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## **Cavity spot**

- Cavity spot is one of the biggest problems for carrot growers in the UK; estimated losses at least £5M per annum
- In the UK principally caused by the oomycete P. violae and to a lesser extent P. sulcatum – incidence of each species was unknown
- Other Pythium species associated with cavity spot include P. intermedium and P. sylvaticum
- Other fungi may cause similar root lesions or invade cavities e.g. Cylindrocarpon destructans and Mycocentrospora acerina







## The problems

- Interaction between components of the cavity spot complex unknown
  - Relative importance of *P. violae*, *P. sulcatum*, *P. intermedium*
- The disease is unpredictable
  - Lack of understanding of key abiotic and biotic factors that enhance disease
  - Lack of understanding of biology of P. violae and timing of infection
- Metalaxyl issues / lack of new actives
  - Microbial degradation of metalaxyl / future withdrawal
  - Field trials to identify new control products / approaches challenging lack of adequate disease levels
  - Lack of artificial inoculation approaches
- Commercial carrot varieties lack good resistance
  - Cv. Nairobi most widely grown low-intermediate resistance





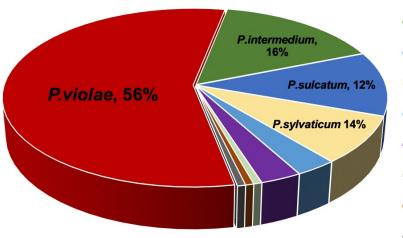




- 1) Identification of Pythium isolates associated with cavity spot
  - Determine frequency of different *Pythium* spp. associated with cavity spot
- 2) Improve molecular detection of *Pythium violae* in soil by PCR
  - Help determine inoculum levels and assess disease risk
- **3) Develop artificial inoculation systems** to induce cavity spot disease to enable:
  - Identification of new crop protection products
  - Identification of new sources of plant resistance

P. violae is the major cavity spot pathogen in

the UK



- P.violae
- P.intermedium
- P.sulcatum
- P.sylvaticum
- Pythium spp.
- P.ultimum
- P.attrantheridium
- P.irregulare
- P.debaryanum

Cavity spot lesion size: P. violae > P. sulcatum > P. intermedium



## Pythium oospore capture



- Pythium often distributed unevenly: DNA extraction from small soil sample kits may give inconsistent detection
- A method of separating *Pythium* oospores from soil was developed to allow quantification from larger soil samples



**Step 1.** Sucrose flotation & centrifugation separates oospores from soil

**Step 2.** Oospores captured on a filter membrane

**Step 3.** Free oospores ready for DNA extraction / quantification

# P. violae detection and quantification by PCR

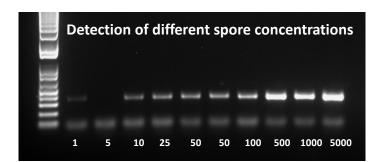
- Published PCR primers amplified other *Pythium* species
- New primers developed were specific to P. violae
- Assay could detect 10 spores in solution; yet to be tested extensively in field







Sul: P.sulcatum, Ult: P.ultimum, Lut: P.lutarium. Irr: P.irregulare, Syl: P.sylvaticum, Int: P.intermedium, Viol1/2: P.violae



# Artificial inoculation of carrots with *P. violae*

- P. violae oospores produced in sand / oat substrate
- Variable disease levels when used to inoculate glasshouse-grown carrots in pots but consistently high cavity spot incidence in field 'macrocosms'
- Millet grain and compost/carrot inoculum more effective for glasshouse experiments

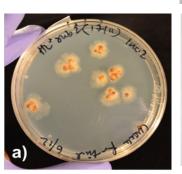




# Inoculation of pot grown carrots with *P. violae* mycelium



- Millet grain inoculum used which is quicker to produced than oospores and resulted in:
  - Seedling damping off at higher levels
  - Reduction in carrot foliage
  - Production of stubby carrots
  - Reduction in root weight
  - Cavity spot incidence of 50%
  - Low cavity spot severity (mean 2-3 lesions / root)

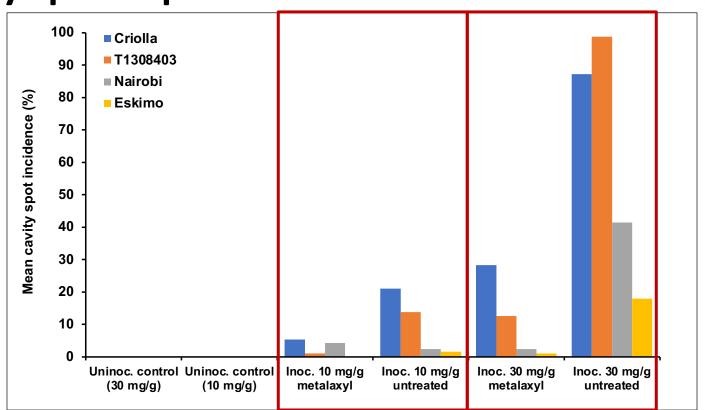






# Metalaxyl and carrot variety reduces cavity spot in pot tests



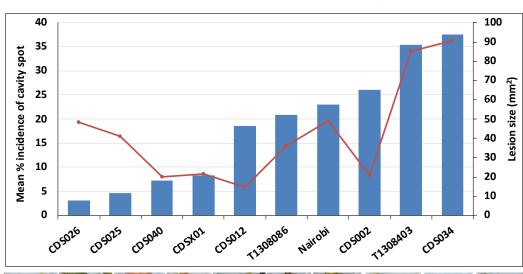


#### **Carrot resistance to cavity spot**



- Glasshouse pot test: millet inoculum
- Differences between different carrot accessions

Some relationship with P. violae agar plug test





# Inoculation of field macrocosms with *P. violae* oospores



- Normal growth of carrots
- Up to 40% cavity spot incidence, severity of 6 lesions / carrot



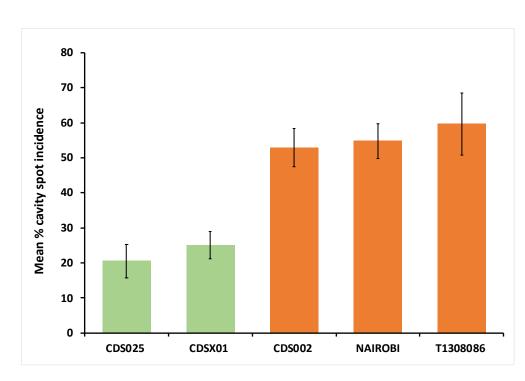


# Carrot resistance to cavity spot



- Macrocosm field test: oospore inoculum
- Differences in resistance observed between carrot accessions

 Similar results to glasshouse pot test



### Sceptre+ trial in progress



- Highly susceptible cv. Criolla used
- Treatments:
  - SL567a (metalaxyl)
  - Previcur Energy (propamocarb and fosetyl Al)
  - Trisoil (*Trichoderma atroviride* I-1237)
  - Ranman (cyazofamid)



## Summary



- P. violae identified as main pathogen causing cavity spot but P. sulcatum / intermedium also present
- New PCR and oospore capture methods developed for P. violae detection and quantification
- Artificial inoculation methods developed for P. violae for pot and field grown carrots which are effective for testing new crop protection products and assessing cultivar resistance
- Tools now in place to investigate other approaches to cavity spot control (e.g. organic / inorganic soil amendments / biocontrol) and combining with new chemistry and carrot cultivars

# Thank you to:



- Nicole Pereira, Kat Hales and Andy Taylor
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