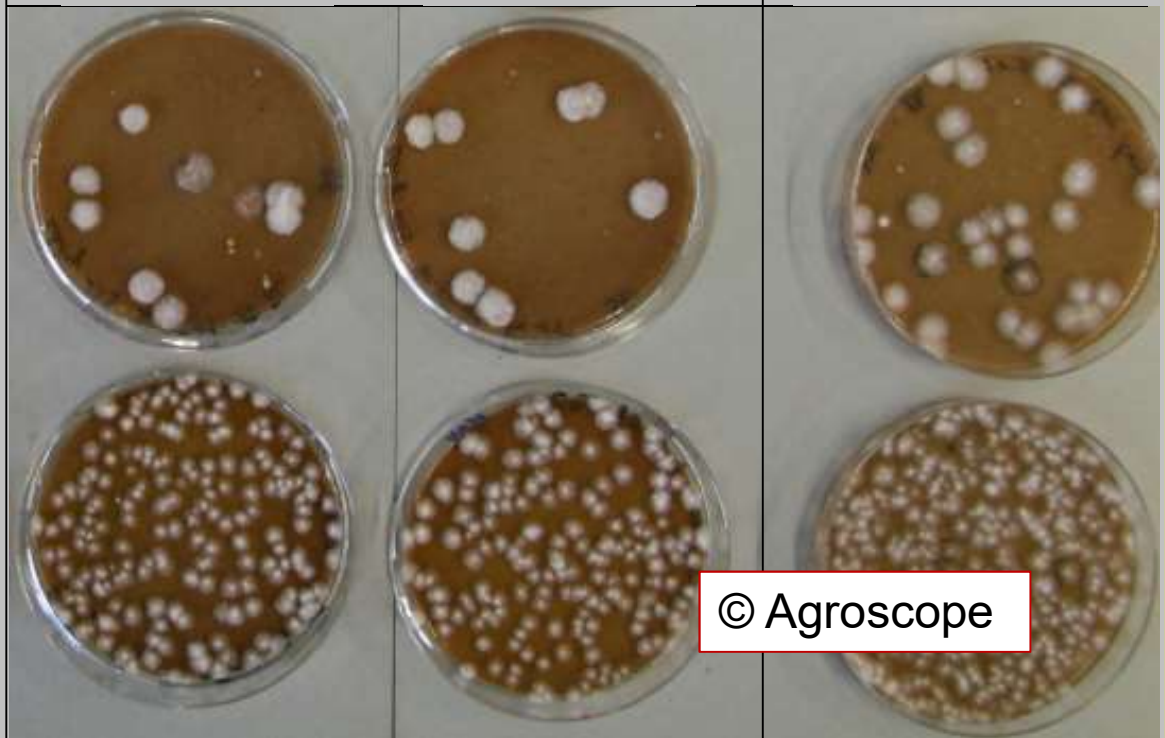


With precrop maize

Spore trap
© H.R. Forrer
Model „T“

Model „L“

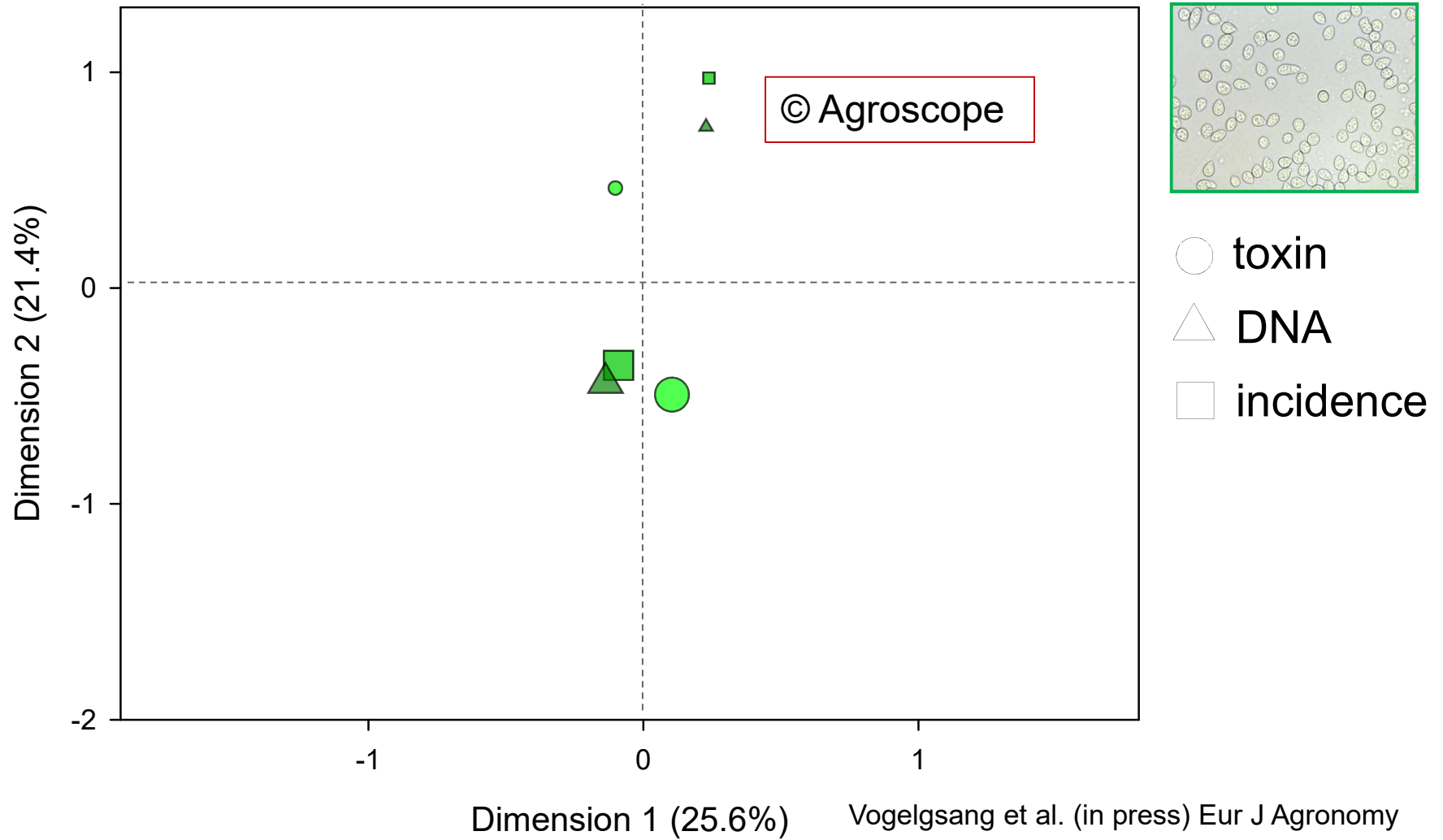
Burkard Jet
Spore sampler



colonies in Petri plates = parameter of spore deposition

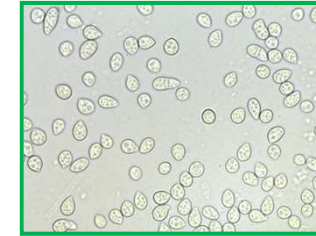
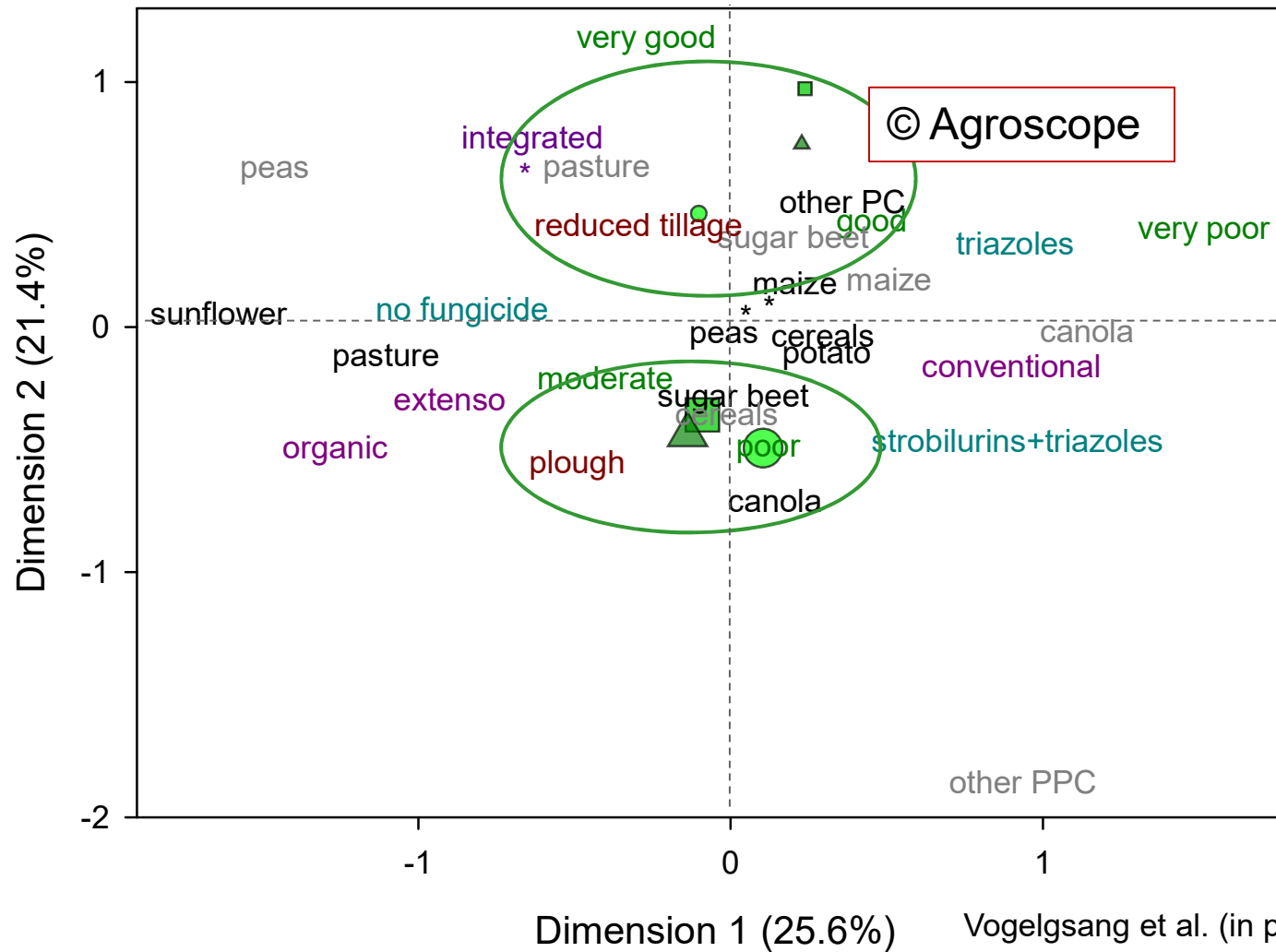


Multiple correspondence analysis FP/NIV





Multiple correspondence analysis FP/NIV





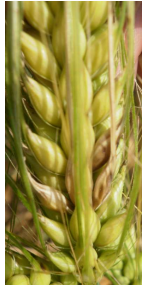
Outline

- **Surveys on *Fusarium* species and mycotoxins in wheat, barley and oats**
- **Our resulting research to reduce the risk in cereals:**
 - Identification of crucial factors and forecasting of *F. graminearum*/DON
 - “Healthy & Safe” cereal varieties
 - “MycoKey”: Biocontrol, cover crops and biofumigation



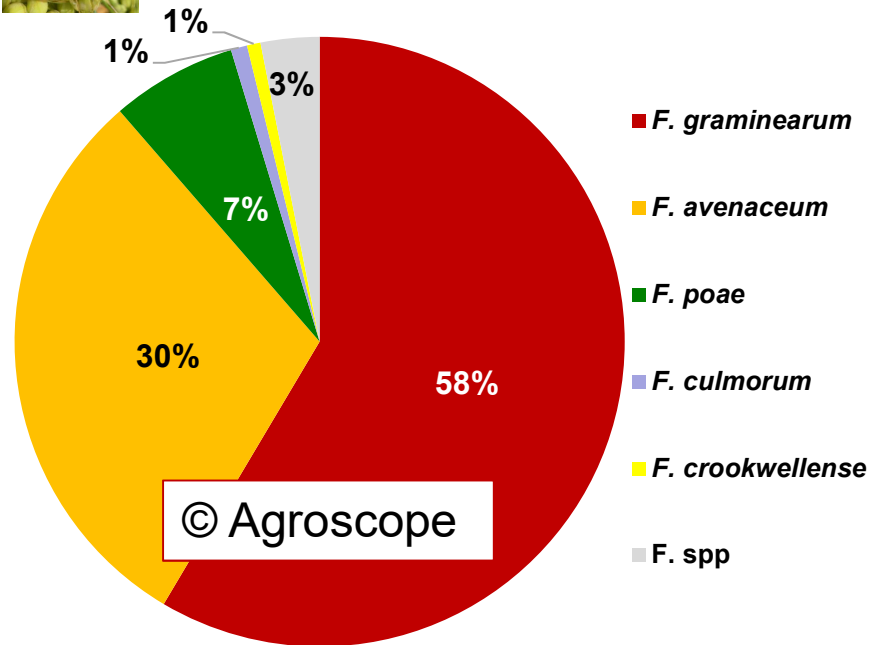


Fusarium occurrence in barley



Schöneberg et al. (2016)
Food Add Cont 33: 1608-6019

2013-2014: n=440



F. graminearum

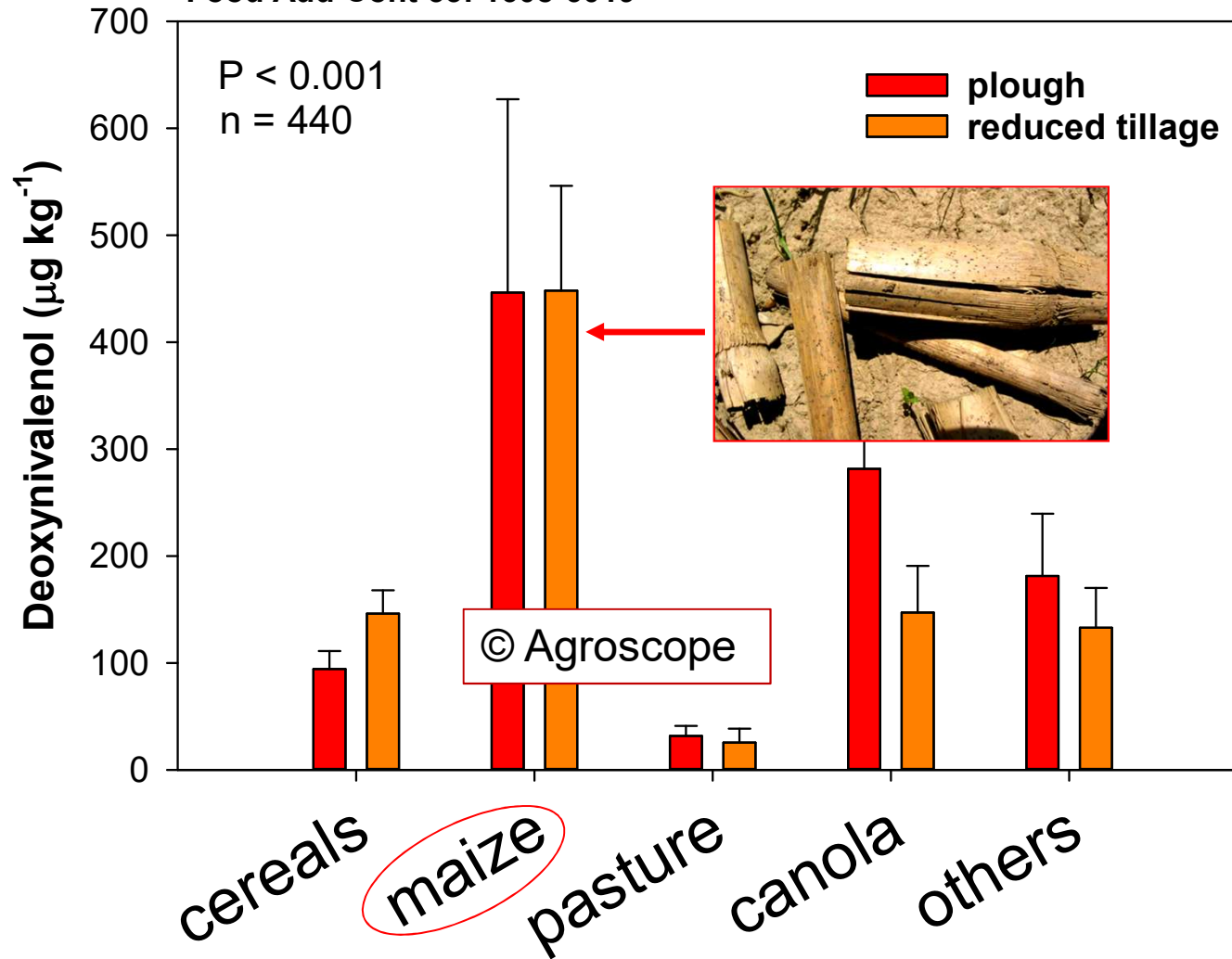


Effect of precrop/tillage on DON in barley



Schöneberg et al. (2016)
Food Add Cont 33: 1608-6019

...work in progress

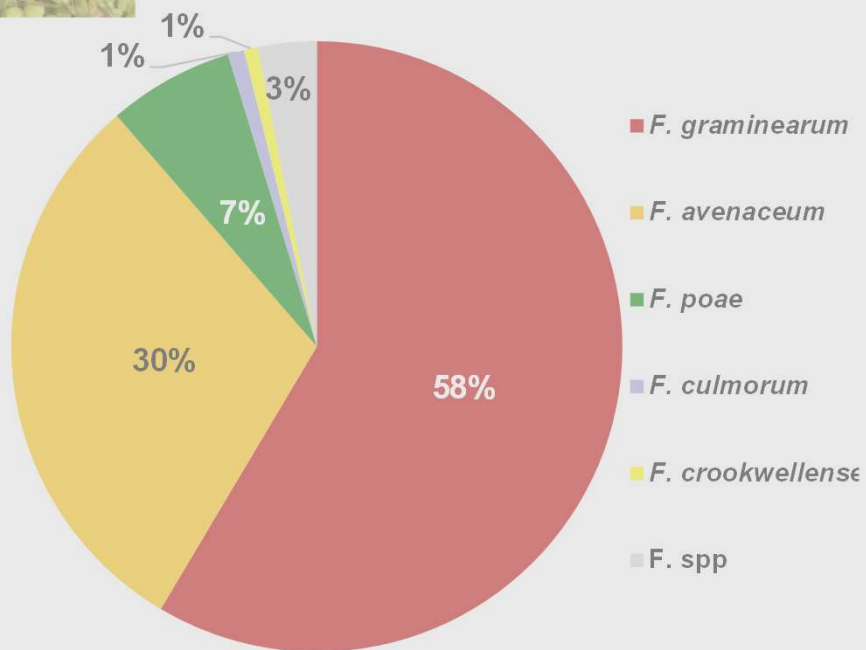


Fusarium occurrence in barley and oats



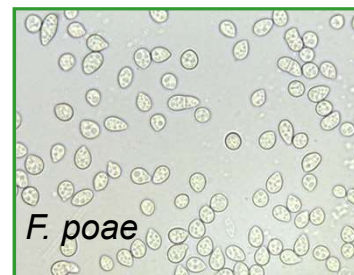
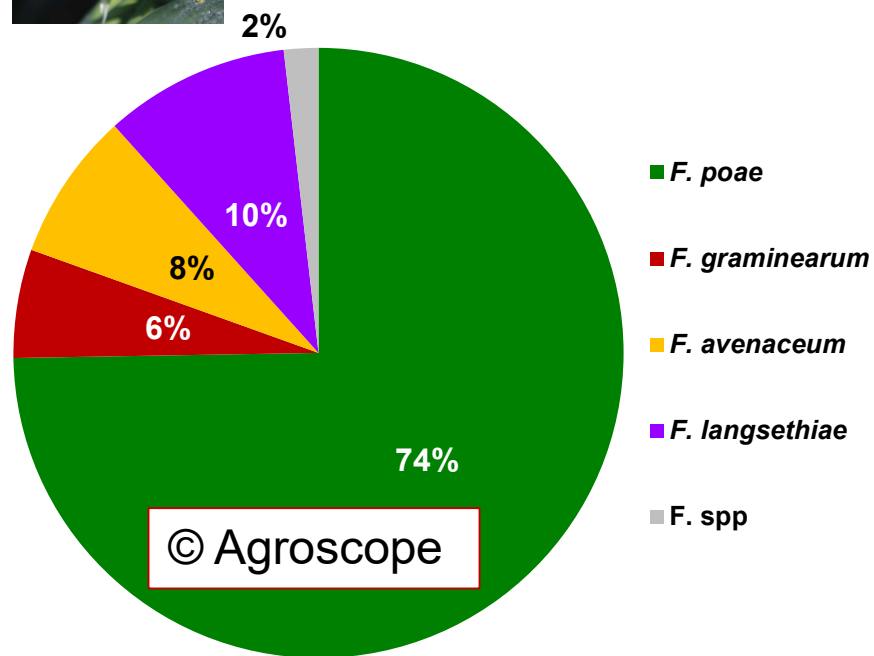
Schöneberg et al. (2016)
Food Add Cont 33: 1608-6019

2013-2014: n=440

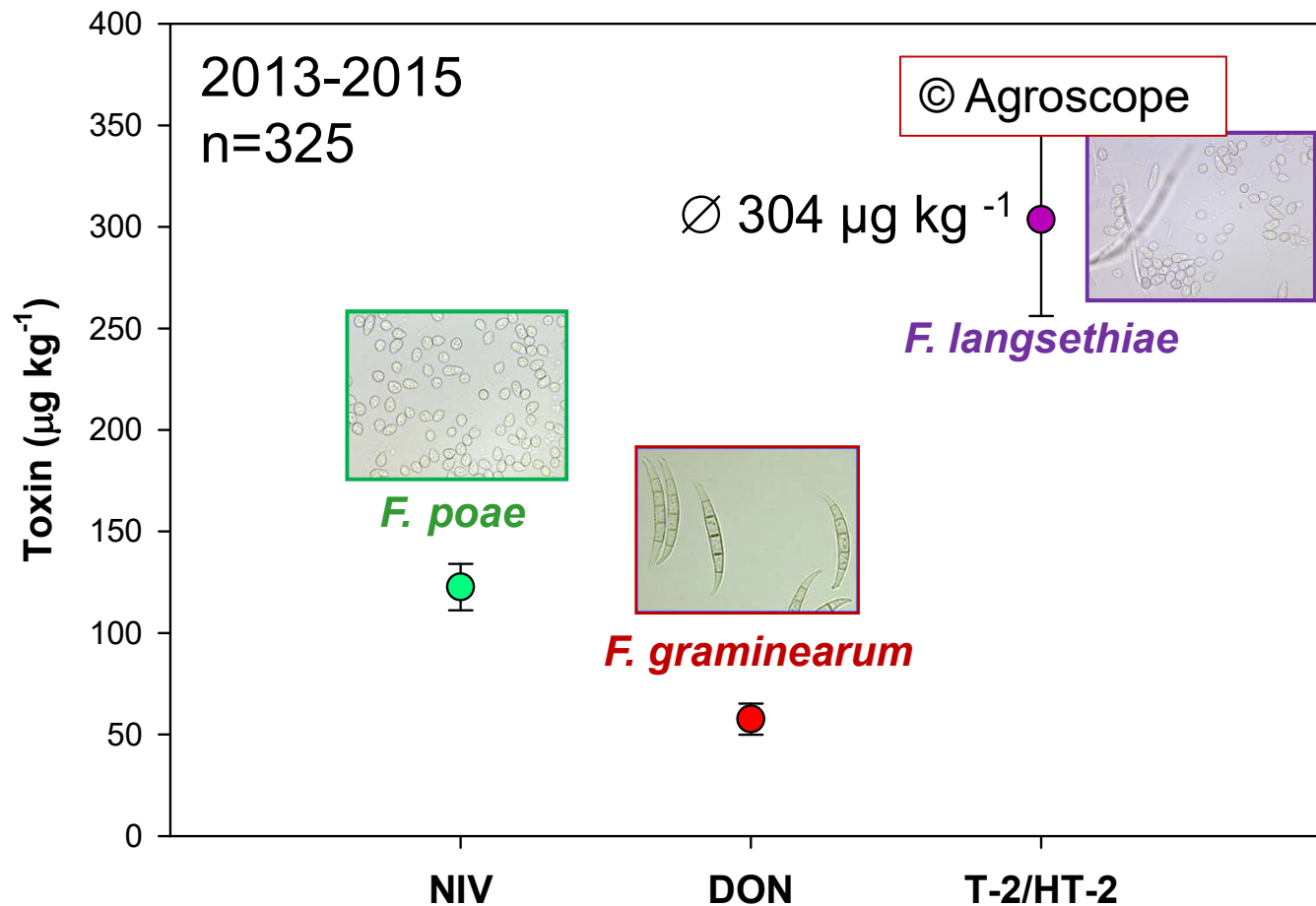


Schöneberg et al. (2018)
Eur J Agron 92: 123-132

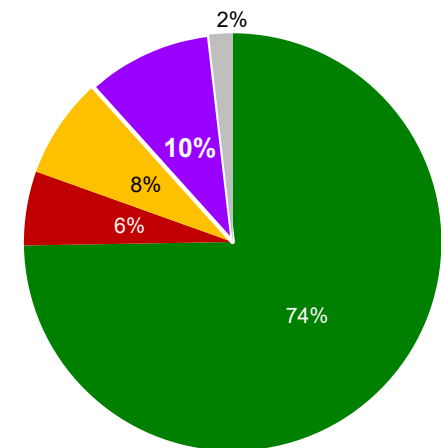
2013-2015: n=325



☝ Toxins in (hulled) **oats** from growers

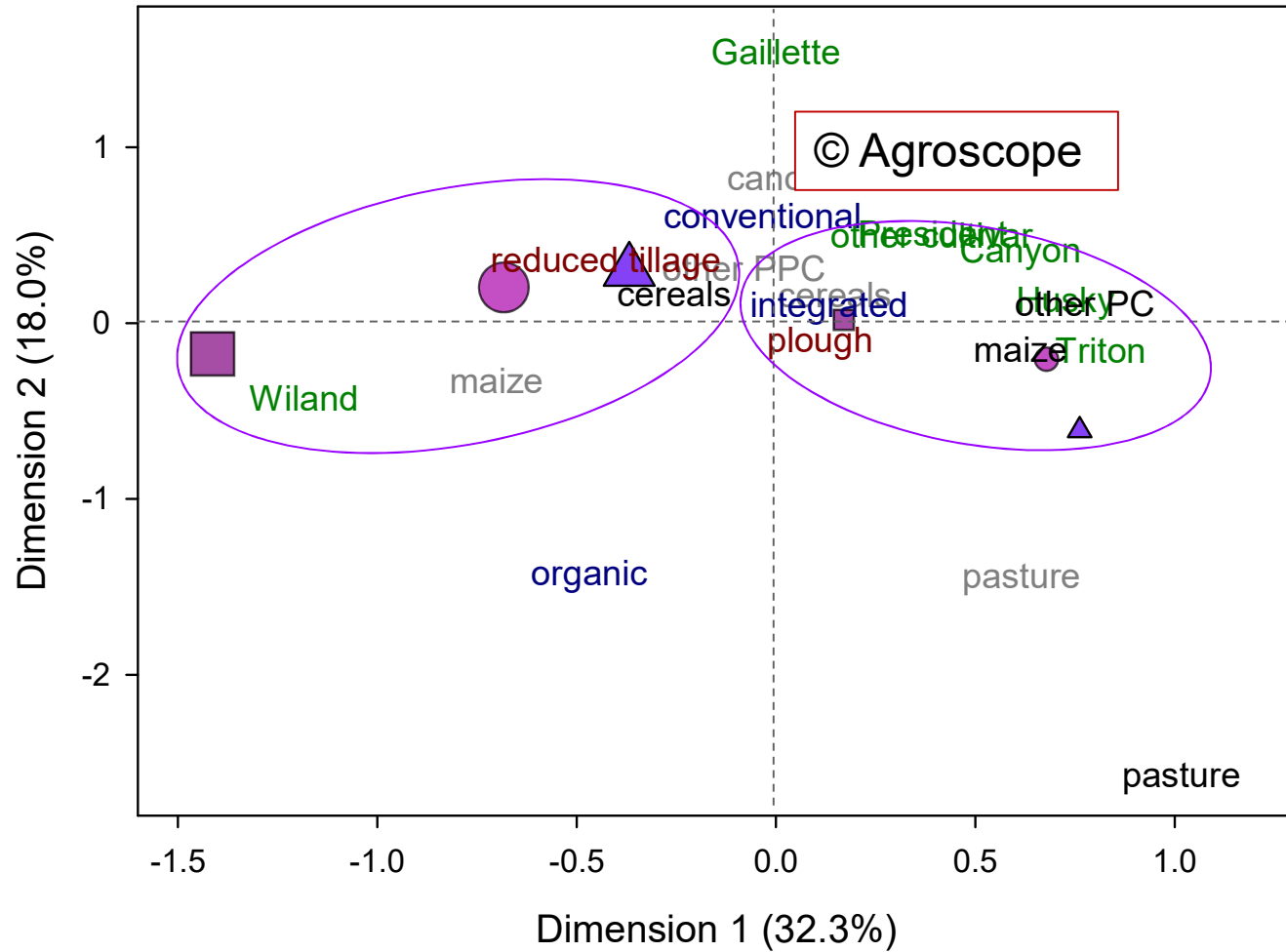


From a ratio of only 10%
→ qPCR: 33%





MCA for oats: *F. langsethiae* & T-2/HT-2





Outline

- Surveys on *Fusarium* species and mycotoxins in wheat, barley and oats
- **Our resulting research to reduce the risk in cereals:**
 - “Healthy & Safe” cereal varieties
 - “MycoKey”: Biocontrol, cover crops and biofumigation



«Healthy & Safe» cereals

- **Wheat, barley and oat cultivars**
Health promoting compounds (HPCs):
anthocyanins, phenolic acids, β -glucans
- **Healthy and safe food**
HPCs might inhibit infection through
toxigenic *Fusarium* species

Research questions

- Dominant species and toxins, impact of cropping factors?
- Epidemiology of the dominant species?
- **Differences in susceptibility of cultivars?**



Romina Morisoli, Charlotte Martin,
Mario Bertossa, Torsten Schöneberg,
Susanne Vogelgsang, Fabio Mascher



Resistance experiments

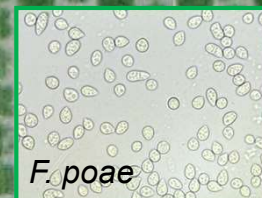


Charlotte Martin

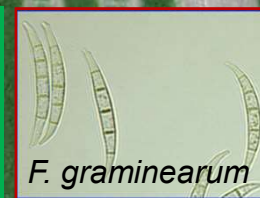


Torsten
Schöneberg

- 3 sites across Switzerland
- Wheat, oat, barley varieties with different contents of HPCs
- Artificial infections with selected *Fusarium* species



F. poae



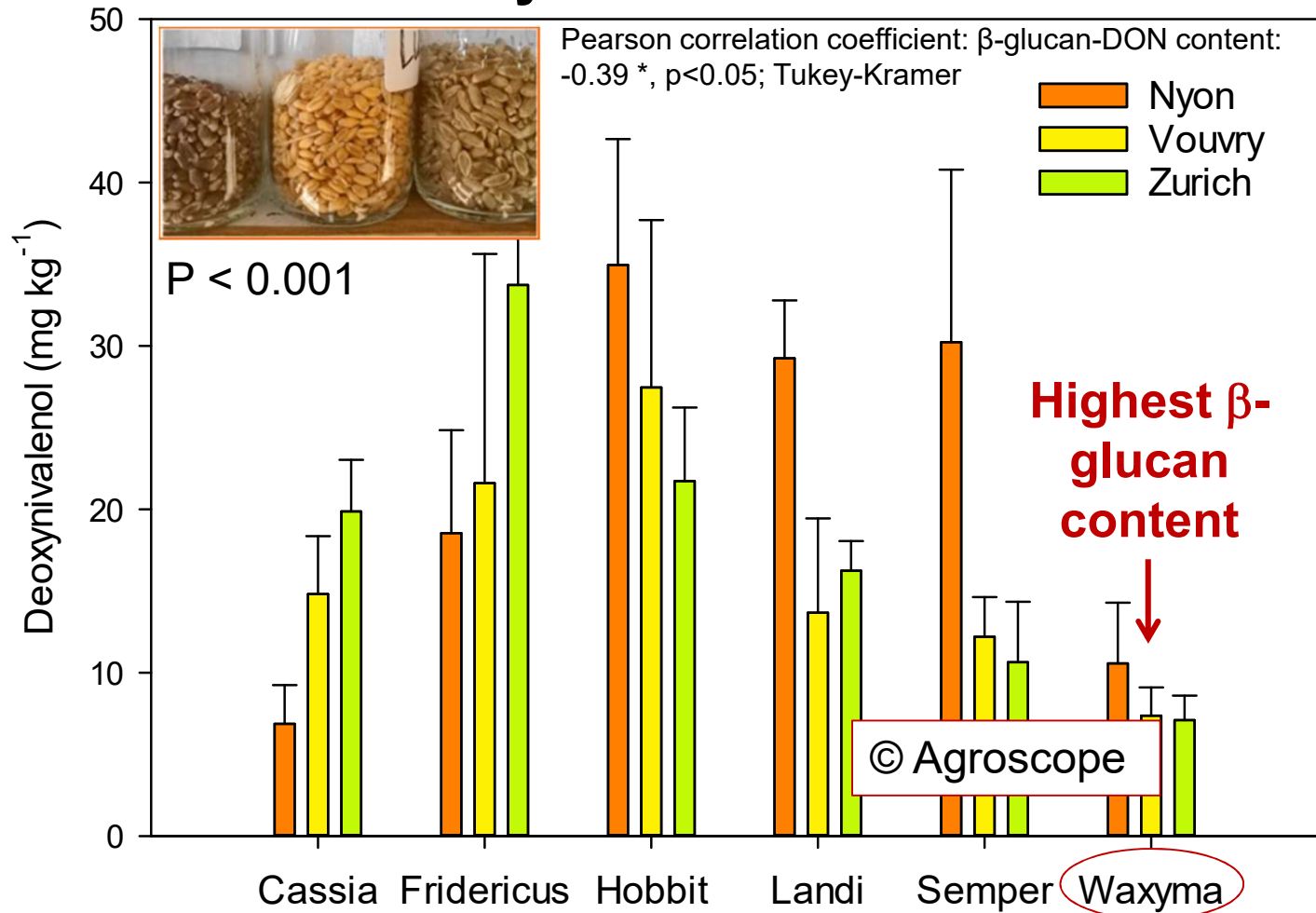
F. graminearum



F. langsethiae

Resistance experiments

DON in barley varieties - 2014





Outline

- Surveys on *Fusarium* species and mycotoxins in wheat, barley and oats
- **Our resulting research to reduce the risk in cereals:**
 - “Healthy & Safe” cereal varieties
 - **...if prevention is not enough:**
Biocontrol, cover crops
and biofumigation



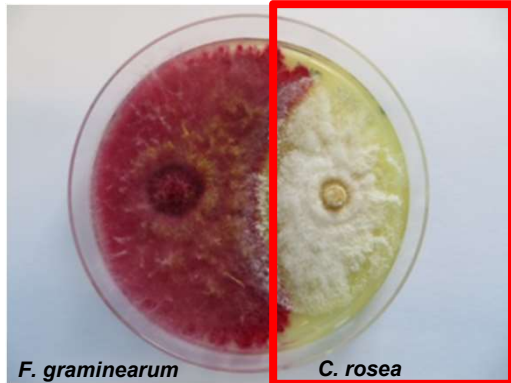


If prevention is not enough...

→ Avoid spore discharge from perithecia

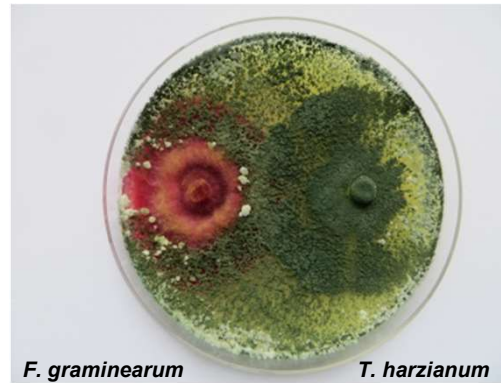
Biological control

- Fungal antagonists on residues from pre-crop



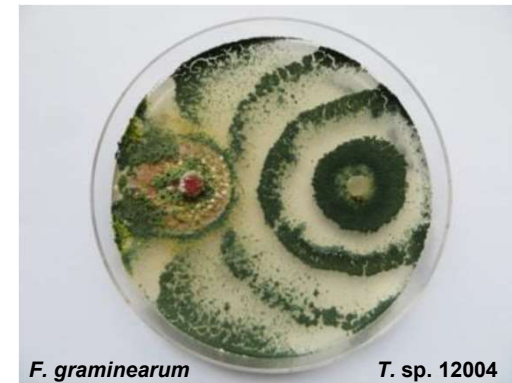
F. graminearum

C. rosea



F. graminearum

T. harzianum



F. graminearum

T. sp. 12004



Anita Schöneberg

*Schöneberg et al. (2015) J Appl Microbiol

... MycoKey comes in



© Agroscope

**Clonostachys rosea* – after the pathogen

Biological control



Fungal antagonist *Clonostachys rosea*

- On residues from pre-crop or directly on wheat heads
- Inoculum production





Biological control



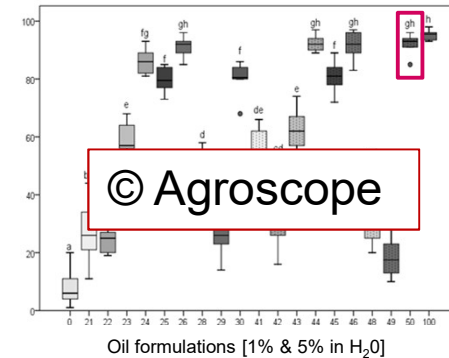
Fungal antagonist *Clonostachys rosea*

- On residues from pre-crop or directly on wheat heads
- Inoculum production
- Formulation (shelf-life, UV resistance)



Formulation development

Controlled environment



Field experiments 2016-2018





Antagonist treatments of infected maize stalk residues



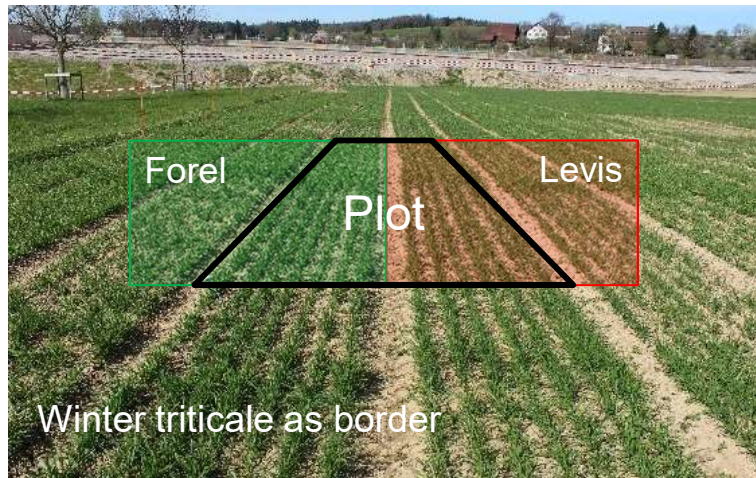
F. graminearum infected maize stalks



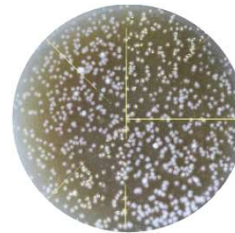
Antagonist suspensions of different formulations



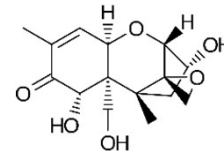
Antagonist treatments of infected maize stalk residues



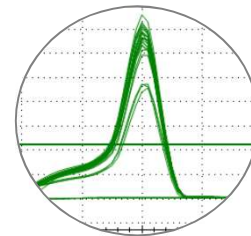
FHB symptoms (%)



Spore deposition



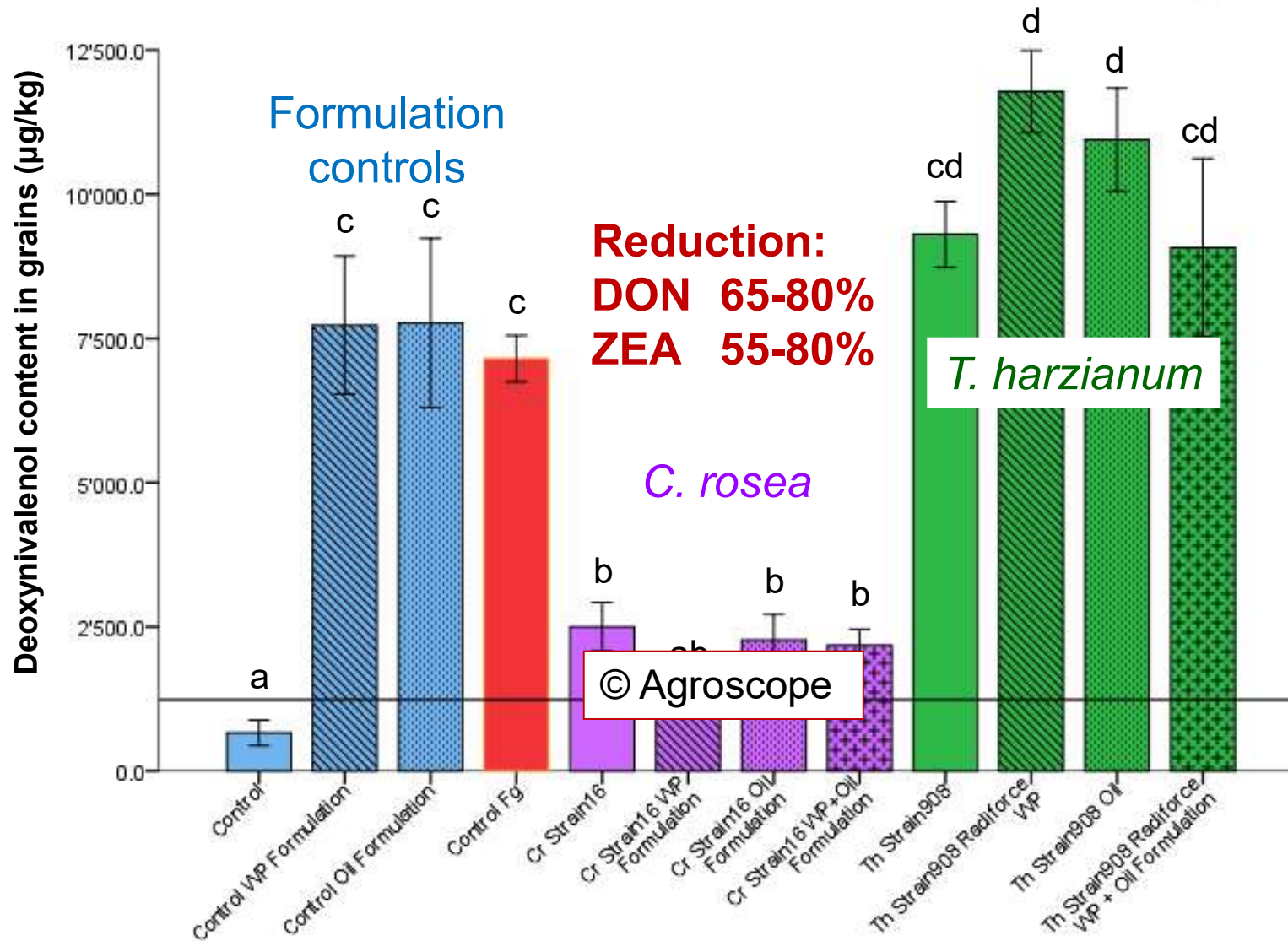
Mycotoxins DON, ZEA



qPCR *F. graminearum*



DON in grains (Levis/Forel)





Suppression of inoculum



- Various **intercrop/cover crop species** on residues from pre-crop: coverage, increased microbial activity or directly anti-fungal
- **Biofumigation**, botanicals



Dimitrios Drakopoulos





Biofumigation of infected maize stalks

White mustard

Sinapis alba

Indian mustard

Brassica juncea

Berseem clover

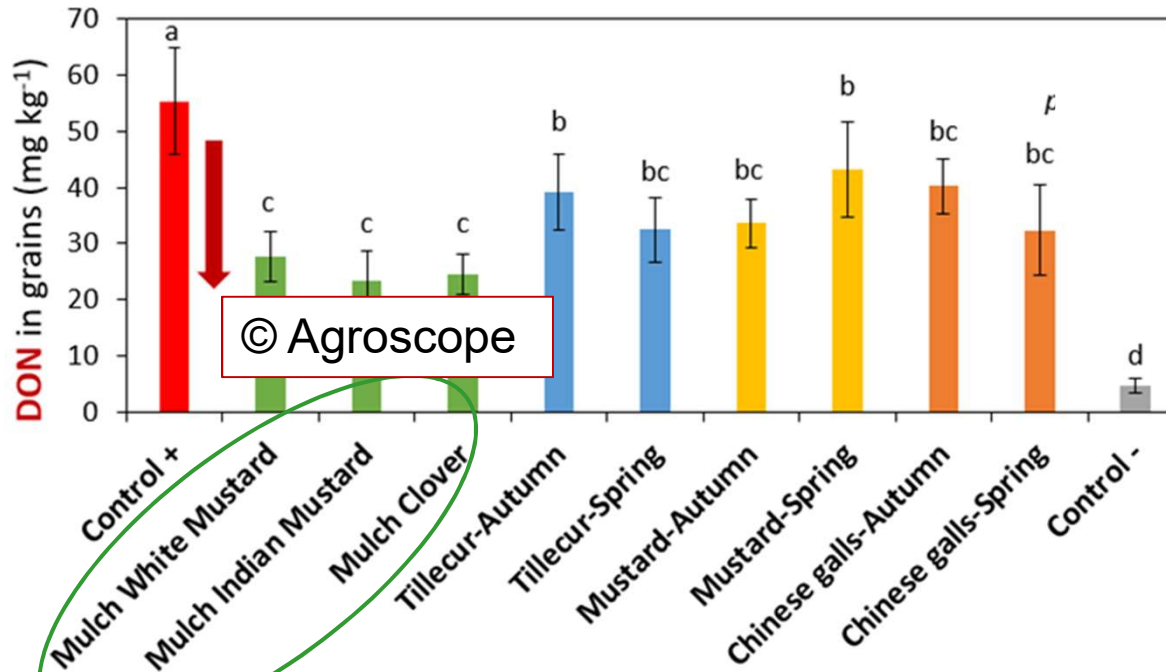
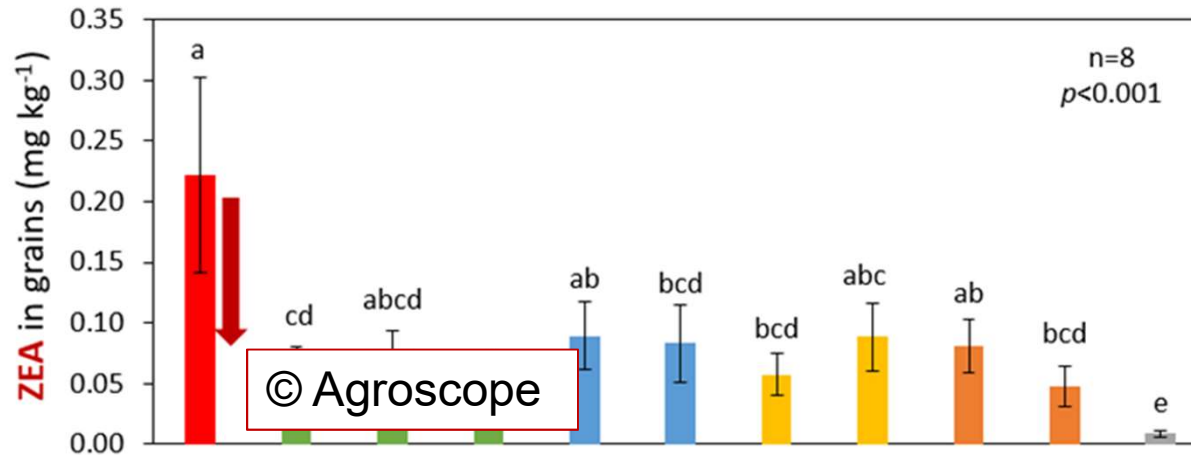
Trifolium alexandrinum

*Application rate 1.9 kg m⁻²
(mustards “plot to plot”)*





Biofumigation of infected maize stalks



Reduction:
ZEA up to 60%
DON up to 75%

Conclusions

- **Significance of mycotoxin surveys?**
 - Crucial to identify dominant *Fusarium* species and influencing cropping factors
- **Wheat = barely = oats?**
 - Extrapolation not possible due to different *Fusarium* species and/or different effects of cropping factors
- **Which measure to choose?**
 - One single measure not sufficient
 - Meaningful combination of puzzle pieces needed



Acknowledgements

- **Research group** “Ecological Plant Protection in Arable Crops”, in particular:
Irene Bänziger, Andreas Kägi, various interns
PhD students Charlotte Martin, Torsten Schöneberg,
Alejandro Gimeno, Dimitrios Drakopoulos
- **Michel Gygax, Sandra Ott, Florian Burkhalter**
«Soil preservation programme», canton Berne
- **Members CPPOs, growers**



Kanton Bern
Canton de Berne



Healthy Nutrition and Sustainable Food Production
National Research Programme NRP 69

Questions?

