

Do Human Activities Negatively Influence Insect-Parasitic Nematodes in the Soil?

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Grade: High school

Observations:

Human activities can have a devastating effect on biodiversity and the ecosystem services it provides. Insect-parasitic nematodes are microscopic roundworms that occur in the soil (**Fig. 1**). As part of the soil foodweb, these nematodes serve as natural regulators (biological control agents) of harmful insects (pests) (**Fig. 2**) and also commercially available for insect pest control (www.oardc.ohio-state.edu/nematodees). However, human activities including lawn management practices such as the use of chemical pesticides and fertilizers may negatively influence these beneficial nematodes, reducing or eliminating the free biological control services they provide.

Question:

Do urban landscape management practices negatively influence beneficial nematodes in the soil?

Hints to form the hypothesis:

Homeowners and lawn care companies routinely apply chemical pesticides and fertilizers to maintain beautiful green turfgrass lawns. The application of these toxic substances to lawns can have non-target effects on beneficial organisms such as nematodes. If the number (abundance) of beneficial nematodes decreases in the soil, the insect control service they provide may be compromised, leading to pest outbreaks.

Hypothesis:

The level of natural biocontrol service provided by insect parasitic nematodes is lower in lawns managed with chemical pesticides compared to the lawns and wooded areas that do not receive such pesticides.

Materials:

- 75-100 Wax worms*
- 15 plastic sandwich bags (Ziploc®)
- A shovel
- 15 plastic cups with lids
- Tap water
- 15 Petri dishes (approximately 150 mm diameter x 25 mm deep) or flat bottom plastic containers with lids

- 15 bottle lids
- paper towel
- A pair of flat-end forceps
- Hand lens or magnifying glass, (magnifying lens can be ordered from websites like www.amazon.com or www.kellycodetectors.com for a price ranging between \$5 to \$20. A 10X magnifying lens would be good.) and a compound microscope (if available in your school's science lab).

*Wax worms can be obtained from commercial sources such as Vanderhoest Canning Company, St. Mary's, Ohio. They can also be purchased from a local gas station or sports shop where they are sold as 'live bait' for fishing.

Experiment:

1. Collect 5 soil samples (about 100 cc each) each from a lawn managed with chemical pesticides, a lawn that does not receive chemical pesticides, and an unmanaged wooded area around your school and bring them to the science lab.
2. Break the soil and remove any stones, plant material including roots as much as possible.
3. Place each soil sample in a plastic cup and release 10 wax worms in each cup (**Fig. 3**). Use only actively moving wax worms.
4. Gently spray a little water on the soil if the soil is too dry. Do not make a puddle of water or press the soil.
5. Replace the lids and place the cups in a dark area in the lab or classroom.
6. Examine the wax worms once daily and record the number of worms killed in each cup.
7. Compare the dead worms with the pictures on the website and select the cadavers that do not emit foul smell, are soft, and have the right coloration (yellow, tan, brown or red) (**Fig. 4**).
8. The infection of wax worms with nematodes can be further confirmed by checking for the emergence of nematodes from the cadavers. For this you will need to set up Water traps (**Fig. 5**) as follows.
 - a) Place a bottle lid upside down in the Petri dish or a flat-bottom plastic container and cover the bottle lid with a piece of paper towel. Cut the paper towel to a size just slightly larger than the bottle lid.
 - b) Carefully transfer the dead wax worms (cadavers) on to the paper towel placed on the bottle lid in the Petri dish/container. Use a pair of forceps to gently place the cadavers on the paper towel. Do not rupture the cadavers.

- c) Add water into Petri dish or the plastic container so that the water reaches mid way to the top of the bottle lid. Make sure the paper towel underneath the cadavers is wet, but not flooded. The wet towel will facilitate the movement of nematodes emerging from the cadavers into the surrounding water in the Petri dish or the plat container.
 - d) Cover the Petri dish or the plastic container with the lid gently.
Keep the prepared Petri dish/container at room temperature for about 7-15 days and check for the emergence of nematodes from the cadavers in the water. Use magnifying glass or compound microscope to check the presence of nematodes in the water. Nematodes that are straight and motionless under the microscope may be dead, and therefore, ineffective. Living nematodes will be seen moving in a wavy, snake like, pattern.
9. The nematodes accumulated in water can be transferred to another Petri dish/container and stored in a refrigerator for counting, conducting bioassays against pests such as white grubs, or for use in the field as biological pest control agents.

Results: Present the result in a bar chart as shown in **Fig. 6**.

Discussion:

Interpret and discuss the results to accept or reject your hypothesis. Think about reasons for such results.

Hints:

1. Think about the potential natural biological control services provided by these nematodes against soil inhabiting insect pests and their potential economic and environmental benefits. Also discuss the importance of other below-ground and above-ground food chains in prevention of pest outbreaks and pest suppression.
2. Think about potential reasons for the absence of nematodes in any of the investigated areas. These may include habitat destruction by human intervention such as excessive soil compaction or use of chemical pesticides that may directly kill the nematodes or eliminate their host insects.
3. Discuss ways to increase populations of naturally occurring insect-parasitic nematodes and how they can be exploited for effective pest management.
4. Formulate conclusions and further questions and hypotheses to investigate.

Further readings and resources:

Insect-parasitic nematodes: A tool for pest management at www.oardc.ohio-state.edu/nematodes
Gaugler, R. (Editor). 2002. *Entomopathogenic Nematology*, CABI Publishing, Wallingford, UK.

Grewal, P. S., Ehlers, R. U, and Shapiro-Ilan, D. I. [Editors]. 2005. *Nematodes as Biocontrol Agents*, CABI Publishing, CAB International, Wallingford, Oxfordshire, UK, 505pp.

Kaya H K and Stock S P (1997) Techniques in insect nematology. *In: (Lacey L A, eds.) Manual of Techniques in Insect Pathology*. Academic Press, New York, pp. 281–324.

Estimated time: Time: 15-20 days

Estimated cost: \$25-\$100

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Fig. 1. Beneficial insect-parasitic nematodes (left) and insect predatory beetle (right)



www.google.com
Insect-parasitic nematodes (~800 x 50 micron)
(Image taken under a microscope at 40x magnification)

www.google.com
Adult lady beetle as seen with naked eye

Fig.2 Japanese beetle larva: An example of target pest insect of insect-parasitic nematodes. It feeds on grass roots due to which it is a harmful insect-pest of lawns and turf grass.



Fig.3. Soil baited with wax worms in a plastic cup



Fig.4. Wax worm cadavers infected with nematodes after three days of baiting in the soil. Note change in color of the worms after infection with nematodes.



Fig.5. A water trap used for collecting the nematodes emerging from the infected wax worm cadavers.



Fig.6. A bar chart showing the mean number of wax worms killed by insect-parasitic nematodes in soil samples collected from three different habitats (Drawn in MS excel).

