

# Effect of a potato bacterial pathogen on barley fungal diseases

Gravouil C.<sup>1,2</sup>, Newton A.C.<sup>1</sup>, Hein I.<sup>1</sup>, Dickinson M.J.<sup>2</sup>

<sup>1</sup>SCRI, Invergowrie, Dundee, DD2 5DA, UK. Email: clement.gravouil@scri.ac.uk

<sup>2</sup>School of Biological Sciences, University of Nottingham, Sutton Bonington Campus, Loughborough, LE12 5RD, UK.

Microbial interactions in the phyllosphere are poorly characterized but could provide a valuable approach to directly or indirectly control diseases. The aim of this work is to assess the effects of the bacterial potato pathogen *Pectobacterium atrosepticum* (Pba) on the severity of two foliar pathogens: *Rhynchosporium secalis* (Rs) and *Blumeria graminis* f. sp. *hordei* (Bgh), in the non-host crop barley.

## Material and Methods

A field trial was undertaken in 2000 where cereals were grown in a field that previously contained Pba-infected potato and natural disease severity was recorded. In a second experiment, survival of Pba on barley (infected with  $10^5$  cells/mL) was monitored with molecular techniques (qrtPCR) and via pathogen isolation. The effects of Pba on Rs and Bgh severity were validated using an *in vitro* plate and a detached leaves assay respectively.

## Results

Pba may affect disease severity of cereal pathogens in the field.

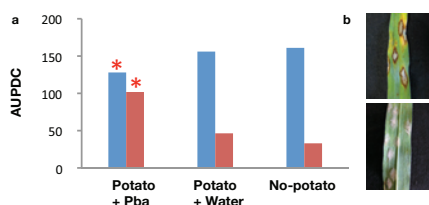


Figure 1: a) Natural disease severity of Rs on barley (blue) and *Blumeria graminis* f. sp. *tritici* on wheat (red) under different field conditions. (ANOVA test,  $P < 0.01\%$ ). b) Rs (top) and Bgh symptoms (bottom).

Pba remains viable at low levels on barley leaves.

time (dpi)	1	3	5
Method enrichment	+	+	+
Method molecular quantification (ng Pba DNA)	0.53	0.045	1.25

Figure 2: Detecting Pba on barley leaves. Isolated Pba was identified using selective medium and PCR approaches. Molecular quantification was based on qrtPCR.

Pba increases mildew susceptibility.

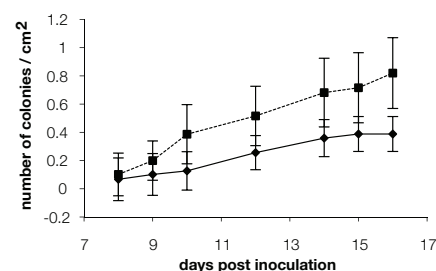


Figure 3: Number of mildew colonies per cm<sup>2</sup> of infected leaves formed on Pba-treated (dash) or control (plain) barley. Standard error are shown.

Pba inhibits Rs sporulation and growth.

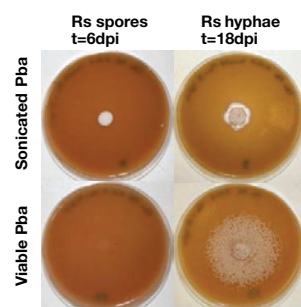


Figure 4: *in vitro* assessment of Rs spores germination ( $10^5$  spores/mL) and Rs hyphal growth in contact with viable or sonicated Pba.

## Conclusions

The presence of Pba in the field was correlated with fewer Rs symptoms on barley but more mildew on wheat. Pba remains viable on a non-host leaf surface. The survival strategies of Pba on non-host crop remain elusive. This work validates the direct interaction between Pba and Rs by inducing spore lysis and inhibiting hyphal growth. Furthermore, Pba treated barley is more susceptible to Bgh.

## Future work

- Utilise Pba pathogenicity mutants to identify the mode of action.

Pathogenicity mutants	Gene	Description
	ehpF	Phenazine antibiotic mutant
	expl	Quorum sensing mutant
	outD	Type 2 secretion mutant
	hrcC	Type 3 secretion mutant
	cta6	Coronafac acid mutant

- Test other enterobacteria and leaf-associated bacteria to determine if the observed interactions are Pba-specific.

- *In vitro* and *in planta* microscopic observations to localize and visualize Pba interference with Rs sporulation and germination.

## Acknowledgement

We would like to acknowledge Dr Ian Toth and his team from the SCRI for providing the Pba strain and helping with the Pba assay. This work is funded by the Scottish Crop Research Institute and the University of Nottingham.