An example of easy diagnostics for *Fusarium oxysporum* f. sp. *melonis* and race determination by using bait plants

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COST Action FA1105 – BioGreenHouse
Towards a sustainable and productive EU organic greenhouse horticulture
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Fusarium wilt of melon: symptoms

- Chlorosis and necrosis of lower leaves

- Xylem necrosis in plant stem

- Gummy exudate

- Epidermal necrosis on the lower stem, with presence of mycellium and sporodochia

Fusarium oxysporum f. sp. melonis

Fusarium wilt of melon: an important worldwide disease

Host Resistance

Environment

Virulence of isolates

Inoculum density

Soil microbiology

Diagnosis

Diagnosis for soils

1- Classical soil analysis and further inoculation.

2- Our proposal: Bait plants.

· A set of soil samples from 3 different areas with a known history. of Fusarium wilt of melon was used and comparatively analyzed.

Soil samples

- Samples from rhizosphere
- 5-20 cm depth
- 10 sample points per soil
- 100 g per sample

- Drying at laboratory
- Analysis at 2-4 weeks after collection

1.

Dried sample

↓

Sieving

Fine fraction (<200 microns)

↓

16 Petri dishes per sample

Komada's medium at 40 °C

Incubation (5-10 days)

↓

Identification of colonies

1.

11 cfu white species

8 cfu orange species

Fusarium oxysporum
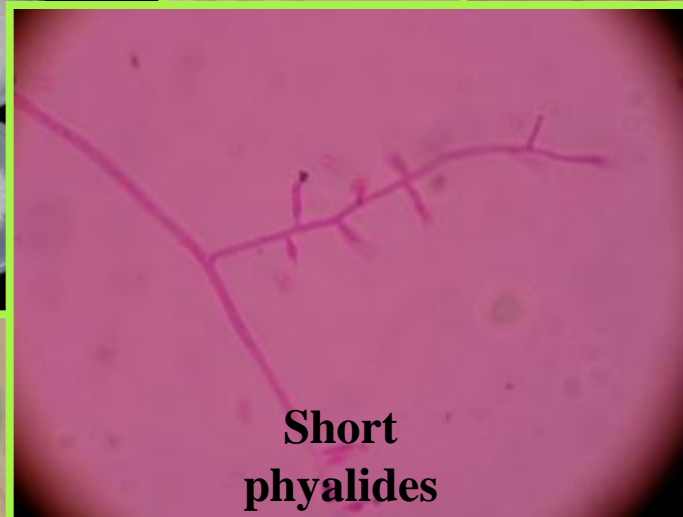
1.

Isolates' identification
Expertise required
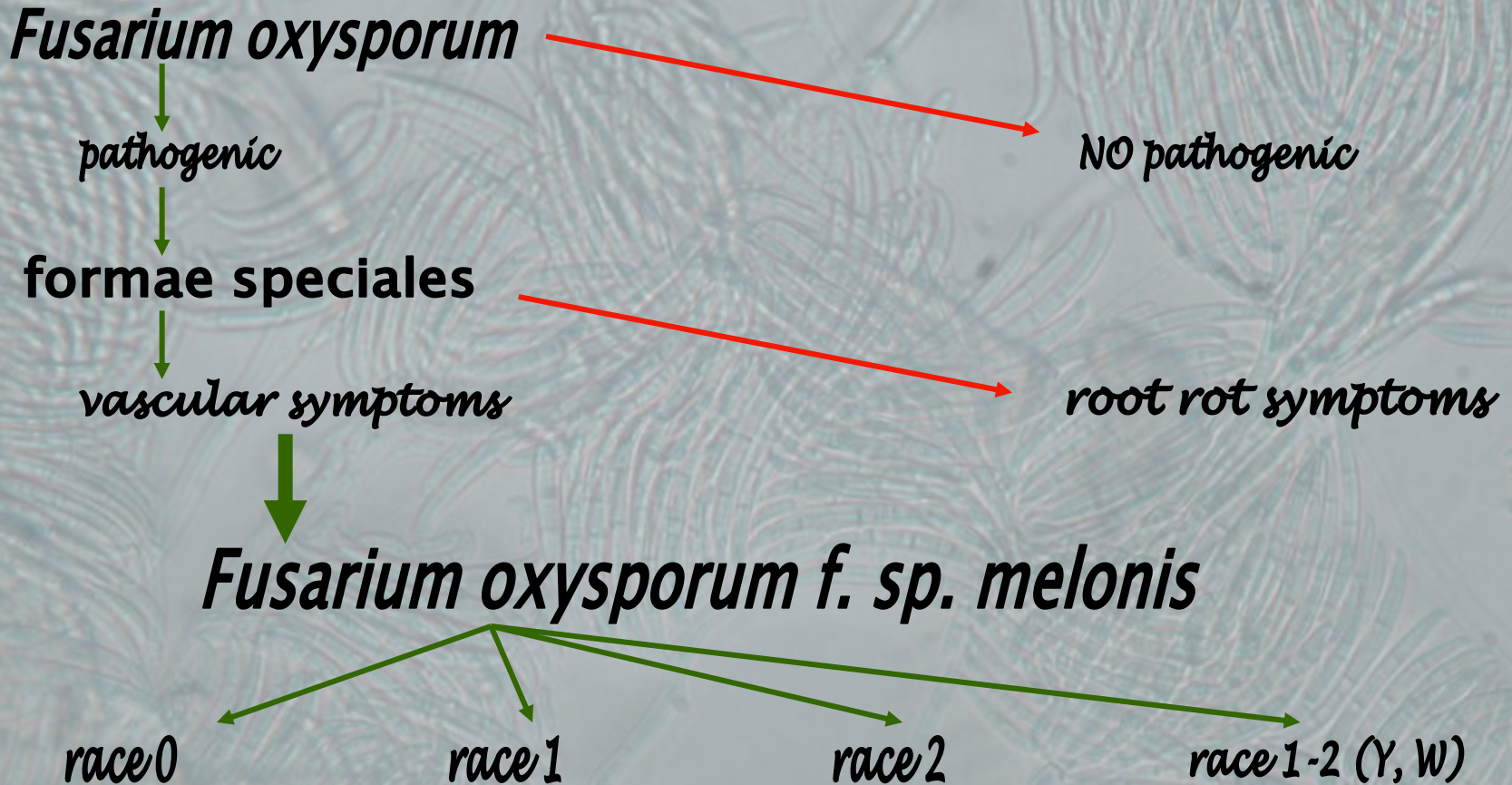(Nelson *et al*, 1983; Gerlach y Nirenberg, 1982)
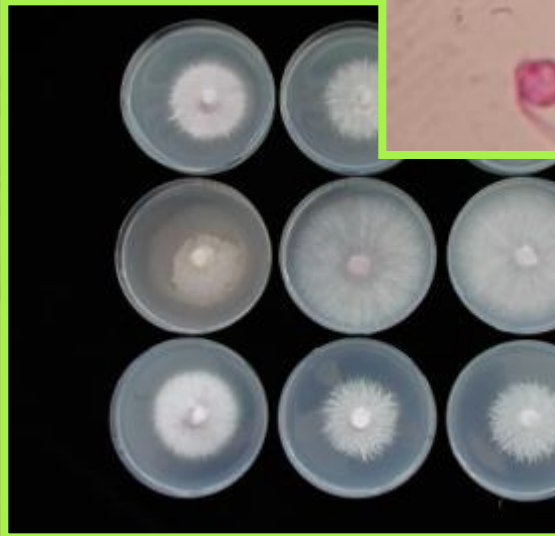
Chlamydospores

Macroconidia

Short
phyalides

1. Isolation and later inoculation to determine the role of *F. oxysporum* isolates in the soil

Fusarium oxysporum ⟶ NO pathogenic

↓ pathogenic

formae speciales ⟶ root rot symptoms

↓

vascular symptoms

↓

Fusarium oxysporum f. sp. melonis

race 0   race 1   race 2   race 1-2 (Y, W)

Soil classical Fusarium analysis (Warcup's technique)

Direct determination of pathotypes
(Risser *et al,* 1976)

Using differential varieties

No genes

Gen *Fom1*

Gen *Fom2*

Genes *Fom1+Fom2*

1.

Positive presence of *F. oxysporum*

67.7% soils from Affected farms group 1   **Range: 10-5378 cfu/g**

46.7% soils from Affected farms group 2   **Range: 2-1153 cfu/g**

100% soils from Affected farms group 3   **Range: 179-2260 cfu/g**

87 isolates were inoculated → **Pathogenic: 0**

14 isolates were inoculated → **Pathogenic: 0**

35 isolates were inoculated → **Pathogenic: 27
(RACE 1)**

Bait plant analysis

Use Autoclaved Vermiculite

Soil sample

⬇

Mixing (25g soil + 100g vermiculite)

⬇

1-Use cvs. With different resistance genes (Fom genes)

2- Disinfect seeds 20 min in bleach (35 g active chloride per liter) and rinse with tap water until do not smell chloride.

3-Plant root-merged seeds
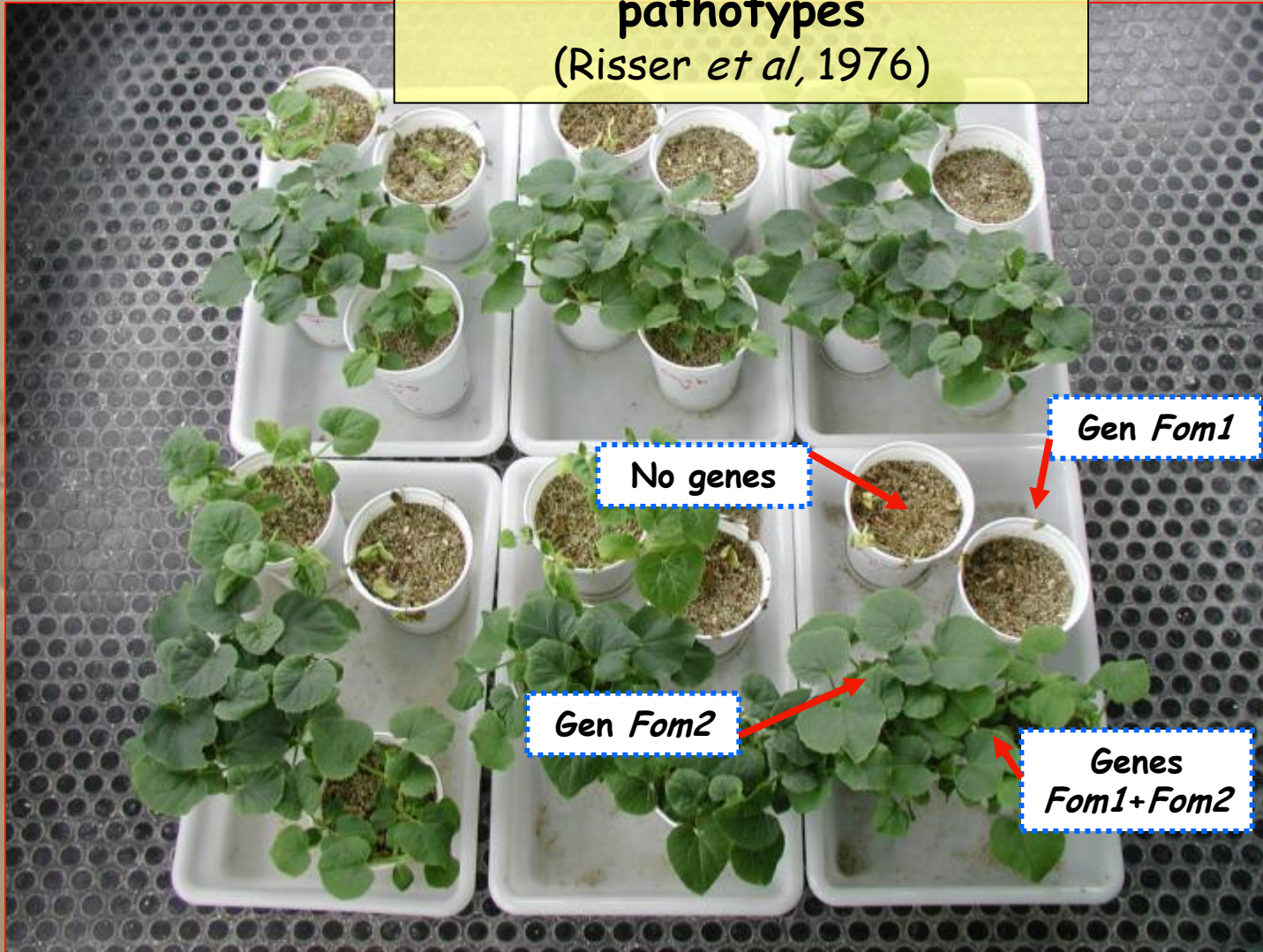
Planting

⬇

Holes to drain at 5cm from the base
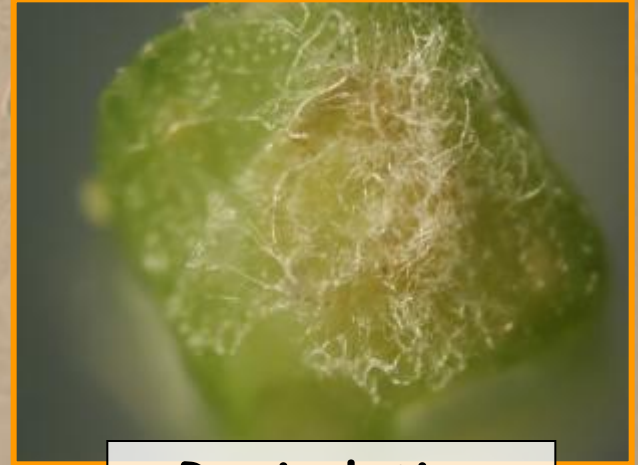

Until observe symptoms

2.

Bait plant analysis

Direct determination of pathotypes
(Risser *et al*, 1976)

No genes

Gen *Fom1*

Gen *Fom2*

Genes *Fom1+Fom2*

Directly planting differential varieties in each different pot

2.

Re-isolation

Pure culture

Results
Bait plant analysis

Symptomatic plants

0% soils from Affected farms group 1

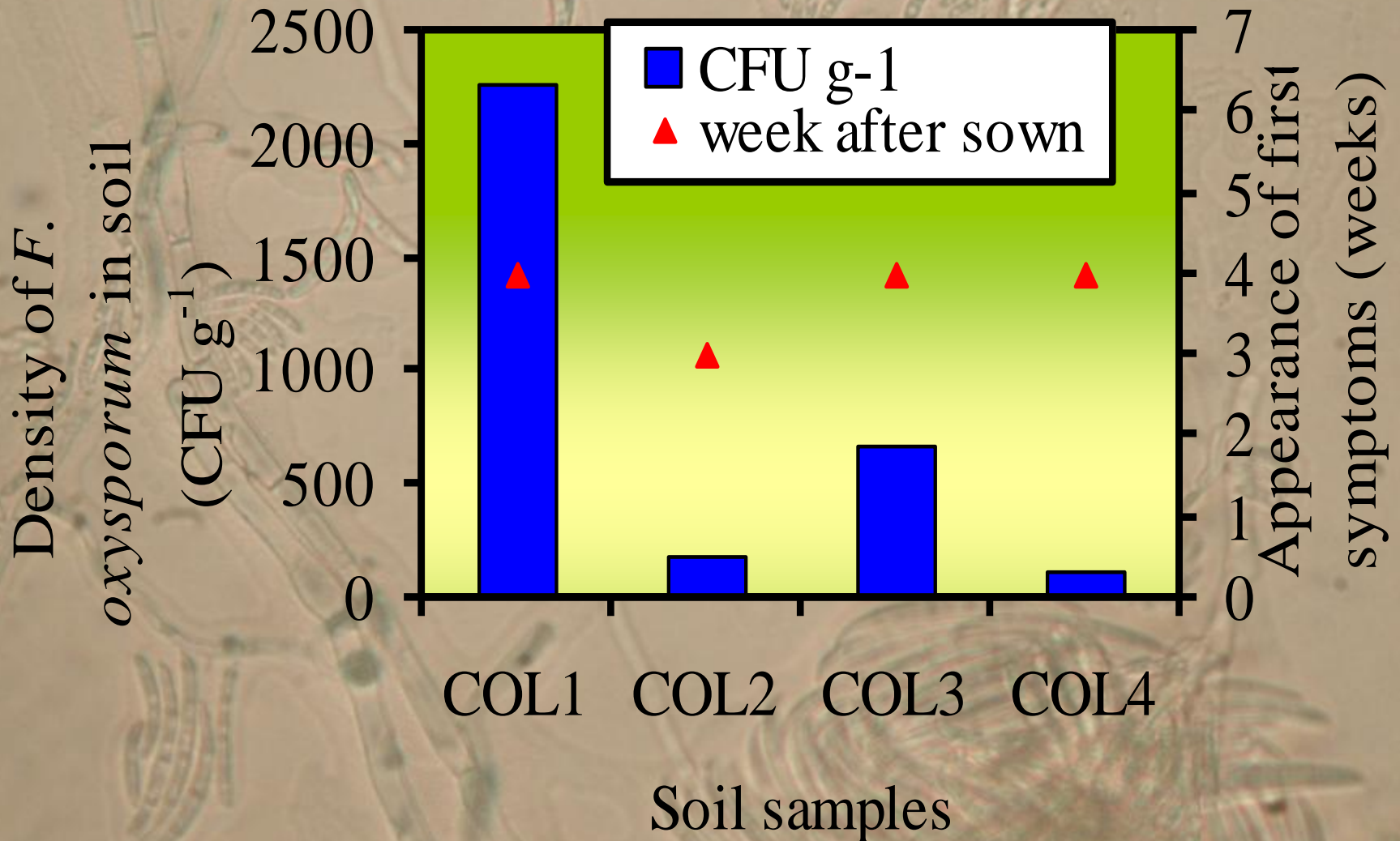**0% soils from Affected farms group 2**

100% soils from Affected farms group 3 (31 isolates RACE 1)

2.

Results
Warcup vs Bait plant

Conclussions

- Bait plant technique for Fusarium Wilt of Melon is:

   - Easily REPRODUCIBLE. Only a kit of seeds is required.

   - COMPARABLE to Classical technique. Not expertise is required.

   - FAST determination of pathotypes. Less than 30 days.

   - Gives INFORMATION about CONDUCIVENESS of the soil, not only about presence of pathogen.

   - EXTRAPOLABLE to other crops with physiological races, such as tomato.

More detailed info in: M. de Cara , F. Diánez , M. Santos , E. J. Fernández , J. Tello , F. J. Estrada & S. Montoya (2006) Presence of *Fusarium oxysporum* f. sp. *melonis* Race 1 in Soils Cultivated with Melon in the State of Colima (Mexico), *Geomicrobiology Journal*, 23:5, 319-322

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