

## Practical Lesson 7

### **Inoculating Plants with Nematodes**

To establish that a nematode associated with diseased plants is the infectious agent causing the disease, four conditions must be fulfilled (Koch's postulates): the nematode must be consistently associated with the disease; a pure culture of the nematode must be obtained from an infected plant; healthy plants must be inoculated with the nematode and in time develop typical symptoms; and the nematode must be reisolated from the inoculated plants and found to be identical to the inoculum. Plant diagnostic clinics rarely perform inoculation trials to determine whether these conditions are met, because the process takes weeks or possibly months to complete. Inoculation tests to determine pathogenicity should be carried out by an experienced nematologist who clearly understands the biology of the nematode and the host plant and the pitfalls of the procedure. Four methods are known to be used for inoculating plants with nematodes :

- 1- Sowing or transplanting in naturally infested soil.
- 2- Inoculation with nematode-infected plant materials.
- 3- Inoculation with nematode eggs.
- 4- Inoculation with infective nematode stages.

### **Inoculating Plants with root-knot nematode**

Root-knot nematode, *Meloidogyne* spp. received the most attention by plant nematologists all over the world. Inoculation of plants with root-knot nematodes could be achieved using either eggs or second-stage larvae (the infective stage). The procedure could be done as follows:

- 1- Grow host plants in a sterilized or pasteurized growing medium, usually a potting mix or a sandy soil, in clay or plastic pots (10 cm in diameter) on a porous greenhouse bench. Clay pots are preferred, because the environment in the pot is more conducive to nematode survival. The greenhouse temperature must be adjusted to suit the requirements of the plant and the nematode.

**CAUTION:** To prevent contaminating potted plants with nematodes and other pathogens from adjoining pots or water on the surface of the bench, place dividers between the pots, and elevate the pots above the bench surface with a saucer or inverted pot, or use a bench with a wire mesh top to prevent water from collecting on the bench surface.

- 2- Extract eggs or live nematodes as described in lessons 1 and 2.
- 3- pour water containing nematode eggs or larvae into small holes dug in the root zone of potted plants. The holes are then covered, and soil moisture is maintained within narrow limits. Drought or flood conditions may kill the nematodes.
- 4- Initial inoculation does not often reproduce symptoms of the disease, because too few nematodes were introduced or because many nematodes in the inoculum were killed by handling. However, large numbers of nematodes are produced in the infected plants, and they can be used to adequately inoculate other plants and reproduce the disease.

## References

- Ayoub, S.M. 1977. *Plant Nematology, An Agricultural Training Aid*. Academic Press. California, 157 pp.
- Shurtleff, M.C. and C.W. Averre III. 2000. *Diagnosing Plant Diseases Caused by Nematodes*. American Phytopathological Society. St. Paul, Minnesota. 187 pp.