

# X-RAY IMAGING OF ROOT SYSTEMS INFECTED WITH ENDOPARASITIC NEMATODES

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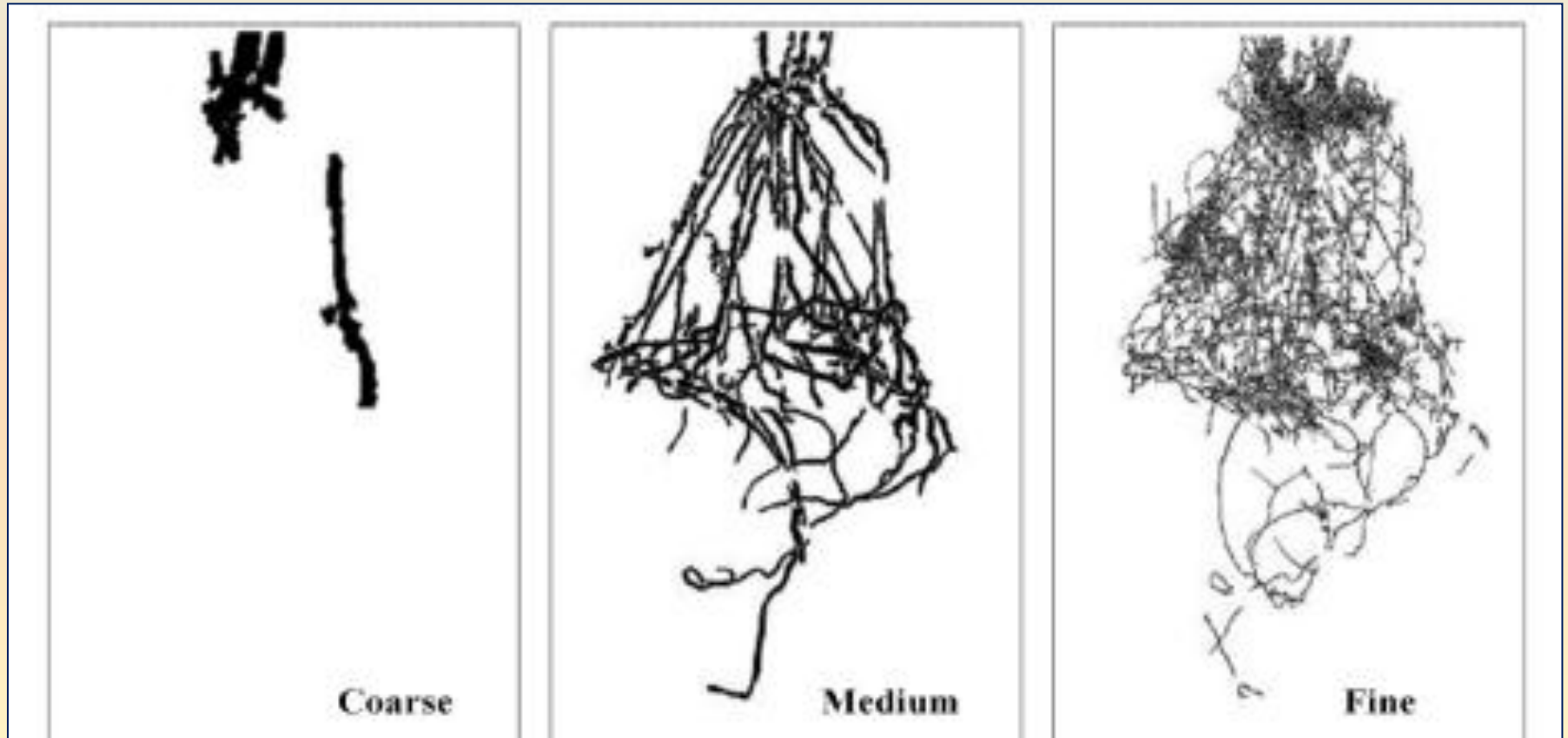
# OBJECTIVES

- ❖ To develop a new tool for characterization and screening of nematode resistance in cotton based upon low energy X-ray imaging technology.
- ❖ To develop and demonstrate new protocols for cotton inoculation, cotton growth monitoring, cotton/nematode interactions, cotton root system infection characterization, and cotton resistance to root-knot nematode (*Meloidogyne incognita*) and reniform nematode (*Rotylenchulus reniformis*) by means of low energy X-ray imaging systems.

# PSC growing System: expanded polystyrene (EPS) beads (0.5-1 mm diameter)



# Separation of root system into size classes using PSC RhizoTraits software.



## Two approaches to X-ray analysis of root systems infected with nematodes:

1. Plants were started in seedling pouches and nematodes or eggs were pipetted onto the roots. After infection, plants were taken to PSC and grown in EPS substrate (mostly tested with *Meloidogyne incognita*).

**RESULTS:** This approach had limited success due to the density of roots in the PSC system, which sometimes obscured galls and egg masses. Second-generation infection was very limited due to the high porosity of the EPS substrate. Results in pouches varied with the plant used (cotton, soybean, sunflower, cantaloupe).

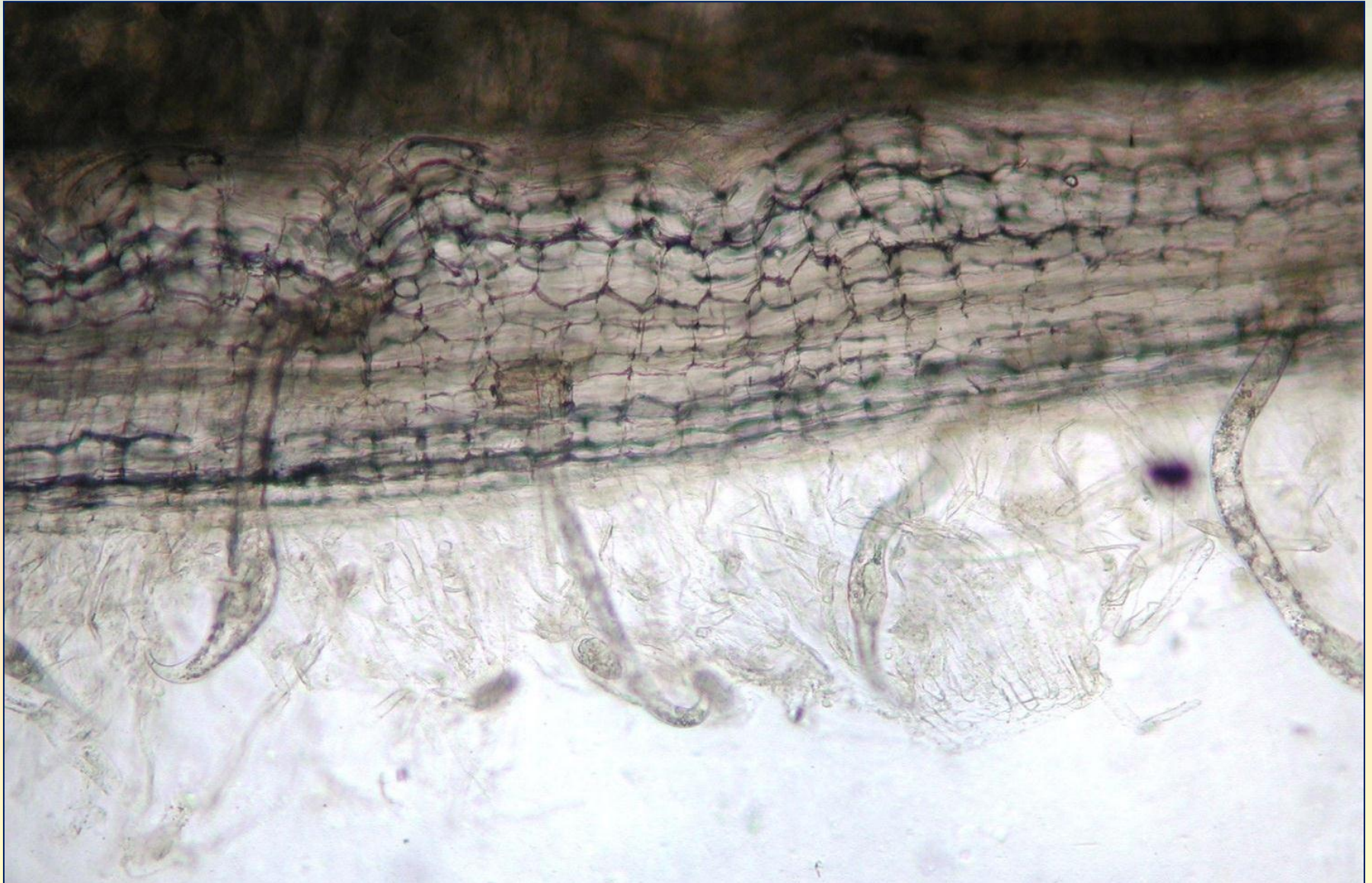
2. Plants were grown in the greenhouse in mineral soil and infected by standard methodologies. Root systems were harvested, cleaned carefully, then taken to PSC for X-ray analysis (mostly tested with *Rotylenchulus reniformis*).

**RESULTS:** This approach was quite successful and has the potential to yield accurate data about nematode establishment and reproduction in relation to root length and quality.

# Early soybean growth in seedling pouches (see Omwega et al. 1988, Atamian et al. 2012).



Young *Rotylenchulus reniformis* females on a cantaloupe root in a seedling pouch.



# Non-destructive X-ray analysis of root architecture and nematode development in the PSC System.

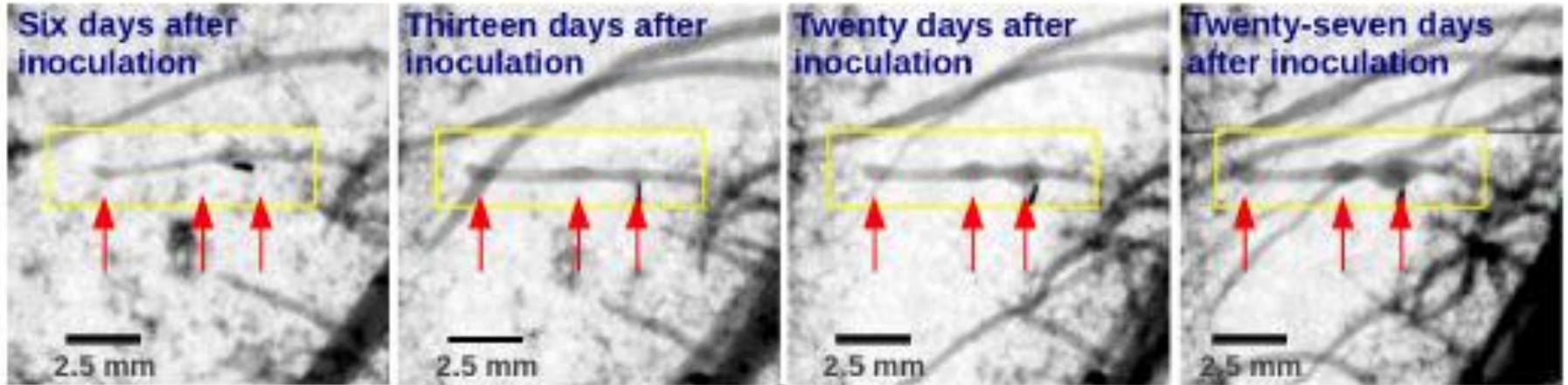


Figure 5: Time Sequence Images of Root-Knot Nematode Feeding Site Development In Cotton

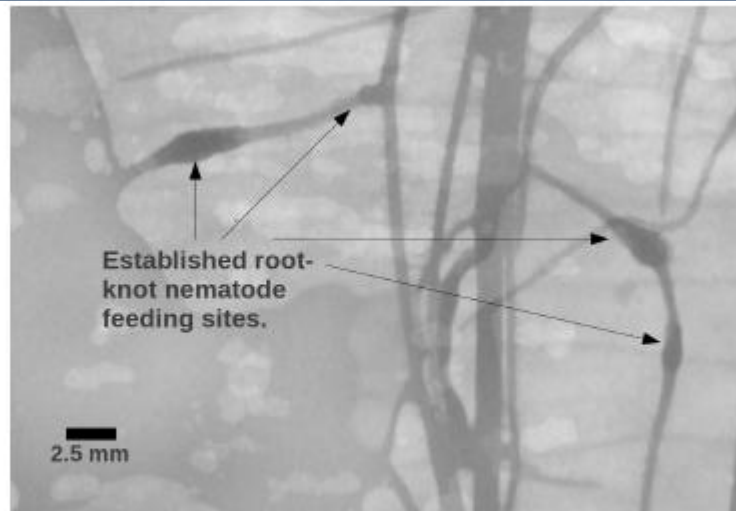
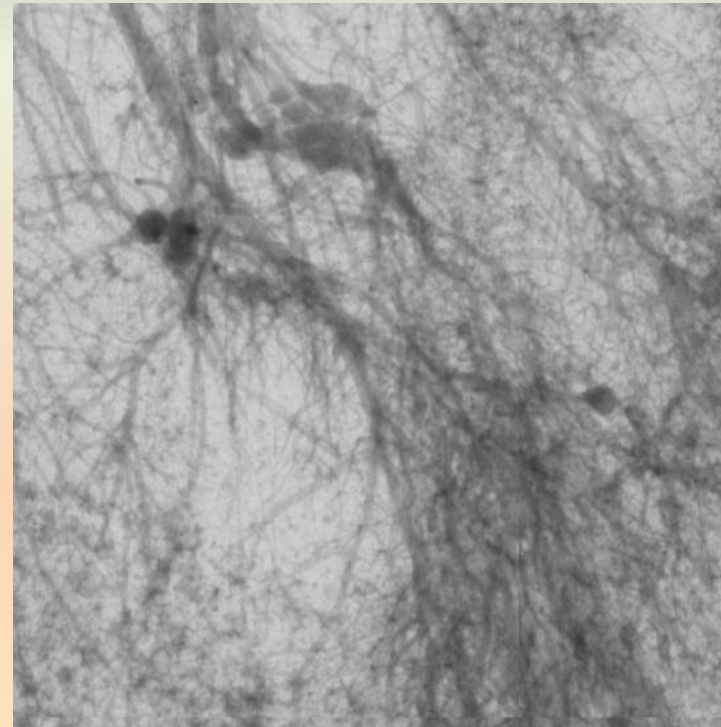
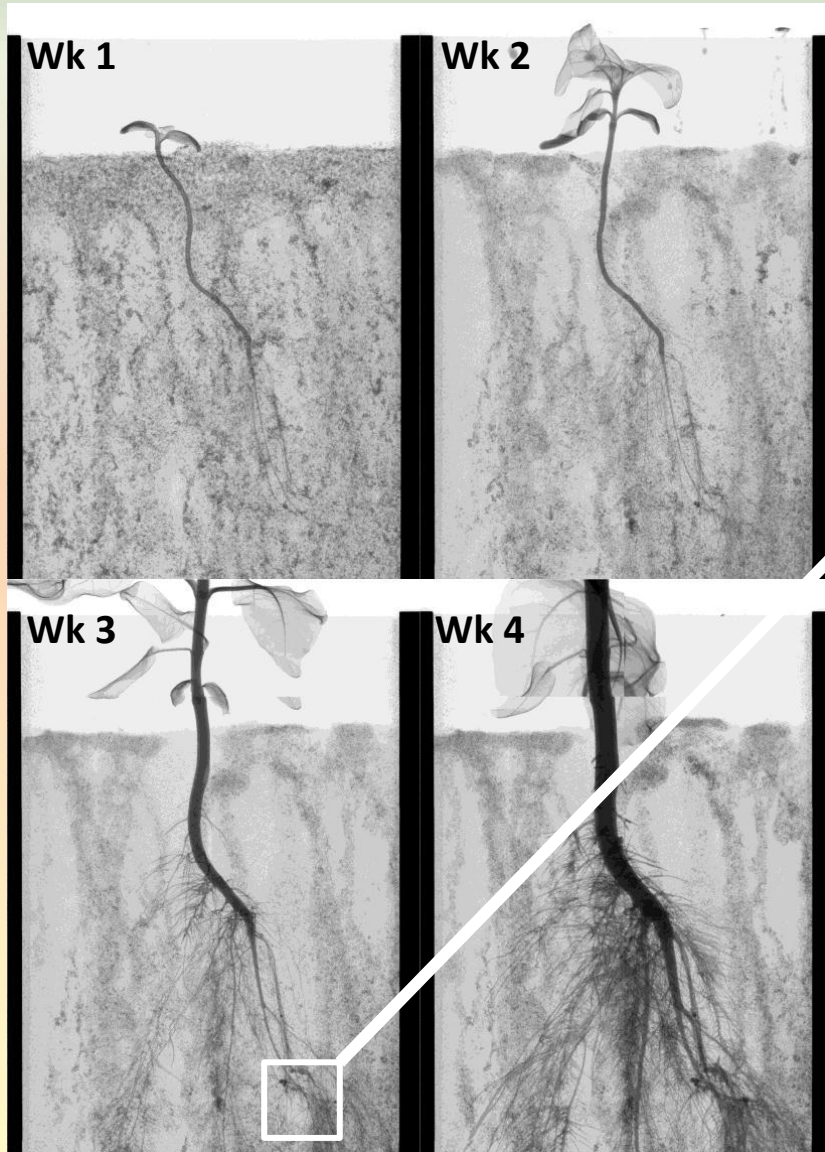


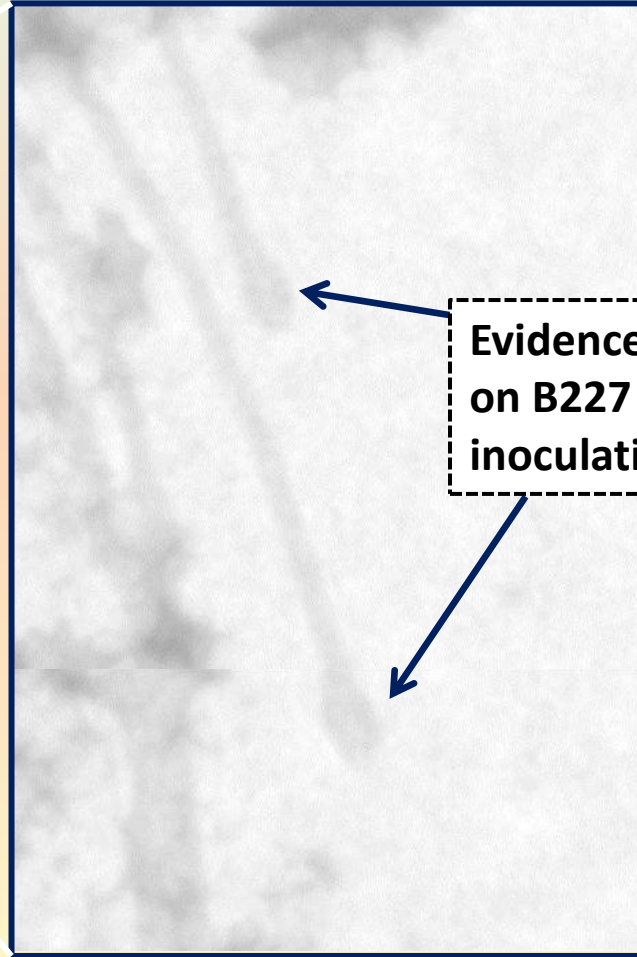
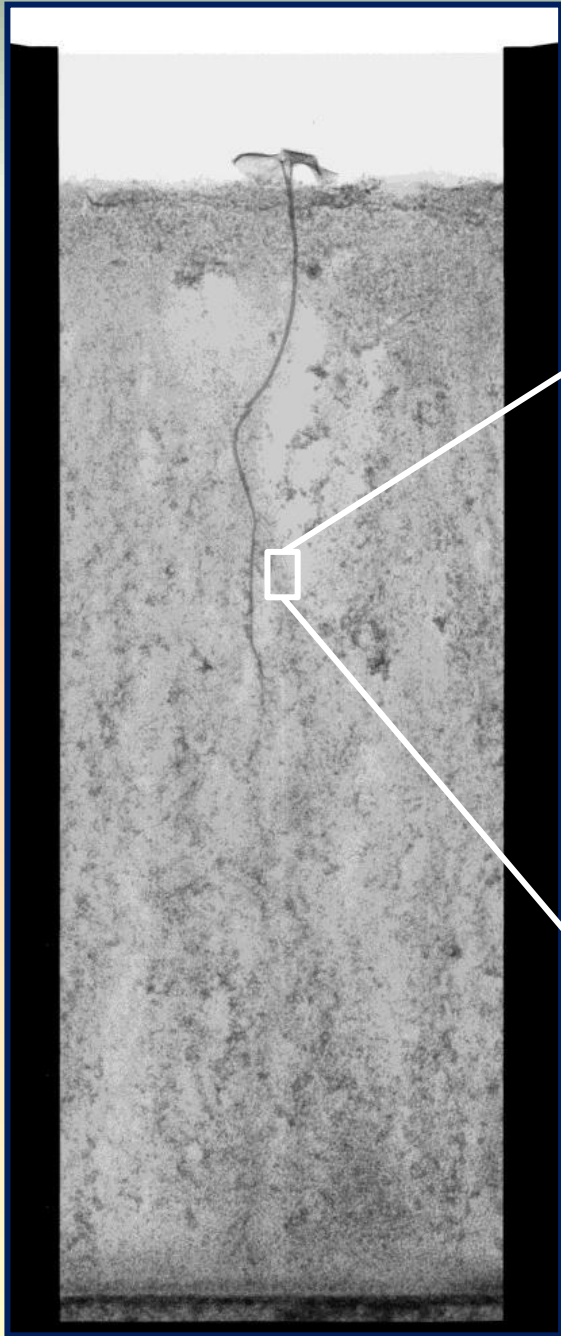
Figure 6: Soft-tissue X-ray Image of Root-knot Nematode Galling Along Sunflower Root Taken in Germination Pouch Six Days After Inoculation with 1200 Second Stage Juveniles.



# Root-knot galling on sunflower after three weeks.

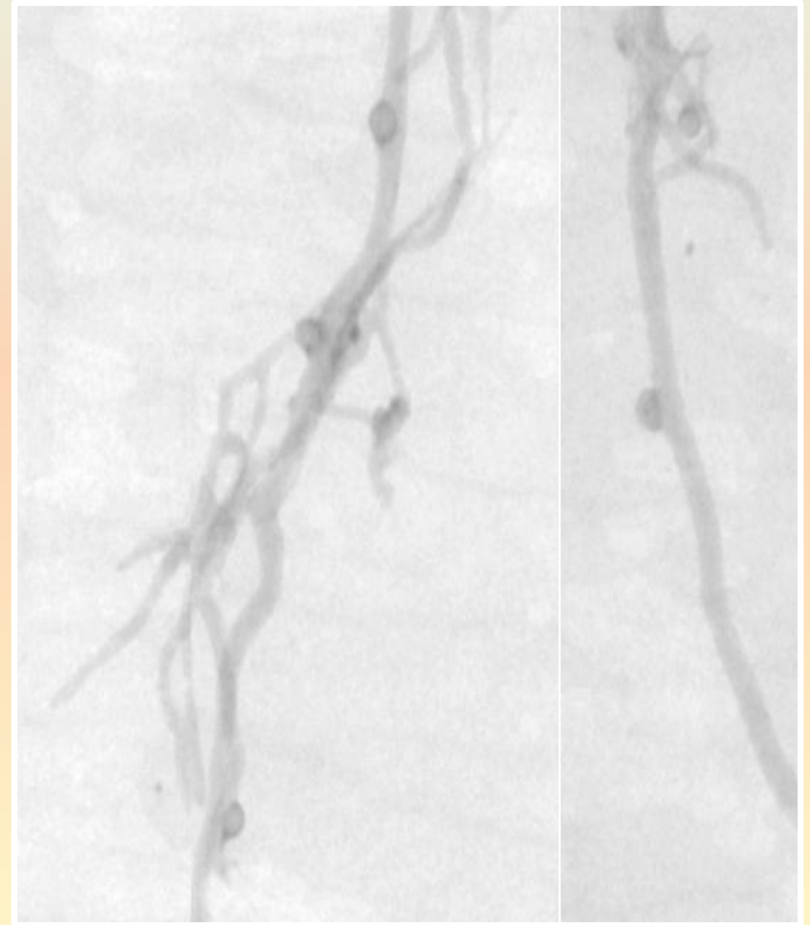


**X-ray image of a root-knot-inoculated  
B227 cotton plant, PSC system.**



**Evidence of early galling  
on B227 six days after  
inoculation with 1200 J2.**

# LM and X-ray imaging of *R. reniformis* egg masses, greenhouse system



# Qualitative assessment of susceptibility of several cotton selections to reniform nematode

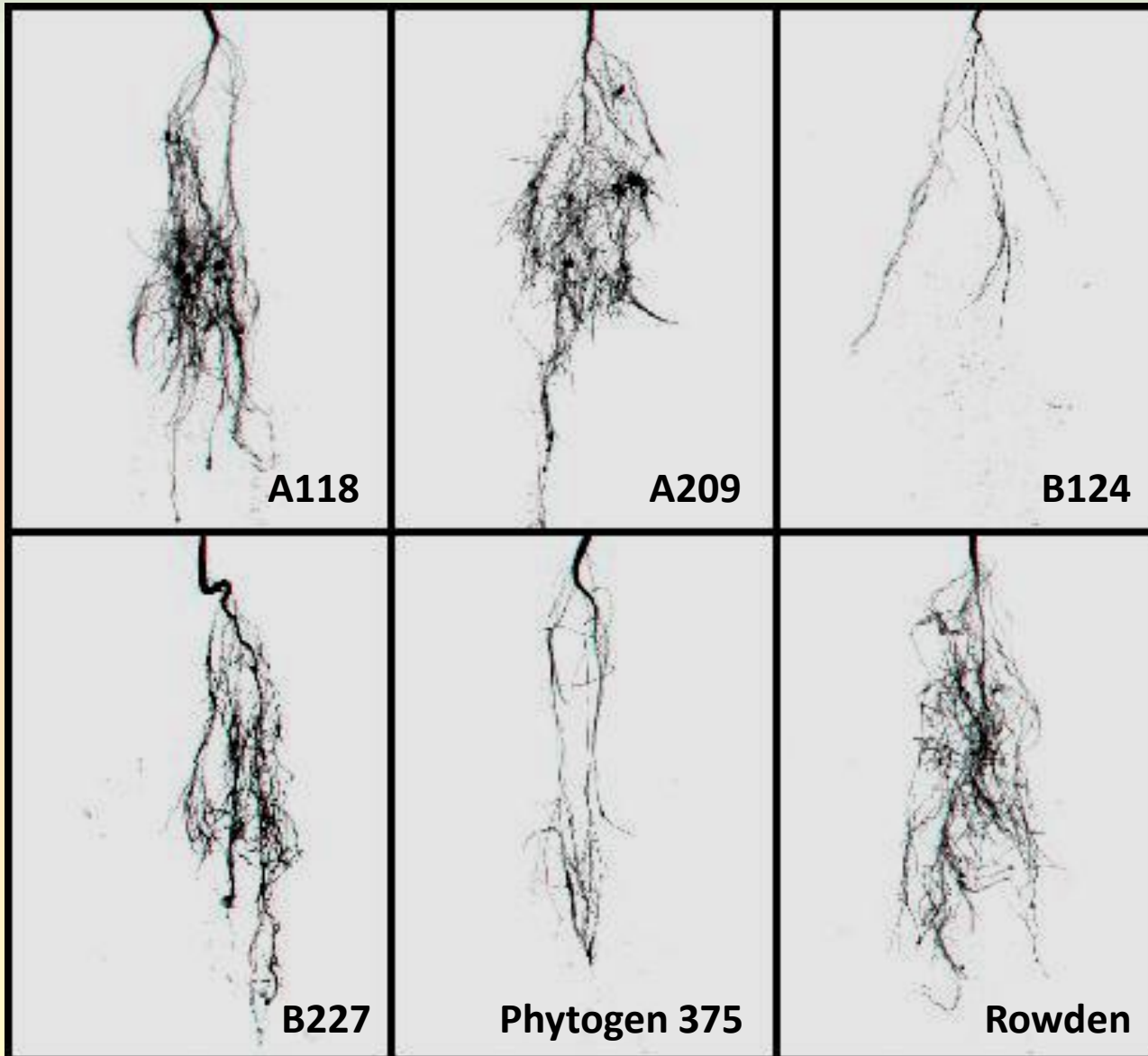
Selection	Rating
GA2004	+++++
Phytogen 375WNR	+++
Rowden	+++
A118	++++
A209	++++
B124	+++++
B227	+++++

Evaluated by E. Bernard,  
University of Tennessee

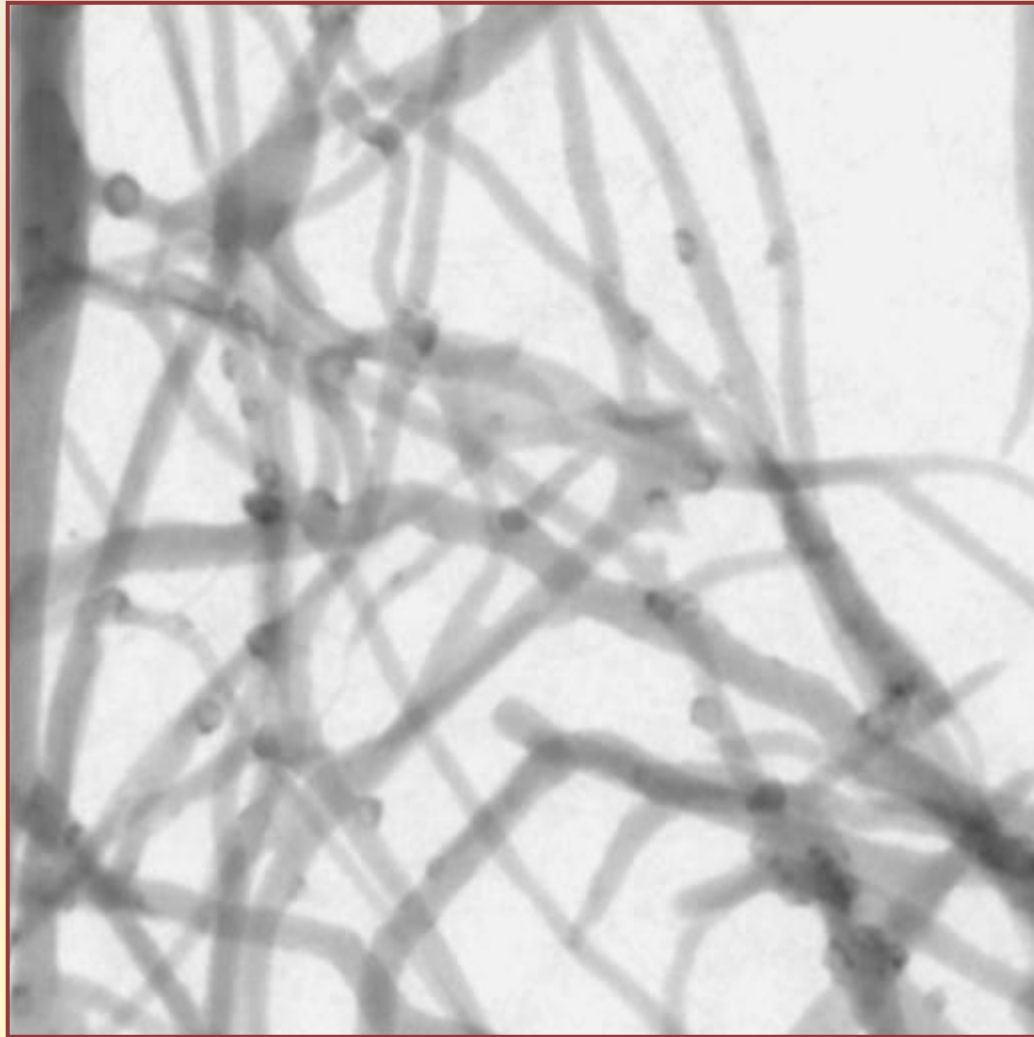
Selection	Assessment
A118	Slightly less susceptible
A209	Very susceptible
B124	Moderately susceptible
B227	Slightly, slightly less susceptible

Evaluated by D. Weaver,  
Auburn University.

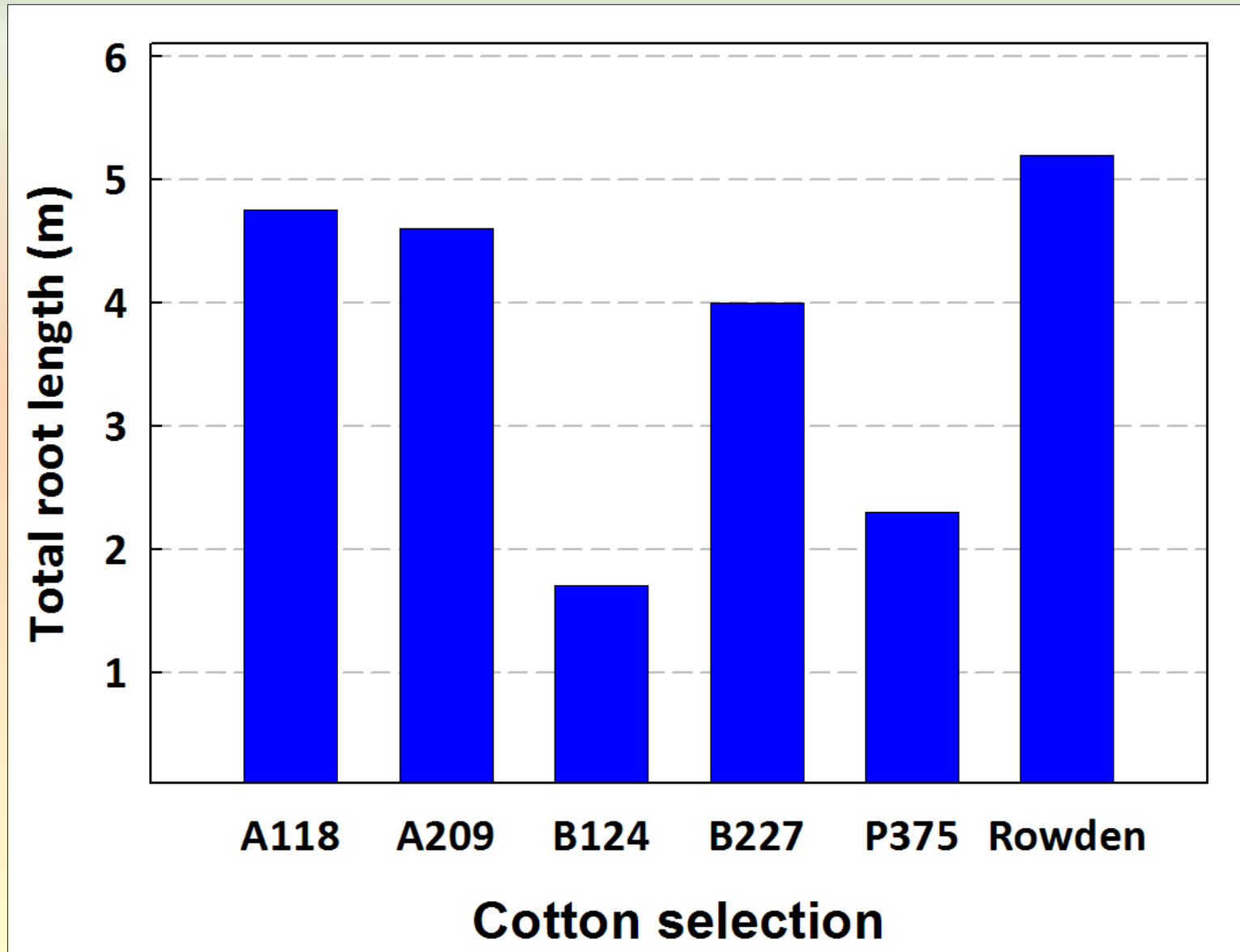
# Root system distribution of six cotton selections, greenhouse system.



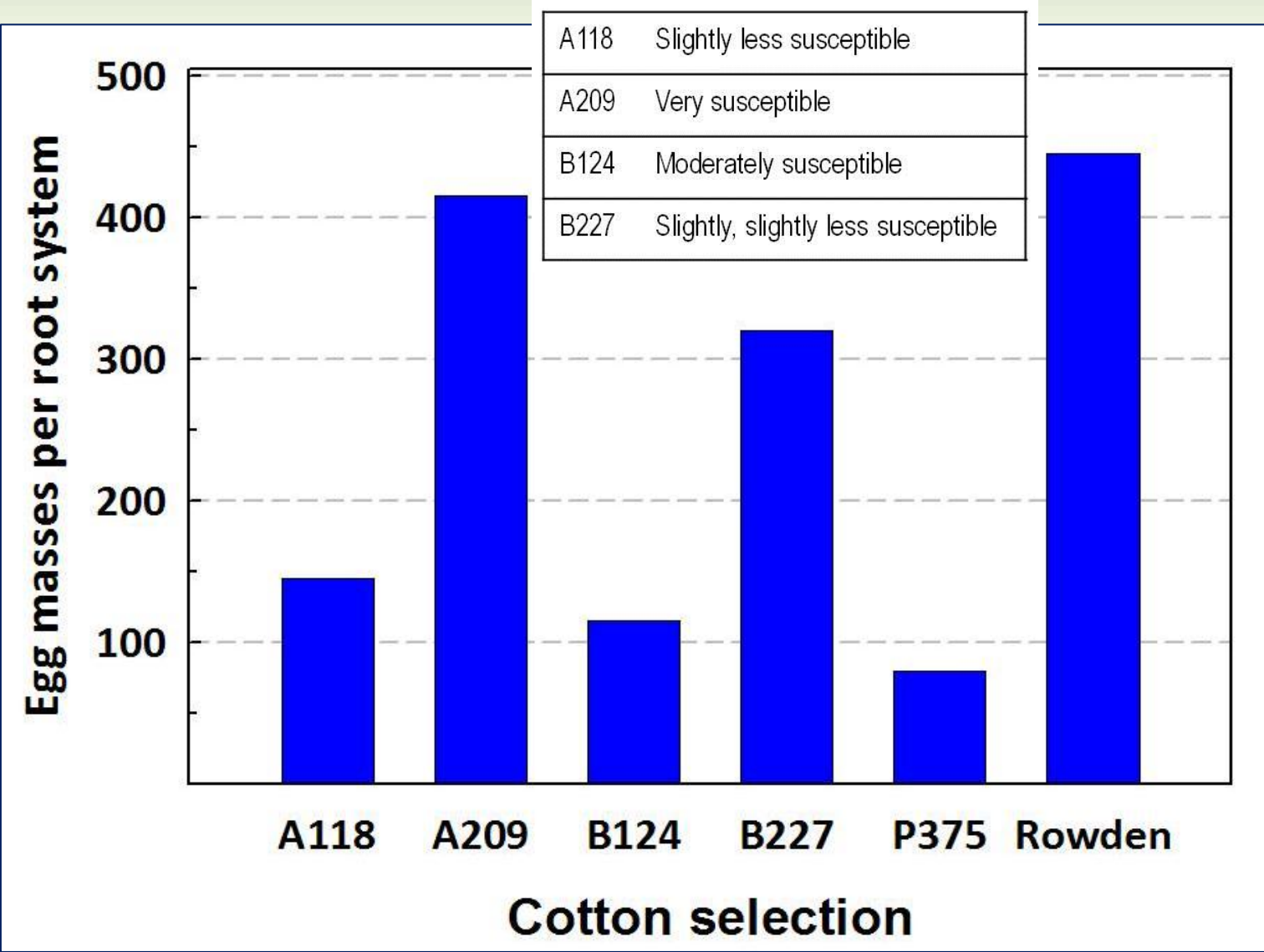
**X-ray image of reniform egg masses on A209 cotton roots grown in greenhouse.**



# Total root length of six cotton plants.



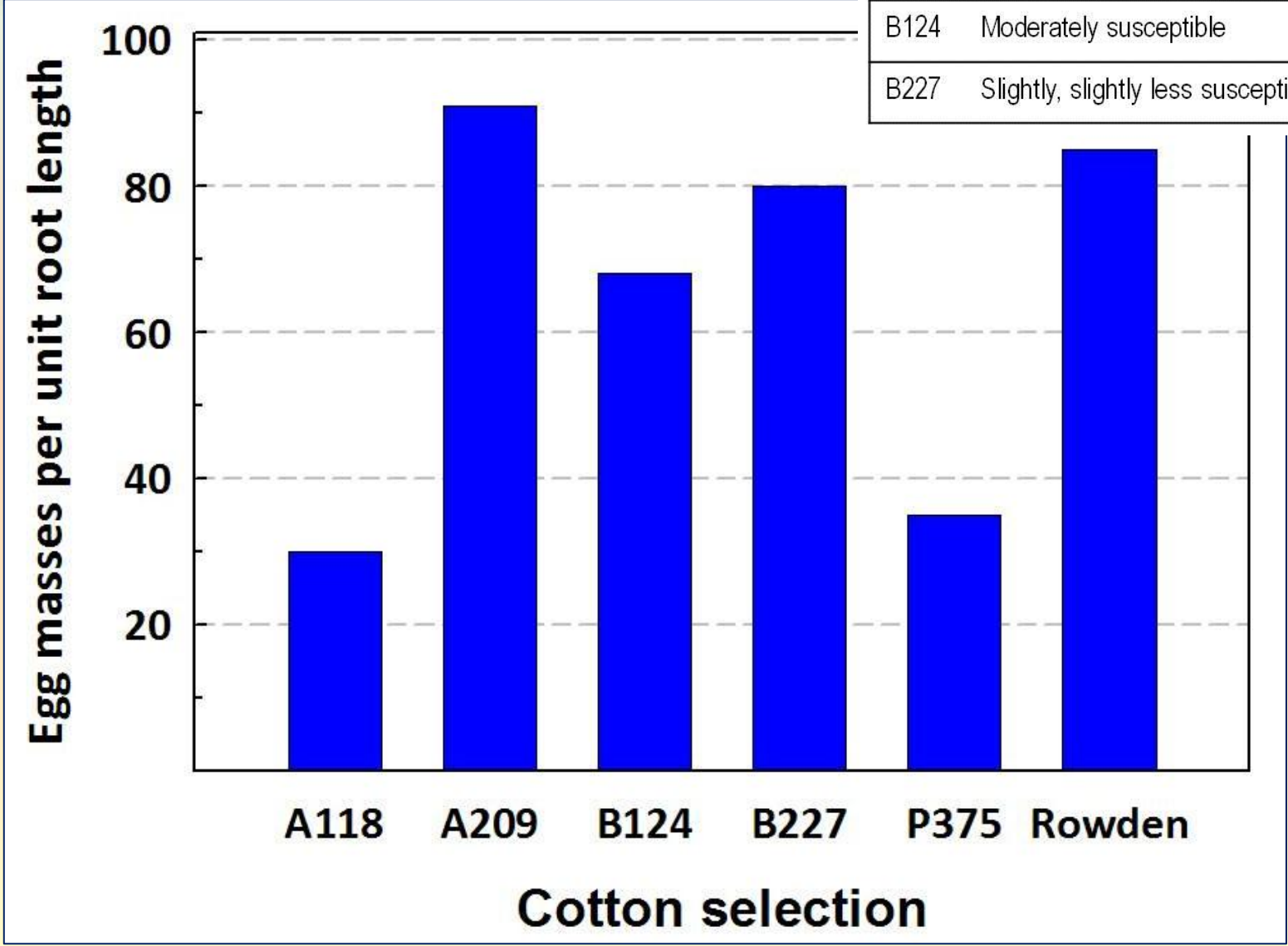
# Reniform nematode egg masses per plant



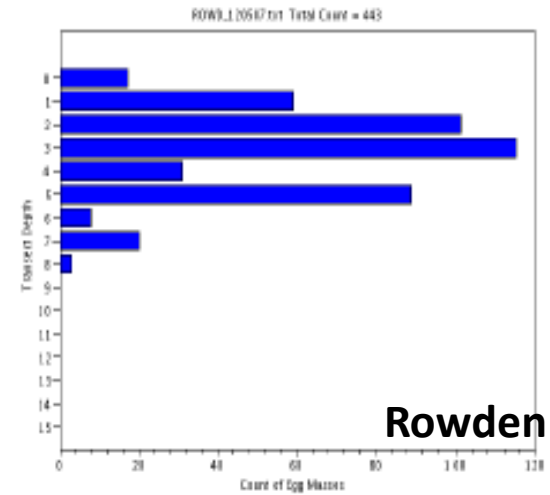
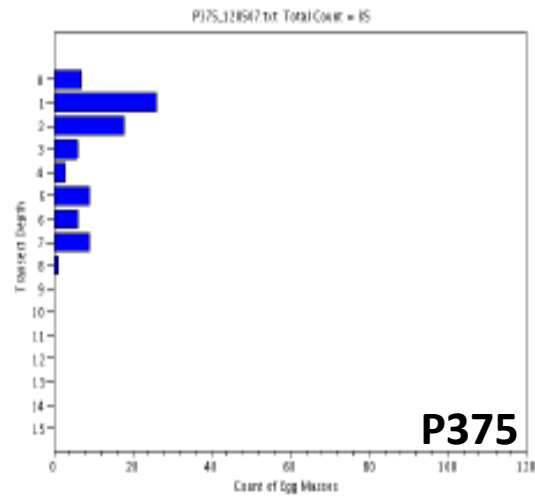
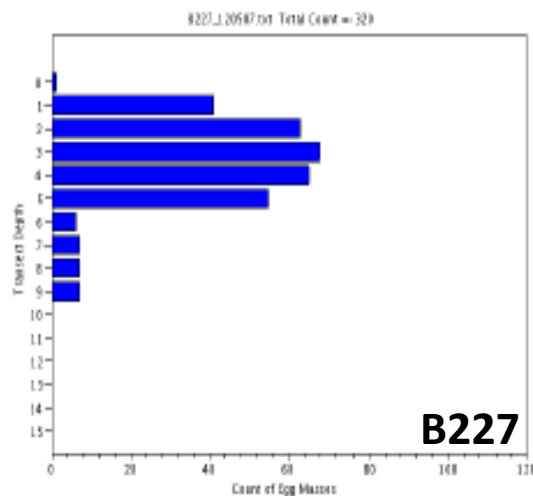
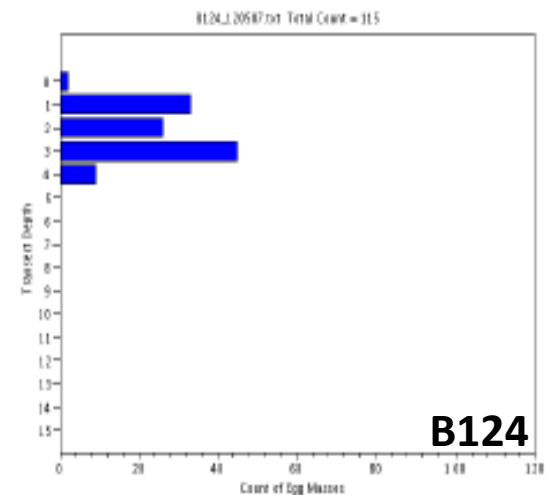
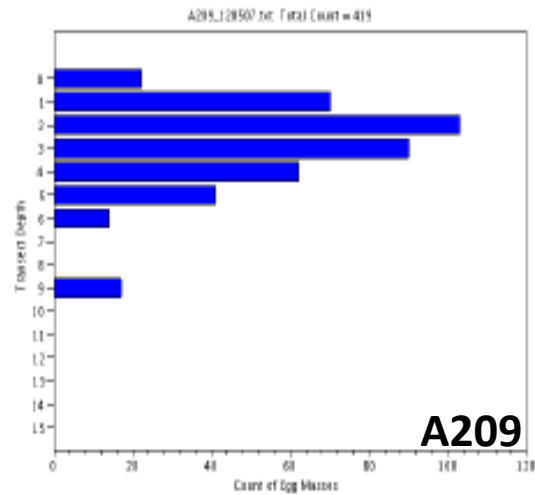
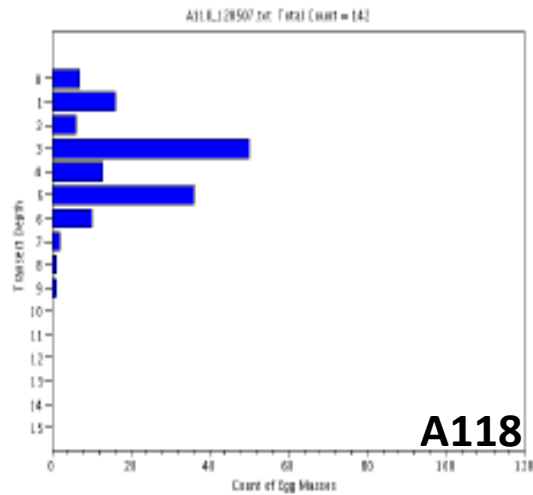


# Nematode infection density.

A118	Slightly less susceptible
A209	Very susceptible
B124	Moderately susceptible
B227	Slightly, slightly less susceptible



# Reniform egg mass distribution as a function of root depth.



**We thank Cotton Incorporated and the University of Tennessee Research Foundation for financial support...**

**...Jim Starr and Terry Kirkpatrick for supplying Rowden and Phytogen345 cotton seed...**

**...and Nancy West for office support at Phenotype Screening.**