Biological disinfestation of tare soils contaminated with quarantine plant pathogens

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Plant pathogen and weed seed eradication from waste streams

- Composting
- Anaerobic fermentation
- Soil tares
- Survival of (quarantine) pathogens in soil tares was identified as ‘knowledge gap’ in our current understanding
  - Relevant for practice
  - Development of disinfection procedures for tare soils

Phytosanitary risks of reuse of waste streams and treated wastes for agricultural purposes , 2011, Plant Research International, Report 382.

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Soil tares

- Industrial wastes from harvested products like potatoes, sugar beets, bulbs etc.,
- Considerable waste streams,
- Soil with crop residues (roots, fragments of tubers/ bulbs), - rich in organic materials-
- BUT... also rich in (quarantine) phytopathogens,
- Never used for agriculture (used for construction of roads, cross-overs, dikes, ... or just dumped into the sea)
- Eradication experiments done by Willemien Runia by heat treatments (aerated steam at 50-60^{o}C, 3 min, complete eradication of *Verticillium*, *Sclerotium* Oomycete spores, and cyst nematodes)
    - Energy demanding process; not sustainable

Experimental design

Soil tares of Potatoes

- **Two soil types:** sabulous clay (21% clay, 2.4 % org matter, pH 7.2) and (cover) sand (3.2% org. matter, pH 5.5)
- **Two pathogens:** *Ralstonia solanacearum* bv 2 & *Globodera pallida*
- **Five soil treatments:** (1) No amendment (control), (2) grass cutting (2-3 cm), (3) Herbie 7025 (Thatchtec, Wageningen), (4) inundation, (5) inundation + Herbie 7025 (first year only)
- **Four repetitions per treatment x soil**
- **Two years:** 2011 (July – September, 12 weeks) and 2012 (May – July, 12 weeks)

Experimental set up (Q facility PPO-AGV)

Globodera pallida cysts in mesh bags

Mixing *Ralstonia solanacearum* through soil

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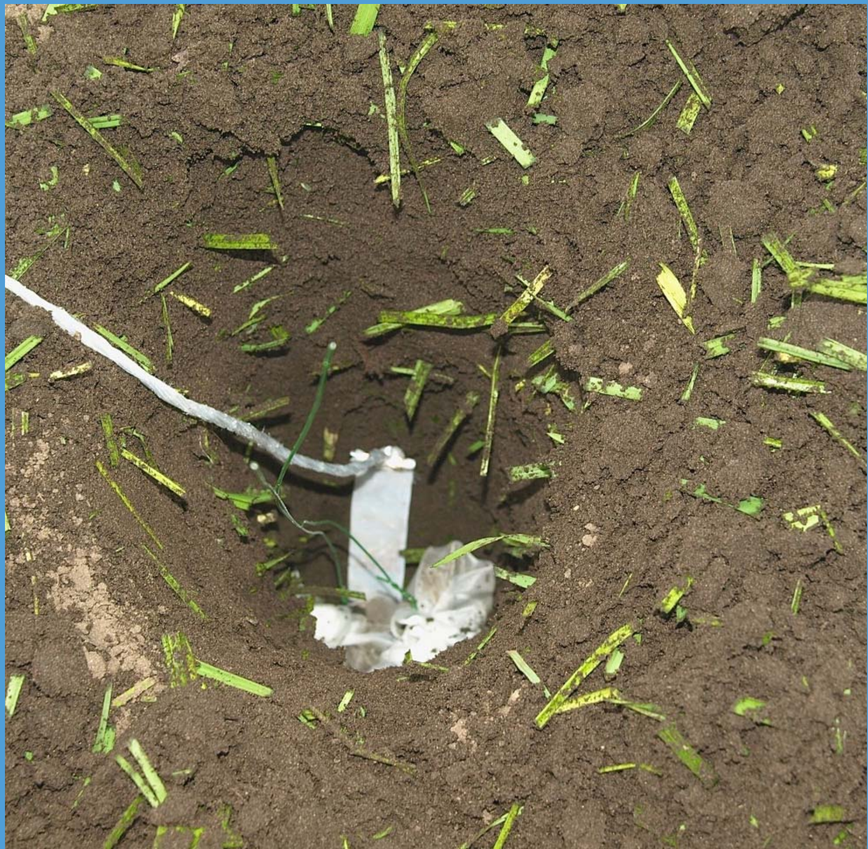
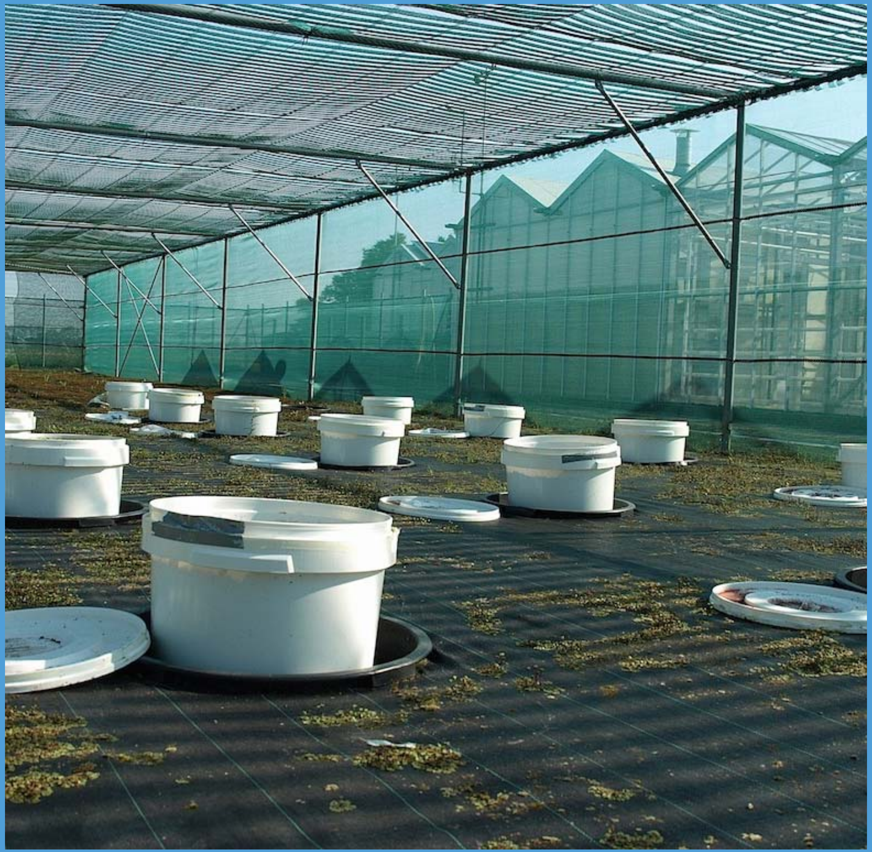
Placing mesh bags (with Rs or Gp) in buckets with soil (sand or clay)



Opening of the buckets after 12 weeks



Soil temperature

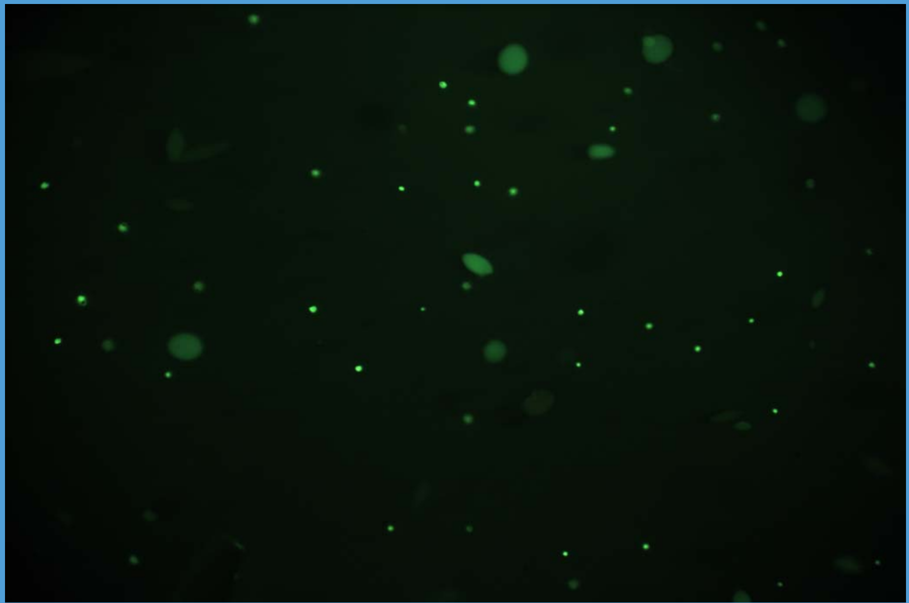
		2011			2012		
soil	**Treatment**	**Min**	**Max**	**Avg**	**Min**	**Max**	**Avg**
Sand	no	9.5	27.2	16.8	7.9	25.4	16.2
	Grass	9.0	27.1	16.5	7.3	26.7	16.1
	H 7025	9.1	23.8	16.6	7.6	24.6	15.6
	Inundation	9.7	25.5	16.3	8.4	25.3	16.8
	H 7025 + inund.	9.4	26.2	16.3	ND	ND	ND
Clay	no	9.0	26.3	16.4	7.9	25.3	16.1
	Grass	9.5	26.2	16.6	8.4	25.7	16.3
	H 7025	9.5	26.4	16.5	6.5	27.1	16.3
	Inundation	9.7	24.5	16.0	8.2	24.3	16.0
	H 7025 + inund.	9.8	26.9	16.4	ND	ND	ND
Ambient temperature		6.8	28.8	16.0	3.1	29.5	15.6

Analyses for pathogen survival

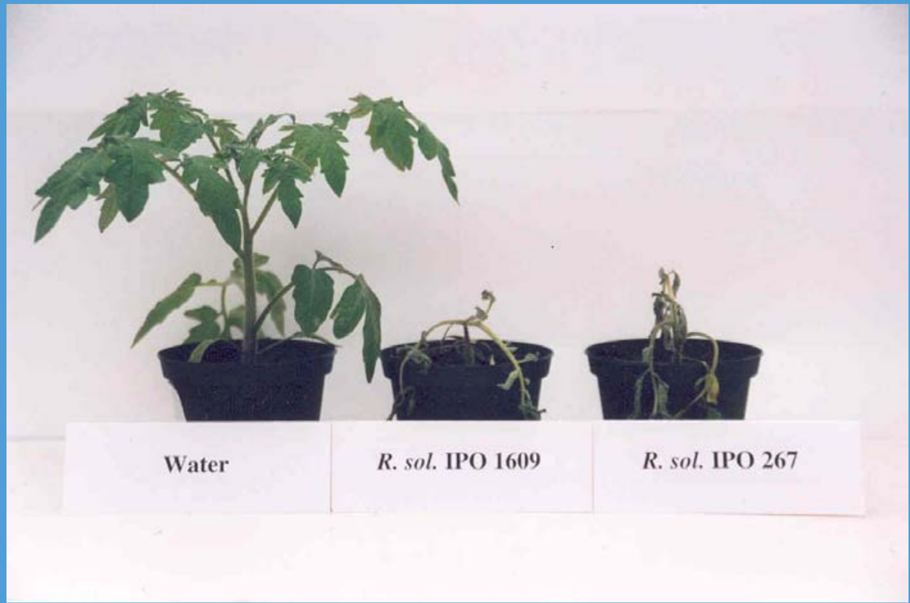
- *R. solanacearum* biovar 2, strain 1609
  - Immunofluorescence colony staining,
  - Test on bacterial wilt in tomato
- *G. pallida* viability test on eggs (luring of larvae)
  - 2000 cysts in mesh bags in soil,
  - After 12 weeks of incubation, cysts were removed from bags,
  - Cysts were soaked in water, ground and eggs were suspended,
  - Juveniles hatched from eggs were lured to potato root exudate (duration of 6 weeks)
  - Number of juvenile nematodes counted

R. solanacearum survival test

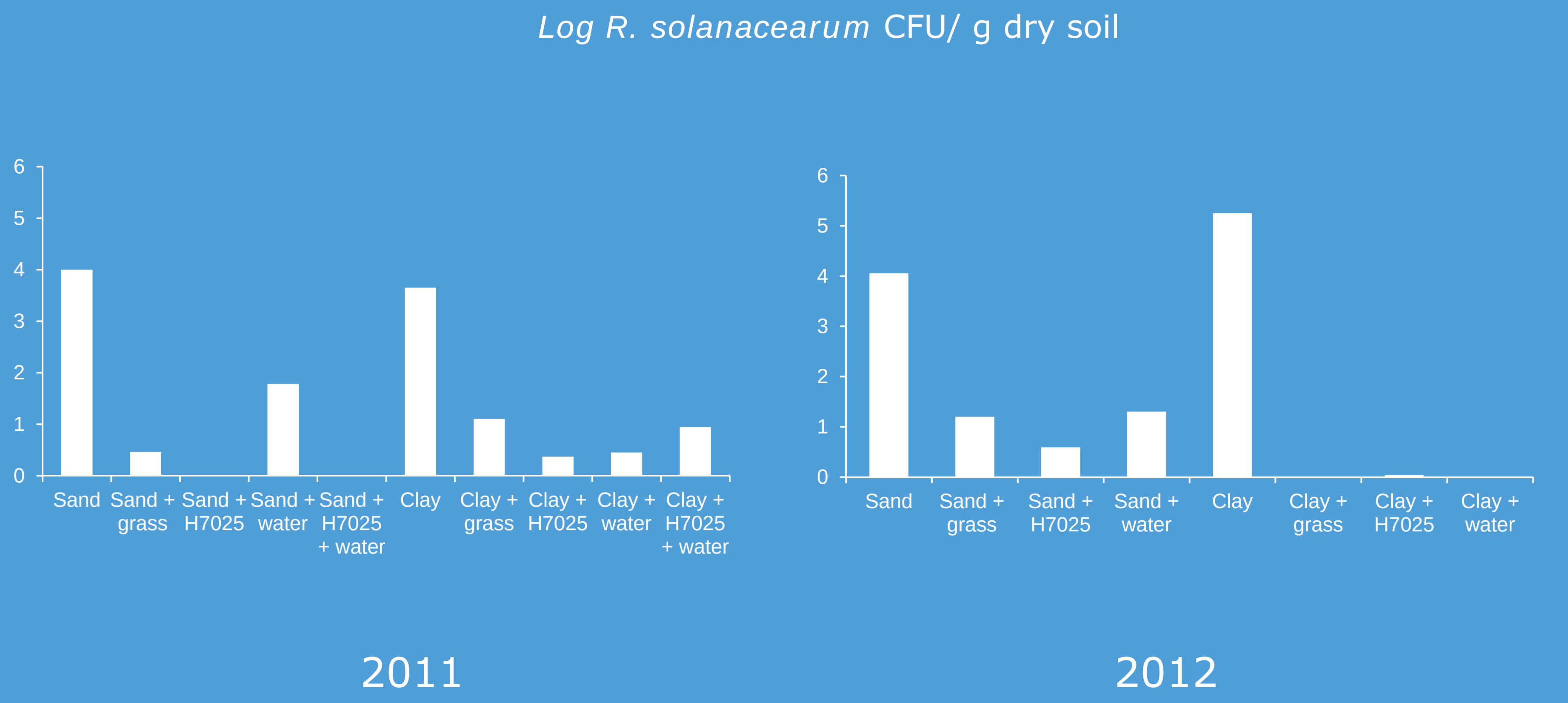
Immunofluorescence colony staining (IFC)



Test for bacterial wilt in tomato



Ralstonia solanacearum bv 2 survival



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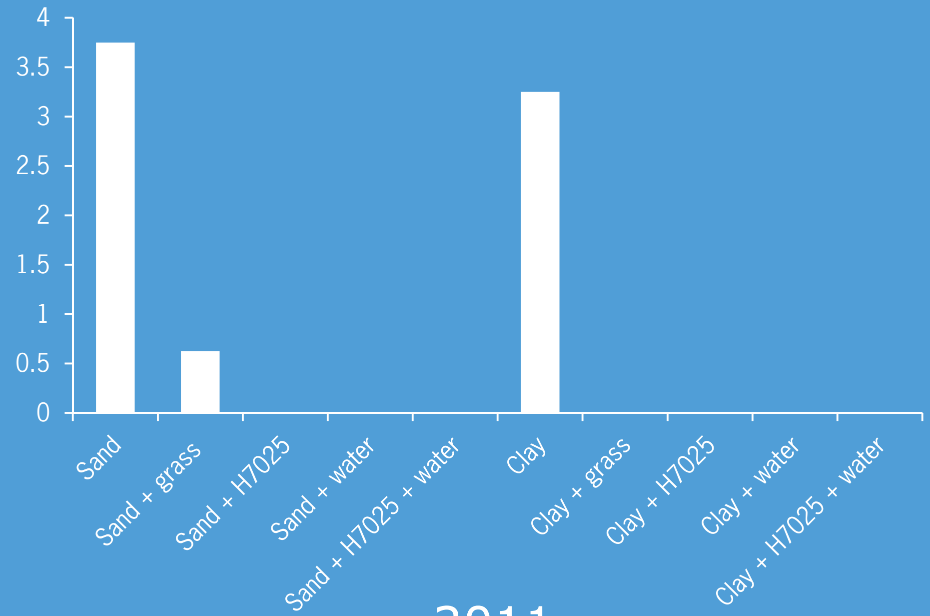
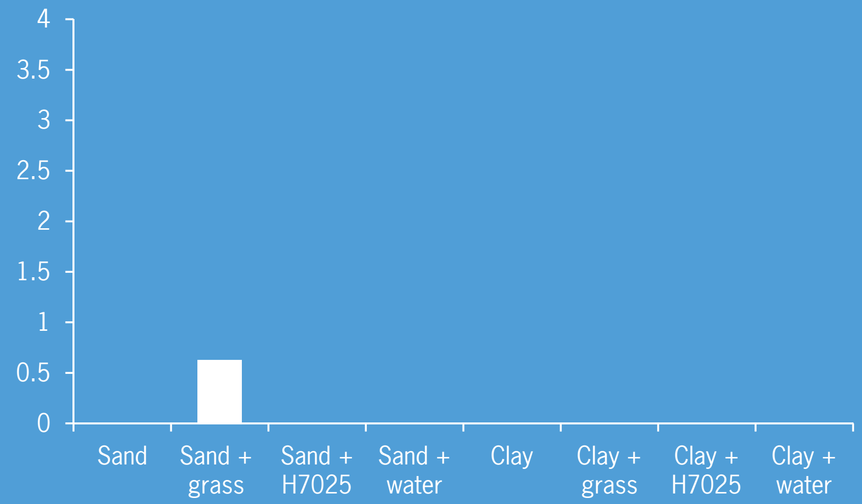
Wilting in tomato (caused by *R. solanacearum*)

Tomato wilting (scale 1-5), *n*=3



2011



2012

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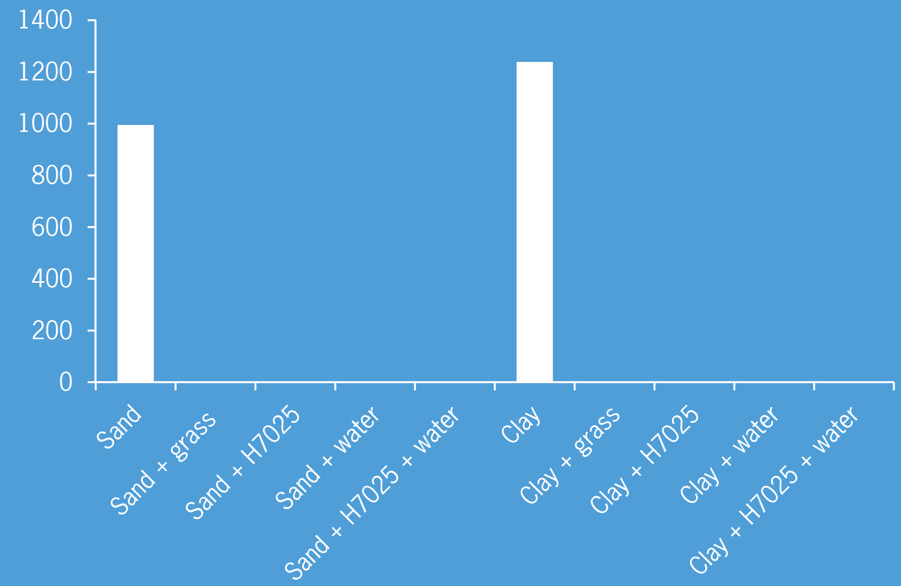
G. pallida survival

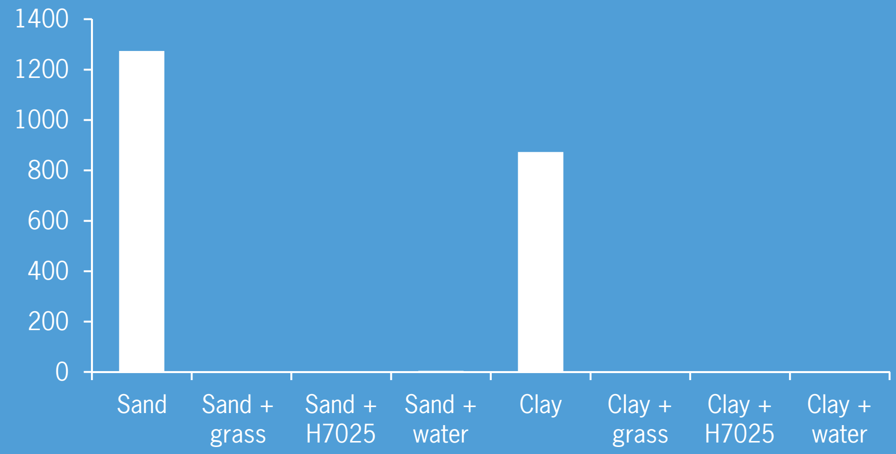
Viability of eggs

of 1200, *n*=2



2011

of 3000, *n*=2



2012

Conclusions

- *Viable R. solanacearum* cells and *G. pallida* eggs consistently dropped in all treated soils.
- No consistent eradication of *R. solanacearum and G. pallida* was found for any of the applied treatments.
- Both species are quarantine pathogens in NL, so complete eradication from soil is necessary for eventual later applications.
- Soil treatment with Herbie 7025 seems most effective against *R. solanacearum*.

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