



Studies on the sensitivity status of *Alternaria solani* towards QoIs, SDHIs and DMIs

EuroBlight Workshop, Lunteren, Netherlands

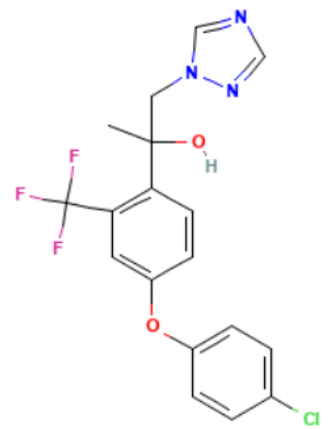
15th Mai 2024

Carolina Schroeder

 **BASF**

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Fungicide control of *Alternaria solani*



Quinone Outside Inhibitors (QoI)

- Inhibitor of respiration in complex III at Qo-site

Succinate Dehydrogenase Inhibitors (SDHI)

- Inhibitor of respiration in complex II at SDH

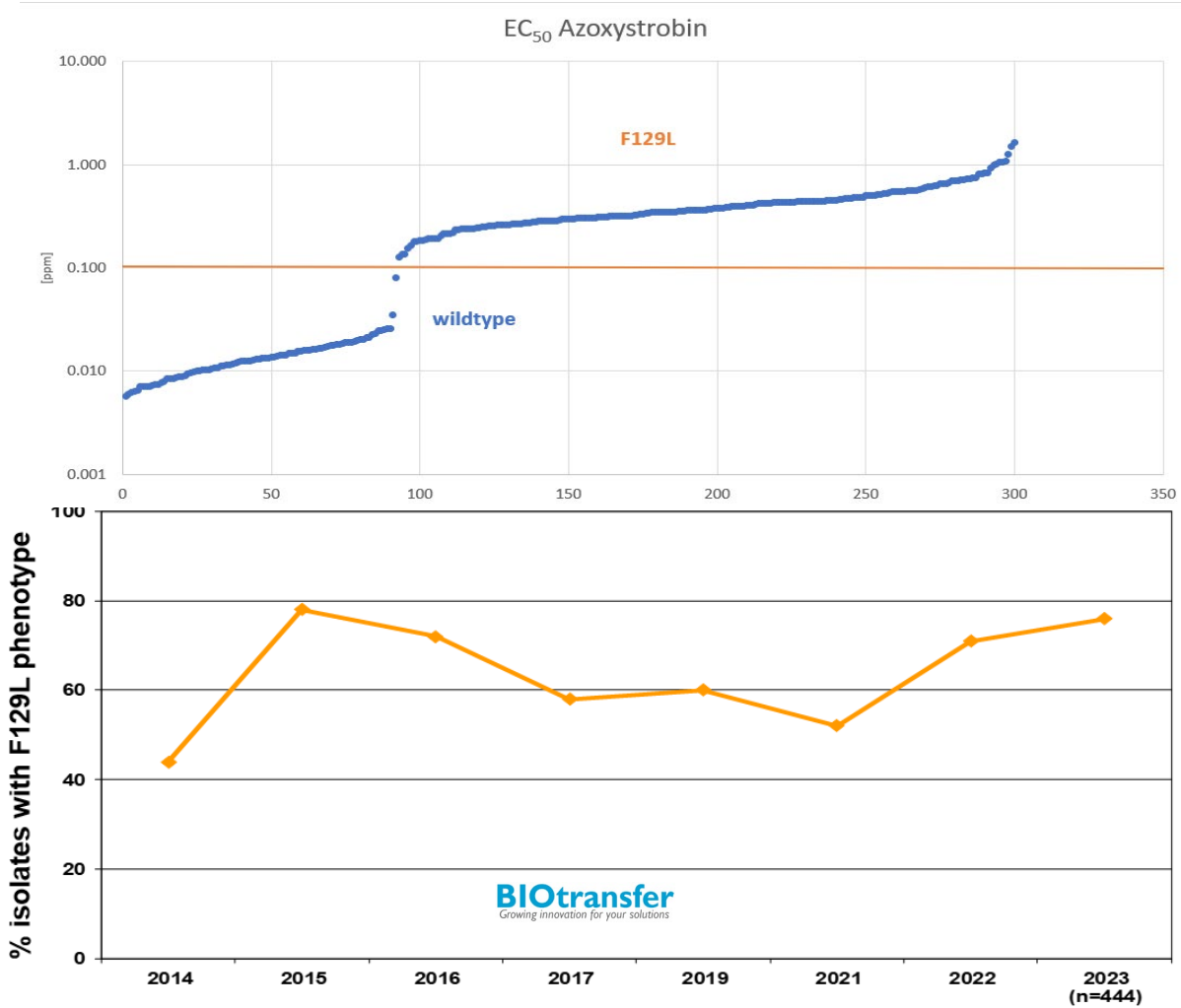
Demethylation Inhibitors (DMI)

- Sterol biosynthesis Inhibitor

Development of the frequency QoI adaptation (F129L) in *Alternaria solani*

BASF European Monitoring (F129L pheno- or genotype)

Latest FRAC statement



Resistance to QoI is associated to the presence of the F129L mutation and molecular information are provided below:

Data from 2023 showed a situation as known from previous years:

High frequency was detected in Belgium, Germany, Netherlands and Sweden.

Moderate frequency was detected in France, Latvia and Poland.

Data from 2022 showed a situation as known from previous years:

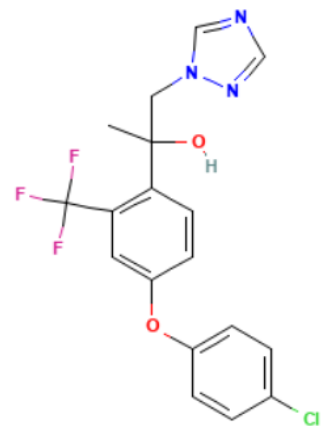
High frequency was detected in Denmark, Netherlands and Sweden.

Moderate frequency was detected in Austria, Belgium, Germany and Norway

➔ Majority of European population with F129L phenotype



Fungicide control of *Alternaria solani*



Quinone Outside Inhibitors (QoI)

- Inhibitor of respiration in complex III at Qo-site

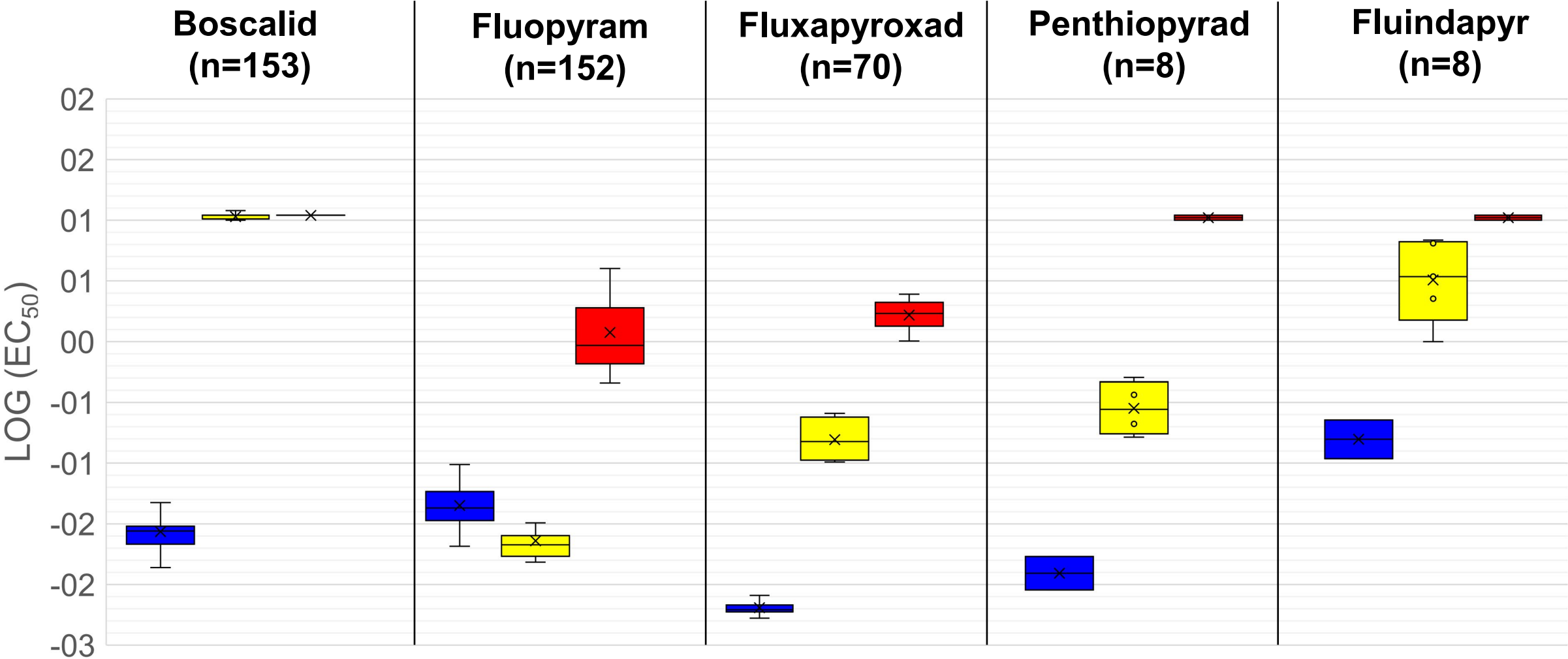
Succinate Dehydrogenase Inhibitors (SDHI)

- Inhibitor of respiration in complex II at SDH

Demethylation Inhibitors (DMI)

- Sterol biosynthesis Inhibitor

EC₅₀ values of wildtype, B-H278Y and C-H134R mutations on different SDHIs

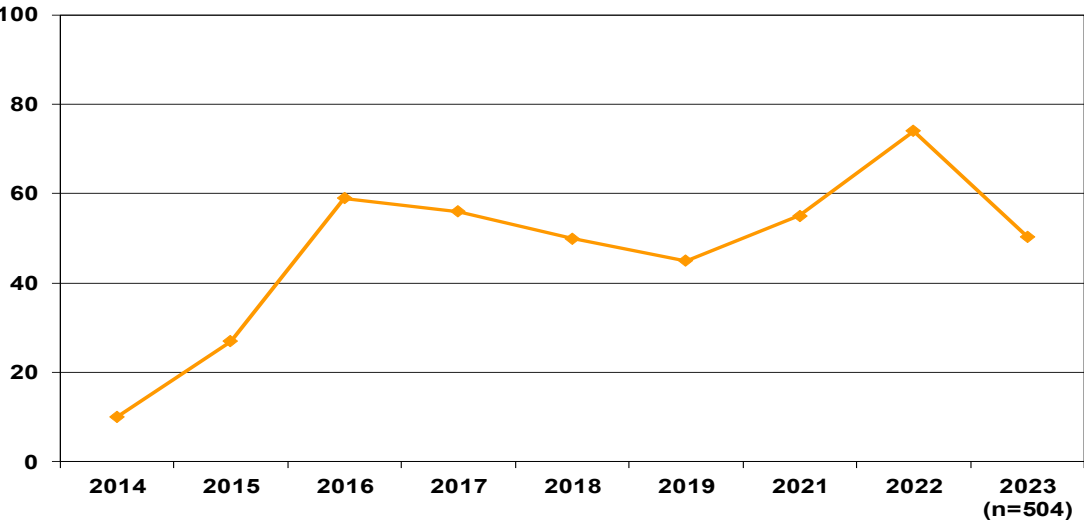


⇒ C-H134R shows strong effect on all SDHIs, H278Y in 2023 not detected, H134R dominates



Development of the frequency of SDHI resistance in *Alternaria solani* in Europe

BASF Monitoring (% SDHI resistant isolates)



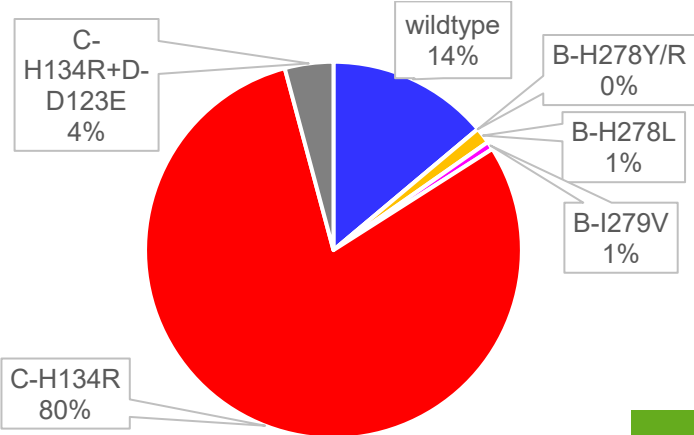
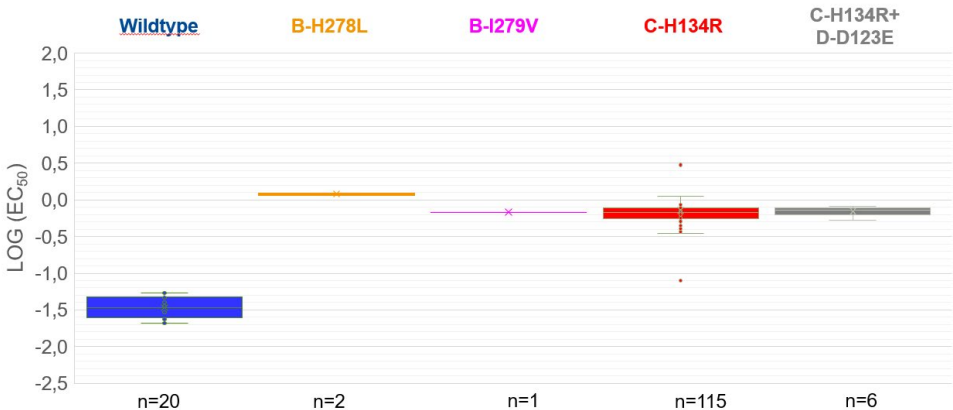
Latest FRAC statement

Potato – Early blight, Alternaria leaf spot (*Alternaria solani*, *A. alternata*)
(Bayer, BASF, Syngenta)

In **2022**, *A.solani* samples were analyzed originating from Austria, Denmark, and Norway.
The following mutations have been detected: B-H278R/Y, C-H134R, D-D123E with B-H278Y and C-H134R being the predominant mutations found.
Low frequencies of target site mutations were found in Norway.
Moderate frequencies were found in Austria.
High frequencies were detected in Denmark.

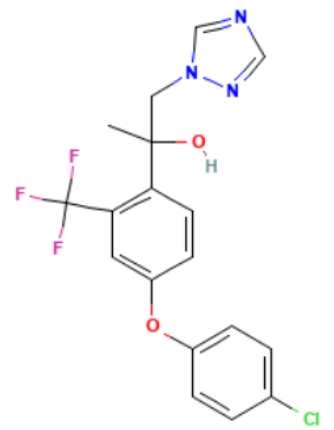
SDHI (Fluopyram) analysis 2023 in NL trial sites

EC₅₀ of **wildtype**, **B-H278L**, **B-I279V**, **C-H134R** and **C-134R+D-D123E** for **Fluopyram**



SDHI resistance heterogenous distributed, over all moderate levels in Europe. Mutation pattern changed in last years.

Fungicide control of *Alternaria solani*



Quinone Outside Inhibitors (QoI)

- Inhibitor of respiration in complex III at Qo-site

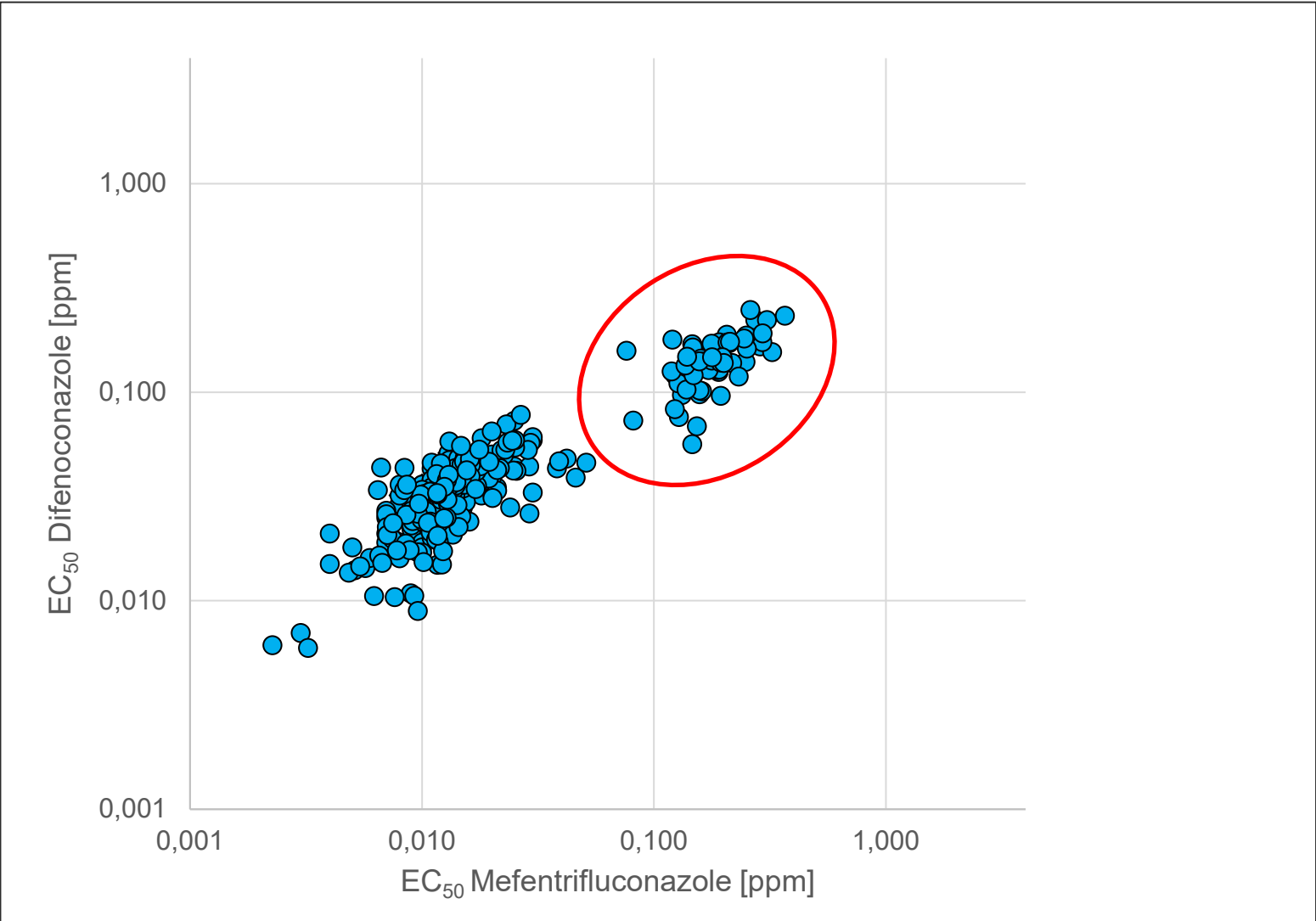
Succinate Dehydrogenase Inhibitors (SDHI)

- Inhibitor of respiration in complex II at SDH

Demethylation Inhibitors (DMI)

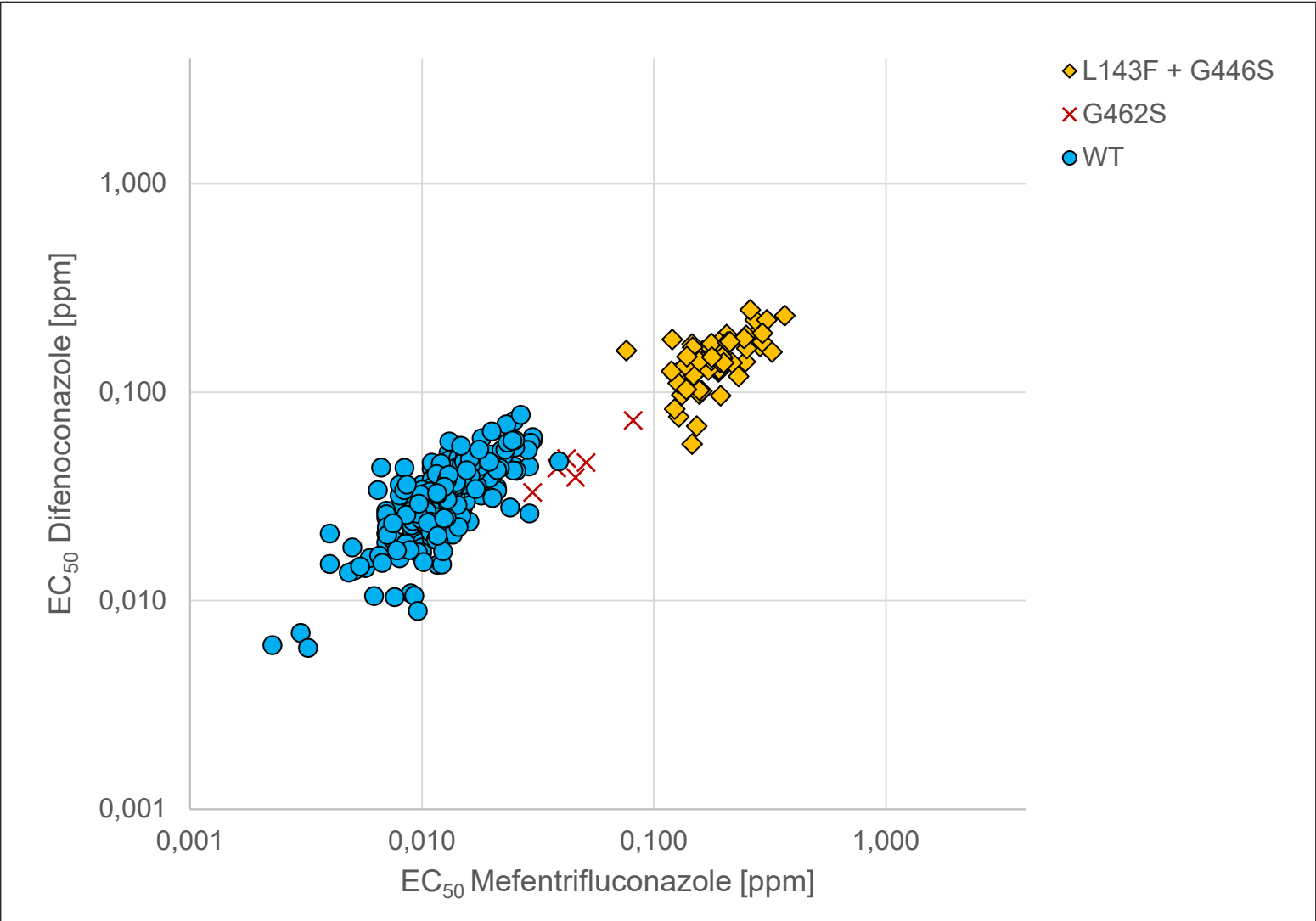
- Sterol biosynthesis Inhibitor

Sensitivity distribution of *Alternaria solani* to Mefentrifluconazole and Difenoconazole in 2021-2023



Isolates with slight adaptation were detected

Sensitivity distribution of *Alternaria solani* to Mefentrifluconazole and Difenoconazole in 2021-2023



Such isolates with slight adaptation harbour target site mutations **L143F+G446S** or **G462S**

Mutations are homologous to mutations in other phytopathogenic species and relate to slight DMI adaptation with limited effects

CYP51 mutations found	homologous to
L143F	L144F (<i>Cercospora beticola</i>) L140F (<i>Venturia inaequalis</i>)
G446S	G444S (<i>Venturia inaequalis</i>)
G462S	G461S (<i>Monilinia fructicola</i>) G460S (<i>Pyrenopeziza brassicae</i>)



Alterations in the predicted regulatory and coding regions of the sterol 14 α -demethylase gene (*CYP51*) confer decreased azole sensitivity in the oilseed rape pathogen *Pyrenopeziza brassicae*

HELEN E. CARTER¹, BART A. FRAAIJE¹, JONATHAN S. WEST², STEVEN L. KELLY³, ANDREAS MEHL⁴, MICHAEL W. SHAW⁵ AND HANS J. COOLS^{1*}

Phytopathology • 2017 • 107:1507-1514 • <https://doi.org/10.1094/PHYTO-02-17>

Genetics and Resistance

The Point Mutation G461S in the *MfCYP51* Gene is Associated with Tebuconazole Resistance in *Monilinia fructicola* Populations in Brazil

Paulo S. F. Lichtemberg, Yong Luo, Rafael G. Morales, Juliana M. Muehlmann-Fischer, Themis J. Michailides, and Louise L. May De Mio[†]

Journal of Plant Diseases and Protection
<https://doi.org/10.1007/s41348-021-00516-0>

ORIGINAL ARTICLE

Mutations in *Cyp51* of *Venturia inaequalis* and their effects on DMI sensitivity

Mascha Hoffmeister¹ · Raffaello Zito¹ · Jan Böhm¹ · Gerd Stammler¹



Research Article

Received: 8 July 2020 Revised: 10 November 2020 Accepted article published: 25 November 2020 Published online in Wiley Online Library:

(wileyonlinelibrary.com) DOI 10.1002/ps.6197

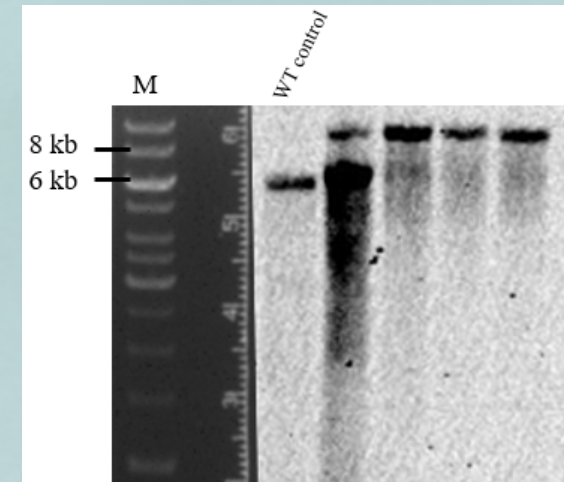
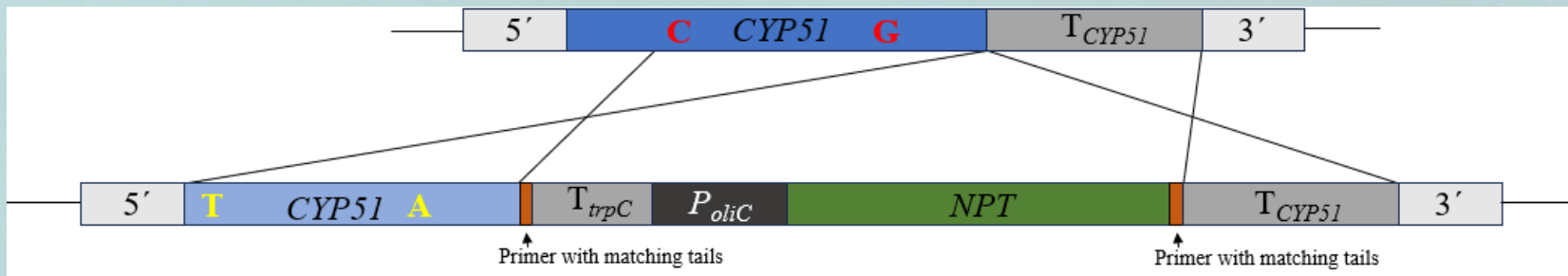
Evidence for the association of target-site resistance in *cyp51* with reduced DMI sensitivity in European *Cercospora beticola* field isolates

Maximilian M Muellender,^{a*} Anne-Katrin Mahlein,^a Gerd Stammler^b and Mark Varrelmann^a

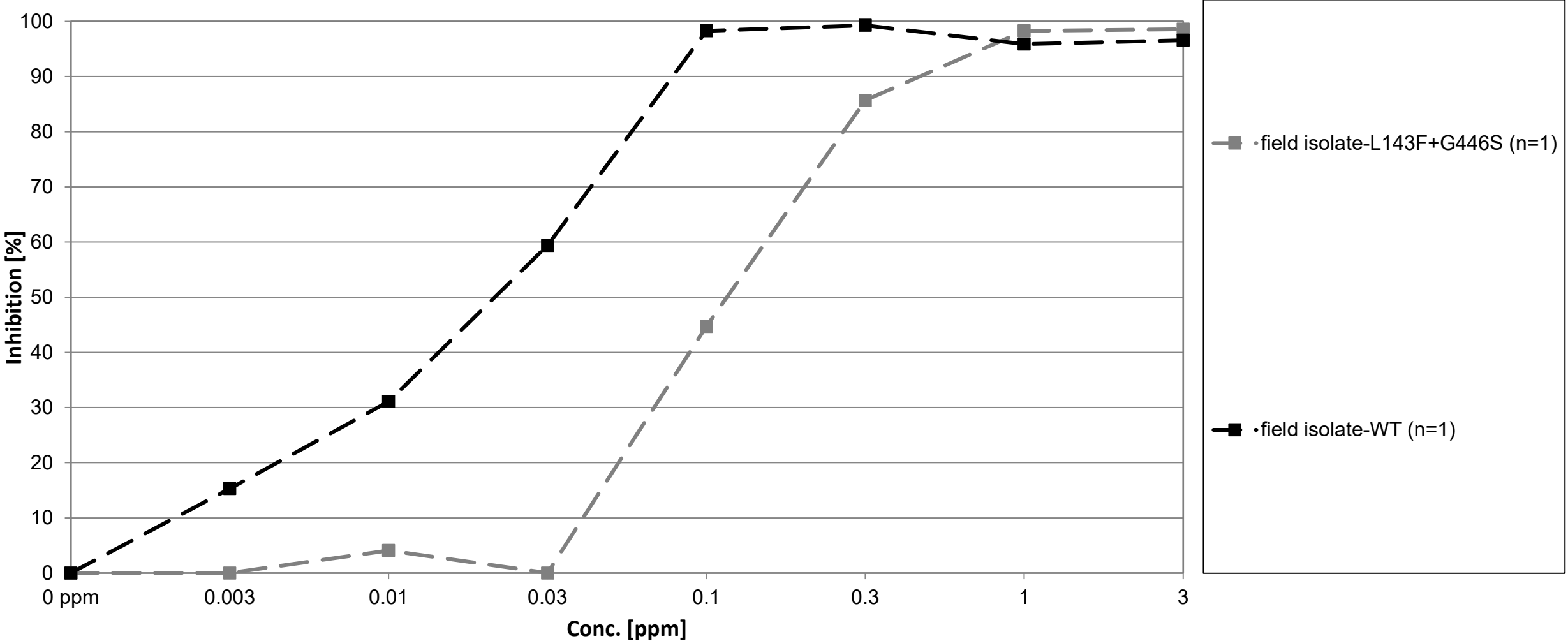
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Proof that mutations are responsible for slight DMI adaptation

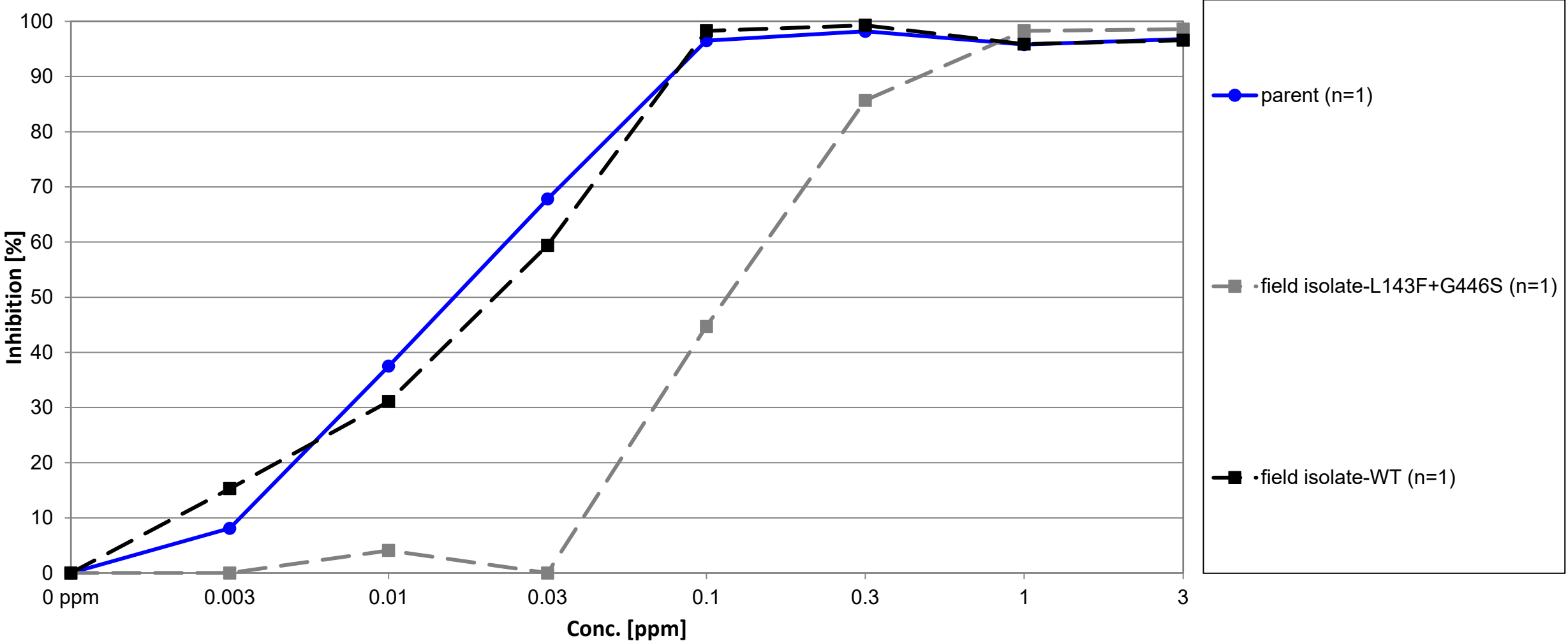
- Transformation of mutations in *A. solani* wildtype
- Transfer of gene cassette with double mutation and one single mutation (L143F or G446S, L143F+G446S) in *Alternaria solani* WT
- Gene transfer in *Alternaria solani* cells was achieved by protoplast transformation and confirmed by southern blot



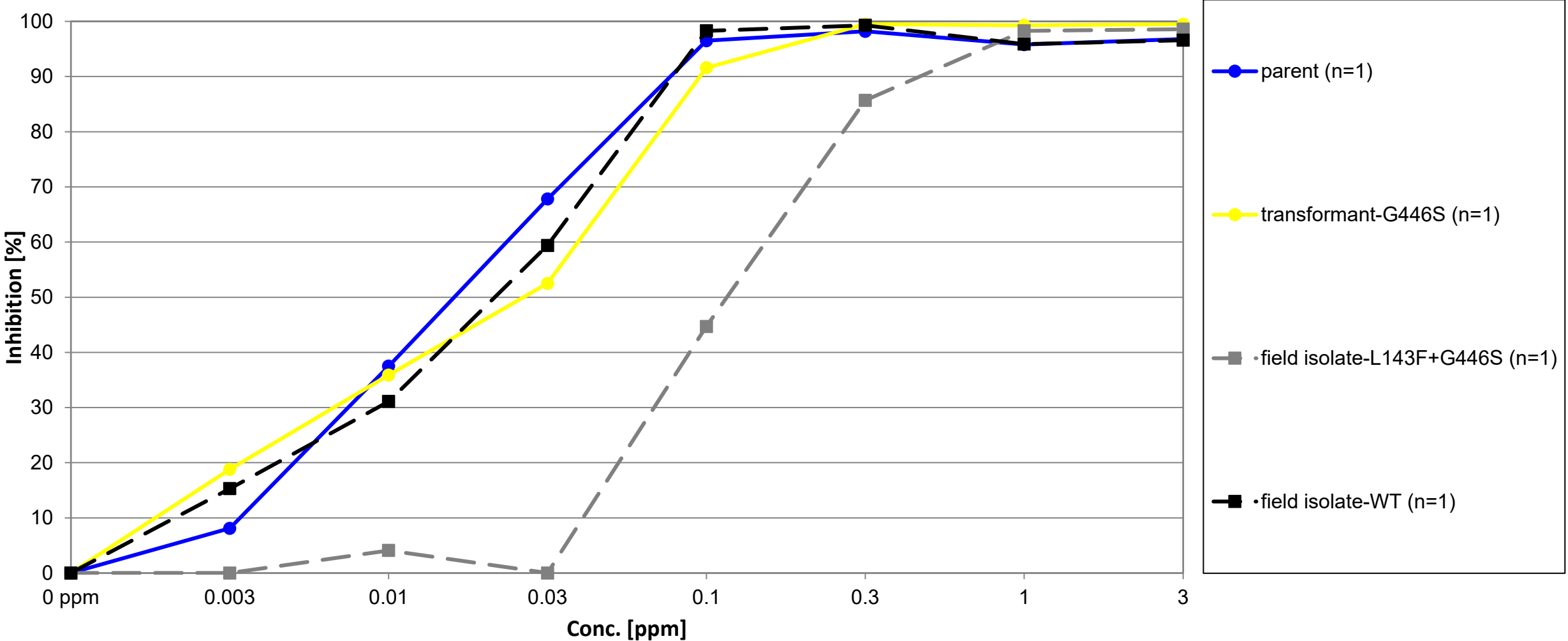
Transformed *A. solani* strains are slightly adapted towards Difenoconazole



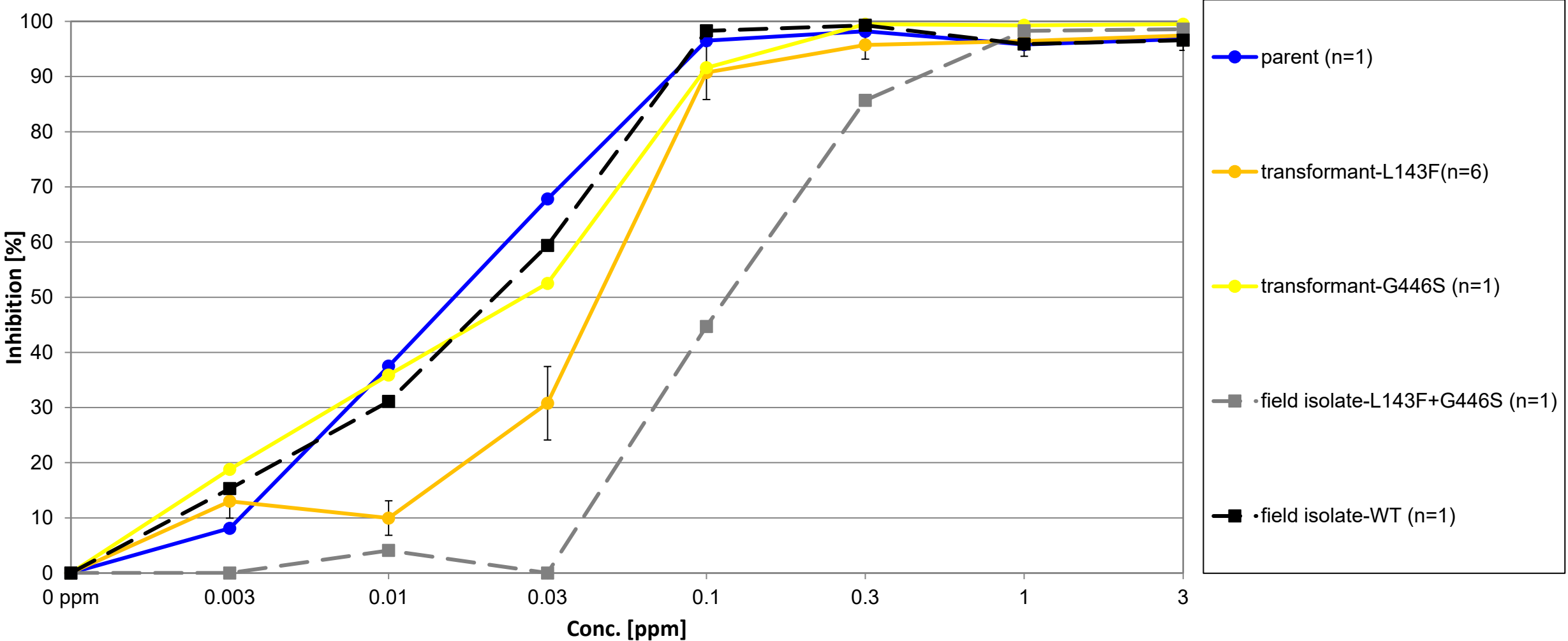
Transformed *A. solani* strains are slightly adapted towards Difenoconazole



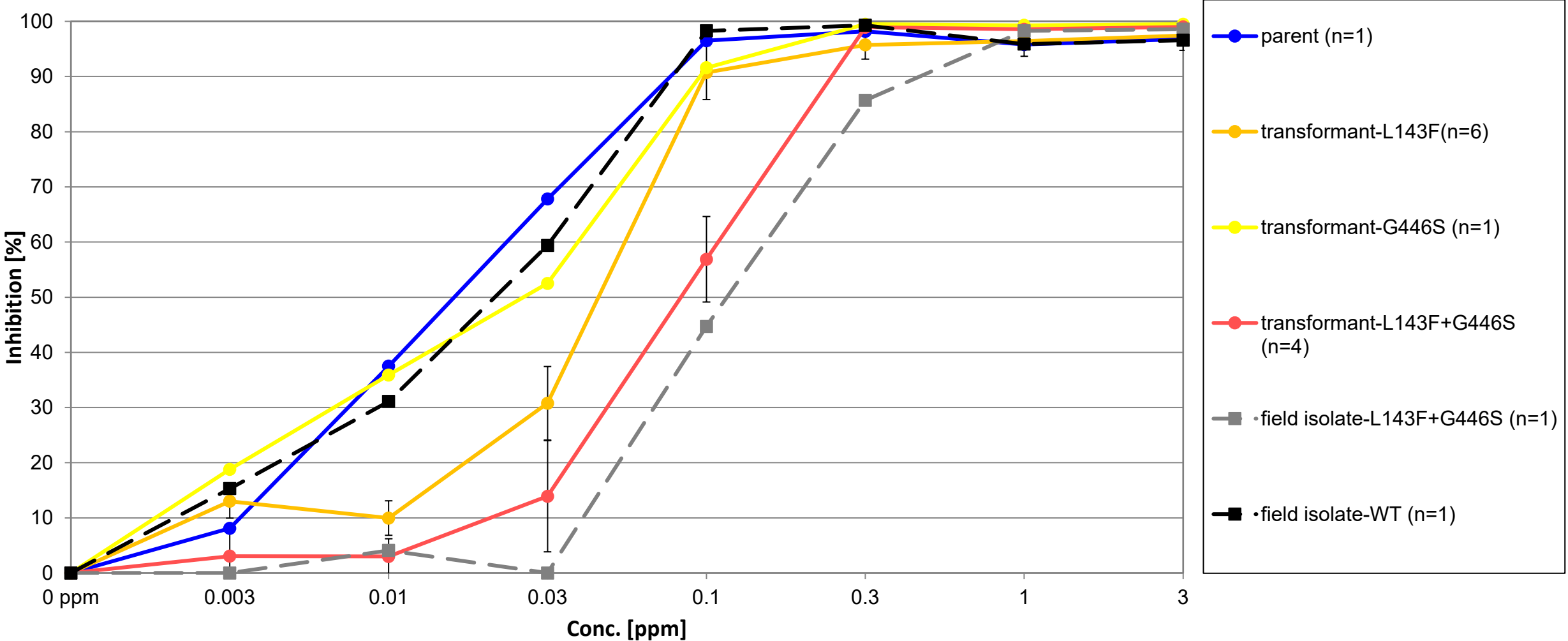
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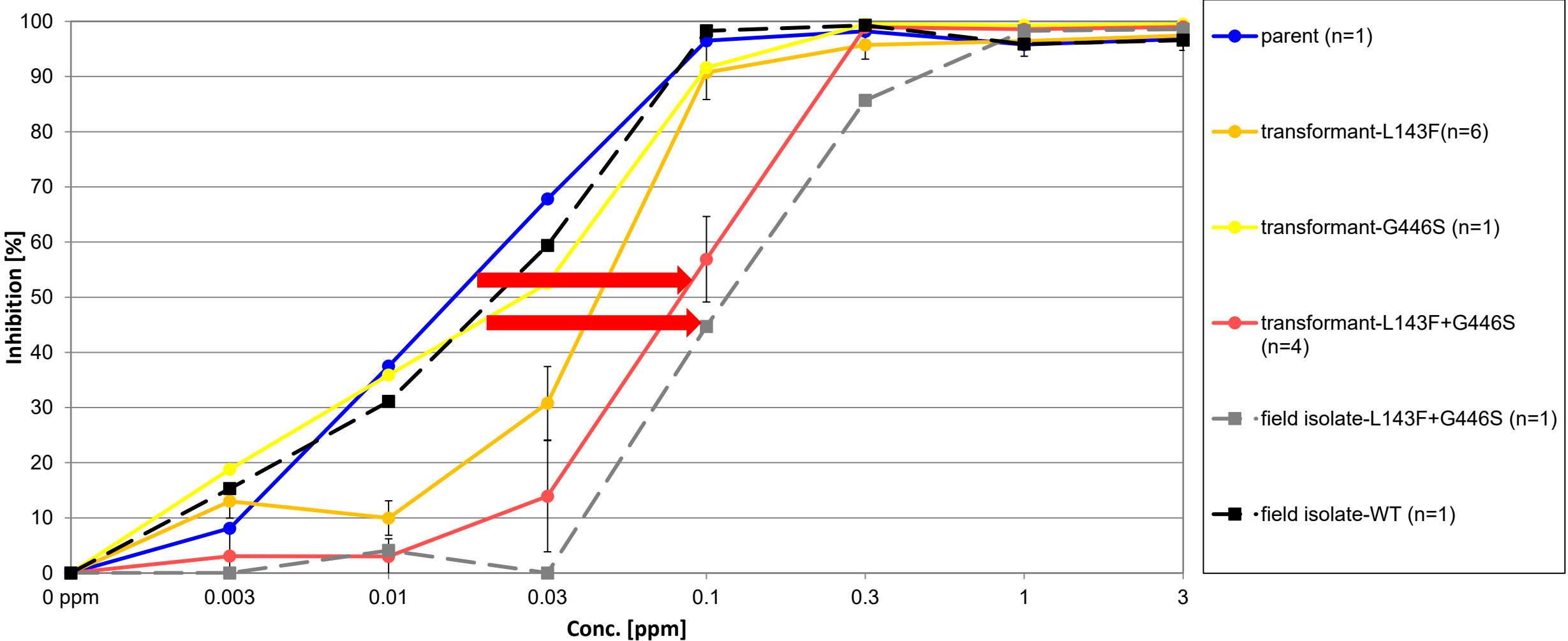
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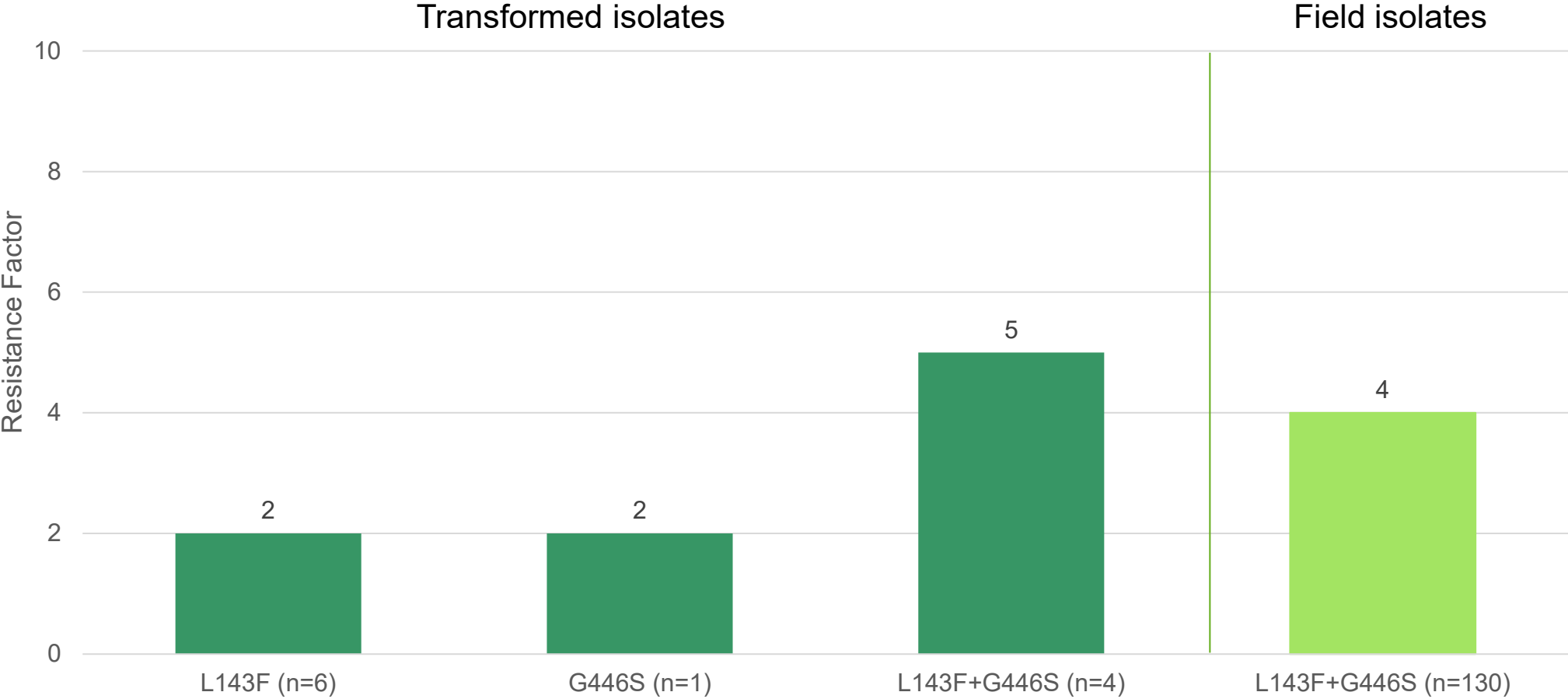
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Transformed *A. solani* strains are slightly adapted towards Difenoconazole

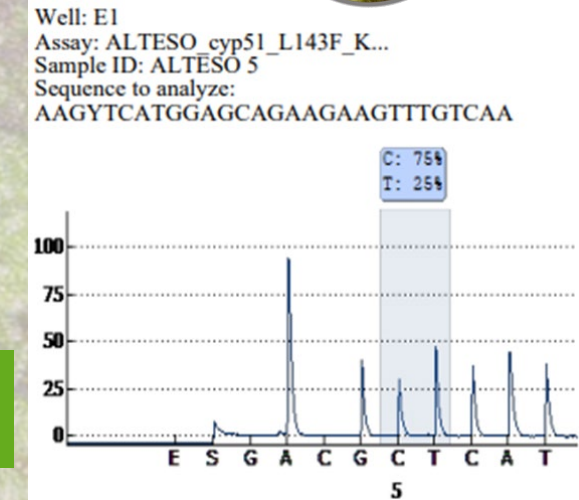
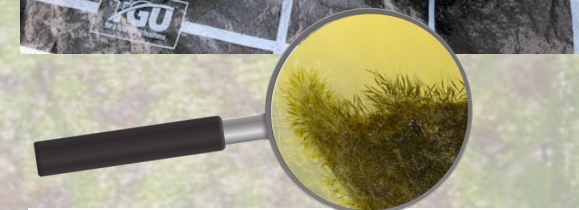
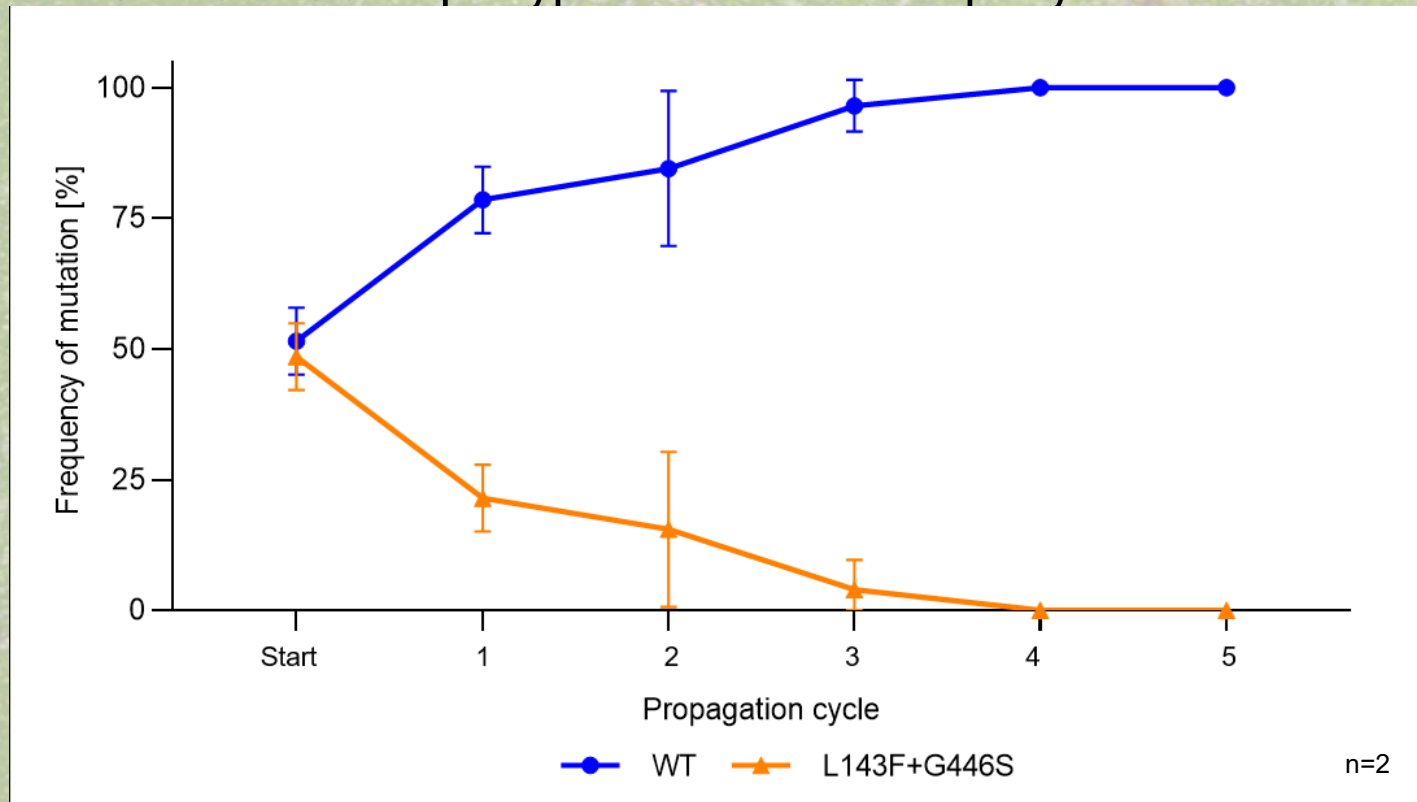


Resistance factor of transformants are in the same low range as for field isolates (Difenoconazole)



Survival of the fittest: L143F+G446S or WT?

- Inoculation of tomato plants with a mix of five L143F+G446S and five wildtypes
- After each cycle, L143F+G446S is quantitatively analysed by pyrosequencing
- L143F+G446S haplotypes decreased rapidly



⇒ Lower competitiveness (fitness) of haplotype L143F+G446S compared to wildtype

Conclusions

■ QoI

- ▶ F129L widespread, but influence on QoI adaptation is limited
- ▶ Contribution to disease and resistance management

■ SDHI

- ▶ C-H134R mutation dominates adaptation to SDHI nowadays
→ limited cross-resistance between SDHIs
- ▶ Significant part of population is still sensitive

■ DMI

- ▶ Stable field efficacy of DMIs
- ▶ Slight adapted isolates with low resistance factors found caused by target site mutations
- ▶ *CYP51* mutants are less fit in competitive experiments
- ▶ Monitoring for evaluation of sensitivity status established and running



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