# Can *Brachypodium distachyon* provide insight into FHB?

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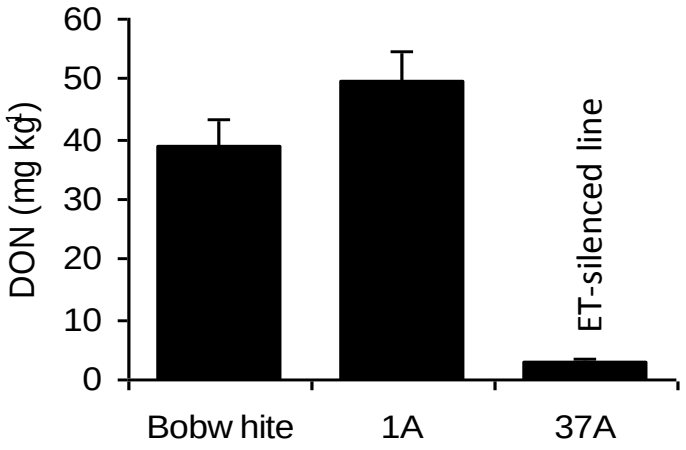
# Genetics and mechanisms of FHB resistance in wheat

- Only partial resistance against FHB exists in wheat.
- Many QTL identified but no gene cloned to date .

Ethylene perception/signalling enhances susceptibility of *Arabidopsis* and wheat to *Fusarium graminearum*

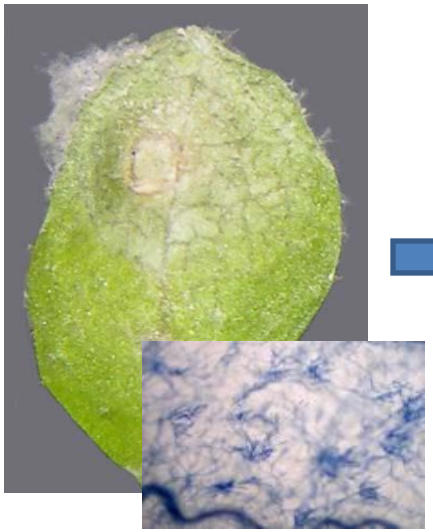
Inhibiting ET perception increases resistance to foliar and floral infection of Arabidopsis


Inhibiting ET signal transduction reduces DON accumulation in wheat grain




(Chen *et al.* 2009 *New Phytologist)*

# Model-crop pathology: Using Arabidopsis to gain insight into *Fusarium graminearum* diseases



We can infect Arabidopsis with *Fusarium*

But:

Are the same pathways/genes always important for resistance in wheat?

When genes controlling resistance in Arabidopsis are identified, how can this inform on identification of orthologous genes in wheat?

Is Arabidopsis the best/most appropriate model?

For example: max. reported DON accumulation is very low (approx. 3 mg/kg)

# *Brachypodium distachyon* (Bd)

*Brachypodium distachyon*.
David Garvin,
University of Minnesota.

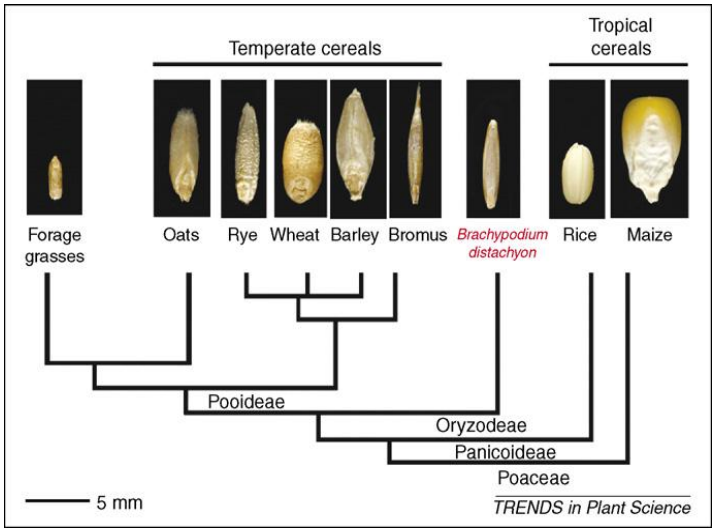
Bd has many qualities that make it a good model for functional genomic studies in temperate cereals.

- Small genome (~272 Mbp).
- Fully sequenced genome.
- Diploid (also tetraploid and hexaploid accessions).
- Small physical stature.
- Self-fertile.
- Short lifecycle
- Simple growth requirements.

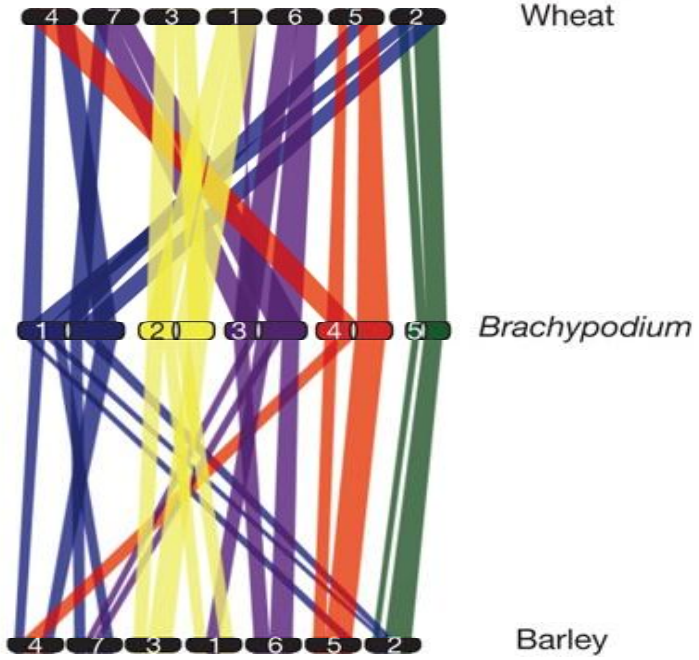
# An attractive model for cereal functional genetics

Genetic advantages:

- Wheat: large hexaploid genome (~ 17,000 Mbp)  Bd: small diploid genome (272 Mbp)
- Synteny between genomes of grass species enables direct translation from a gene in *Brachypodium* to a candidate gene in cereals.
- Mutant populations available: EMS, fast neutron, T-DNA insertion




(Opanowicz *et al.*, 2008)




(The Int. Brachy. Init., 2010. *Nature*)

# Components of resistance to FHB

Type I resistance

Resistance to initial penetration of the pathogen within host tissue.

Can be studied using spray inoculation

Type II resistance

Resistance to spread of the fungus within adjacent host tissues.

Can be studied using point inoculation

Barley has an inherent type II resistance to *Fusarium*

# Characterisation of the *Fusarium-Brachypodium* pathosystem

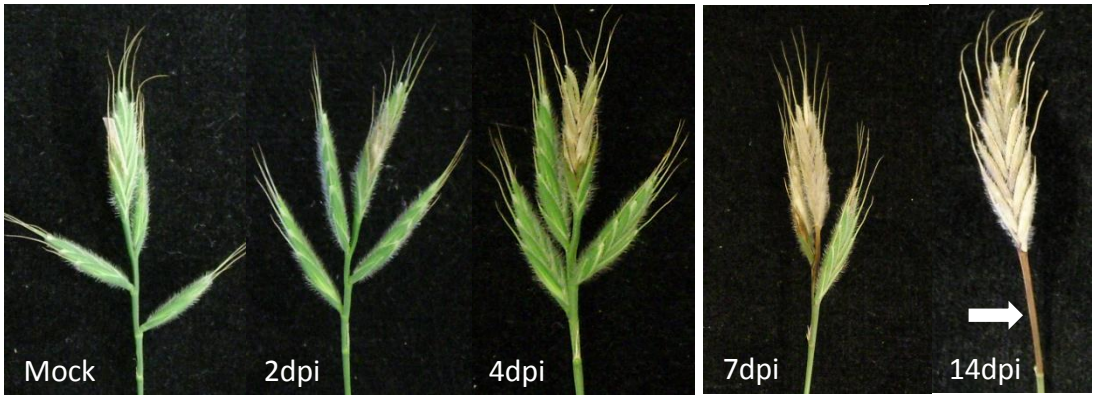

Bd21 flowers sprayed with Fg conidia 3 dpi.

Bd plants sprayed with *Fg* or *Fc* conidia display symptoms which resemble closely FHB on wheat:

Necrotic lesions appear from about 3 days post inoculation (dpi) and are rapidly surrounded by a chlorotic area.

High DON accumulation (>1,500 mg/kg)

Spray inoculation
Assess type I + II resistance

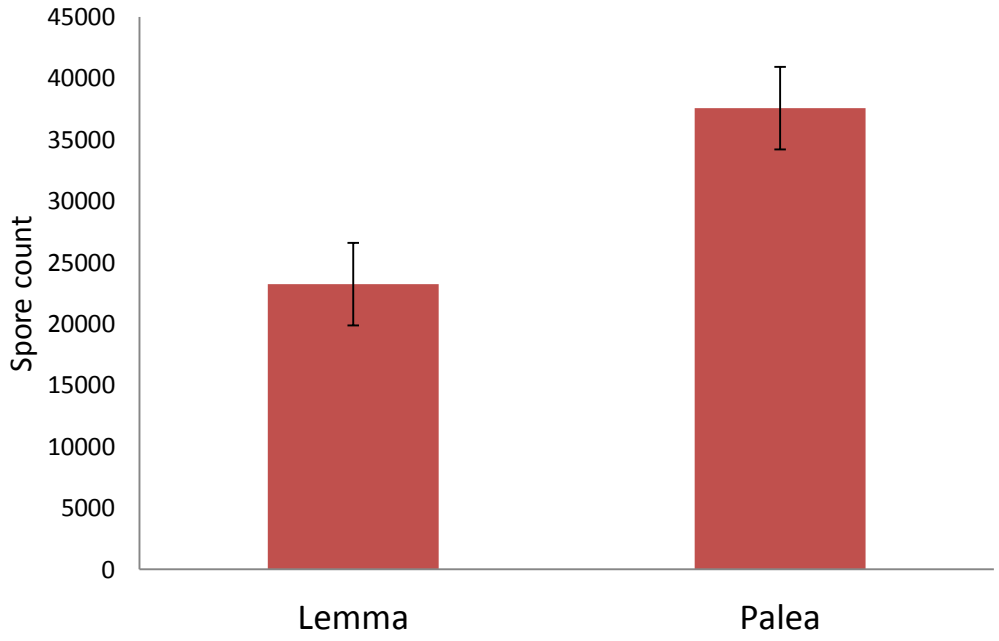



Point inoculation
Assess type II resistance

Compatibility of interaction with predominant *Fusarium* spp.

(**Peraldi** *et al*. 2011 *BMC Plant Biology*)

# Differential susceptibility of *Brachypodium* floral tissues to *Fusarium* infection



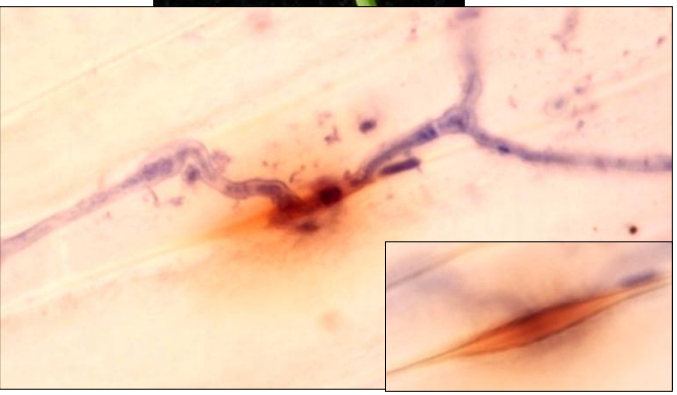

Bd21 Palea

Bd21 Lemma

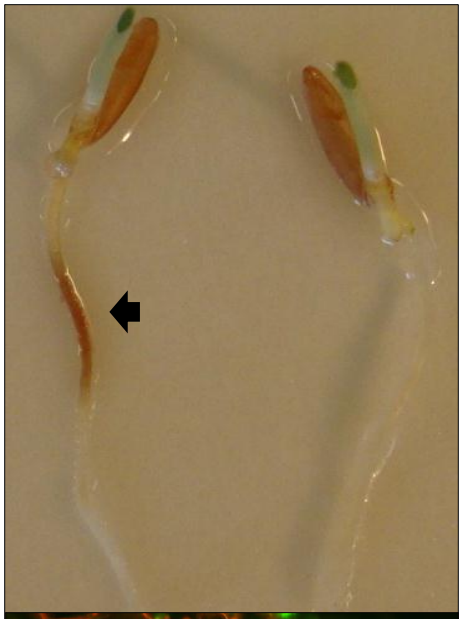
Similar findings reported in barley. (**Lewandowski** *et al* (2006) *Phytopathology*)

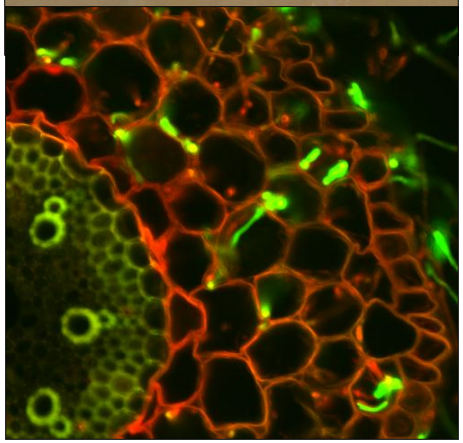
# Prospects for modelling other *Fusarium* diseases

Stem/leaf sheath infection
-> Crown rot



Root infection
-> Root rot

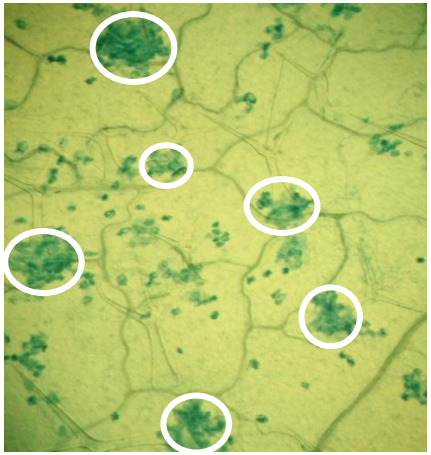


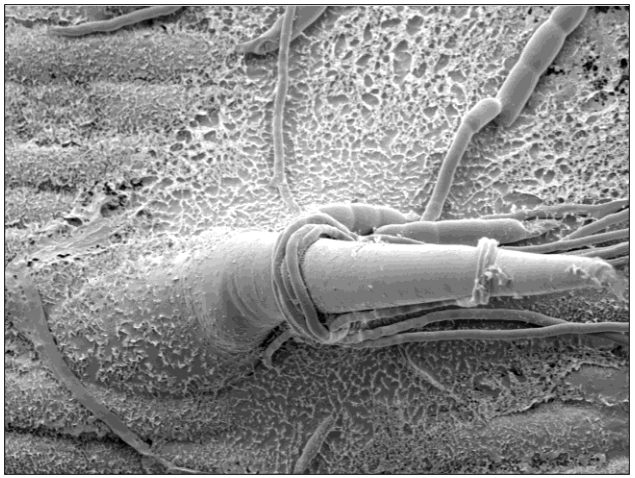


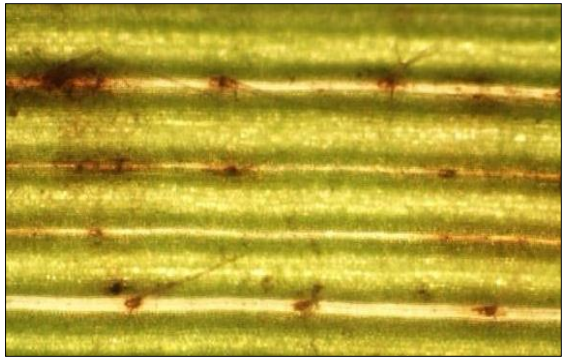
Leaf infection

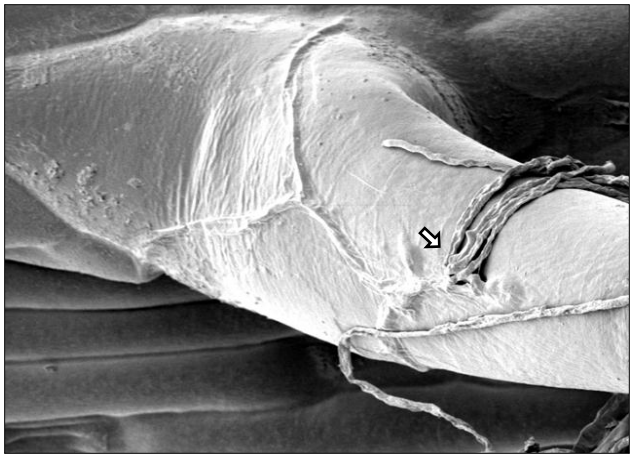
# Trichomes as sites of Fusarium infection

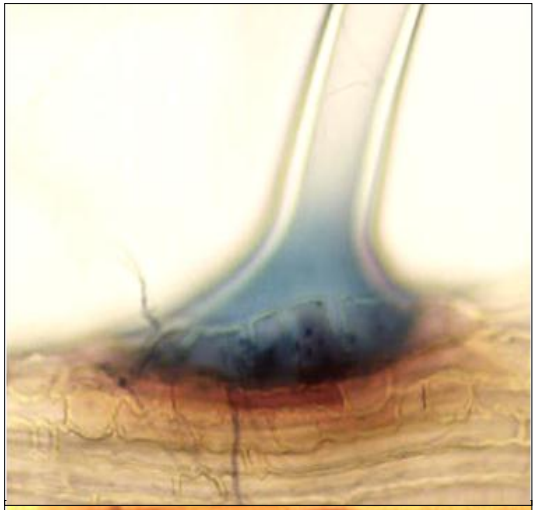
Arabidopsis trichome cell death

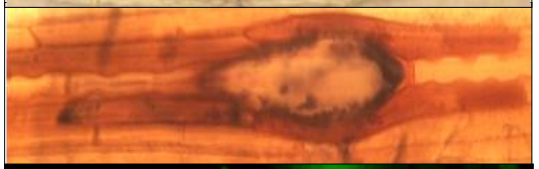


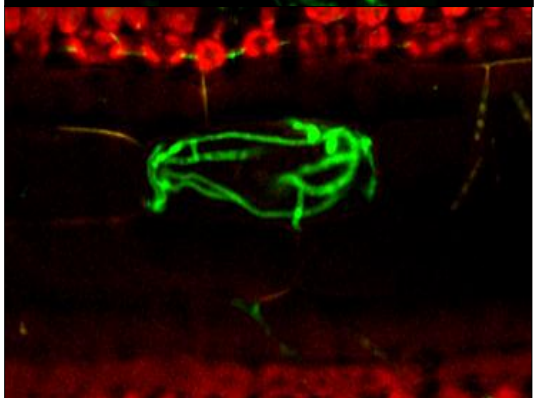




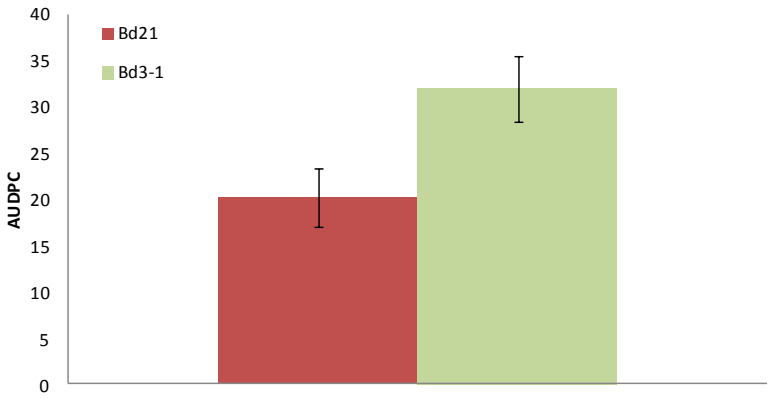




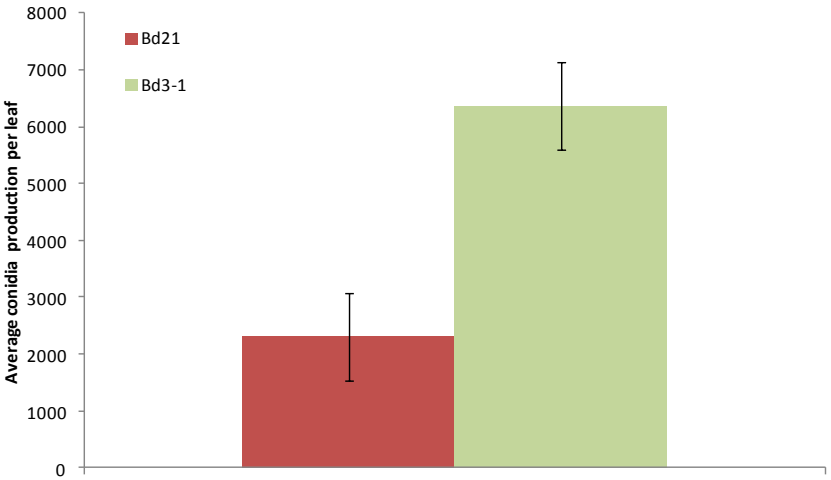




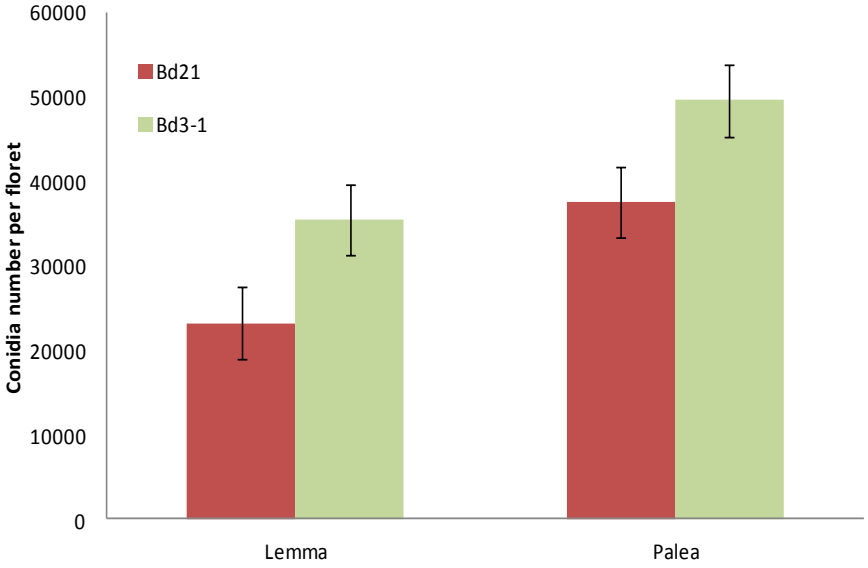
# Differential susceptibility of Bd accessions

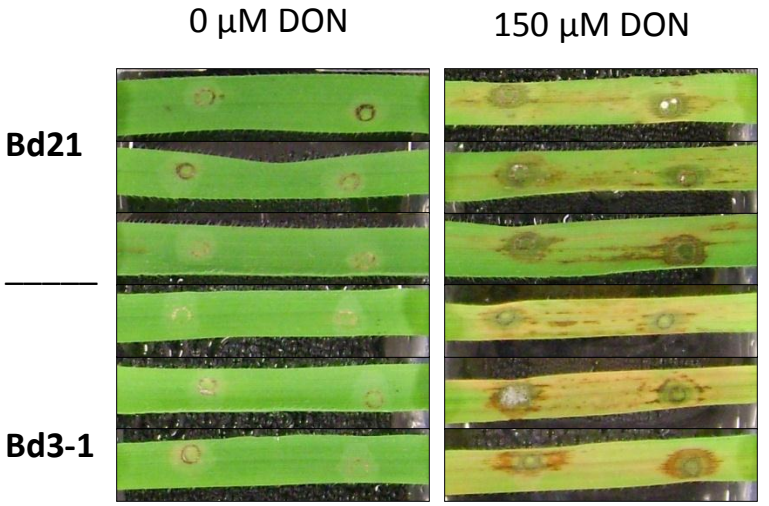



Disease (AUDPC) of Bd21 and Bd3-1 flowers following point inoculation



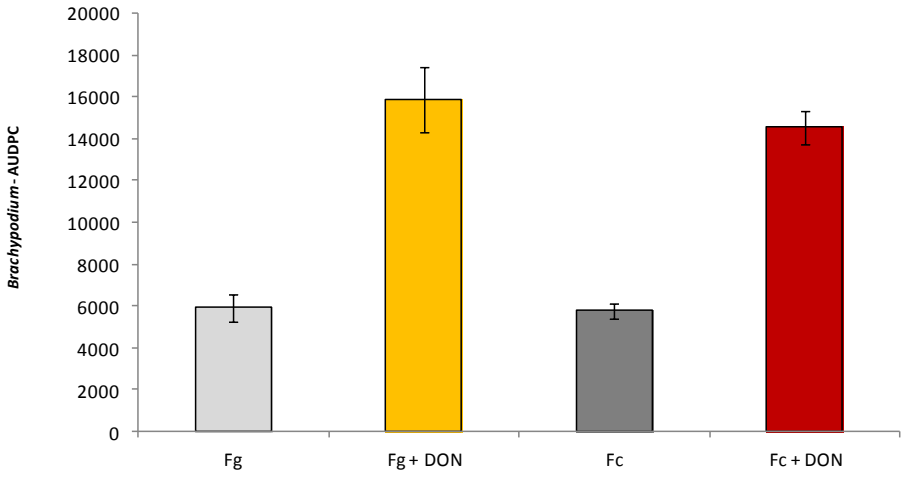

Conidial production on detached leaves of Bd21 and Bd3-1



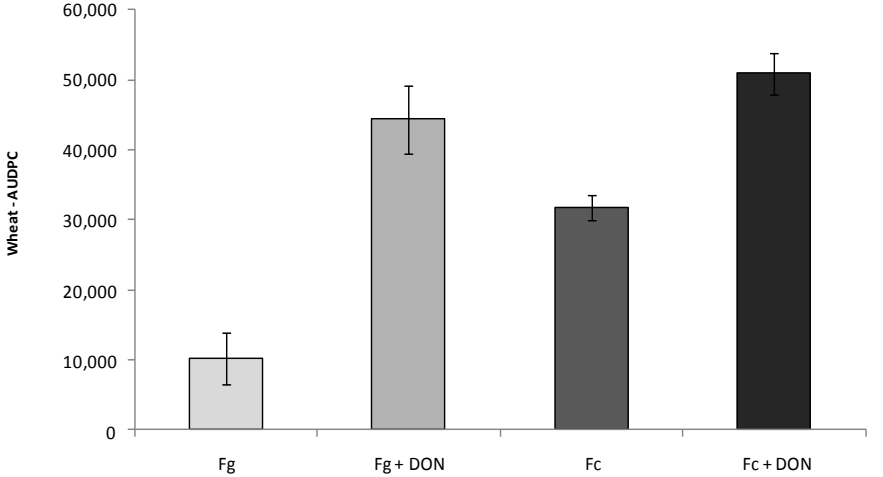



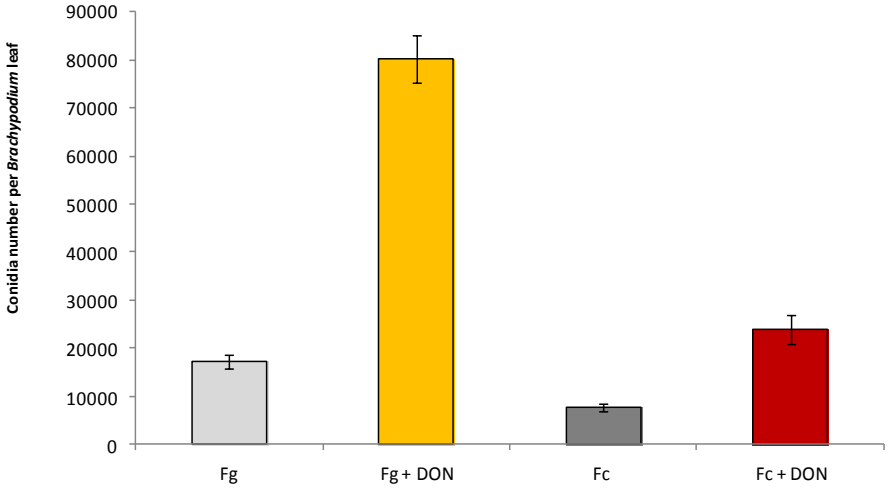
# Influence of DON on disease progress and conidial production

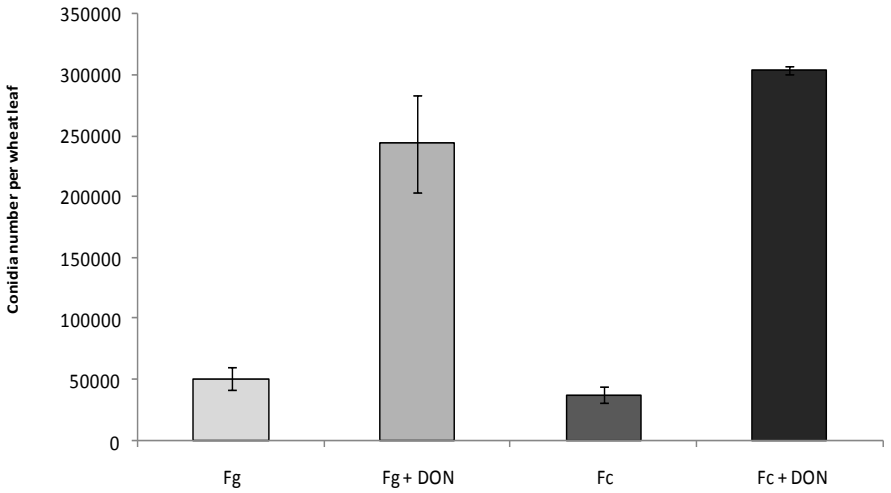
## Brachypodium




## Wheat



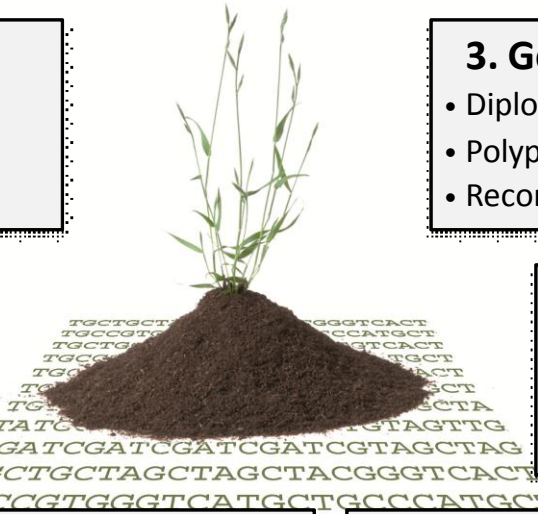



# Bd: Functional genomics resources

**1. Plant attributes**
- Small plant stature
- Simple growth requirements
- Rapid life cycle

**2. Genome sequence**
- Diploid inbred line Bd21 (~272 MB)
- Bd21-3, Bd1-1, Bd3-1, Koz-3, BdTR12C, Bd30-1 (2011) and ~50 accessions (2011+)

**9. Other resources**
- EST, cDNA, BAC libraries
- Molecular & cytogenetic markers
- Microarrays …




**3. Germplasm collections**
- Diploid (2n=10)
- Polyploid / hybrids (4n, 6n)
- Recombinant inbred lines (RILs)

**8. Online resources**
- Brachypodium.org
- Genomics / bioinformatics
- Functional genomics

**4. Mutant collections**
- T-DNA: BrachyTAG, WRRC
- Sodium Azide: BrachyTIL
- EMS: BTI

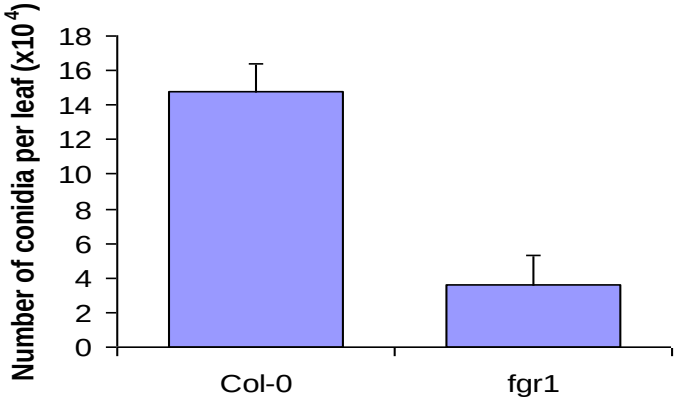
**7. Genome synteny**
- Temperate cereals
- Biomass grasses
- Grassland species

**6. Biological traits**
- Pathogen resistance
- Drought tolerance
- Cell wall…

**5. Genetic transformation**
- Overexpression, RNAi
- High-throughput transformation
- Complementation

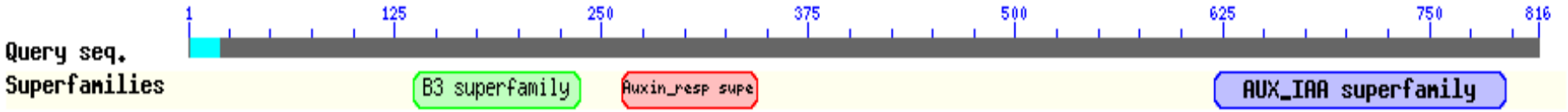
# Identification of a new Fusarium resistance

- *fgr1-1* confers resistance to *Fusarium* infection
- *fgr1-1* is an allele of auxin response factor 2 (*ARF2*). Enhances leaf chlorophyll content and delays senescence


## Orthologue of At *ARF2* (Bradi4g07470) BdAA724

Predicted protein domains:

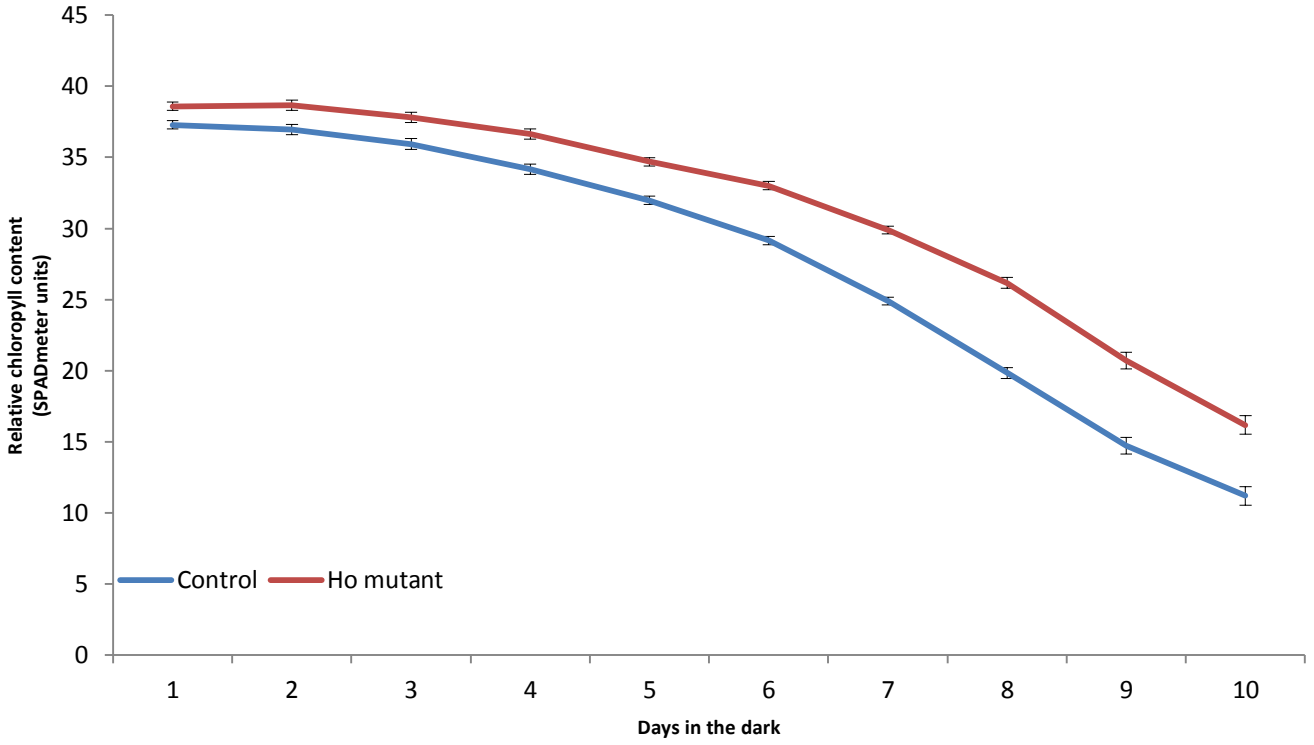



EnsemblPlant orthologue prediction:

| Species | identifier | Target % identity | Query % identity | Reference |
|---|---|---|---|---|
| *A. thaliana* | AT5g62000 | 46 | 49 | ARF2 |
| *O. sativa* | Os12g29520 | 71 | 73 | Novel Ensembl prediction |

# Orthologue of At *ARF2* (Bradi4g07470)

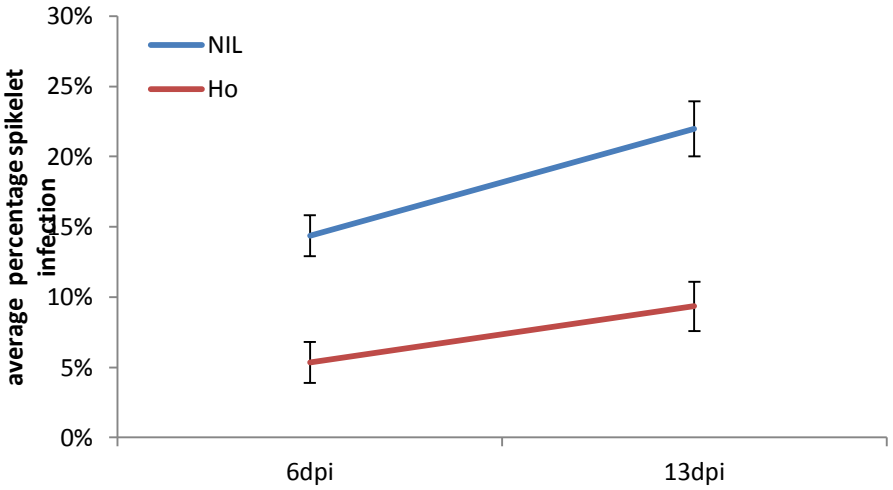
The Bradi4g07470 predicted protein shares high sequence homology with *Arabidopsis* (AtARF2, 49%) and rice (OsARF24, 75%). Found in T-DNA collection (Dr P. Vain).



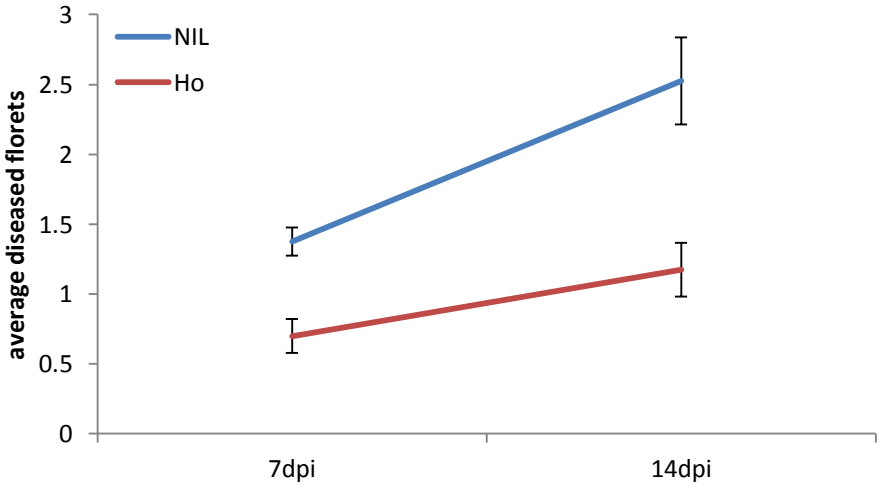

*Bdarf2* exhibits delayed senescence (from dark-induced senescence test)

# Bradi4g07470 contributes to susceptibility to FHB infection

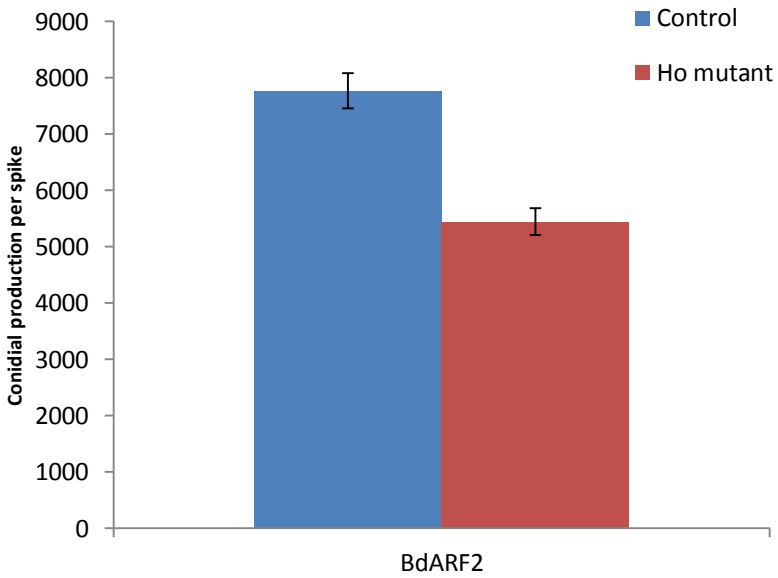
Spray inoculation (type 1 & 2 resistance)




Point inoculation (type 2 resistance)



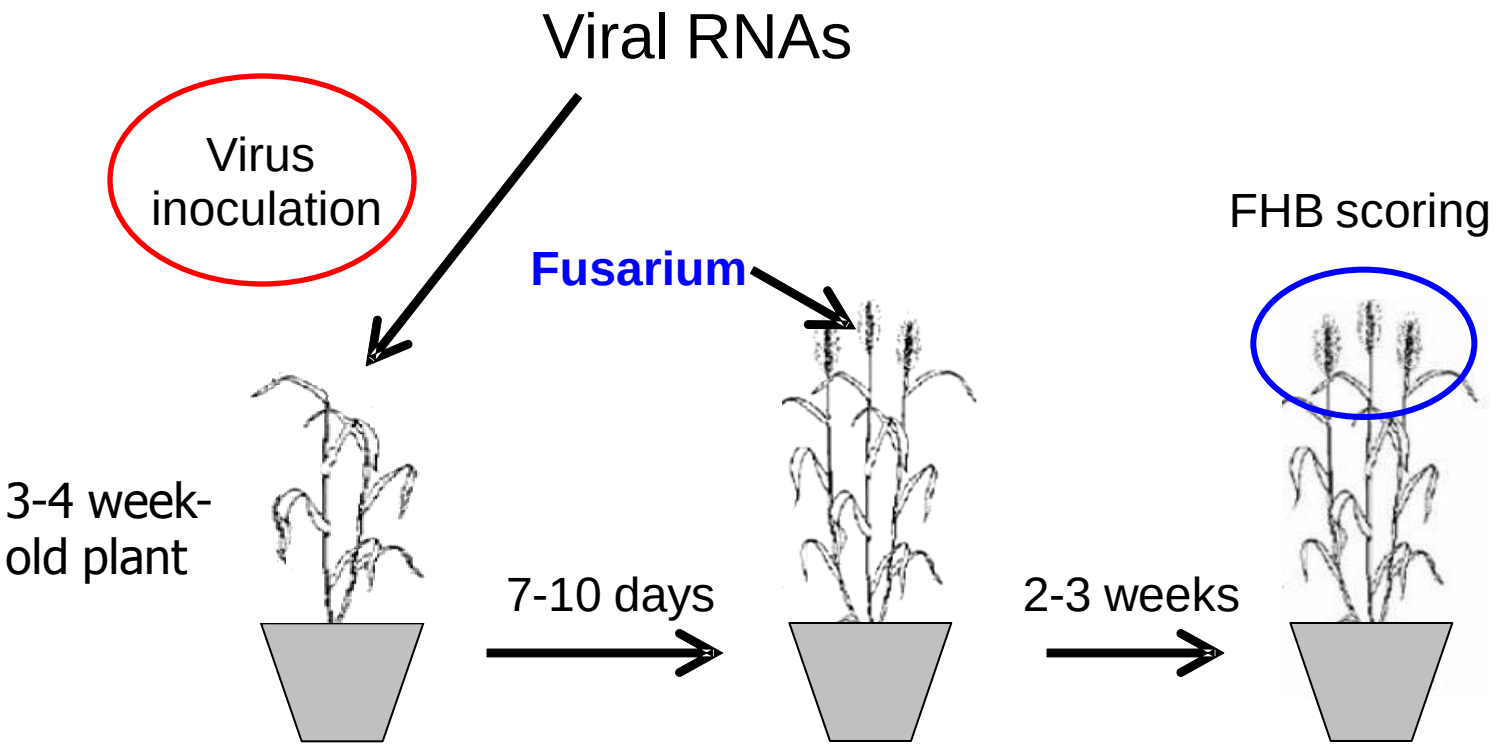

*Bdarf2* reduces Fg conidial production (from spray inoculation test)



# Translation to wheat

Virus-induced gene silencing (VIGS) targeting *ARF2* in wheat:

(Wanxin Chen & Patrick Schweizer, unpublished)

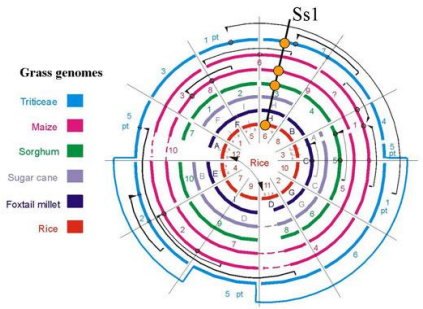



*TaARF2* silencing resulted in about 20% reduction in disease severity

# Summary

## Brachypodium: a model species for direct translation to wheat

- *Brachypodium* potentially provides an excellent pathosystem for a number of important cereal diseases.
- We (and others) have demonstrated the potential of Bd for model-to-crop translation for FHB and other *Fusarium* diseases.
- The availability of functional genomics resources has allowed the characterisation of a first gene candidate, (*ARF2*).


We can infect Brachypodium with *Fusarium*

John Innes Centre

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Movember team photo

…and thank you for your attention.


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