Actinobacteria vs. Fusarium: Battle of the toxins

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Introduction

Fusarium head blight is a devastating disease in small grain cereals caused by the Fusarium Head Blight species complex, of which *Fusarium graminearum* is the most important. Next to deteriorated quality of the grains, **mycotoxins** are a major health concern that challenges the global food chain. In order to control Fusarium Head Blight, an integrated crop management system is needed.

In this study, we screened a collection of **Actinobacteria** for their use as biocontrol agents. After an initial screening using dual culture assays, we looked into the potential of the most promising strains for application at different stages of the Fusarium Head Blight disease cycle.

Overview of screening results

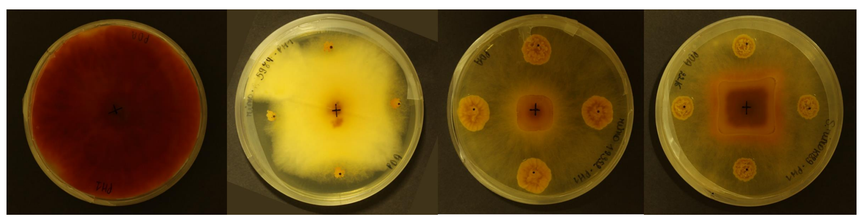
62% of 53 screened strains showed significant growth inhibition of *F. graminearum* PH1 ($p<0,05$)

2 *S. rimosus* strains caused almost complete growth inhibition and one strain showed clear growth reduction. These 3 strains were further studied.

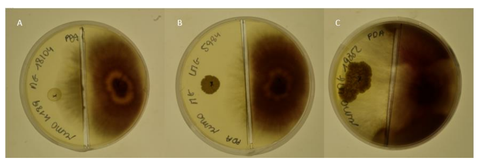
23% of biological repeats was not normally distributed and showed high variation in pigmentation, sporulation and growth of the fungus, indicating an interaction between pathogen and biocontrol agent.

Screening for growth inhibition in a dual culture assay

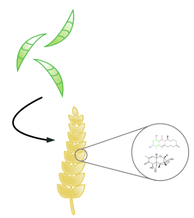
	F. graminearum PH1 w/o treatment	*F. graminearum* PH1 vs. *S. rimosus* LMG 5984	*F. graminearum* PH1 vs. *S. rimosus* LMG 19352	*F. graminearum* PH1 vs. *S. rimosus* K189
Relative growth inhibition		26 ± 10 %	71 ± 20 %	66 ± 10 %



Two out of three screened *S. rimosus* strains showed high growth inhibition of *F. graminearum* PH1, while one showed clear growth reduction. When a slot was cut out off the agar plates, this inhibition diminished, indicating the plausible use of (a) diffusible component(s)



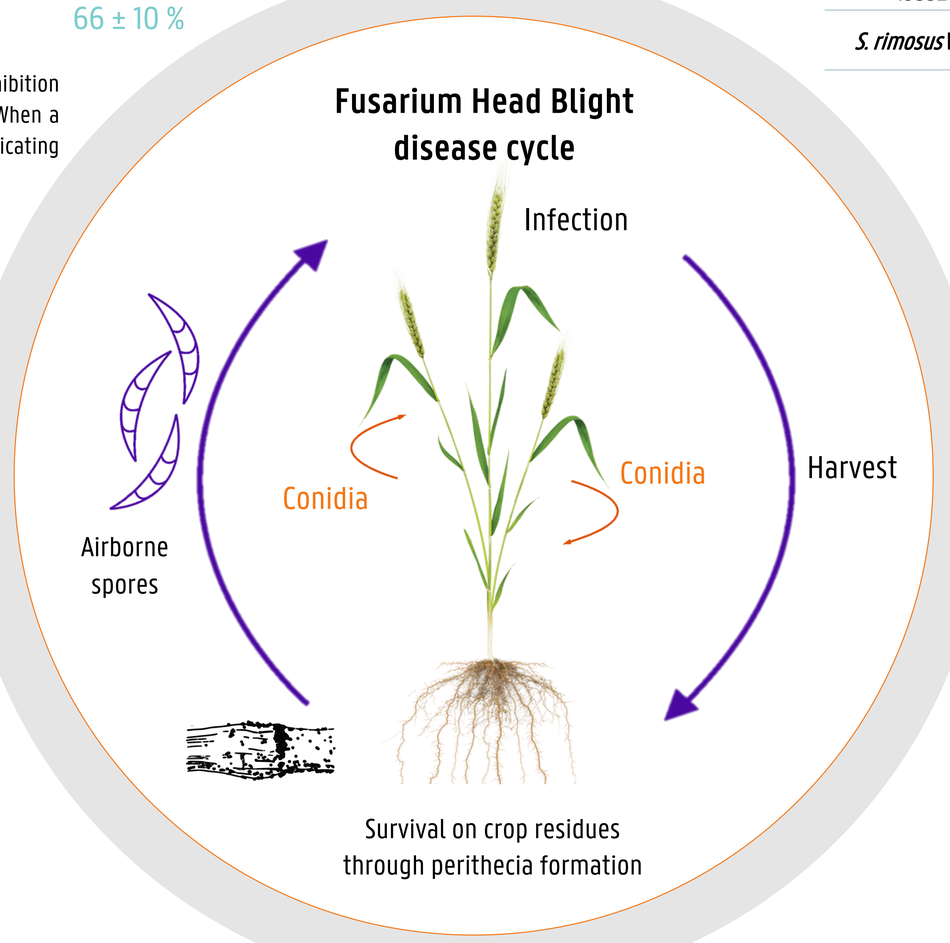
Degradation of mycotoxic virulence factors and defence mechanisms



	Zearalenone		Deoxynivalenol	
5 mg/L in LB After 72 h	Degradation (LC-MS/MS)	Detoxification (BLYES/BLYR)	Degradation (HPLC-UV)	Detoxification (*Lemna minor*)
***S. rimosus* LMG 5984**	100%	98%	0%	n.a.
***S. rimosus* LMG 19352**	100%	99%	0%	n.a.
***S. rimosus* K189**	100%	98%	0%	n.a.

All three strains were able to degrade zearalenone (ZEN), a molecule produced by *F. graminearum* as part of its defence against mycoparasites. However, the strains did not degrade deoxynivalenol (DON), an important virulence factor.

This shows that the biocontrol and loss of virulence observed in the other assays, is most likely not related to degradation of DON. The strains are however able to hijack *F. graminearum*'s defensive efforts.

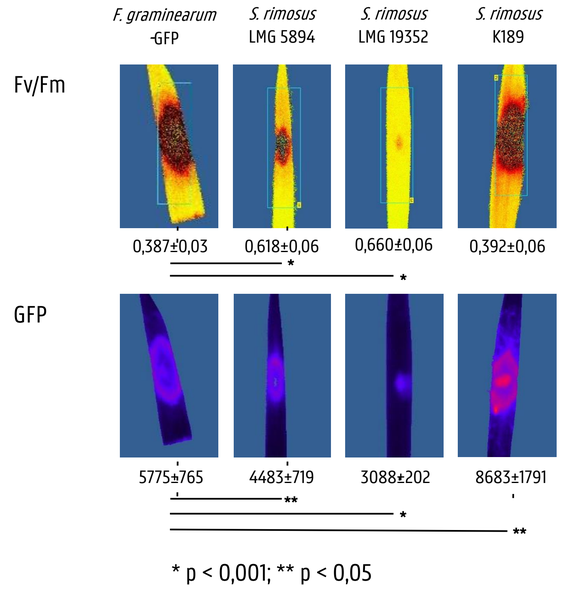


Loss of perithecia formation on wheat stubble

The application of the three *S. rimosus* strains on wheat stubble was tested.
First, the soaked and sterilized wheat stubble pieces were inoculated with *F. graminearum.* After 48h, the stubble was treated with an overnight grown and washed *S. rimosus* culture. For all three strains, we could visually see a loss of perithecia formation.

Reduced virulence and fungal growth on detached wheat leaves

The effect of the three *S. rimosus* strains on *F. graminearum* virulence was tested on detached wheat leaves. A GFP-tagged *F. graminearum* strain was used. Virulence of infection was measured through Fv/Fm and active fungal biomass was assessed as GFP fluorescence. All images were made with the PathoViewer phenotyper.



* $p < 0,001$; ** $p < 0,05$

Take-home messages

- The screened Actinobacteria were highly bioactive against *F. graminearum* PH1, with 62% causing significant growth reduction.
- Two *S. rimosus* strains show high potential as biocontrol agents at different stages of the *F. graminearum* life cycle.
- Interaction between PH1 and the biocontrol agents gave way to a variety of phenotypes (sporulation, pigmentation), showcasing an interactive battle between the two parties.

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