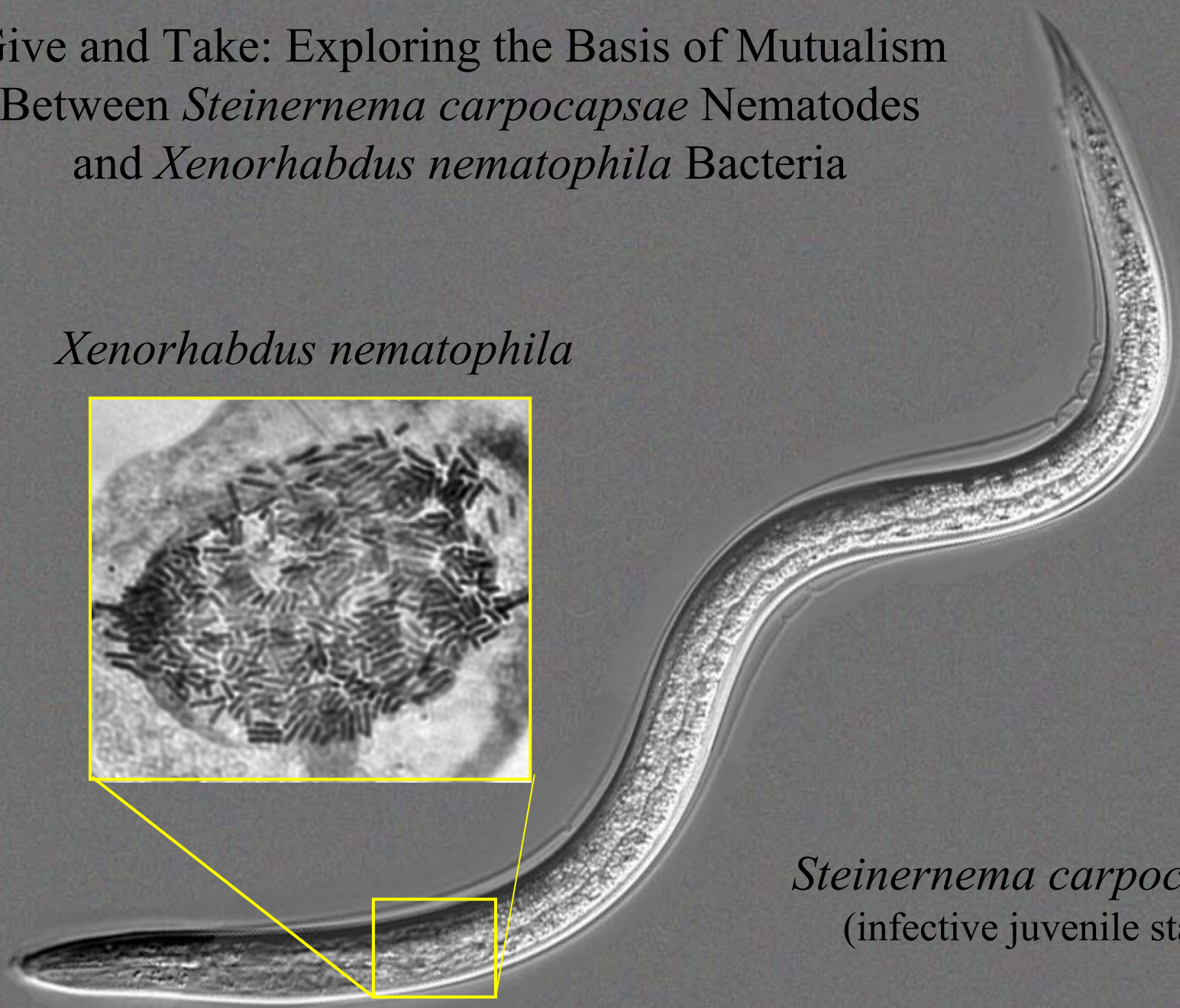
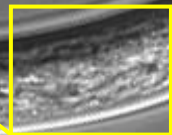
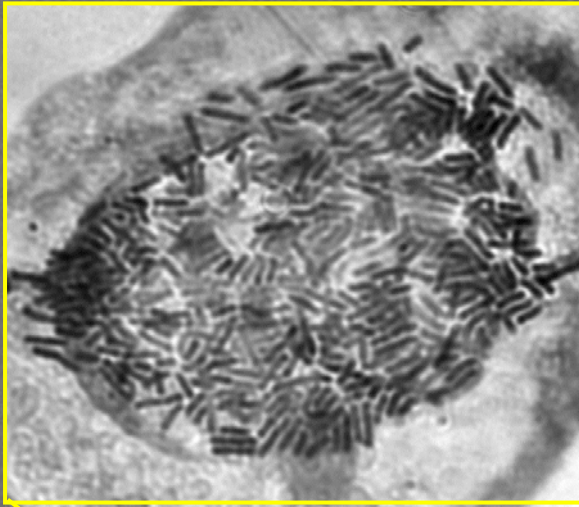


Give and Take: Exploring the Basis of Mutualism
Between *Steinernema carpocapsae* Nematodes
and *Xenorhabdus nematophila* Bacteria

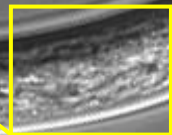
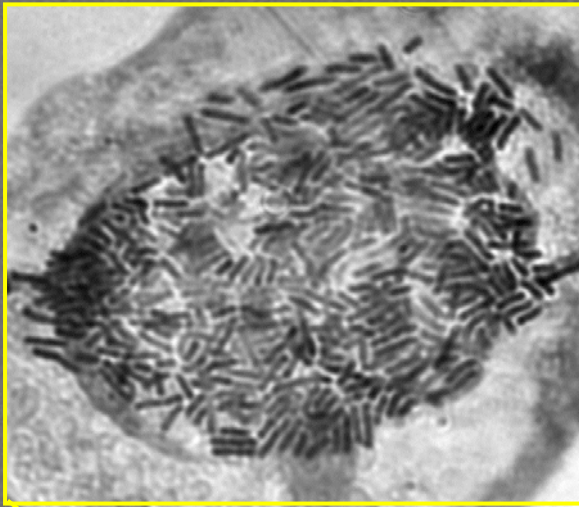
Xenorhabdus nematophila



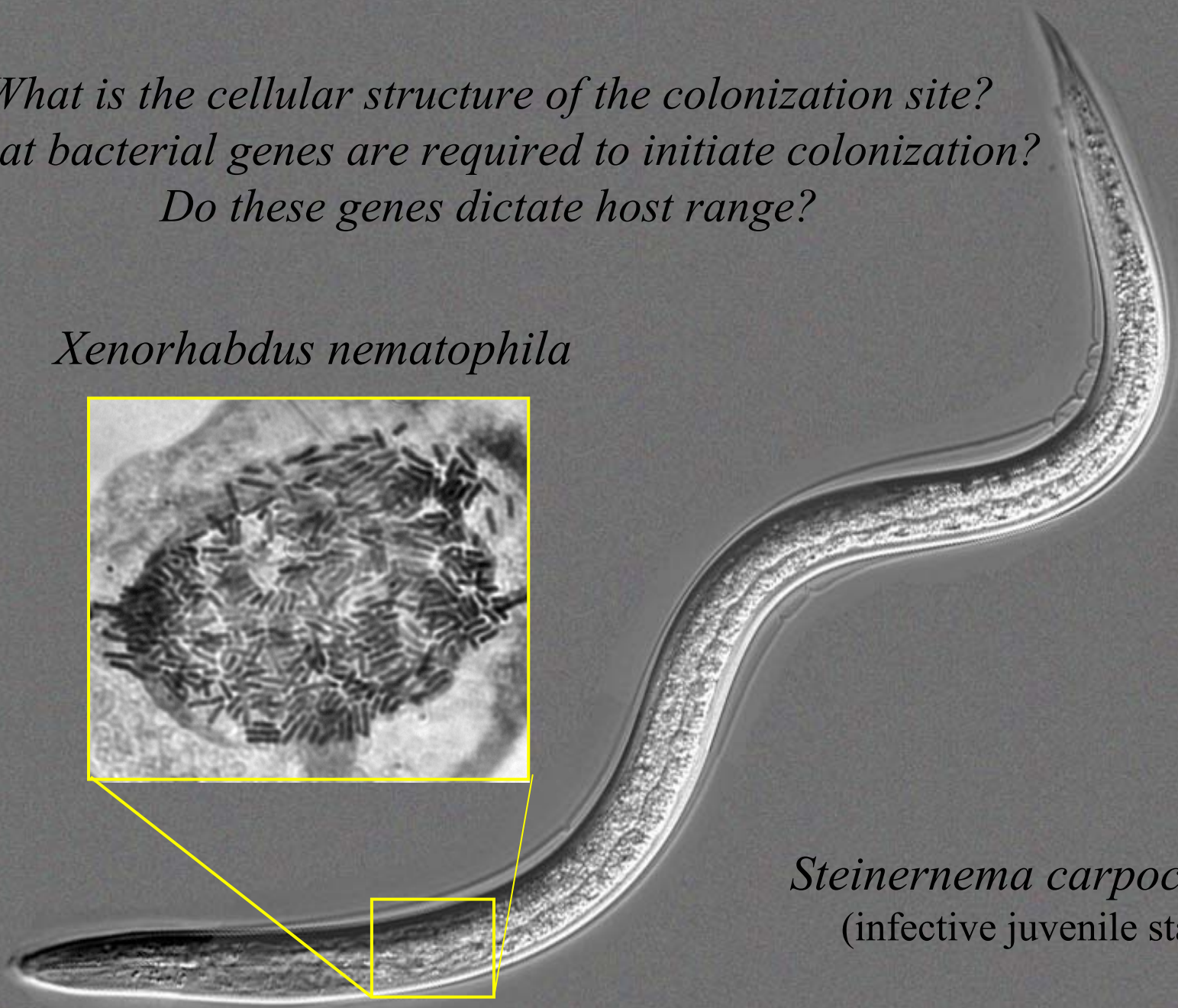
Steinernema carpocapsae
(infective juvenile stage)

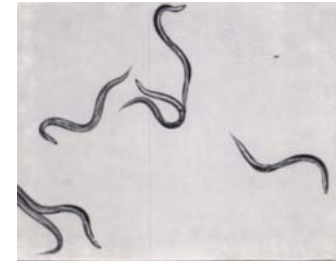
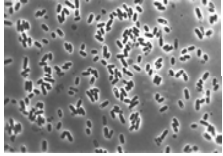
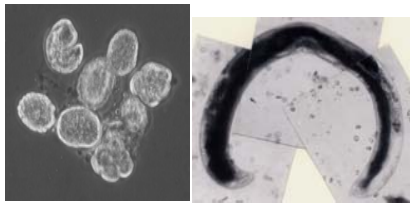
*What is the cellular structure of the colonization site?
What bacterial genes are required to initiate colonization?
Do these genes dictate host range?*

Xenorhabdus nematophila



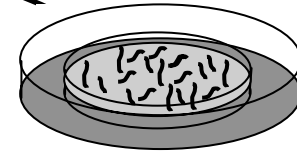
Steinernema carpocapsae
(infective juvenile stage)





Association/Migration

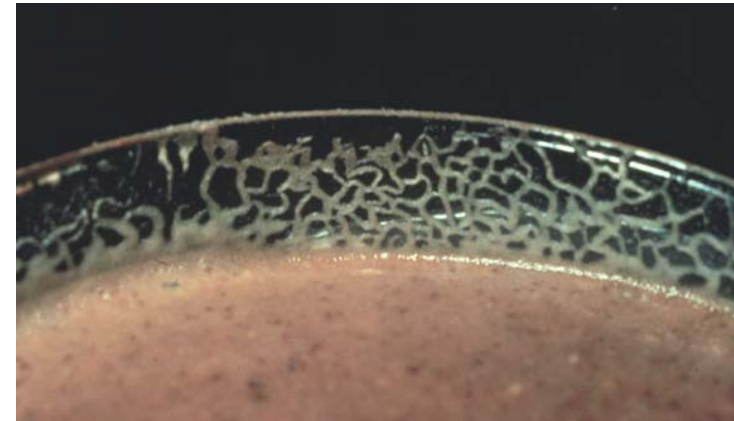
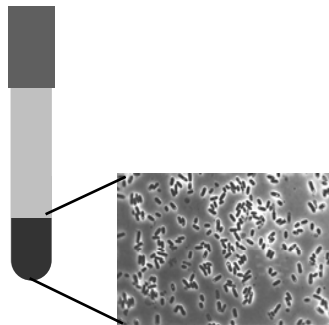
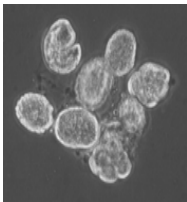
**Growth/
Development**



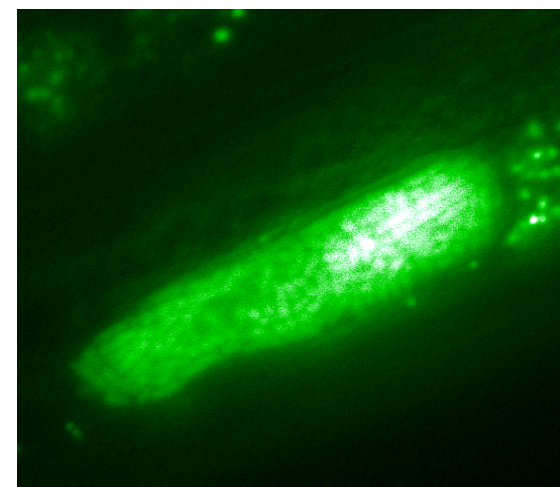
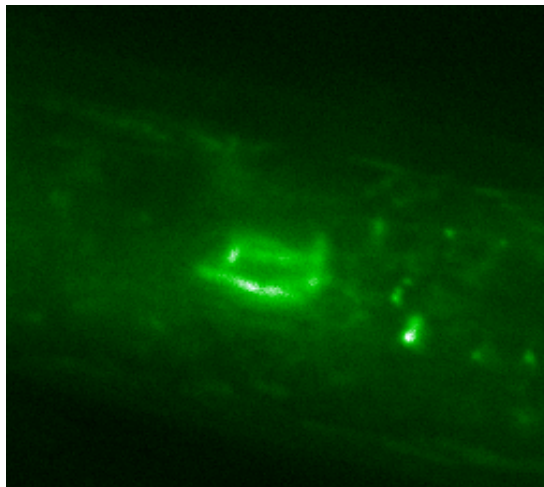
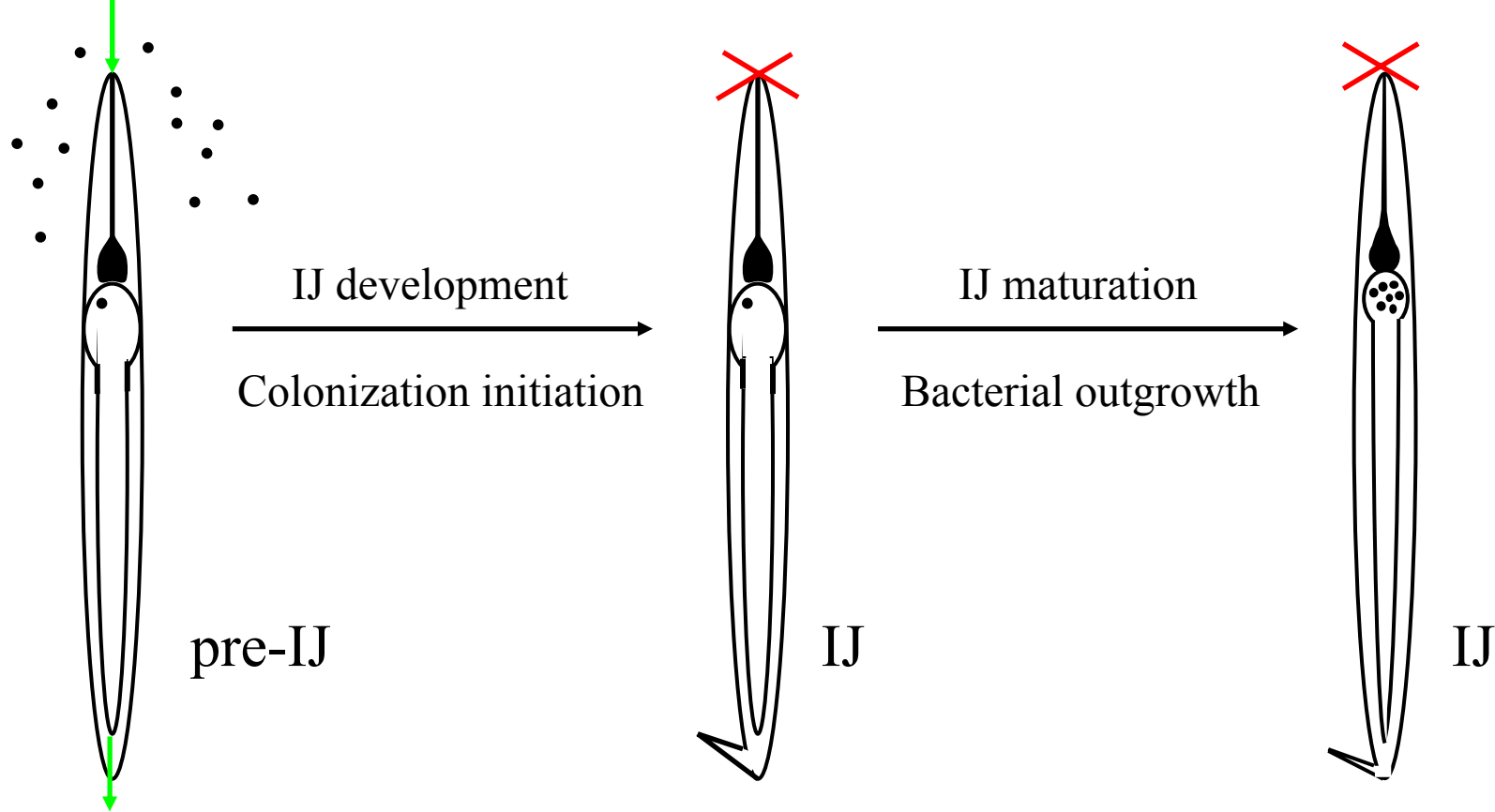
**Nematodes/
Nematode eggs**

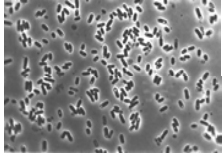
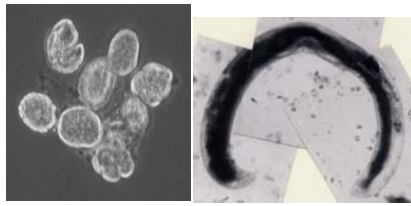
Bacteria

**Bacteria / Nematode
Complex**



picture courtesy Dr. Harry Kaya

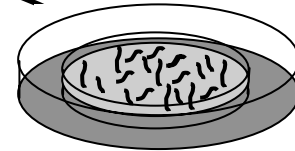
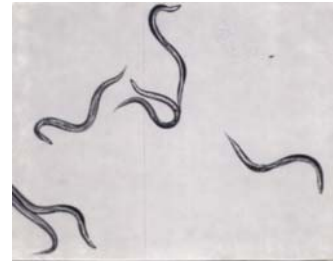




**Growth/
Development**

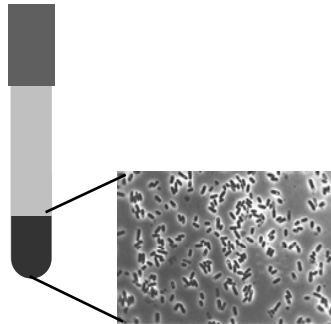
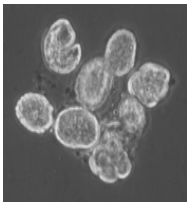


Association/Migration

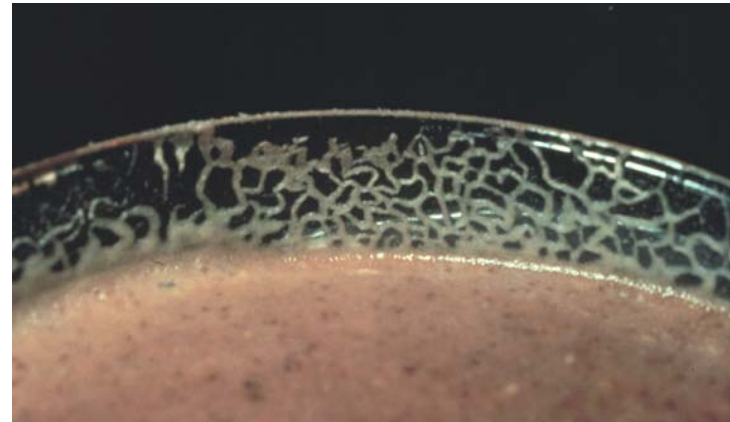


**Colonization-defective
Bacteria**

**Nematodes/
Nematode eggs**



**Uncolonized
Nematodes**



Colonization-deficient mutants:

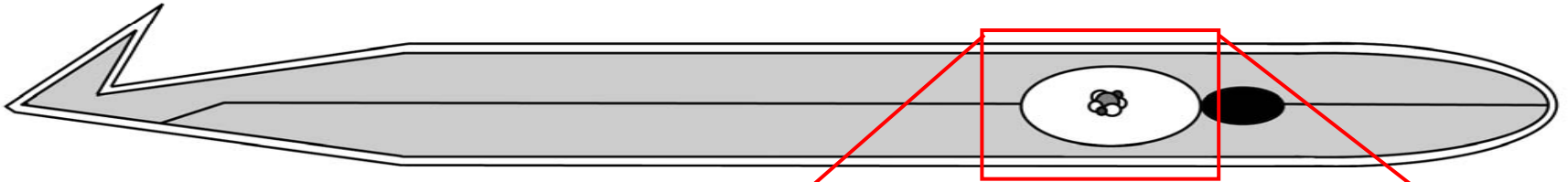
Das and Goodrich-Blair. 2001 *J. Bacteriol.* 183(16): 4687-4693

de Boer, et al. 2002 *Mol. Microbiol.* 45(5): 1337-1353

de Boer, et al. 2002 *J. Bacteriol.* 184(10): 2670-2680

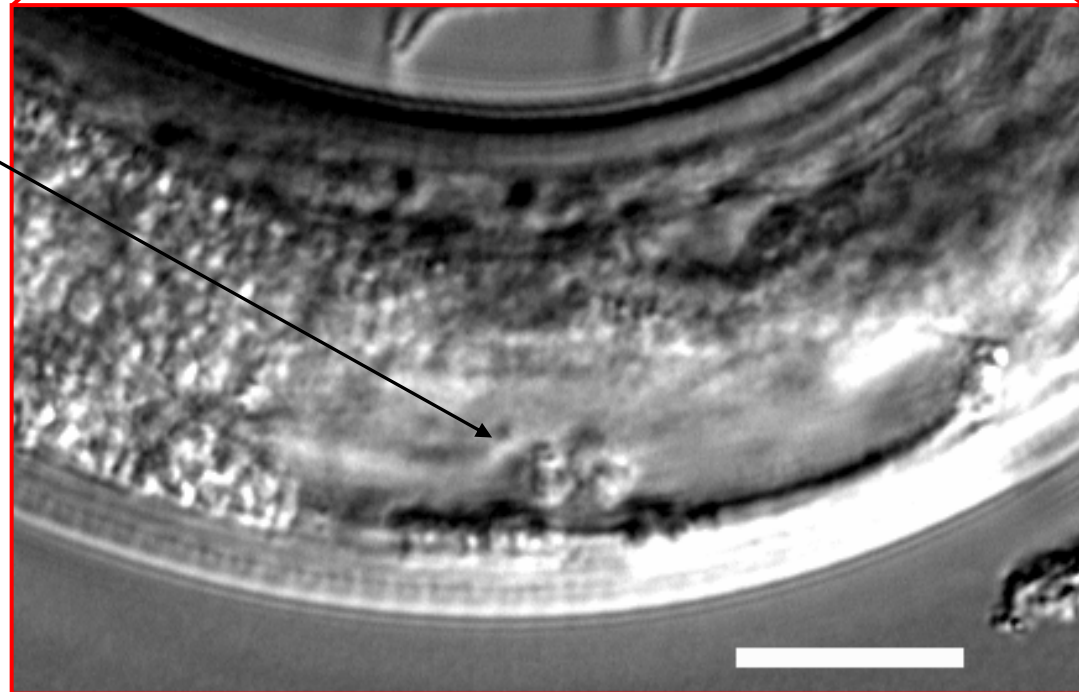
picture courtesy Dr. Harry Kaya

Intestinal vesicle and sub-structure

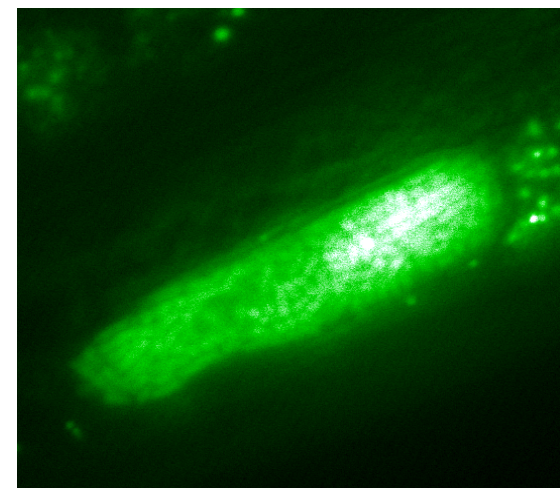
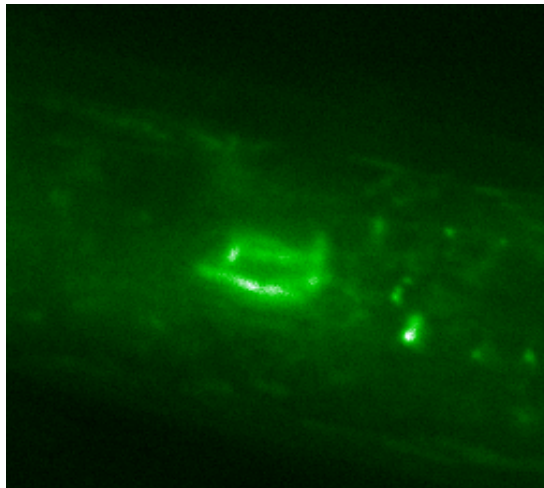
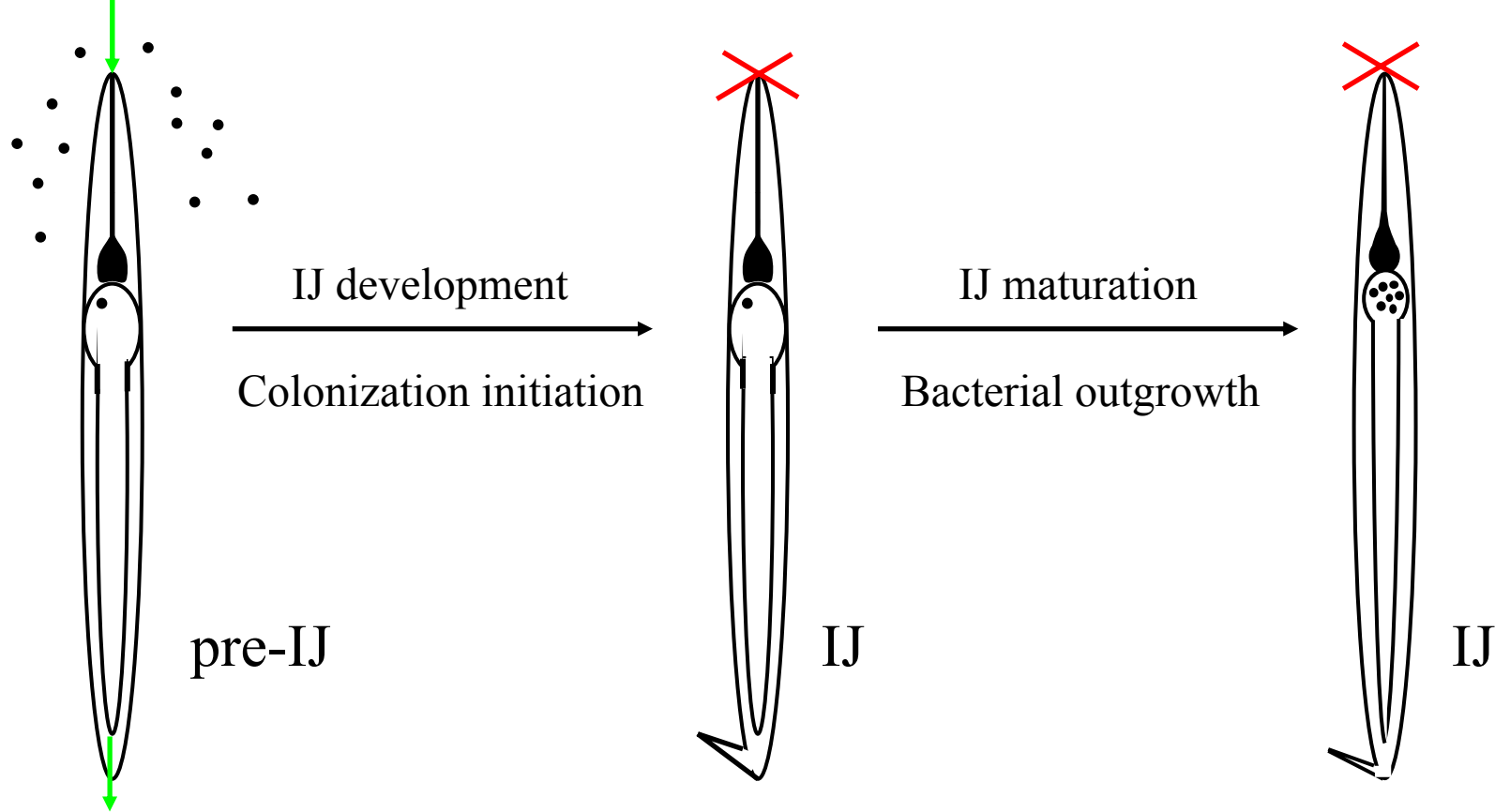


Intra-vesicular structure (IVS)

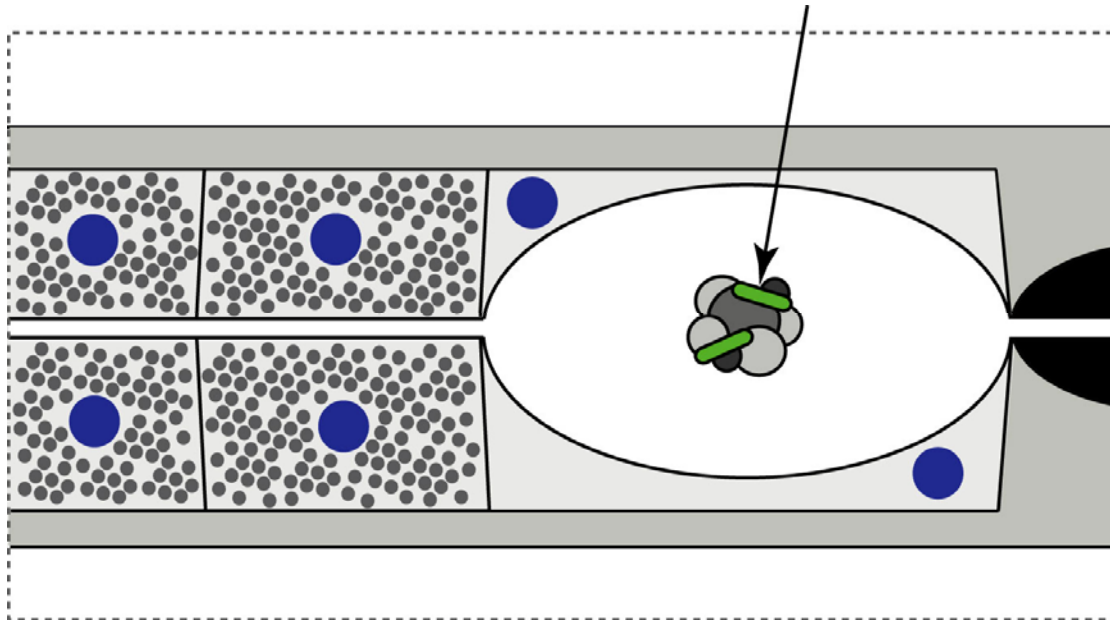
1 or 2 per IJ
~4-6 μm diameter
cluster of smaller spheres
moves in vesicle



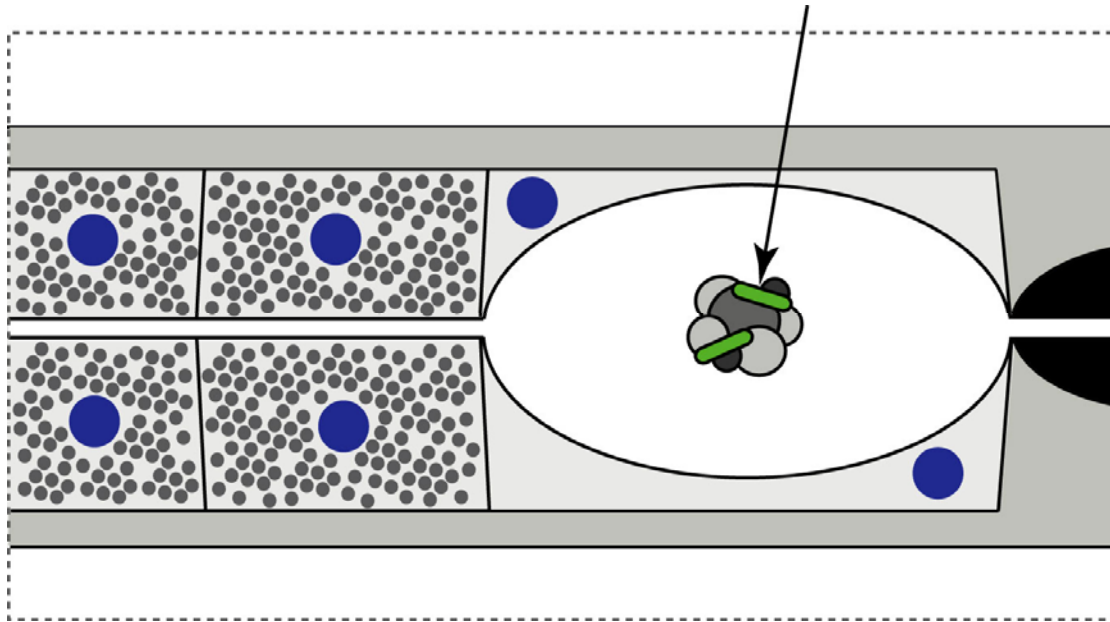
10 μm



X. nematophila association (adherence?) with IVS



X. nematophila association (adherence?) with IVS



What genes does *X. nematophila* require to colonize infective nematode intestines?

What genes does *X. nematophila* require to colonize infective nematode intestines?

- mutagenize bacteria
- screen for loss of colonization
- characterize disrupted loci

Colonization-deficient mutants:

Das and Goodrich-Blair. 2001 *J. Bacteriol.* 183(16): 4687-4693

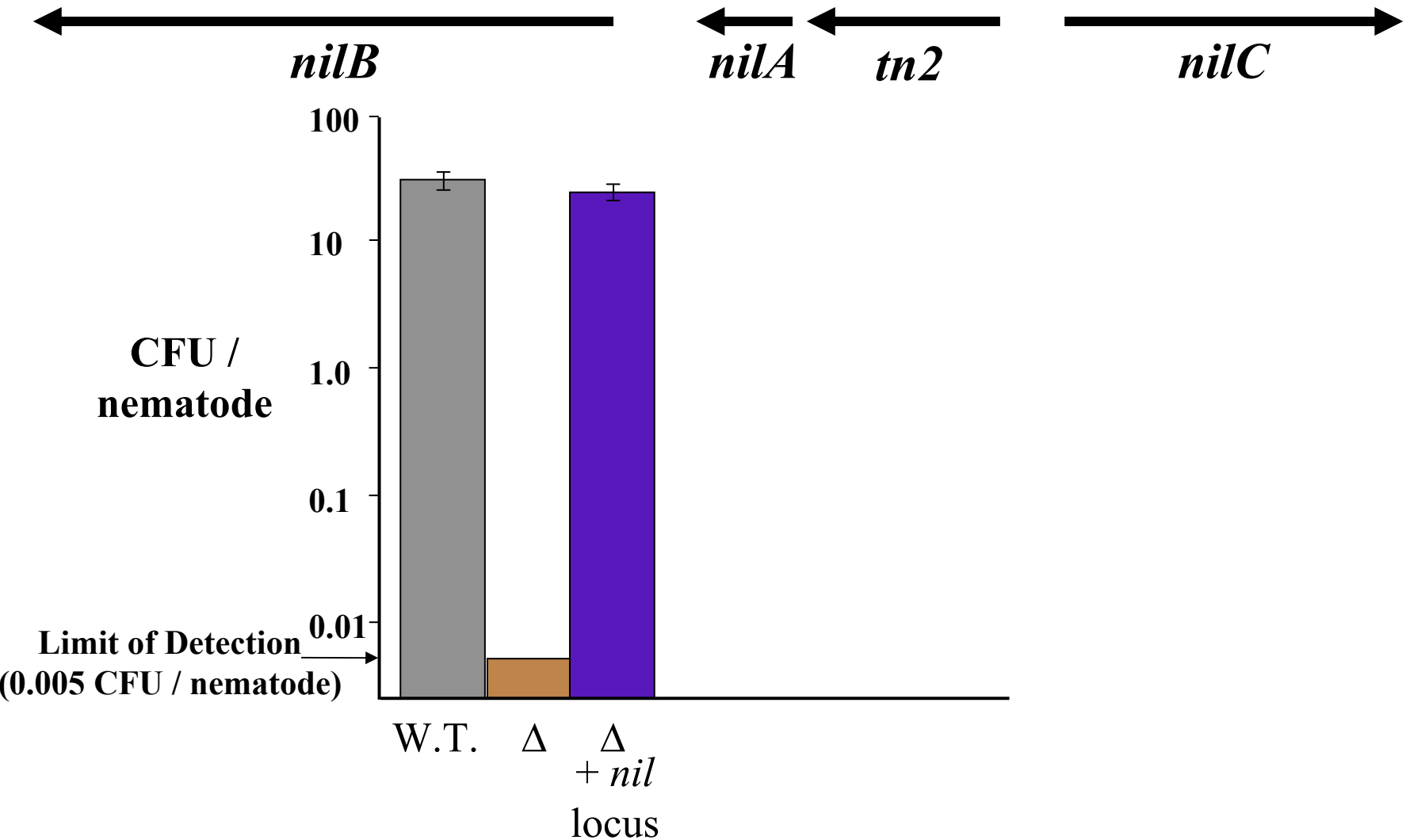
Reungens, et al. 2002 *Mol. Microbiol.* 45(5): 1337-1353

Das et al. 2002 *J. Bacteriol.* 185(12): 3670-3682

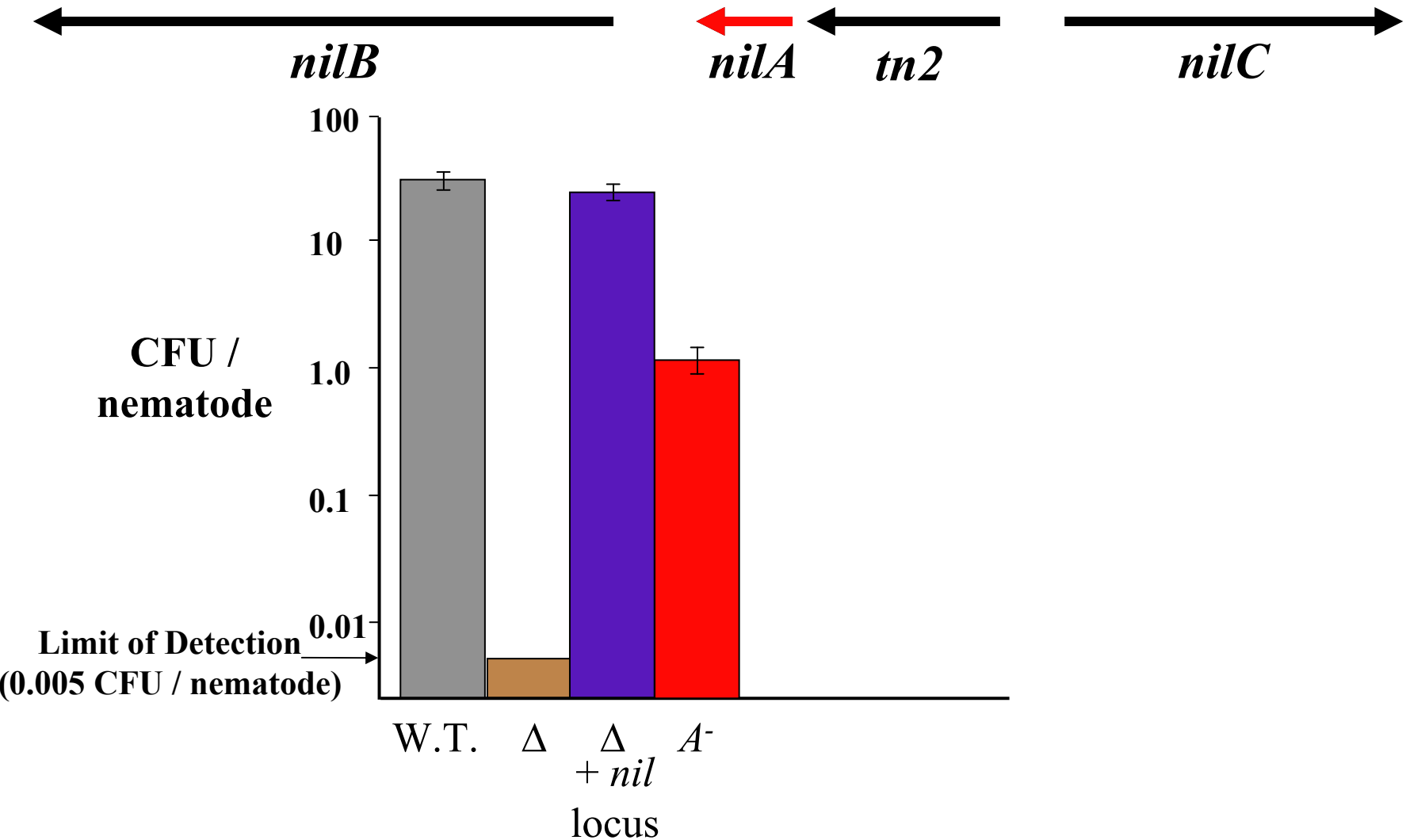
***X. nematophila nilA, B, C* mutants are impaired
in colonization**



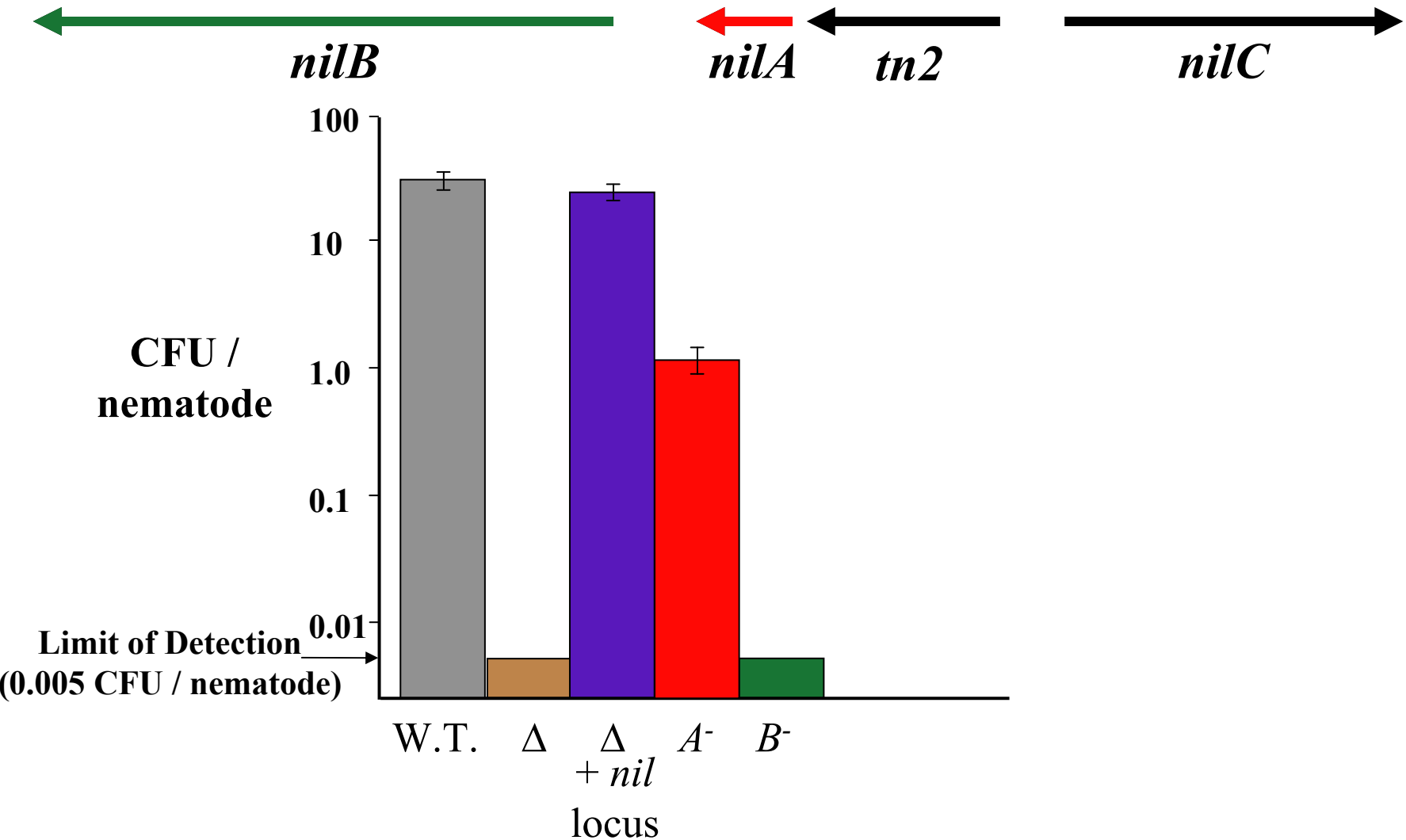
X. nematophila nilA, B, C mutants are impaired in colonization



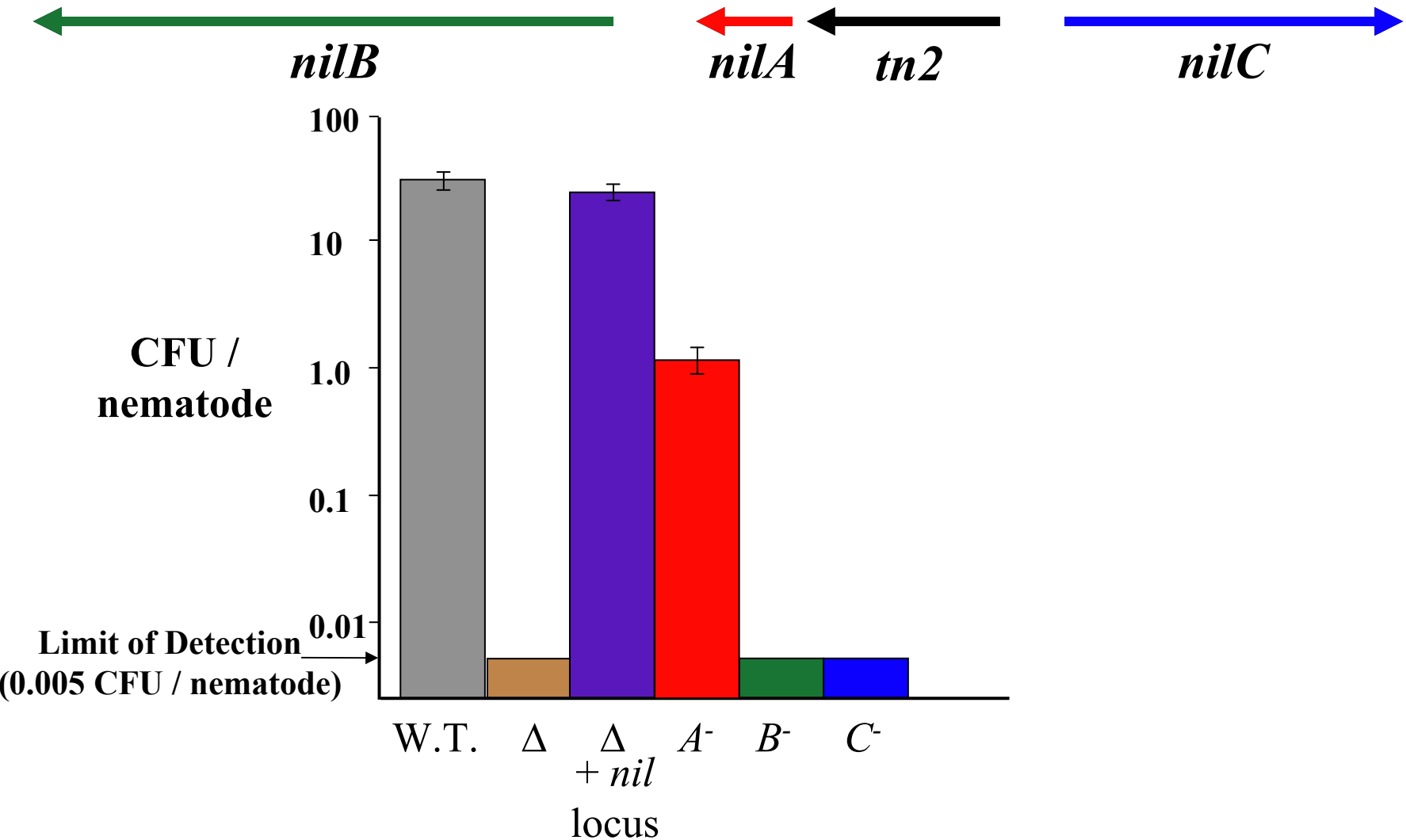
X. nematophila nilA, B, C mutants are impaired in colonization



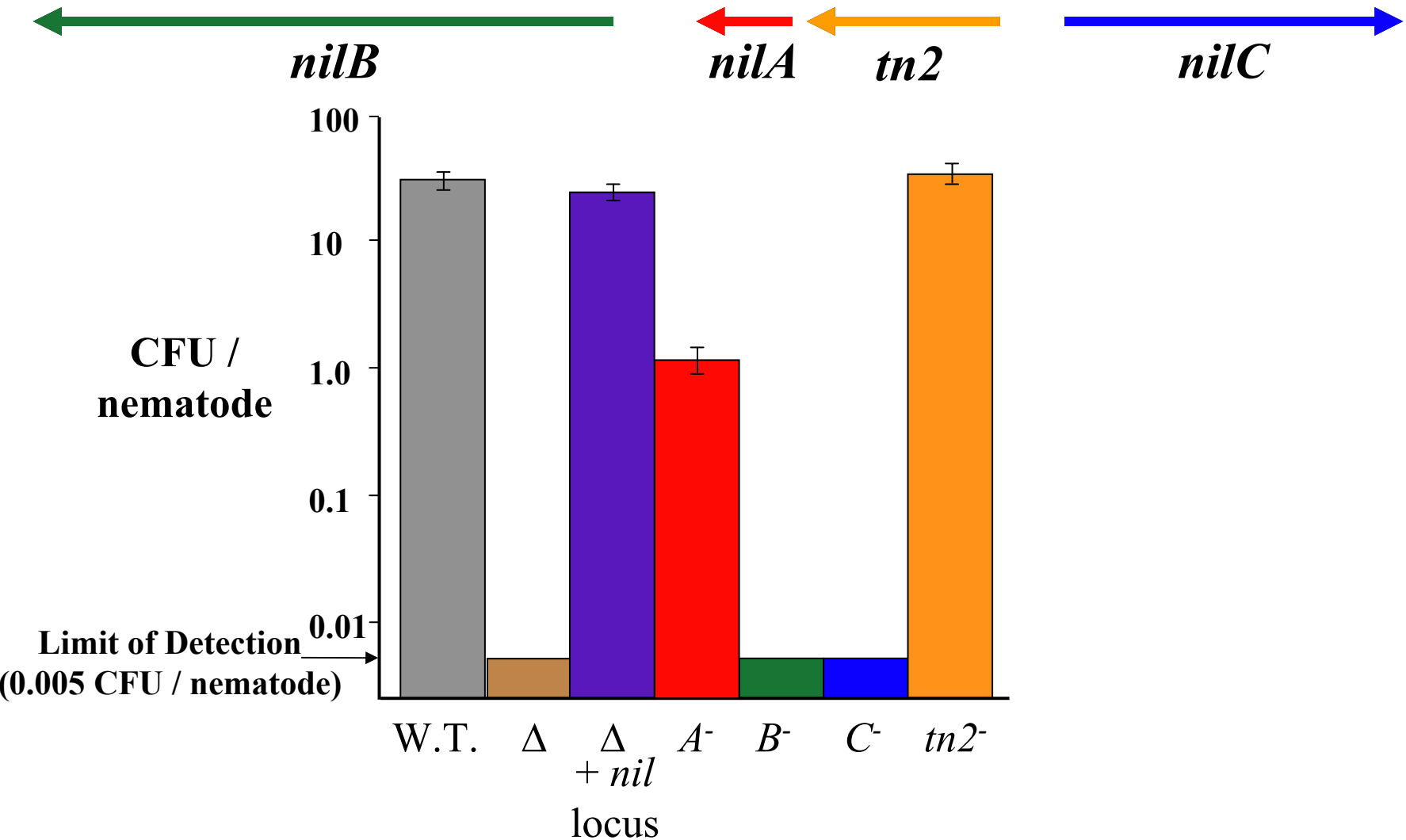
X. nematophila nilA, B, C mutants are impaired in colonization

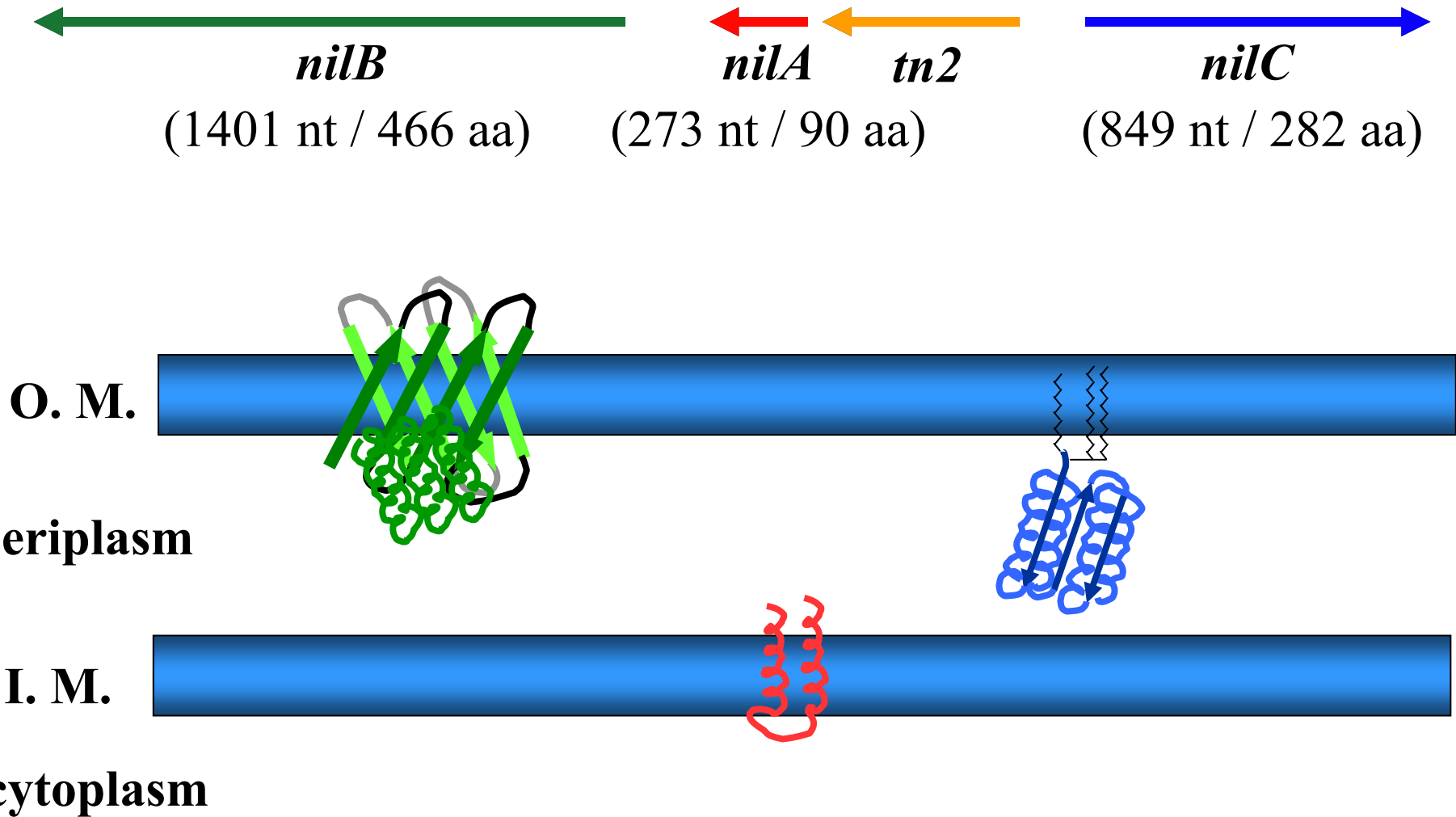


X. nematophila *nilA*, *B*, *C* mutants are impaired in colonization



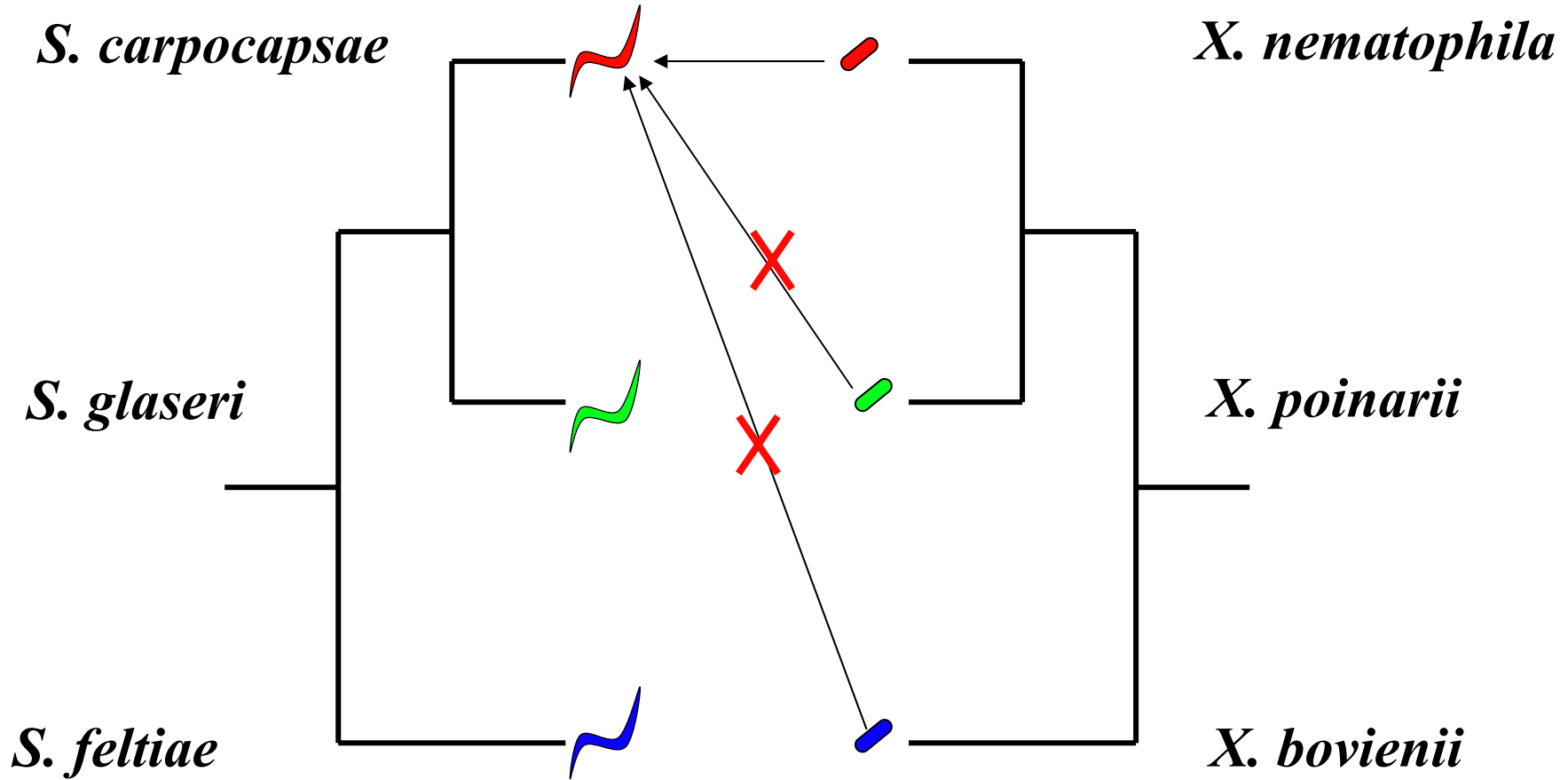
X. nematophila *nilA*, *B*, *C* mutants are impaired in colonization





**Do *nilA*, *B*, and *C* contribute to
species-specificity?**

Do *nilA*, *B*, and *C* contribute to species-specificity?

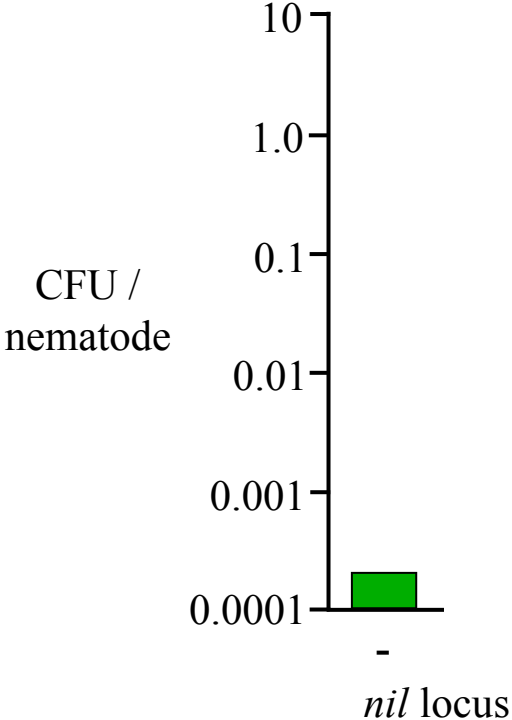


Do *nilA*, *B*, and *C* contribute to species-specificity?

- 1) *nilA*, *nilB*, and *nilC* are essential for nematode colonization by *X. nematophila*.
- 2) NilC and NilB are (or are predicted to be) outer-membrane-localized proteins, possibly enabling a direct interaction with the nematode host.
- 3) *nilA*, *nilB*, and *nilC* are not found in *X. poinarii* or *X. beddingii*, as determined by low-stringency Southern hybridization and PCR. No *nil* gene-similar sequences are present in the *X. bovienii* genome sequence.

The *nilA, B, C* locus confers species cross-specificity

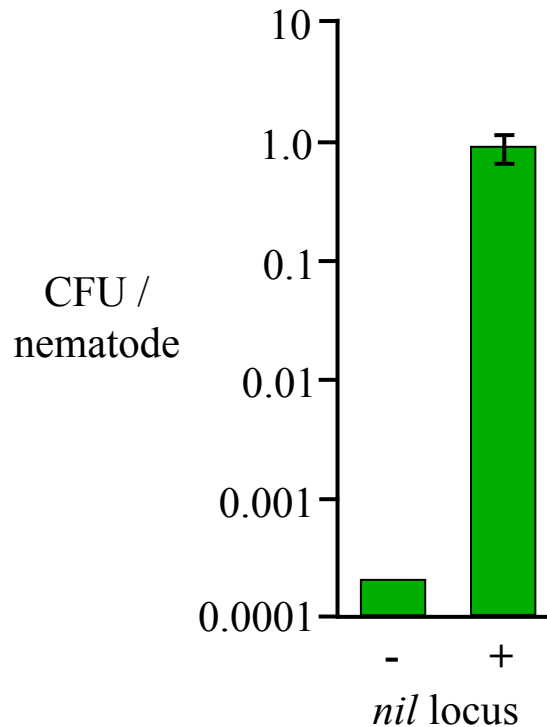
X. bovienii



***Limit of Detection:
0.0002 CFU / nematode**

The *nilA, B, C* locus confers species cross-specificity

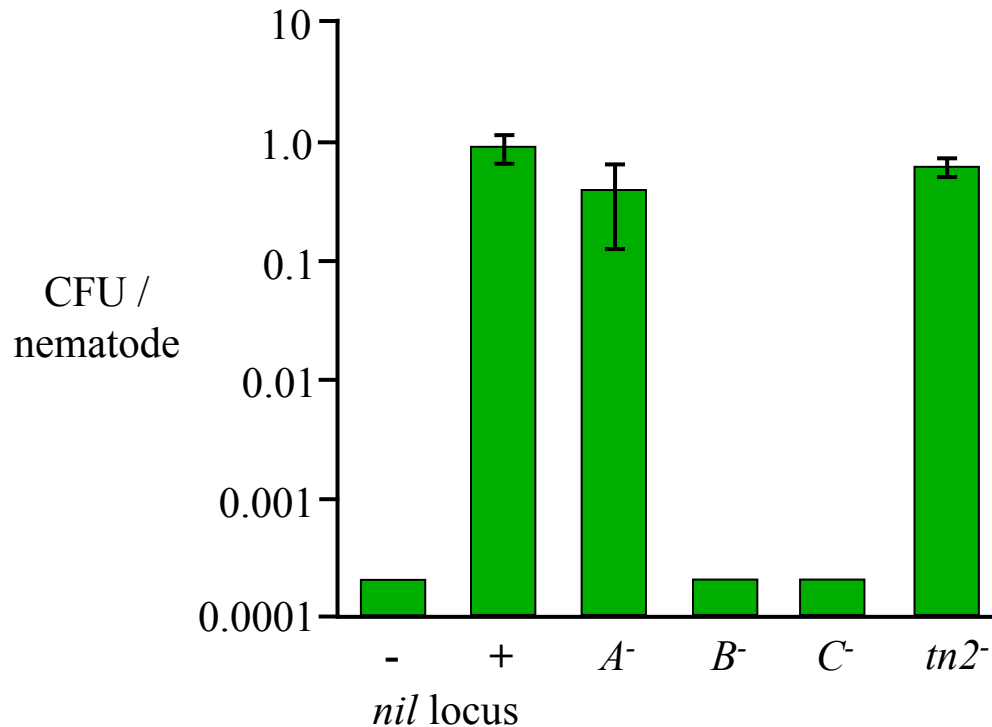
X. bovienii



***Limit of Detection:
0.0002 CFU / nematode**

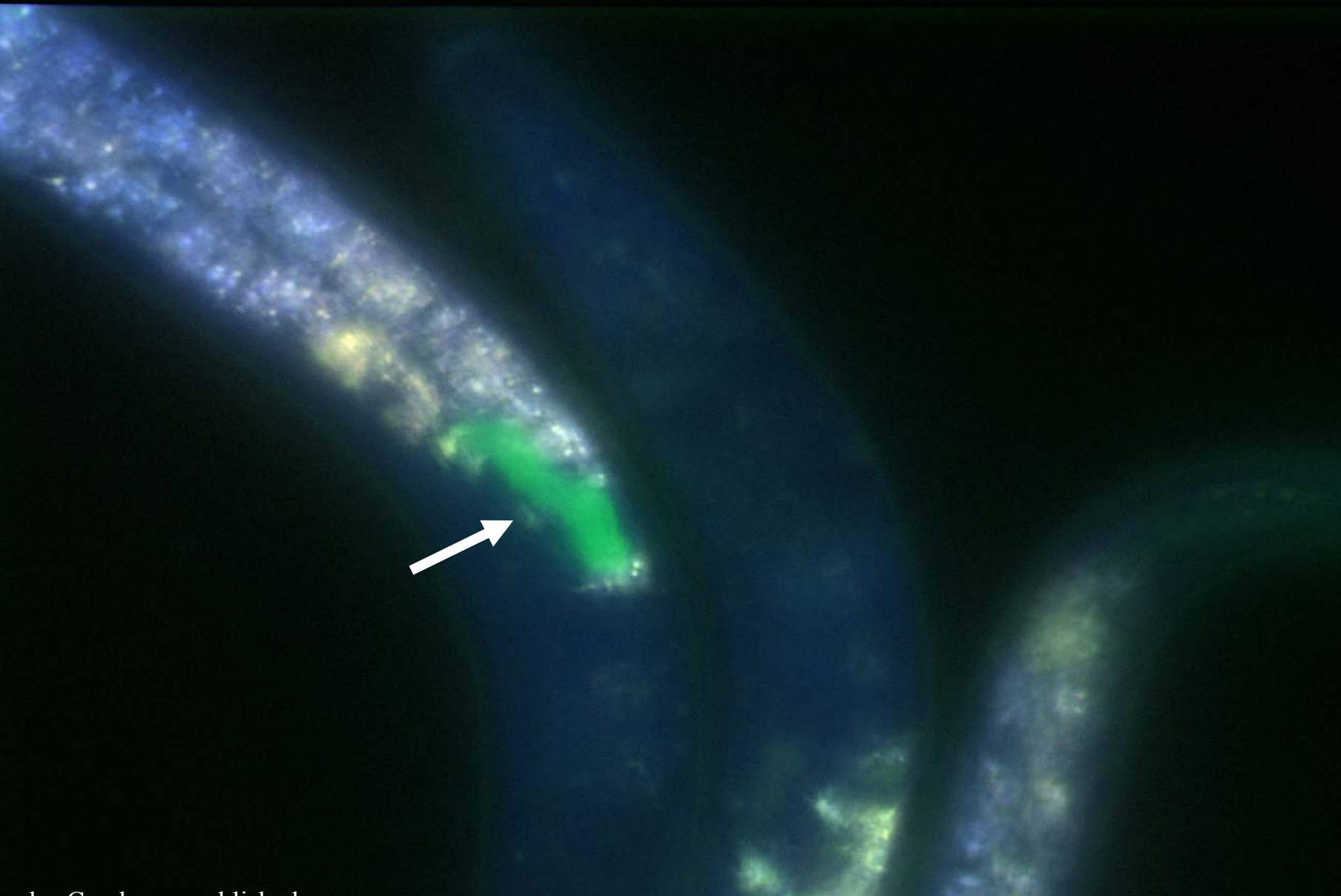
Point mutations in *nilB* and *C* impair species cross-specificity

X. bovienii

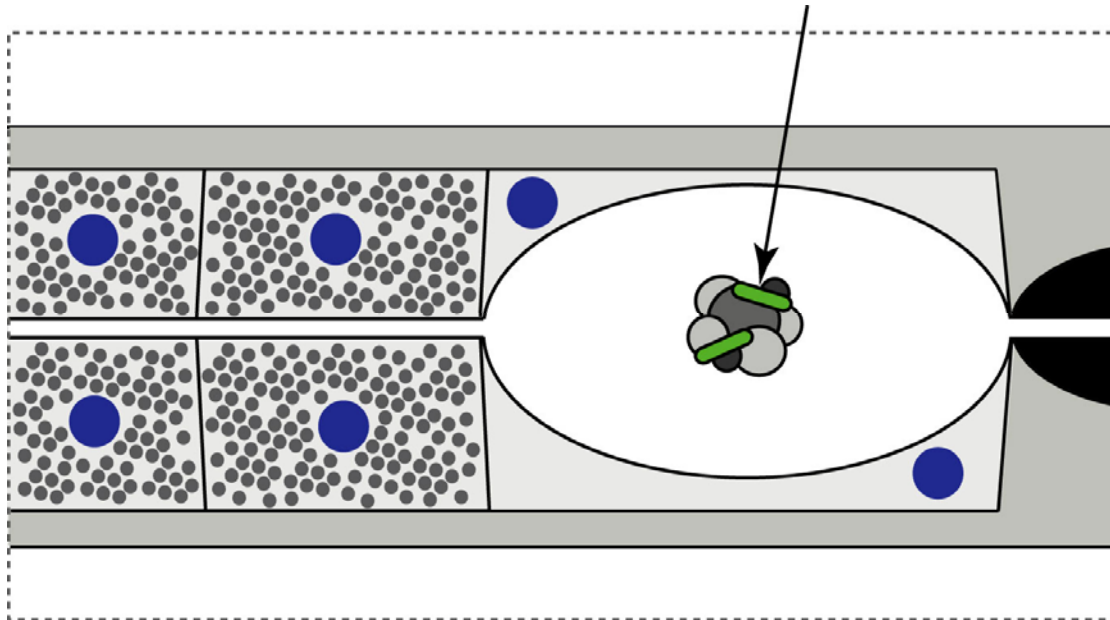


*Limit of Detection:
0.0002 CFU / nematode

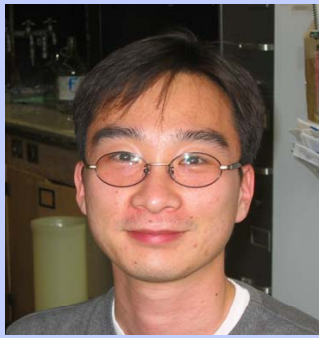
X. bovienii colonizes the *S. carpocapsae* nematode intestinal vesicle.



X. nematophila association (adherence?) with IVS



Are *nilB* and *nilC* responsible for adherence to IVS?



Dr.
Youngjin
Park



Kimberly
Cowles



Erin
Herbert



Carolyn
Lipke



Greg
Richards



Dr.
Archna
Bhasin



Aaron
Andersen



Charles
Cowles



Eric
Martens



Samantha
Orchard