

Managing
Sclerotinia
sclerotiorum
in high tunnels
with biofumigation
and solarization

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Outline

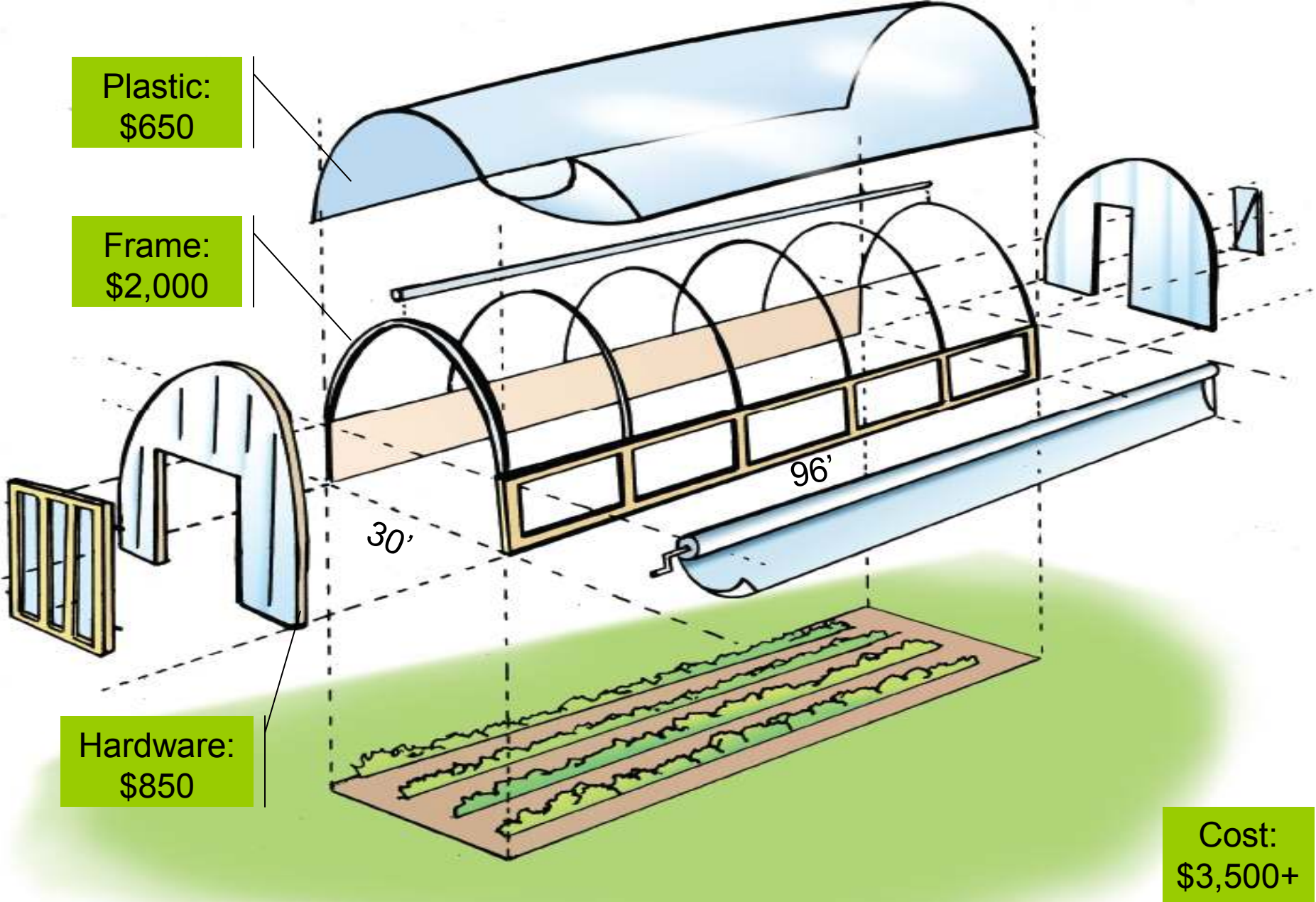
- High tunnels
- *Sclerotinia sclerotiorum* and *Phytophthora capsici*
- Biofumigation and solarization
- Experiments
 - Glucosinolate production
 - Effects of genetics, growing conditions, planting date
 - Bioassays
 - Petri dish, soil-based
 - High tunnel field study

High Tunnels



Solar

- ~~Un~~heated greenhouses
- Metal quonset frame
- Plastic cover
- Passive ventilation
- Soil-based production
- Simple
- Cheap





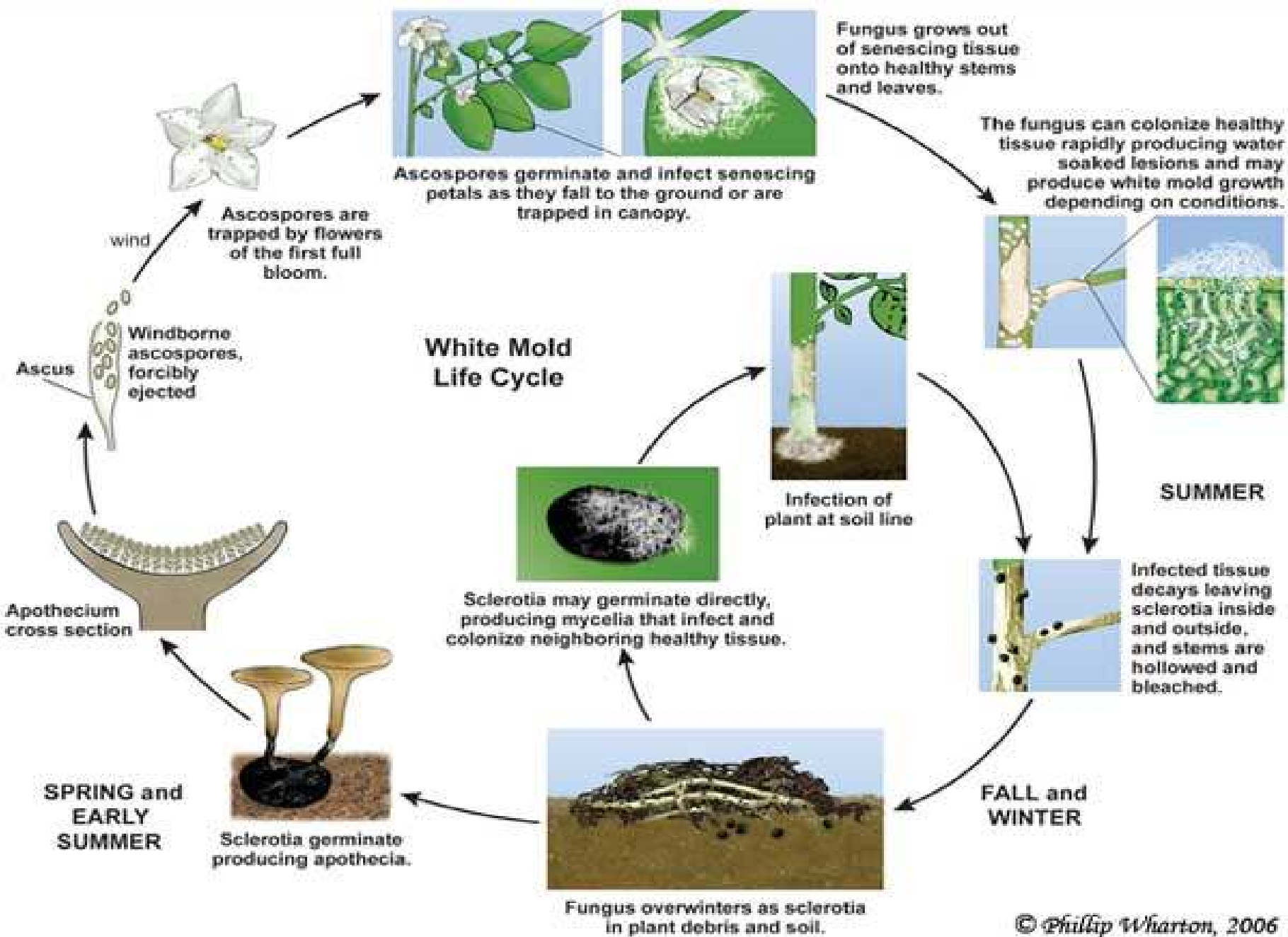


Sclerotinia sclerotiorum

- Thrives in cool, moist conditions
- Persists in soil as sclerotia



- White mold of lettuce
- Broad host range
- Problem in high tunnels



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Phytophthora capsici

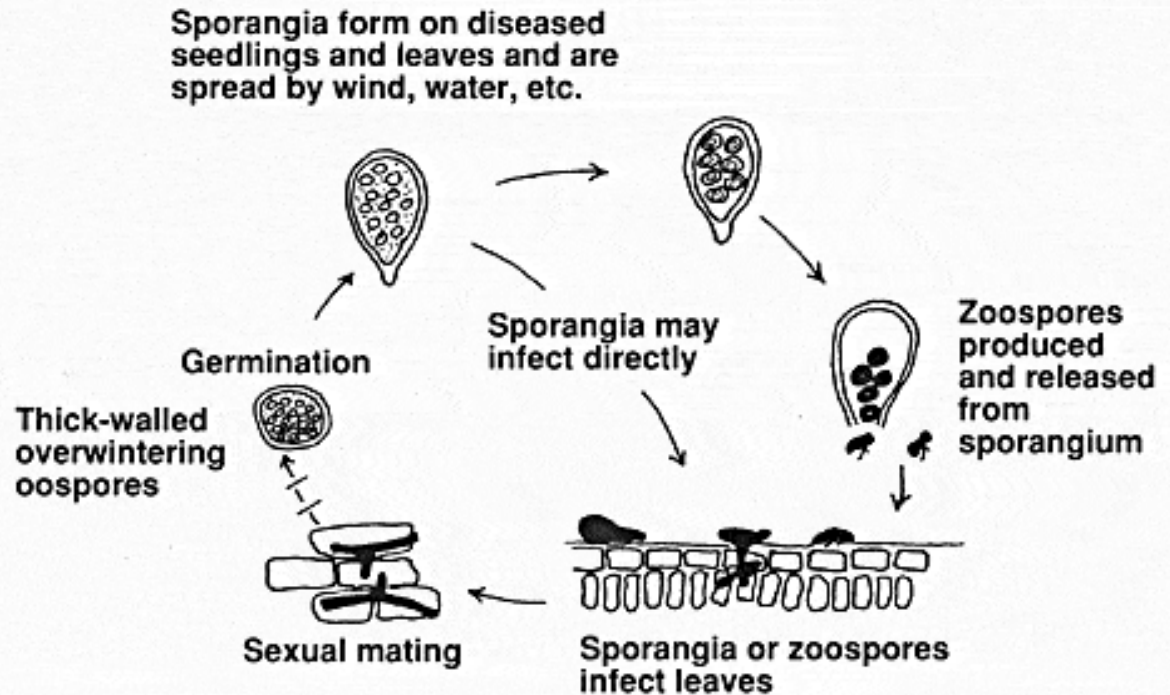
- soil-borne fungal pathogen of peppers, tomatoes, melons, cucumbers, and squash
- severe losses
- recent problem in Kentucky field vegetable production



Phytophthora capsici

- Grows best between 25-30°C
- Forms microscopic spores called sporangia
- Forms thick-walled overwintering oospores after mating

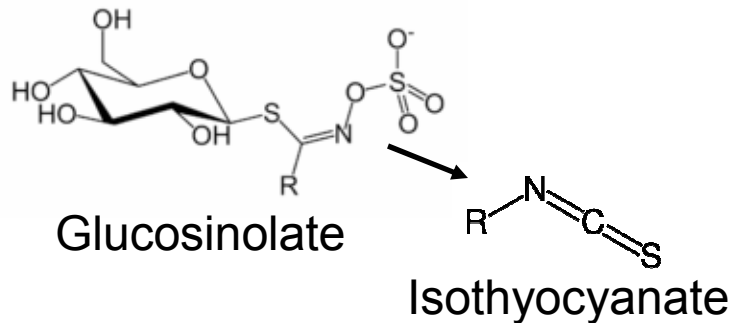
Fig. 8 Disease cycle of Phytophthora blight of pepper caused by *Phytophthora capsici*.



Organic management options

- Biofumigation

- General: Use of volatile chemicals produced by plants to control or suppress soil-borne pests and diseases
- Specific: Soil incorporation of brassicas, which contain glucosinolates that break down into volatile isothiocyanates



- Solarization

- Using clear plastic over the soil surface to capture solar energy, heat soil, and kill weed seeds and soil-borne pests and diseases



Glucosinolate production

- Objective
 - Identify suitable brassica cover crop
- Methods
 - Evaluated 47 brassica accessions for suitability as a cover crop in high tunnel (winter) and field (spring)
 - *Brassica juncea* (Indian mustard),
 - *B. napus* (Rape),
 - *B. carinata* (Ethiopian mustard),
 - *Eruca sativa* (Arugula)
 - Recorded survival, days to maturity, biomass production
 - Determined glucosinolate content of most promising accessions (Antonious et al 2009)

Materials & Methods

Ten accessions that demonstrated relative cold tolerance, rapid maturity, and superior biomass production were selected from 48 accessions of the National Germplasm Repository:

Ames 8660

Brassica juncea

Ames 8674

Brassica juncea

Ames 8709

Brassica juncea

Ames 8887

Brassica juncea

PI-120923

Brassica juncea

PI-603015

Brassica juncea

Pacific gold

Brassica juncea

PI-169083

Brassica napus

PI-633215

Eruca sativa

Ida gold

Brassica campestris

Indian mustard

Oil seed rape

Arugula

Field mustard

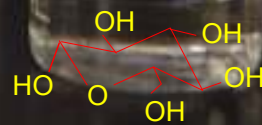
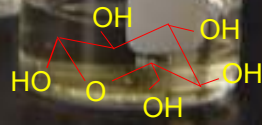
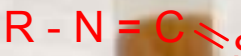
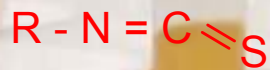
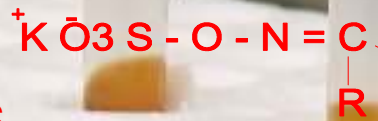
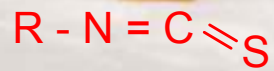
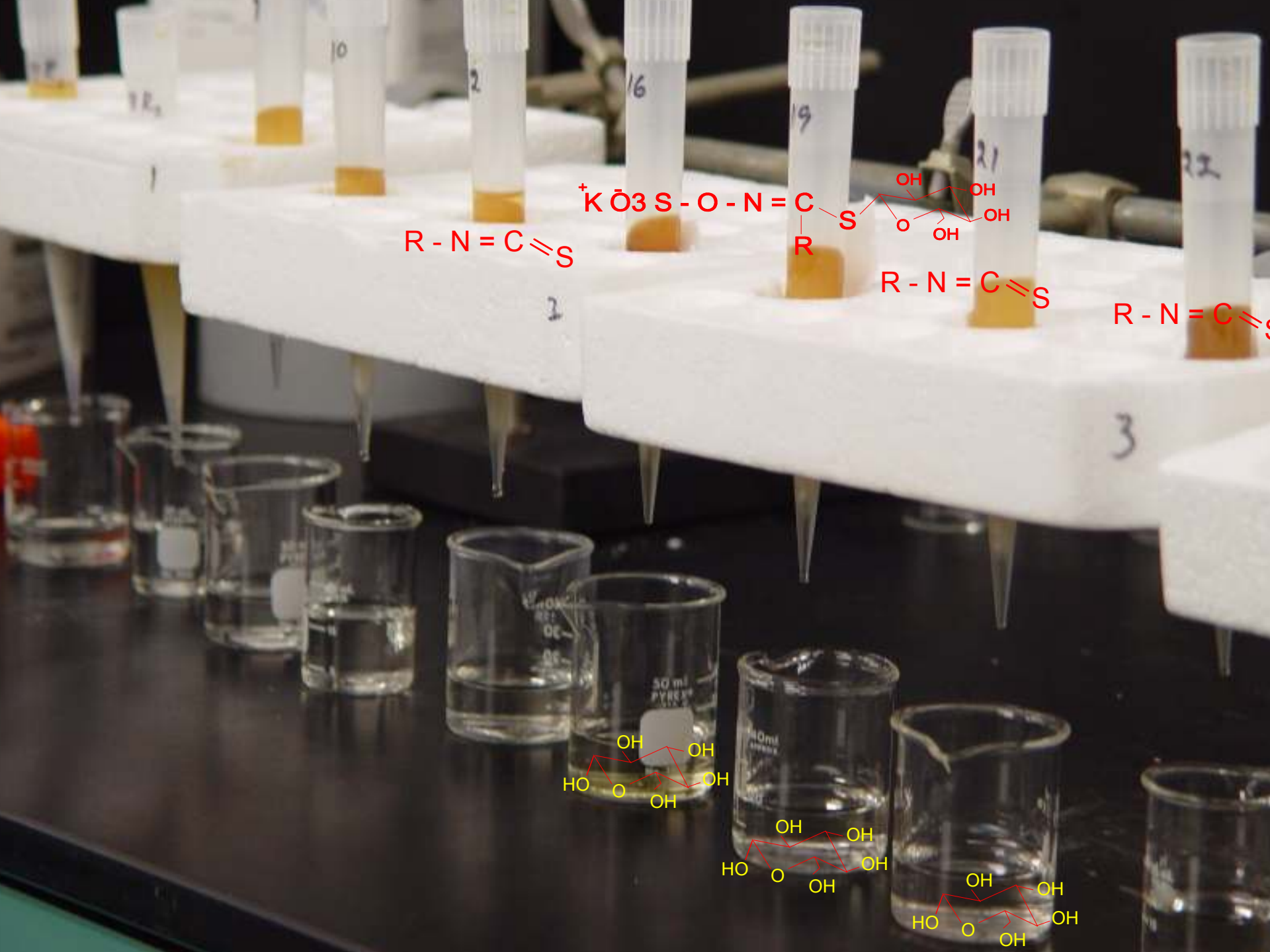
Greenhouse

High Tunnels

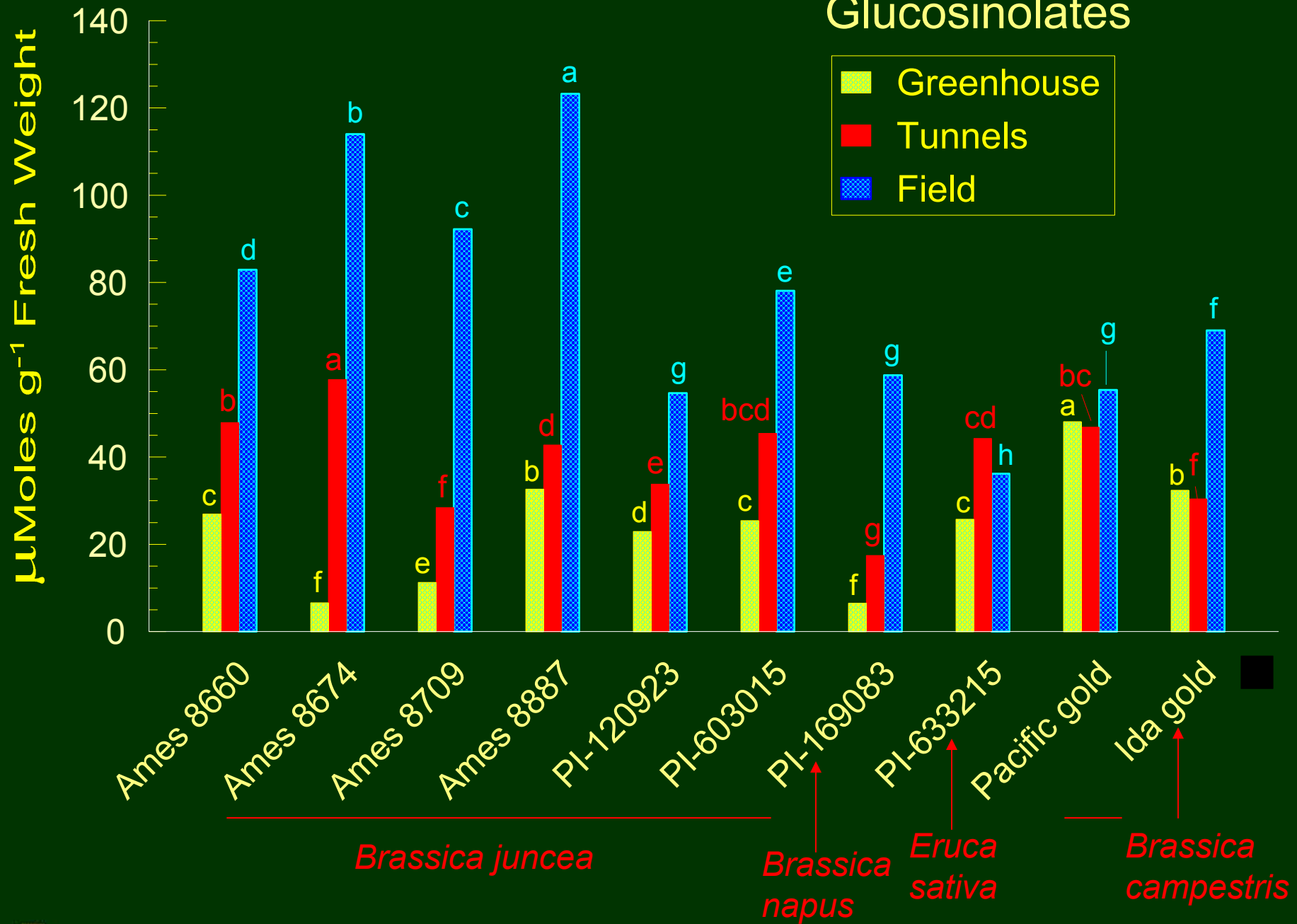
Field

Analysis of Glucosinolates

- ① Extraction using boiling methanol
- ① Clean-up using celite column
- ① Separation by anion-exchange resin “Sephadex”
- ① Hydrolysis with Thioglucosidase
- ① Quantification

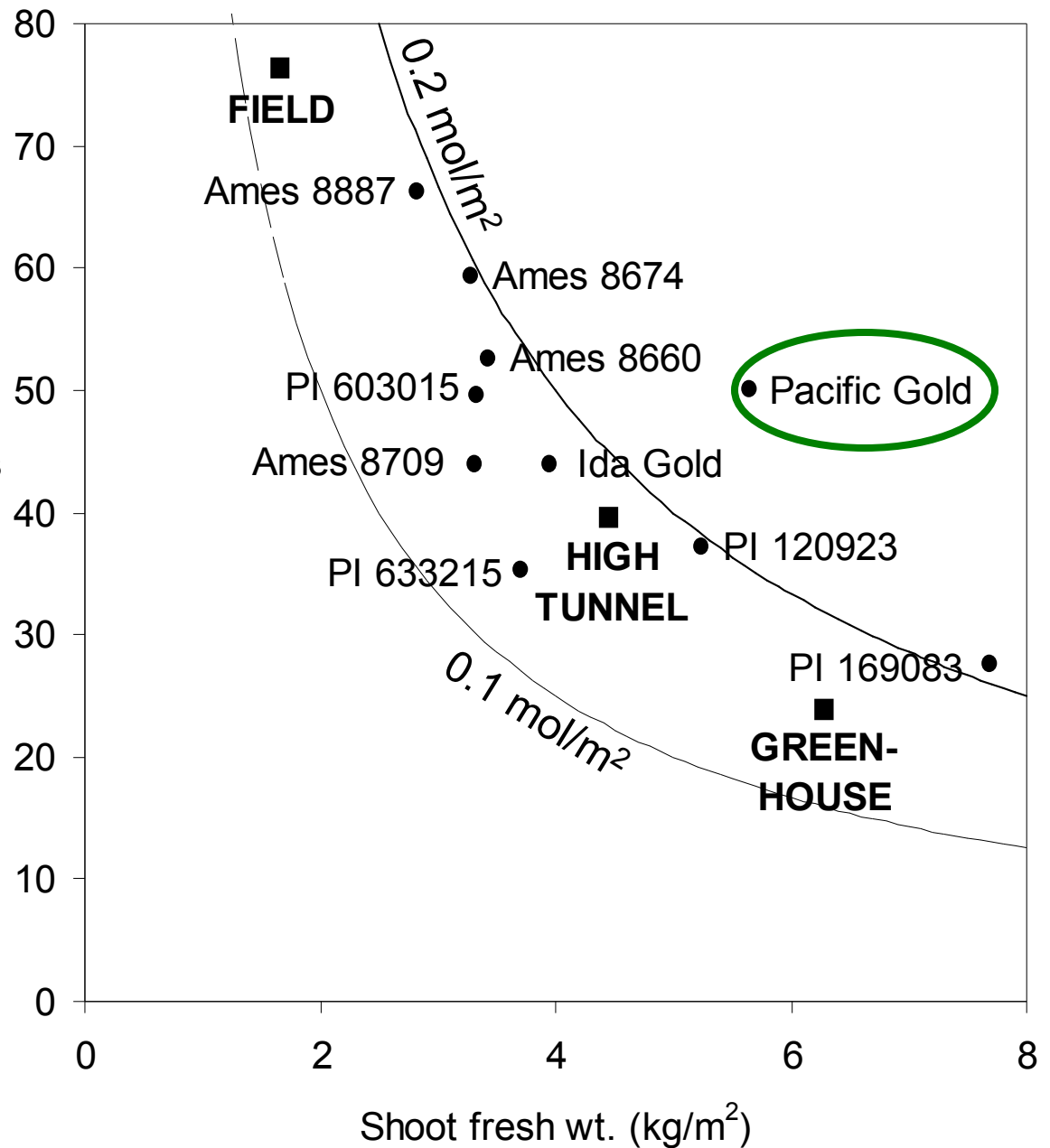


Glucosinolates



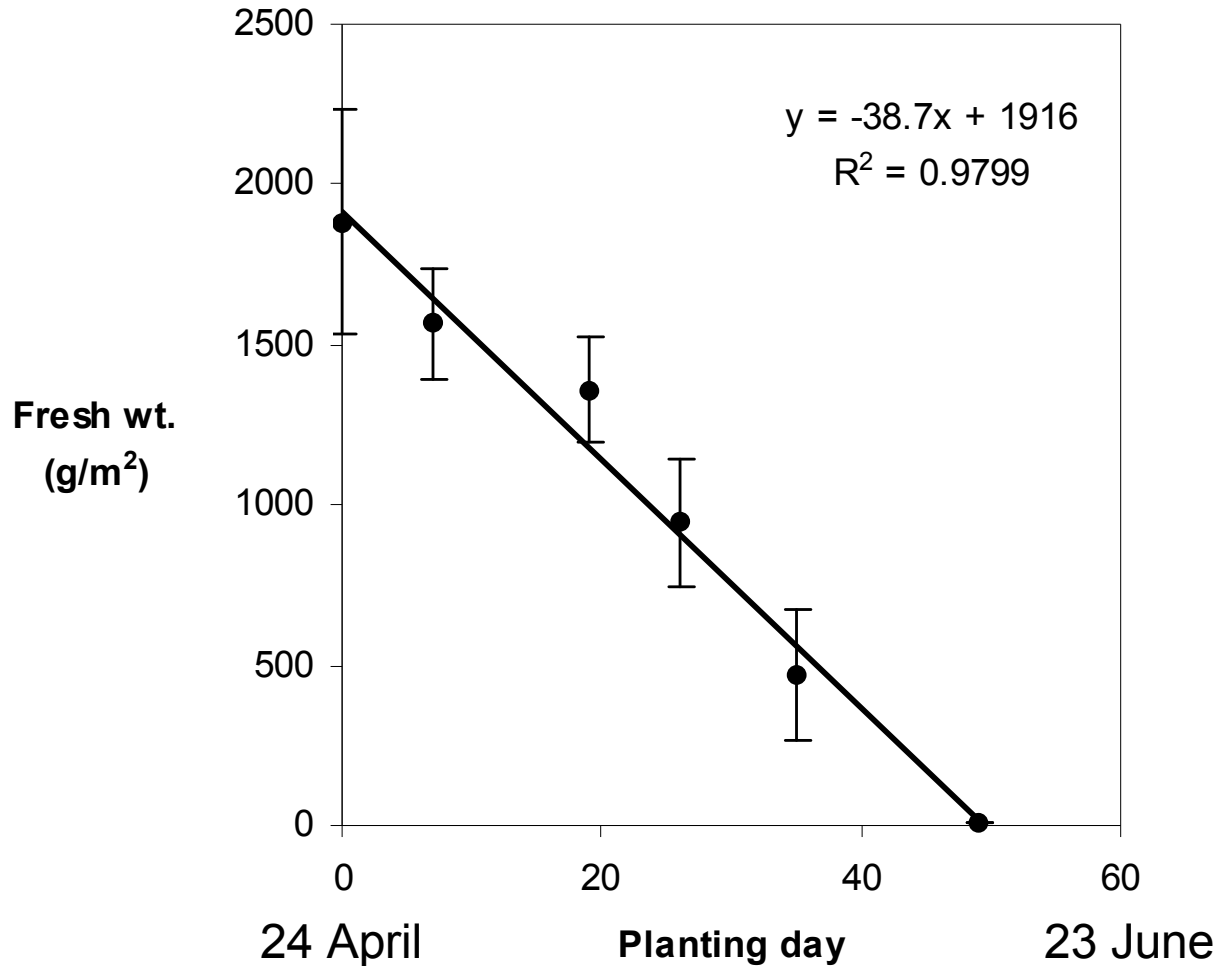
Glucosinolate production

Glucosinolates
($\mu\text{mol/g}$ fresh weight)



Effect of planting date on field biomass production

- Pacific Gold mustard direct-seeded in field at weekly intervals, weather permitting
- Above-ground biomass collected and weighted at flower initiation



Germination assay

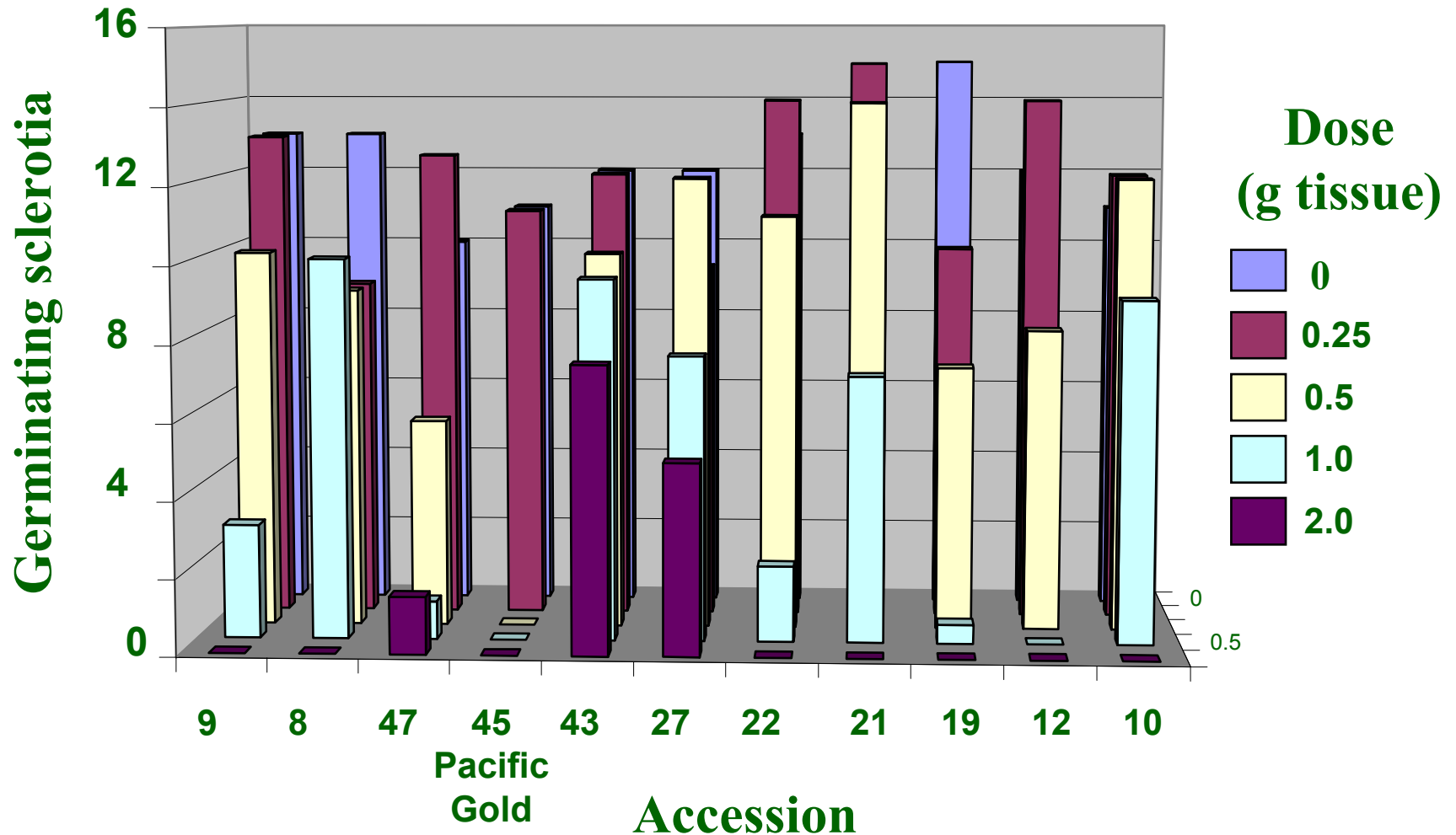
- MeOH tissue extract
 - 0, 0.25, 0.5, 1.0, and 2.0 g f.w.
- Placed in scintillation vials for 24 h with
 - equivalent amount of myrosinase
 - 15 *S. sclerotiorum* sclerotia
 - sterile soil (4 mL total)
- Plated onto Petri dishes with 40 mL sterile soil for 6 wk germination at 16 °C



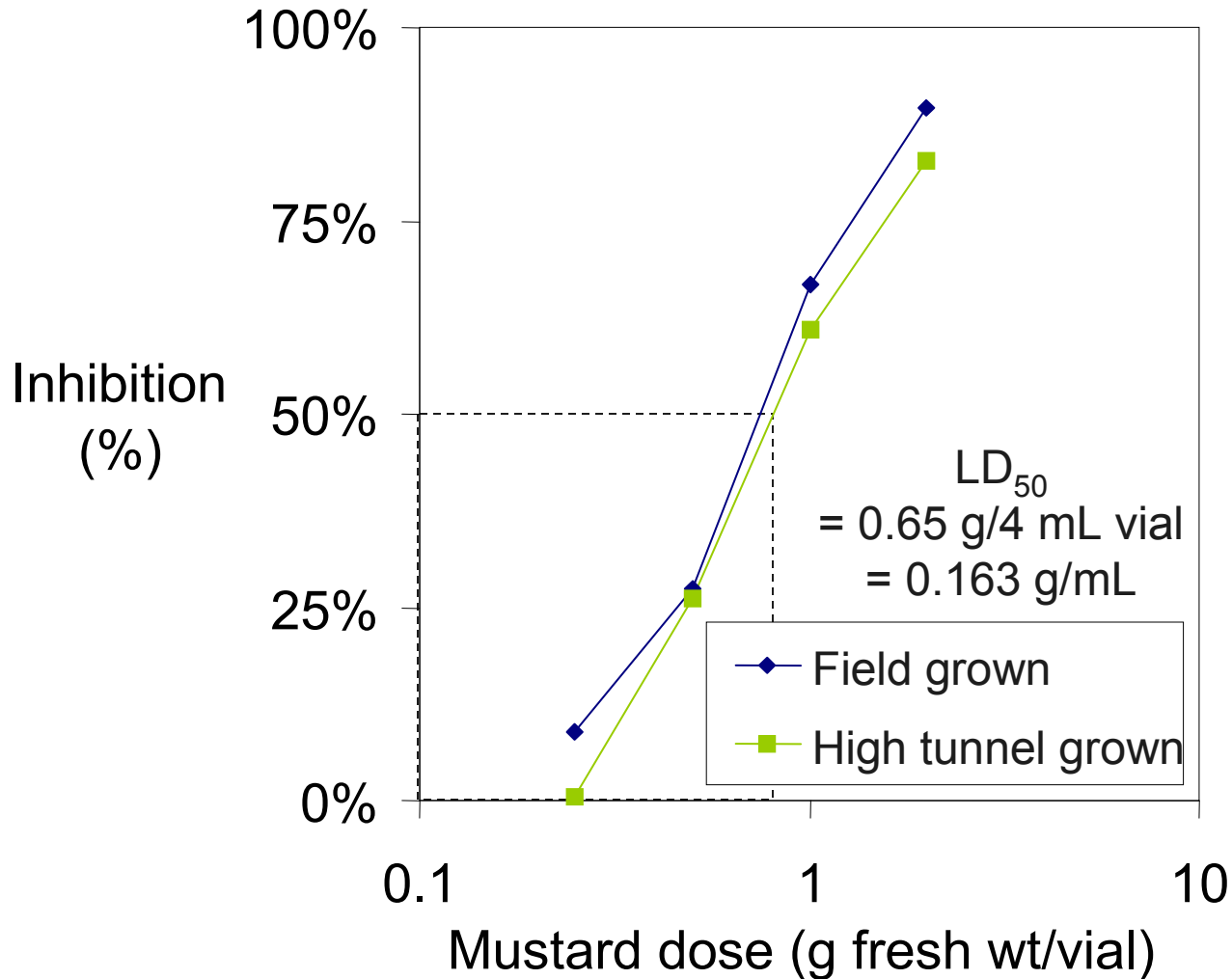
ID	Accession	Species
8	Ames 8657	<i>B. juncea</i>
9	Ames 8660	<i>B. juncea</i>
10	Ames 8674	<i>B. juncea</i>
12	Ames 8754	<i>B. juncea</i>
19*	PI 120923	<i>B. juncea</i>
21	PI 180417	<i>B. juncea</i>
22	PI 603015	<i>B. juncea</i>
27*	PI 169083	<i>B. napus</i>
43*	PI 633215	<i>Eruca sativa</i>
45	Pacific Gold	<i>B. juncea</i>
47*	Ida Gold	<i>B. campestris</i>

*Field-grown accession tested in addition to high tunnel grown accession

Results: *S. sclerotiorum* Germination (of 15) After Exposure to Mustard Extracts



Results: *S. sclerotiorum* Inhibition



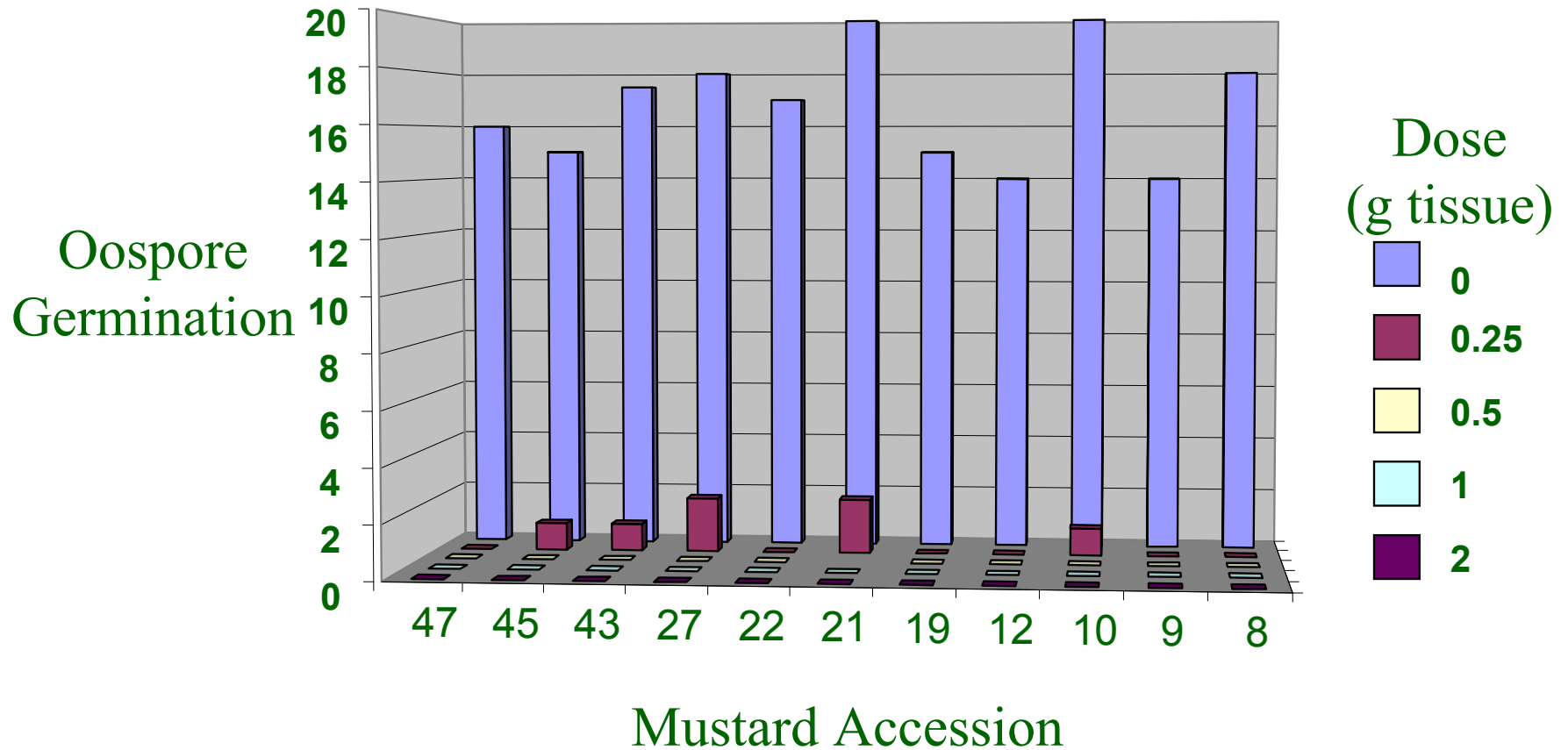
Phytophthora capsici assay

- MeOH tissue extract
 - 0, 0.25, 0.5, 1.0, and 2.0 g f.w.
- Placed in dark sealed culture flasks for 2 wk with
 - equivalent amount of myrosinase
 - 20 *P. capsici* oospores
 - sterile soil (4 mL total)
- Oospore germination tallied

ID	Accession	Species
8	Ames 8657	<i>B. juncea</i>
9	Ames 8660	<i>B. juncea</i>
10	Ames 8674	<i>B. juncea</i>
12	Ames 8754	<i>B. juncea</i>
19*	PI 120923	<i>B. juncea</i>
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45	Pacific Gold	<i>B. juncea</i>
47*	Ida Gold	<i>B. campestris</i>

*Field-grown accession tested in addition to high tunnel grown accession

Results: *P. capsici* Oospore Germination (of 20) After Exposure to Mustard Extracts

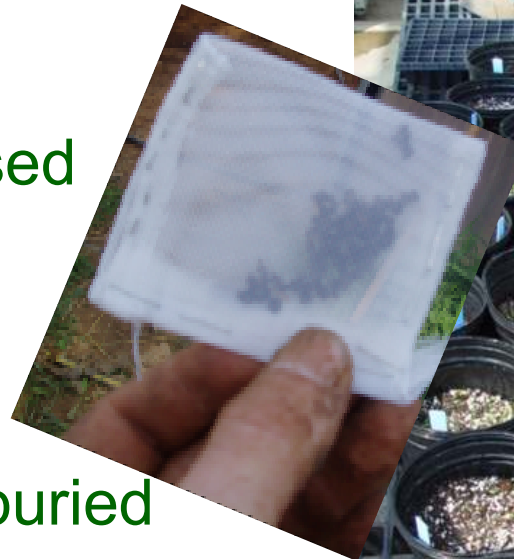


Petri dish assay conclusions

- 24 h exposure to extracts from 0.65 g mustard tissue per 4 mL vial (0.163 g/mL) reduced *S. sclerotiorum* sclerotial germination by 50%
- 'Pacific Gold' gave the best reduction in *S. sclerotiorum* germination
- *P. capsici* oospore germination was completely inhibited by exposure to extracts > 0.5 g mustard tissue / 4 mL substrate (0.12 g/mL).
- *P. capsici* oospores are more sensitive to mustard extracts than *S. sclerotiorum* sclerotia.
- More research is needed to apply these results in the field.

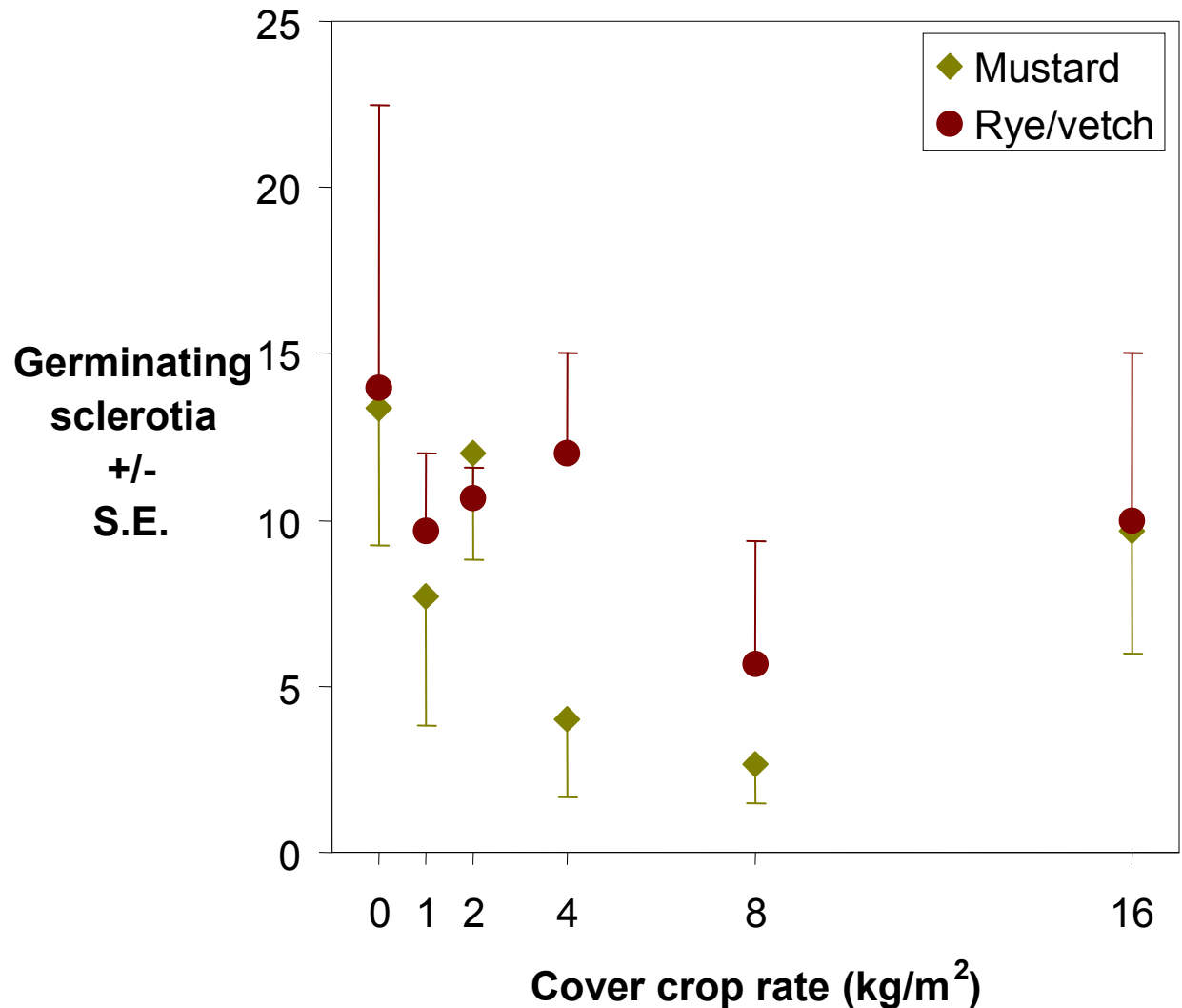
Greenhouse Potting Soil Assay: Methods

- Pacific Gold mustard greens and rye/vetch cover crop harvested and macerated at first flower, 06/12/08
- Cover crops separately incorporated into peat-based potting mix, 15 cm deep
 - 0, 1, 2, 4, 8 and 16 kg m⁻²
- 3 replicates, completely randomized
- Mesh bag of 50 sclerotia buried in each pot for 4 days
- Sclerotia plated onto Petri dishes with 40 mL sterile soil for 6 wk germination at 16 °C



Greenhouse Potting Soil Assay: Results

- Low germination
- No significant effects:
 - Cover crop
 - Rate (significant if highest rate excluded)



Soil-based *P. capsici* assay: Methods

- Split plot design:
 - Main effect – Presence/Absence of Organica Plant Growth Activator
 - Blend of micro-organisms intended to promote seedling and plant establishment
 - Sub-plots – Presence/Absence of glucosinolates from green manure at 6 incorporation rates
 - Presence: Biofumigation with Pacific Gold mustard greens
 - Absence: Incorporation of rye (*Secale cereale*)/vetch (*Vicia villosa*) cover crop to add organic matter but no glucosinolates
 - Rates: 0, 13.5, 27, 54, 108 and 216 g/L (0-16 kg/m²)



Methods

- 16 liters of organic planting mix thoroughly mixed with 80 ml of *Phytophthora capsici* inoculant (0.5% volume/volume) and divided into 12 equal portions



Methods

- Green manure harvested at first flower, finely chopped and mixed into planting media at 0, 13.5, 27, 54, 108 or 216 g/L





- Each mixture randomly assigned to fill two of 24 evenly spaced 65 ml cells in each of the 10 seedling plug trays
- A pre-germinated yellow squash (*Cucurbita pepo*) seed planted in each cell

Methods

- Trays randomly assigned to flood irrigation with untreated tap water or tap water treated with 4.2 g/l of Organica Plant Growth Activator



Methods

- Trays were partially submerged in the liquid to ensure thorough drenching and incubated for 5 days at 26°C .



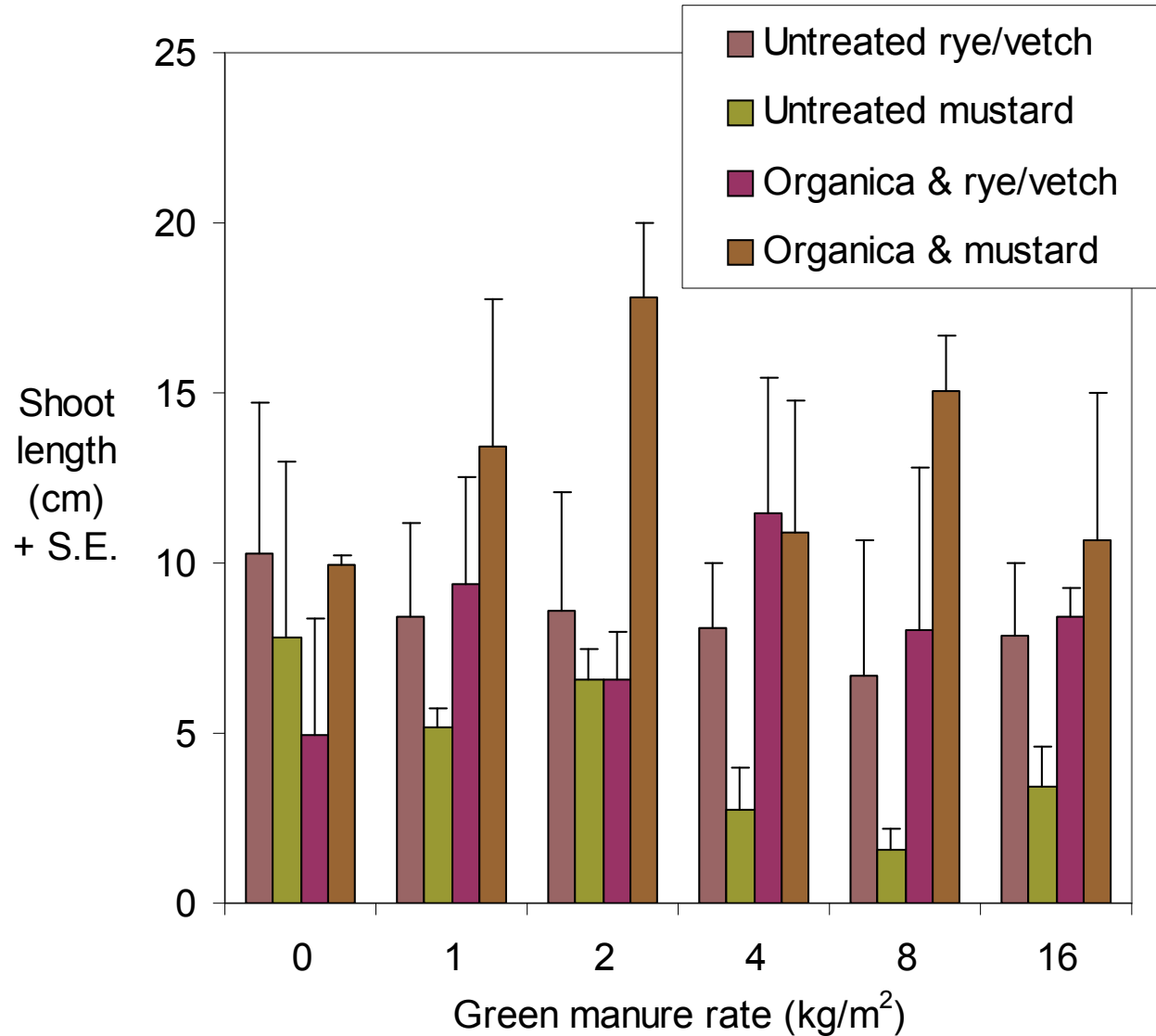
Methods

- Seedling survival and shoot length were recorded for each treatment combination in each of 5 replicates



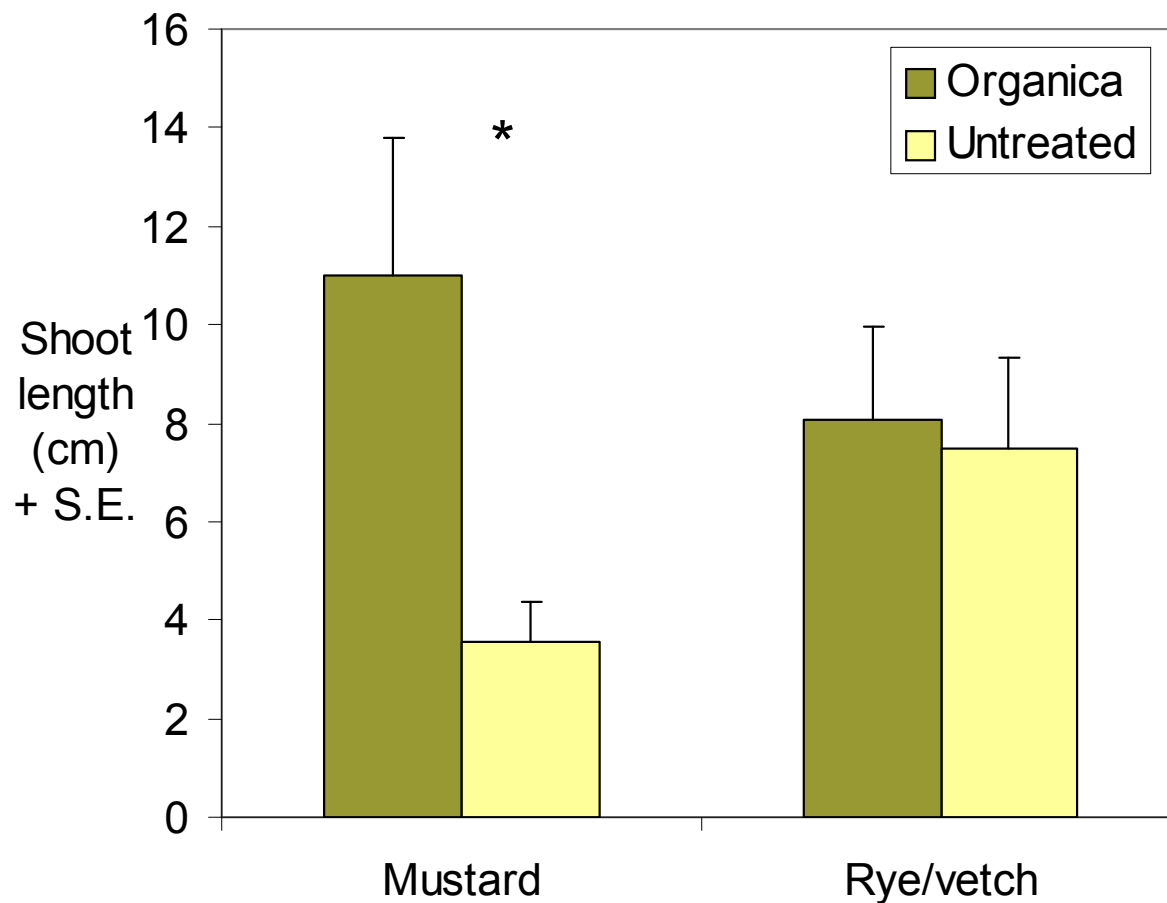
Results

- No significant effect:
 - Cover crop
 - Rate



Results

- Interaction:
 - Cover crop x Organica
- Superior seedling growth with combination of mustard and Organica
- Repeatable?



See video:

<http://organic.kysu.edu/SclerotiniaVideo.shtml>

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- Southern SARE

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