

# Recent advances in understanding PCN decline and their use in developing novel control strategies

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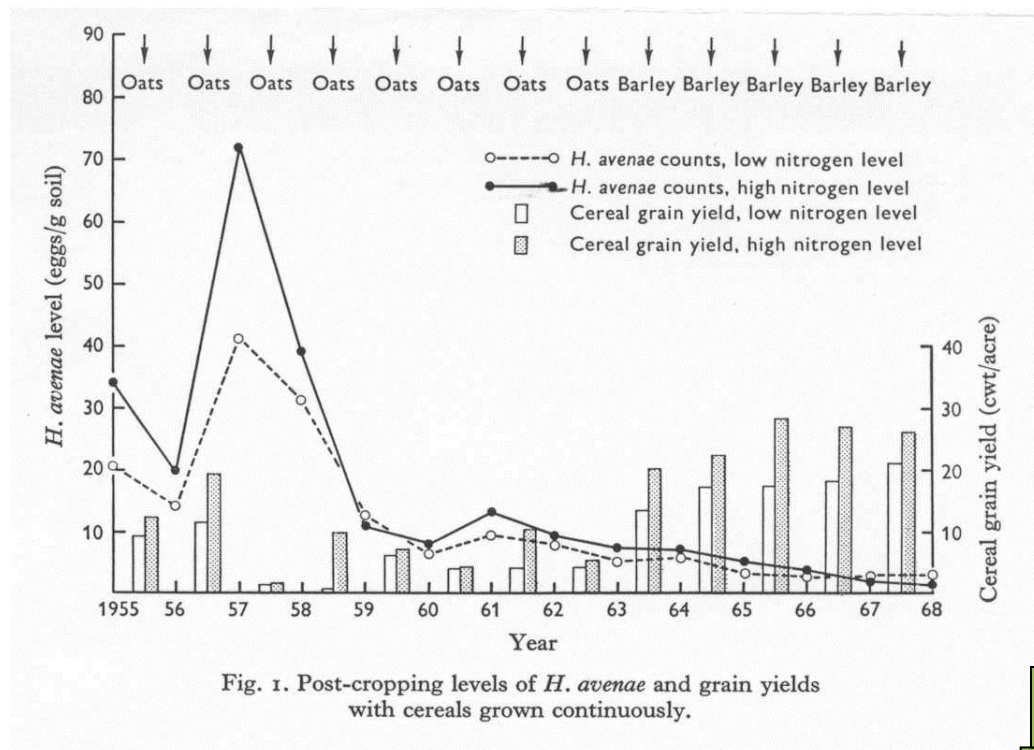


ROTHAMSTED  
RESEARCH

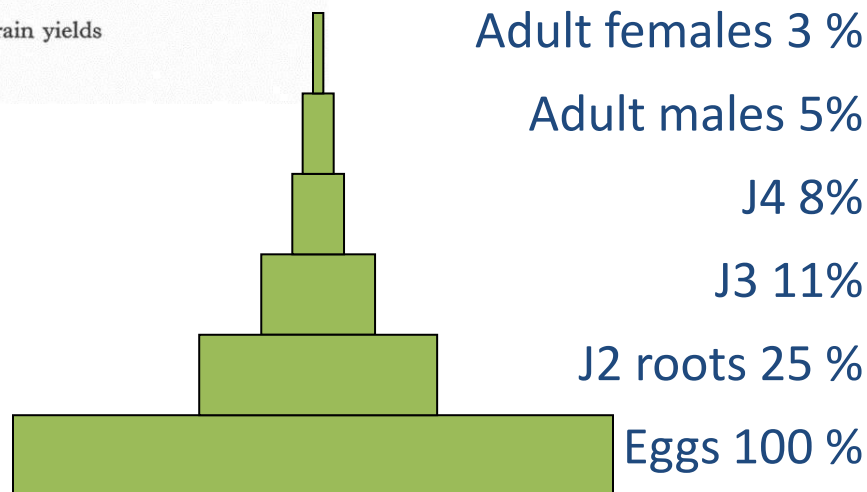
Rothamsted Research, Harpenden, Hertfordshire, AL5 2JQ



# Biological Control of Plant – parasitic Nematodes



(Gair *et al.*, 1969)



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# Biological control organisms

## 1) Facultative parasites

*Pochonia chlamydosporia*

*Paecilomyces lilacinus*

*Monographella cucumerina*

*Cylindrocarpon destructans*

*Arthrobotrys oligospora*

## 2) Obligate parasites

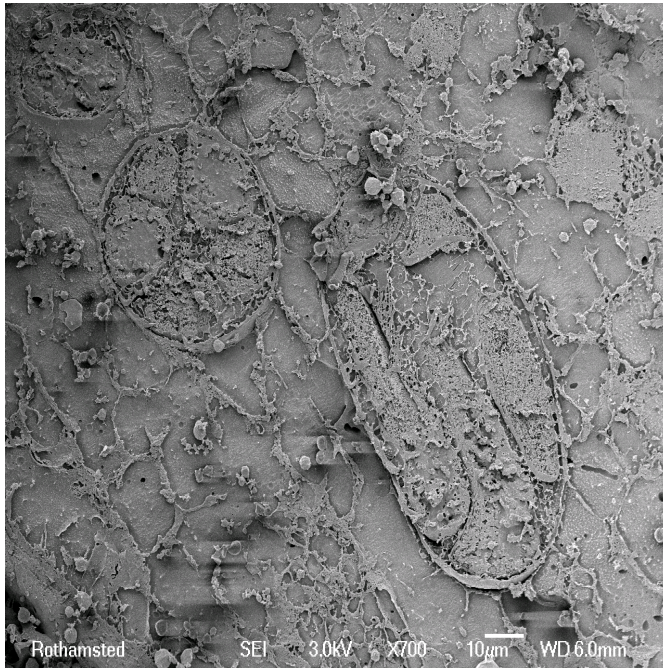
*Pasteuria penetrans*

*Nematothrora gynophila*

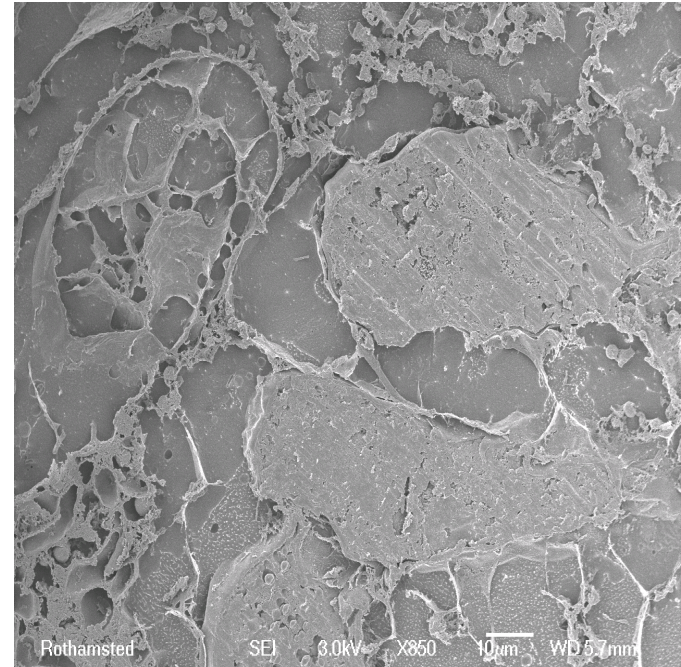
*Hirsutella rhossiliensis*



# Measurement of colonisation of cysts by egg parasitic fungi using cryo-scanning electron microscopy at 8 weeks



Control



Fungal treated



## PCN decline key objectives

- Objective 1.** To identify key factors that affect PCN suppression and decline rates
- Objective 2.** To develop new protocols for measuring decline rates in field soils
- Objective 3.** To investigate the influence of edaphic factors and cropping regimes
- Objective 4.** Assess effects of fungicides & herbicides on the associated microbes
- Objective 5.** Investigate the biotic and abiotic factors that influence fungal growth
- Objective 6.** Investigate potential formulation and delivery of fungal agents
- Objective 7.** Investigate effects of carbon and nitrogen acquisition on fungi
- Objective 8.** Assess the importance of fungal parasites in the patch dynamics of PCN
- Objective 9.** Assess the effects of different nematicides formulations





## PCN decline key objectives

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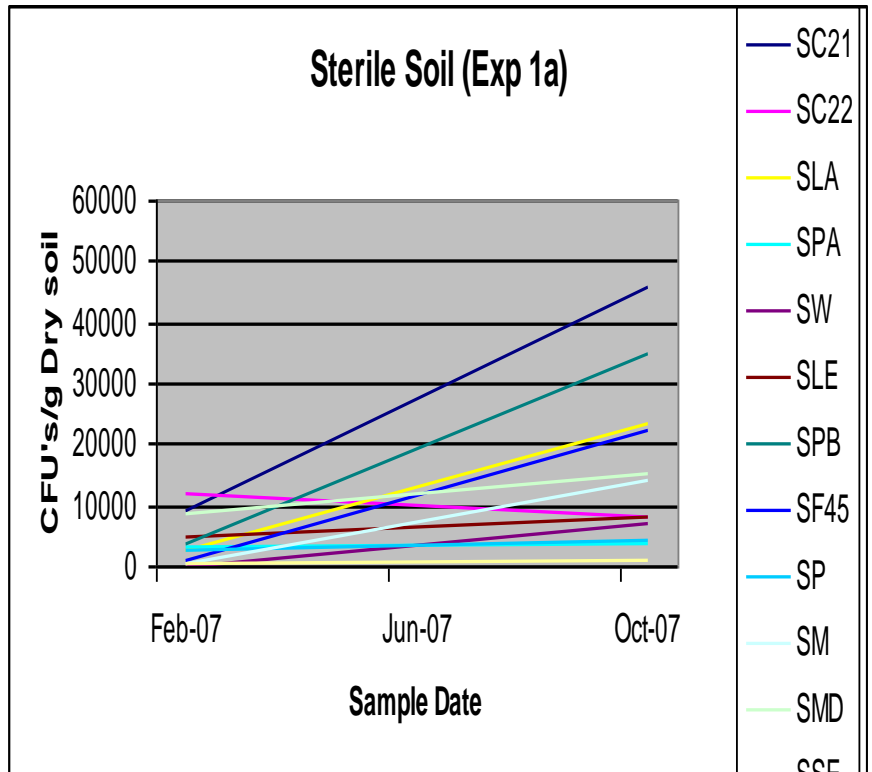
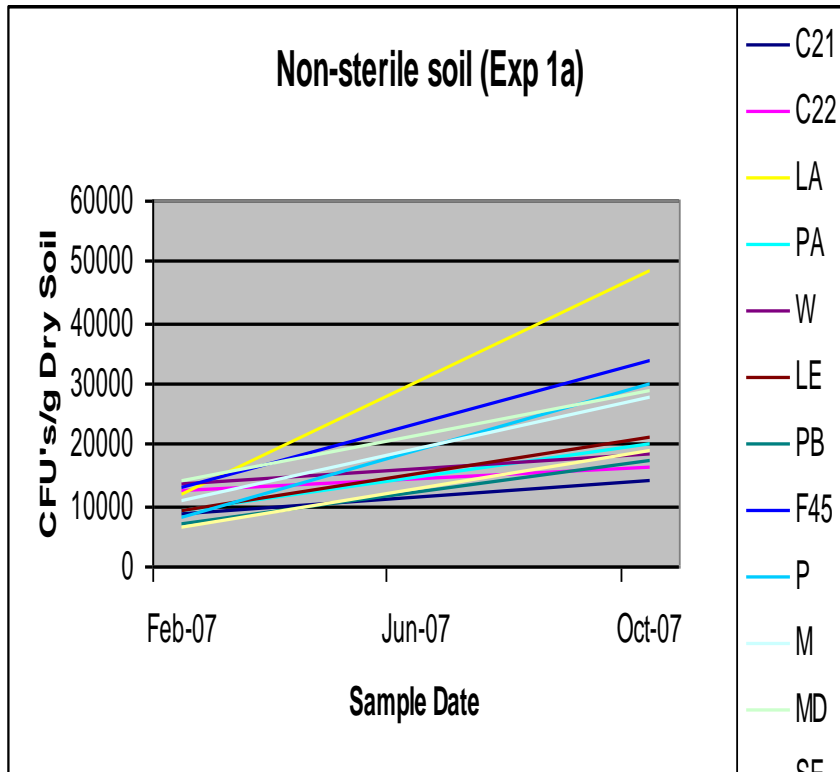


## Objective 1: Assess the rates of decline in a range of soils of different textures and cropping histories

- Ten different soil types
- Samples divided into two one sub-sample sterilised
- PCN cysts placed in soil in mesh bags
- Sampling beginning and of each season over 3 years
- Egg counts done



## Objective 1: Assess the rates of decline in a range of soils of different textures and cropping histories

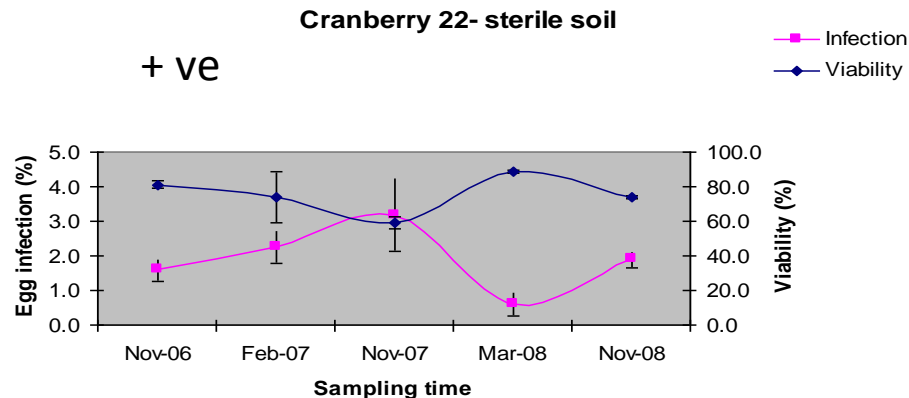
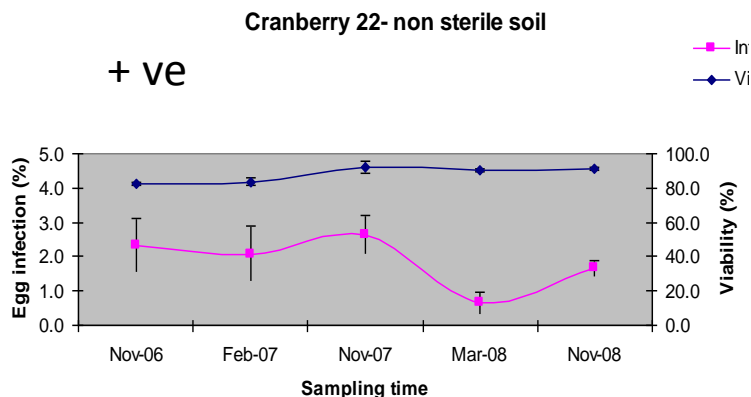
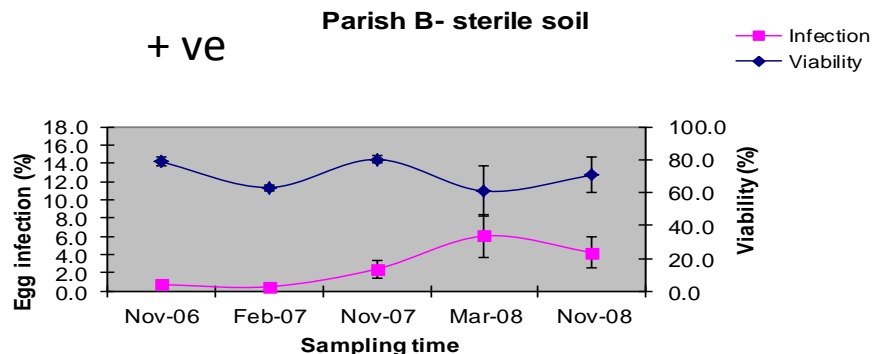
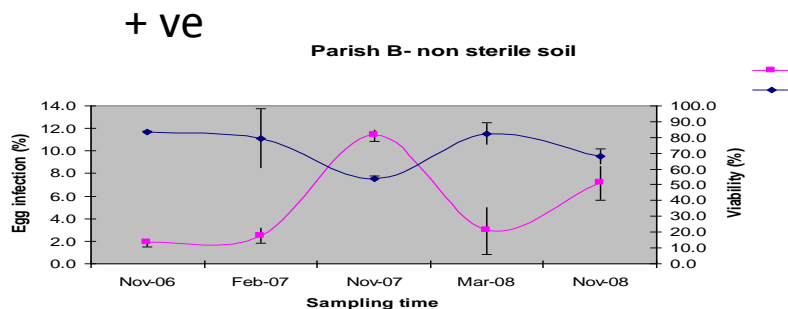


Does sterilization have an effect on fungal communities?

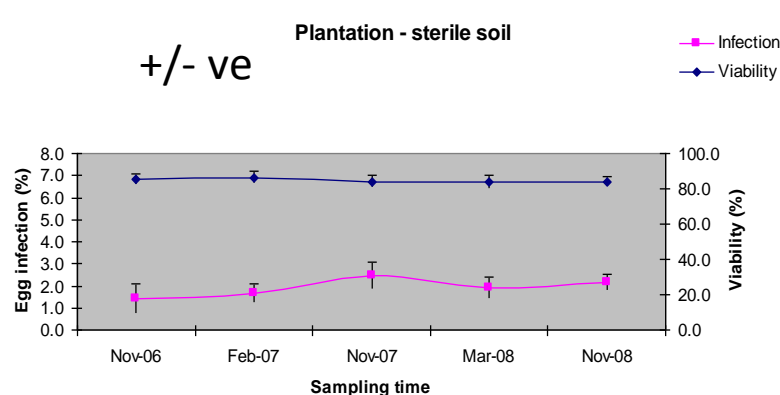
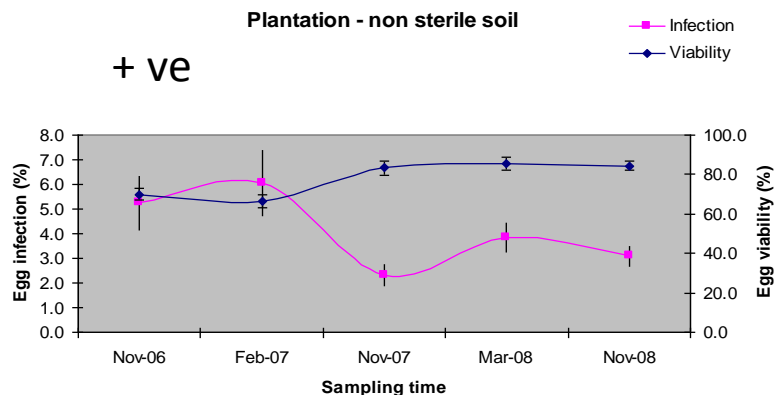
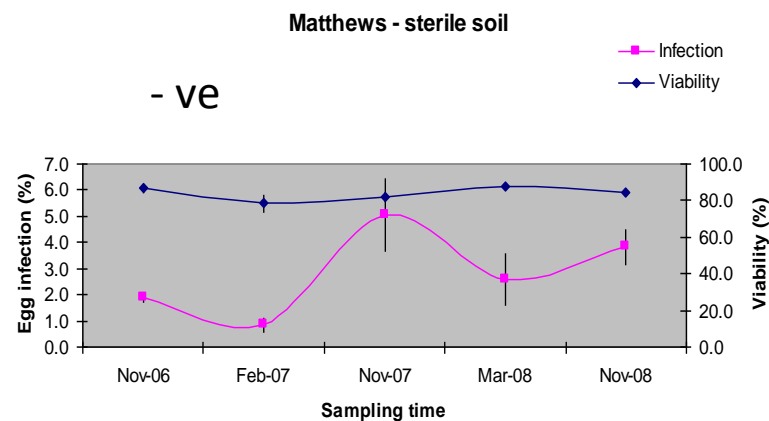
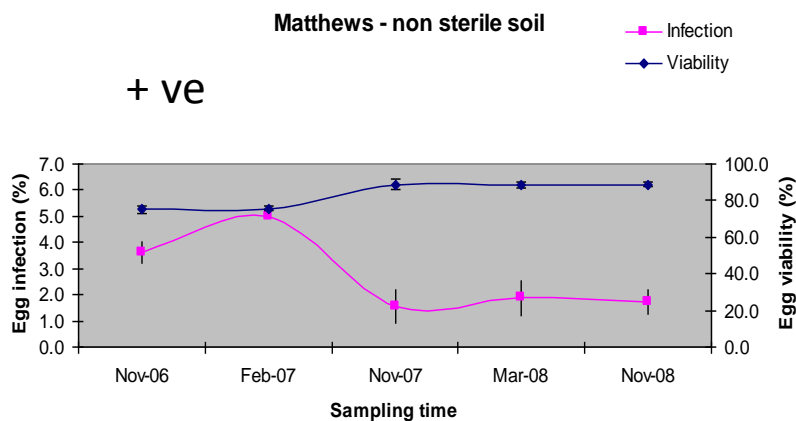




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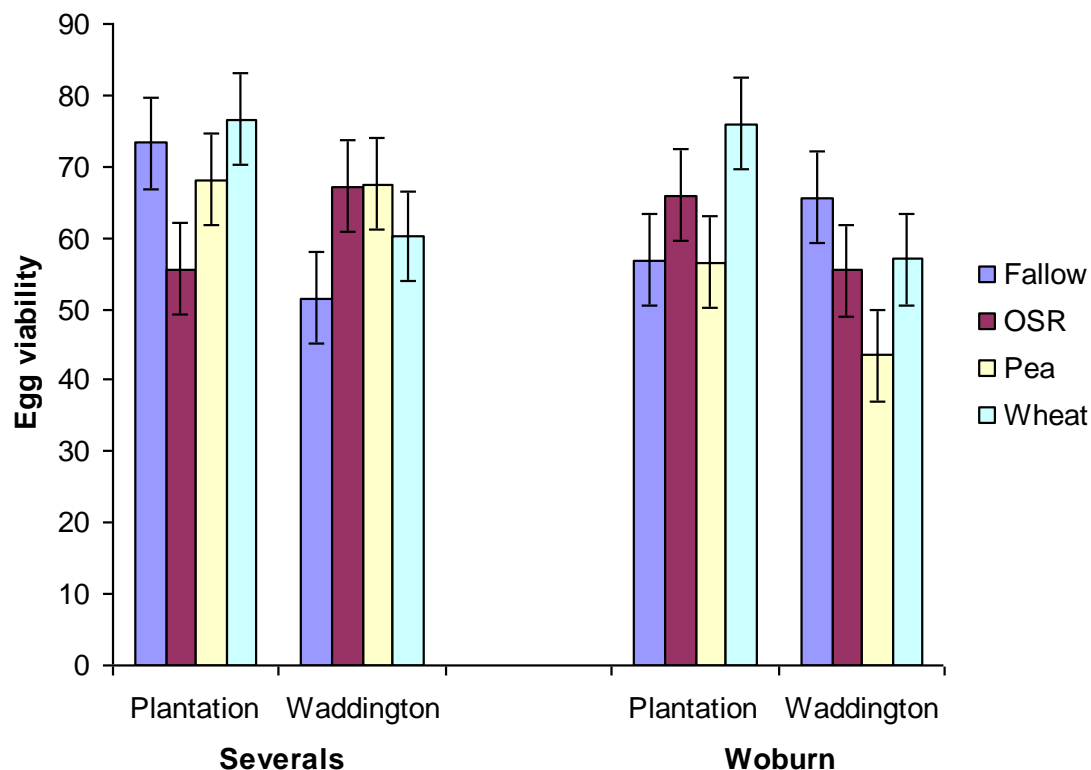


## **Objective 1:** Assess the rates of decline in a range of soils of different textures and cropping histories

### **Summary**

- Large amounts of variation makes interpretation problematic
- 40 % of the soils where the sterile soil showed the same +ve trend as non-sterile soil
- 10 % showed a negative trend between sterile vs non-sterile

## Objective 3: Investigate the influence of various cropping regimes on PCN decline and potato yields



### Effect of break crops

2 soils: Severals & Woburn

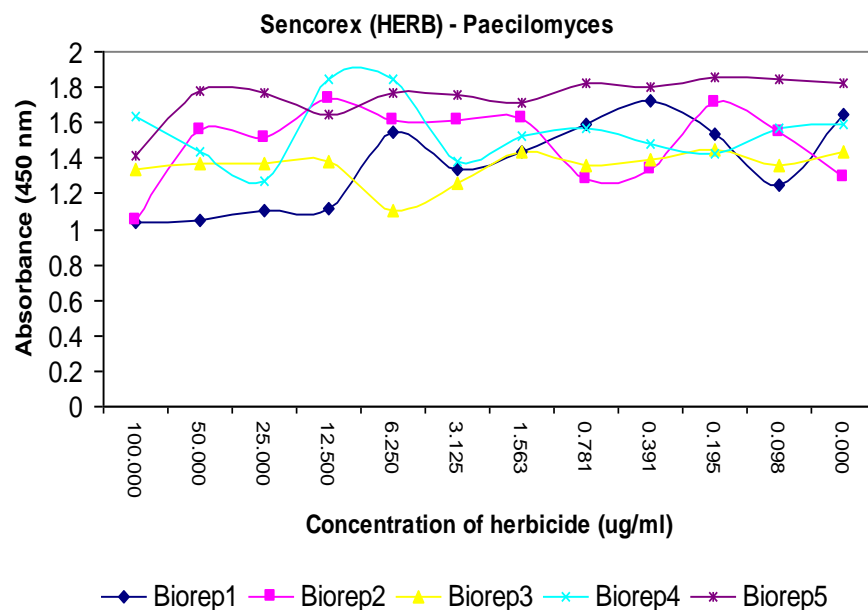
2 PCN pops: Plantation & Waddington

Very weak interaction  $P < 0.05$   
as measured by egg viability  
which was between 43.5 to 76.6 %



## Objective 4: . Assess effects of fungicides & herbicides on the associated microbes

The effect of herbicide Sencorex (Metribuzin; photosynthesis inhibitor) on the growth of *Paecilomyces lilacinus*.

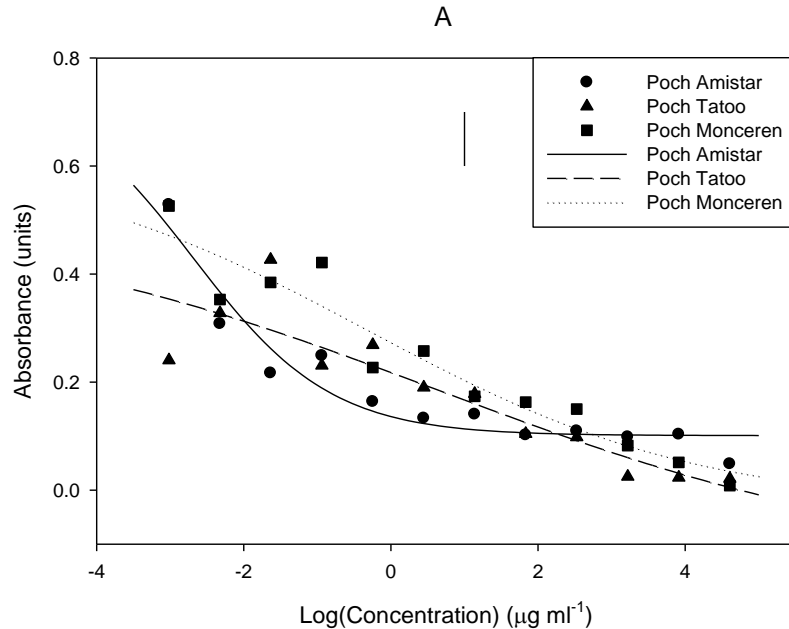


- The different concentrations of herbicide produced no significant effects on the growth of the fungus.
- Similar results were found for Basagran and Stomp, in both *Pochonia chlamydosporia* and *Paecilomyces lilacinus*.

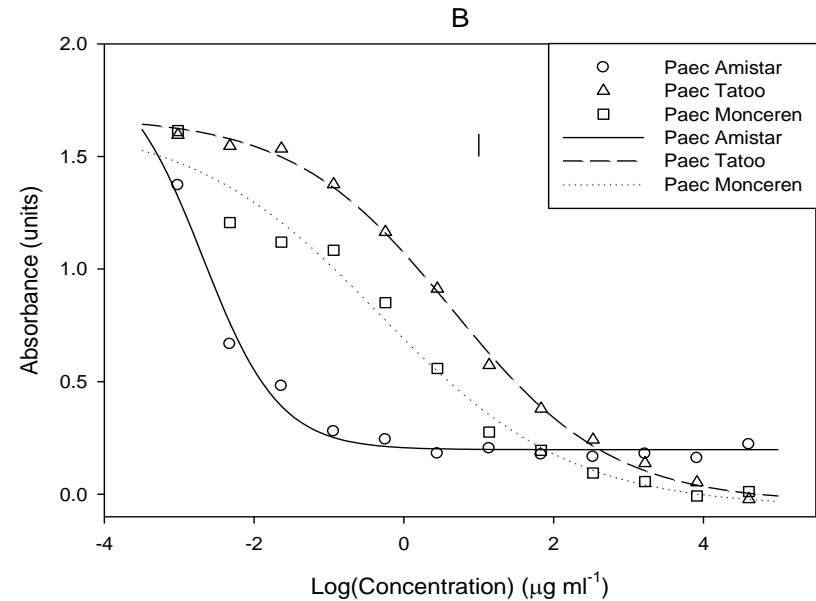
Herbicides had no measurable effect on the growth of either biological control fungi in ELISA plate assays

## Objective 4: . Assess effects of fungicides & herbicides on the associated microbes

**Impact of three fungicides (Monceren, prothioconazool; Amistar, azoxystrobin; and Tatoo, carbamate) dose response curves**



*Pochonia chlamydosporia*



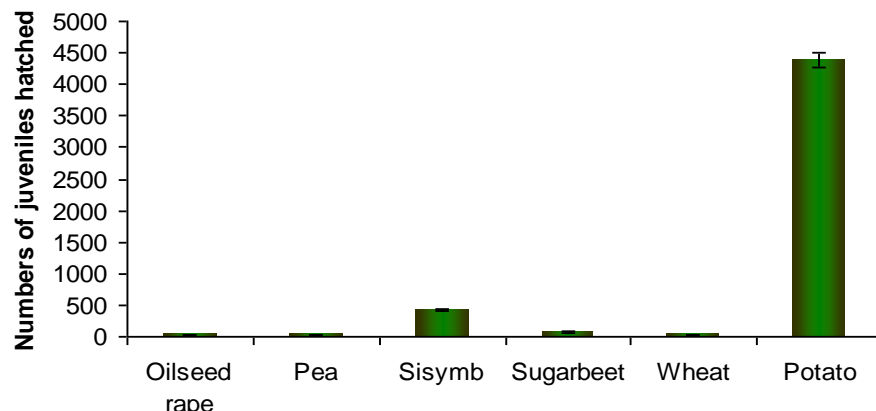
*Paecilomyces lilacinus*

**Fungicides did have a major effect on *Paecilomyces lilacinus* but did not have an effect on *Pochonia chlamydosporia***

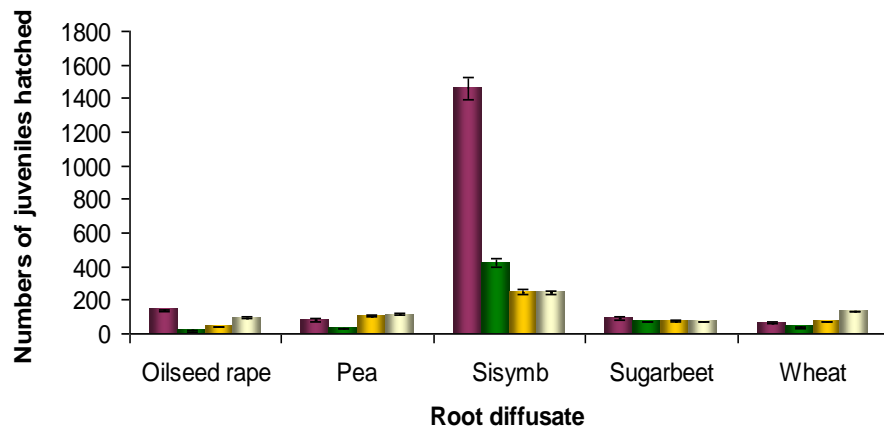


## Objective 5: Investigate the biotic and abiotic factors that influence fungal growth

Root diffusate 4 wks old - Total

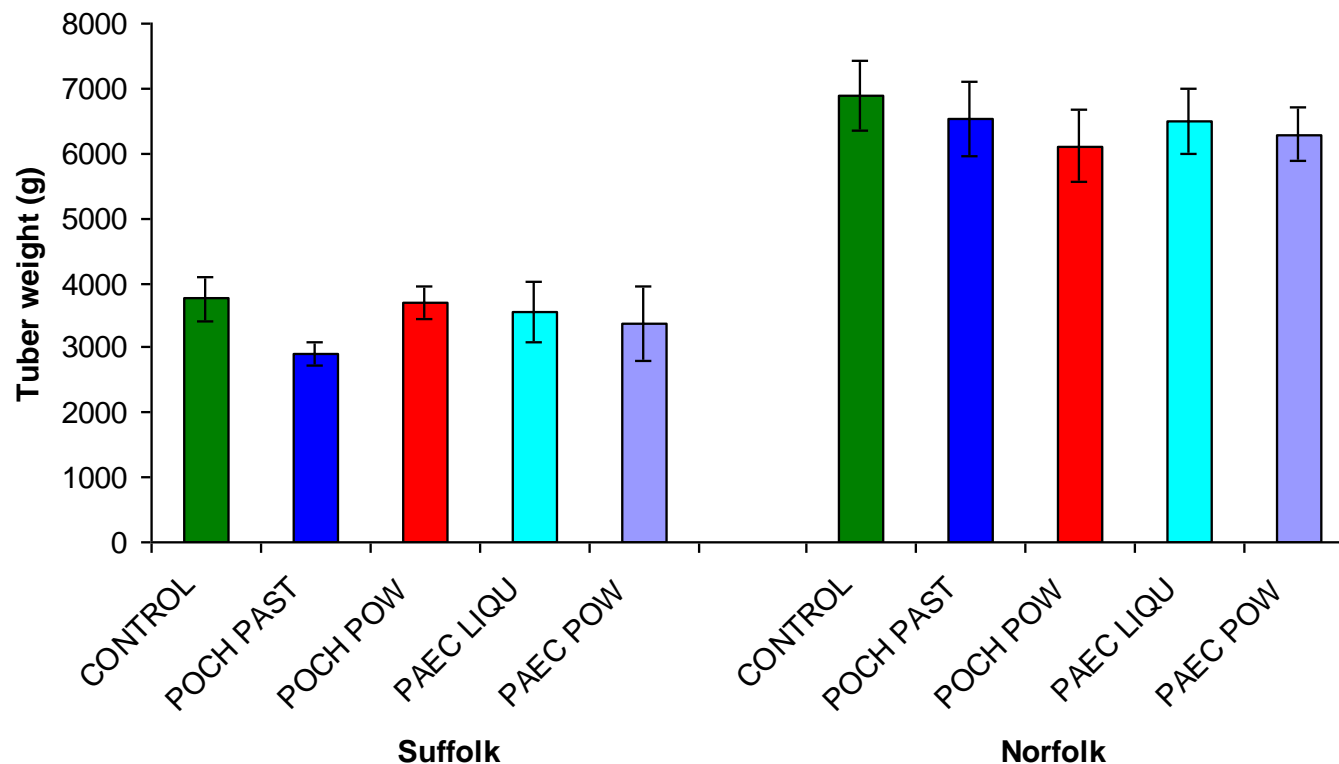


Potato root diffusate collected from 4 week old plants.



Potato root diffusate collected from a range of different aged plants.

## Objective 6: Investigate potential formulation and delivery of fungal agents

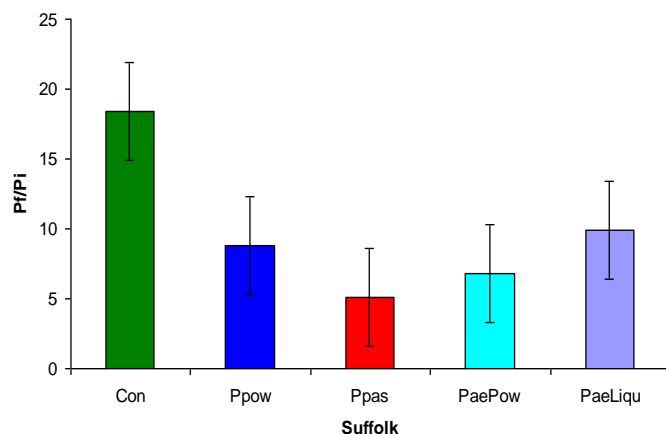


Yield of tubers per plot at two sites, Suffolk and Norfolk, comparing and untreated control with the biological control agents *Paecilomyces lilacinus* & *Pochonia chlamydosporia* with different formulations

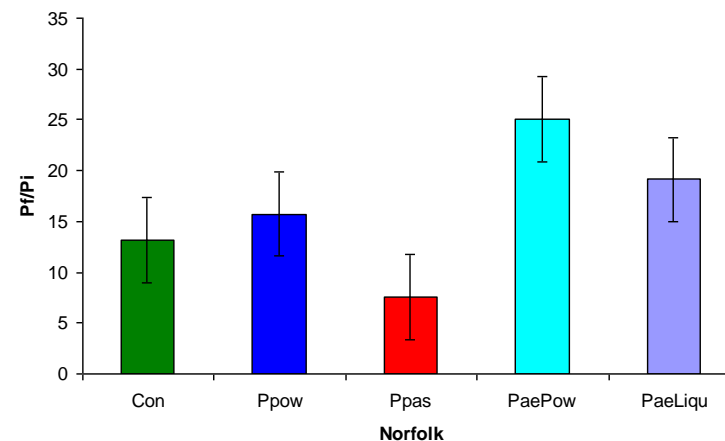


## Objective 6: Investigate potential formulation and delivery of fungal agents

High *Paecilomyces*  
X2 applications  
0.1 gram inoculum per m<sup>2</sup>



Low *Paecilomyces*  
X1 application  
0.1 gram inoculum per m<sup>2</sup>

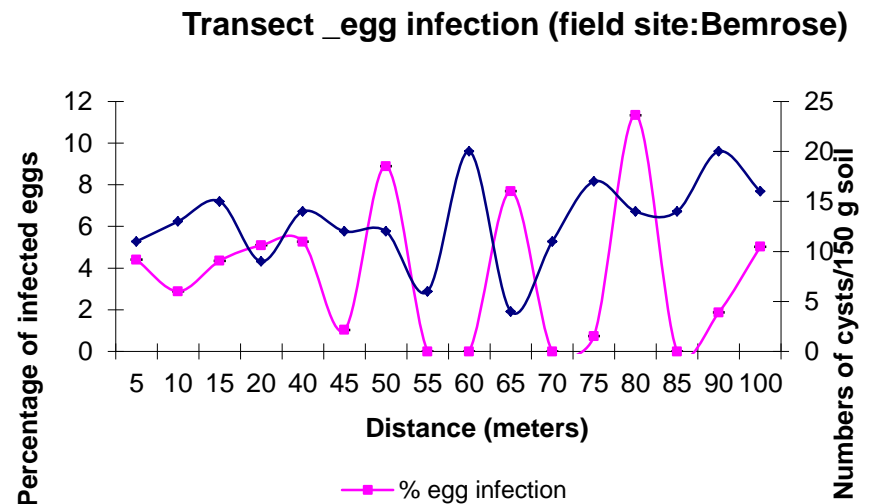
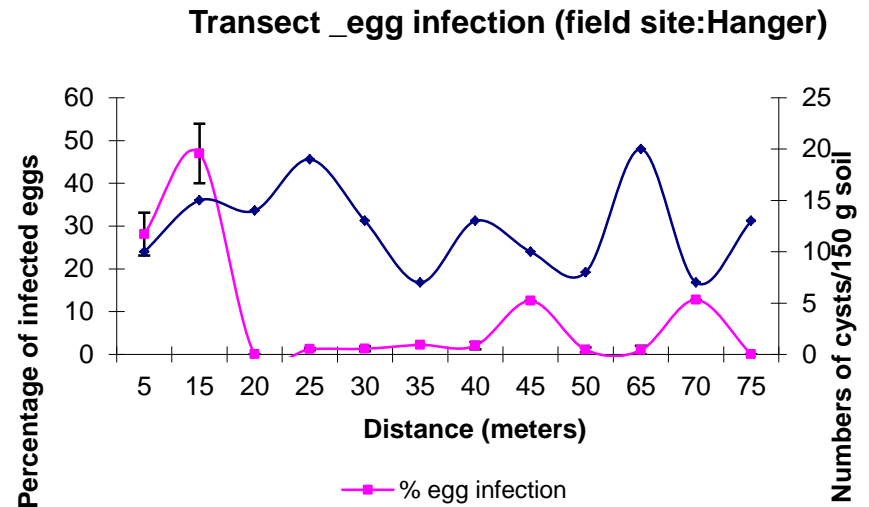
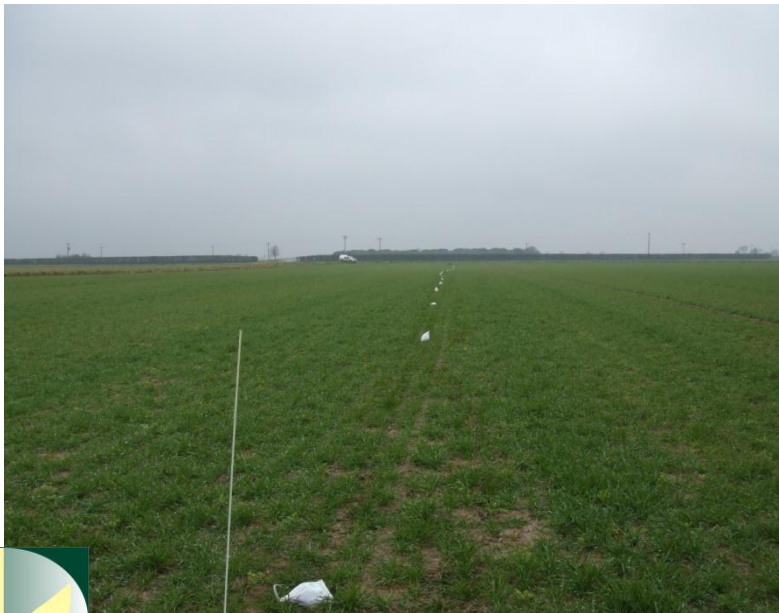


Pf/Pi ratio at two sites, Suffolk and Norfolk, comparing and untreated control with the biological control agents *Paecilomyces lilacinus* (PAEC), as a liquid (LIQU) or a wettable powder (POW) formulation, and *Pochonia chlamydosporia* (POCH) as a paste (PAST) of wettable powder (POW), (ANOVA between treatments  $P < 0.05$ ).



## Objective 8: Assess the importance of fungal parasites In patch dynamics

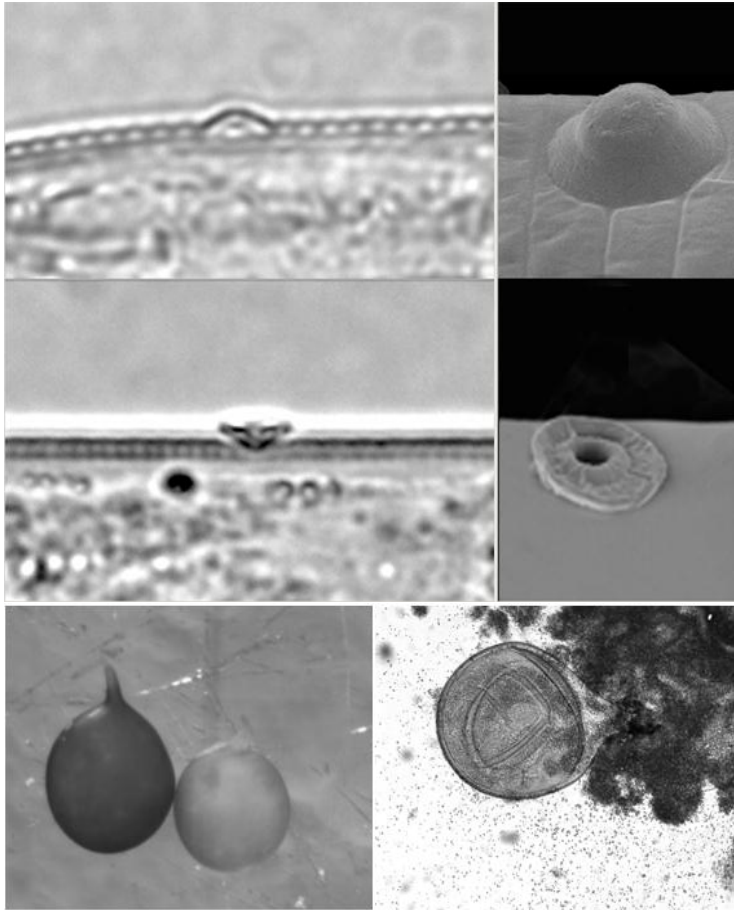
- Field PCN mapped
- Transect across a patch of PCN
- Cyst number and infection quantified



## Summary and some key findings

- An inverse relationship was observed between eggs per cyst and fungal egg infection but this was dependent on soil type
- Break crops had little effect on decline, however, *S. sisymbriifolium* increased hatch
- Fungi were tolerant to herbicides but sensitive to fungicides
- Although application of fungi did not increase tuber yield it had a an effect on PCN multiplication rates
- The type of formulation of fungi did not have a significant effect

## Multitrophic interactions in the rhizosphere



*Pasteuria* sp. found to be  
Parasitising *Heterodera cajani*  
also attaches to *Globadera*  
*pallida*

It germinates and infects  
*G. pallida*

Female becomes a cadaver  
full of spores

Sharad Mohan *et al.*, submitted





# *Globodera pallida* genome sequence

Sanger Centre, Leeds University, SCRI & Rothamsted

## Contigs

Size Mb

Number

*Globodera pallida*

112.1

26,637

*Meloidogyne hapla*

53.6

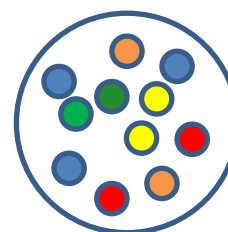
NA

*Meloidogyne incognita*

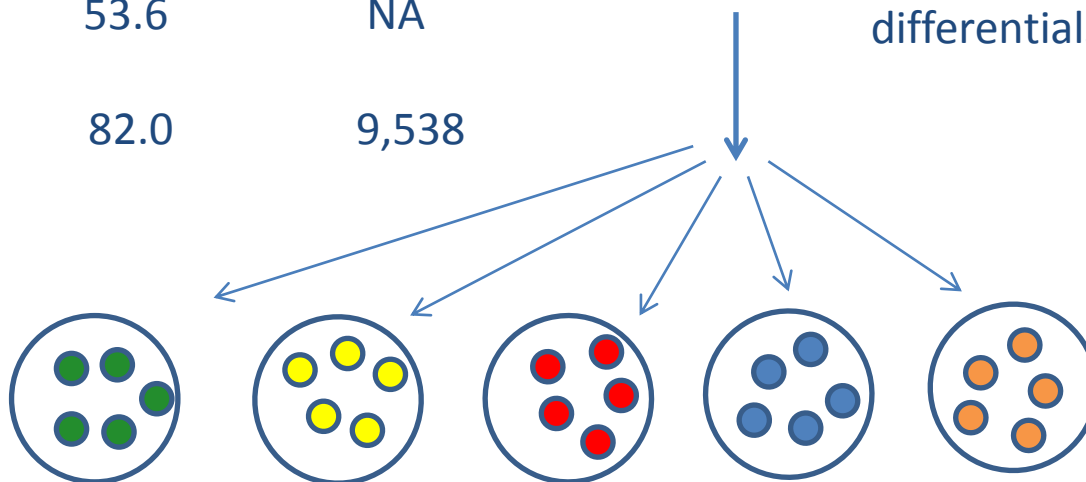
82.0

9,538

*G. pallida*  
Population  
*Lindley*



Select  
on  
differentials



# Acknowledgements

**UKIERI**  
UK-India Education  
and Research Initiative



Bayer CropScience



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