INFLUENCE OF THE CASTOR BEAN (*RICINUS COMMUNIS*) LECTIN (RICIN) ON MOTILITY OF *MELOIDOGYNE INCOGNITA*[1]

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Rich, J. R., G. S. Rahi, C. H. Opperman y E. L. Davis. 1989. Influencia de la lectina (ricina) del ricino (*Ricinus communis*) en la movilidad de *Meloidogyne incognita*. Nematrópica 19: 99–103.

Se realizon bioanálisis de laboratorio para evaluar el efecto del la lectina (ricina) del ricino (*Ricinus communis*) en la movilidad de los segundos estados juveniles de *Meloidogyne incognita* utilizando una modificación de la técnica "Moje". Los resultados indicaron que la ricina redujo la movilidad de *M. incognita* a concentraciones entre 10 y 20 μg/ml. El valor CE_{50} de la ricina se calculó a 33 μg/ml, mostrando, por lo tanto, una actividad similar a la nicotina.

Palabres claves: lectina, *Meloidogyne incognita*, nicotina, ricina, *Ricinius communis*.

Some plant-derived products are known for their pesticidal properties. Compounds such nicotine, a tobacco alkaloid, have been in use since the 1600's (16). Various other plant and oil cake extracts have been shown to reduce populations or the development of plant-parasitic nematodes (4). Among products tested, Masood and Hussain (10) found that castor bean (*Ricinus communis* L.) seed meal was quite effective in reducing damage to tomato (*Lycopersicon esculentum* Mill.) roots by *Meloidogyne incognita* (Kofoid & White) Chitwood. Other authors reported that castor bean meal significantly reduced populations of *Meloidogyne* spp., *Hoplolaimus indicus* Sher and *Tylenchorhynchus brassicae* Siddiqi (6,15).

The seed meal of castor bean contains a highly toxic protein originally thought to be a strong hemagglutinin (8). Later, however, it was determined that the toxic and agglutinating properties of the extract

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from castor beans reside in separate and distinct lectins termed ricin and *Ricinus* agglutinin, respectively (7). Ricin is the toxic moiety and has a percutaneous LD_{50} to mice of 0.006 mg/kg (3). *Ricinus* agglutinin, a strong agglutinating agent (17), has been shown to suppress growing mammary tumors in rats (14).

The high mammalian toxicity of ricin would indicate that it might be a principal compound in castor bean meal adversely affecting plant-parasitic nematodes. The present studies were conducted to determine the effect of ricin on activity/motility of *M. incognita* and to compare its effect with that of nicotine.

Laboratory bioassays were conducted using a modified Moje technique (12). *Meloidogyne. incognita* eggs were collected from the roots of tomato cv. Rutgers by a modification of the method of Hussey and Barker (5). The eggs were hatched in a modified Baermann funnel and second-stage juveniles (J2), collected within 24 hours after hatching, were used as the test organisms in these experiments.

Stock stolutions of ricin and nicotine (Sigma Chemical Company, St. Louis, MO, U.S.A.) were prepared using distilled water. The concentrations for ricin were 40, 20, 10, and 5 μg/ml. The solutions were diluted 1:1 (v/v) with suspensions of J2 in distilled water for each determination. The final concentrations of ricin were 20, 10, 5, and 2.5 μg/ml plus a distilled water control. Nicotine was diluted similarly and final concentrations were 100, 50, 20, 10, and 5 μg/ml.

Four ml of stock solution and 4 ml of water containing *M. incognita* J2 were added to individual 25 ml glass scintillation vials. Each vial was covered with Miracloth® (Chicopee Mills, Milltown, NJ, U.S.A.) screens containing 17 μm openings and incubated at room temperature for 24 hours. Vials then were inverted over petri dishes containing 8 ml of the solution of the same concentration. The number of juveniles that migrated into each petri dish within a period of 24 hours was counted and recorded. Numbers of nematodes migrating into the petri dish were expressed as a percentage of the nematode numbers obtained in the distilled water treatments. Each treatment was replicated six times with vials arranged in a completely randomized design. The experiment was repeated once.

Motility of *M. incognita* J2 was reduced as the ricin concentration increased. The highest ricin concentration, 20 μg/ml, reduced the number of *M. incognita* migrating through the screen by 45% in relation to the distilled water controls. At a ricin concentration of 10 μg/ml, motile nematode numbers were reduced approximately 25%. The dosage response plots of percent motility (control = 100%) versus $\log_{10}$ of toxin concentration of ricin (Fig. 1) and nicotine (Fig. 2) exhibited significant ($P \leq 0.01$) linear relationships. The extrapolated EC_{50} values for motility inhibition of both ricin and nicotine were 33 μg/ml. In similar

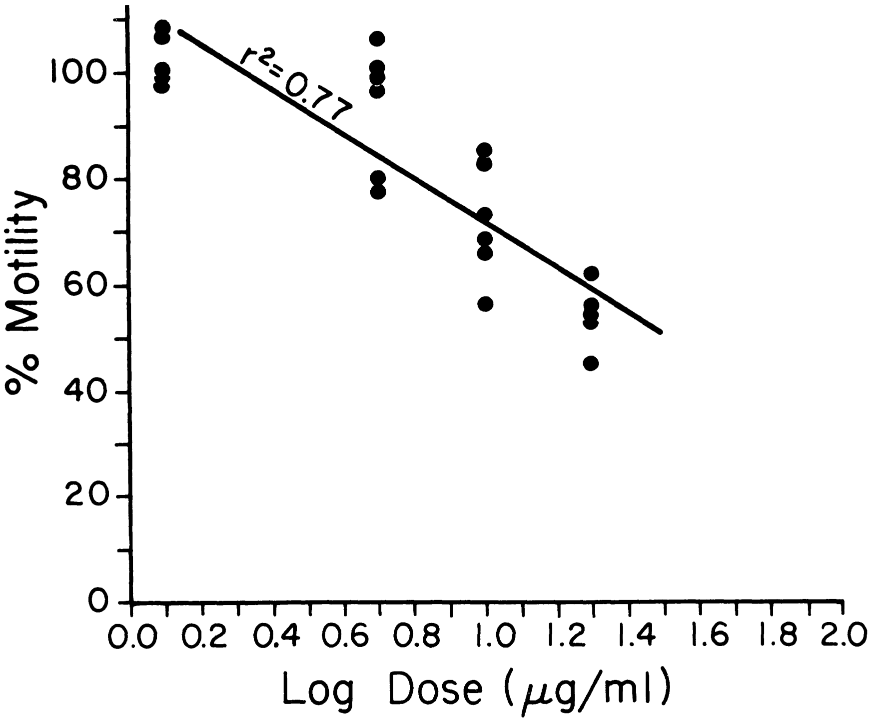

Fig. 1. Dosage-response relationship between ricin and motility of *Meloidogyne incognita* second-stage juveniles.

experiments, Davis and Rich (2) calculated lower EC_{50} values for nicotine than those found in the present studies. However, the data reported herein, compare unfavorably with commercial nematicides such as aldicarb and fenamiphos which were found to have EC_{50} values of less than 1.00 µg/ml in similar tests (Opperman, unpubl.).

The results of these studies show that ricin has an adverse effect on the motility of *M. incognita*. The influence of lectins on nematode activity have been reported by others (9,19). However, the mode of action of ricin, is not understood. Ricin is a large, hydrophilic molecule which has affinity for D-galactose residues and its activity is inhibited by N-acetyl-D-galactosamine (14). The large size and hydrophilic nature of the ricin molecule could prevent significant penetration through the nematode cuticle. Generally, neutral, nonpolar hydrophobic molecules move through the cuticle with most ease (1). Researchers, however, have demonstrated strong and reversible binding of fluorescent *R. communis* agglutinin to the amphids of *M. incognita* (11). Even though experiments were not designed to test modification of chemotatic behavior, this could be another explanation. For example, the lectin concanavalin A binds

Fig. 1. Noninfected (ni) and root-knot infected (i) Japanese mint *Mentha arvensis* cv. MAS–1.

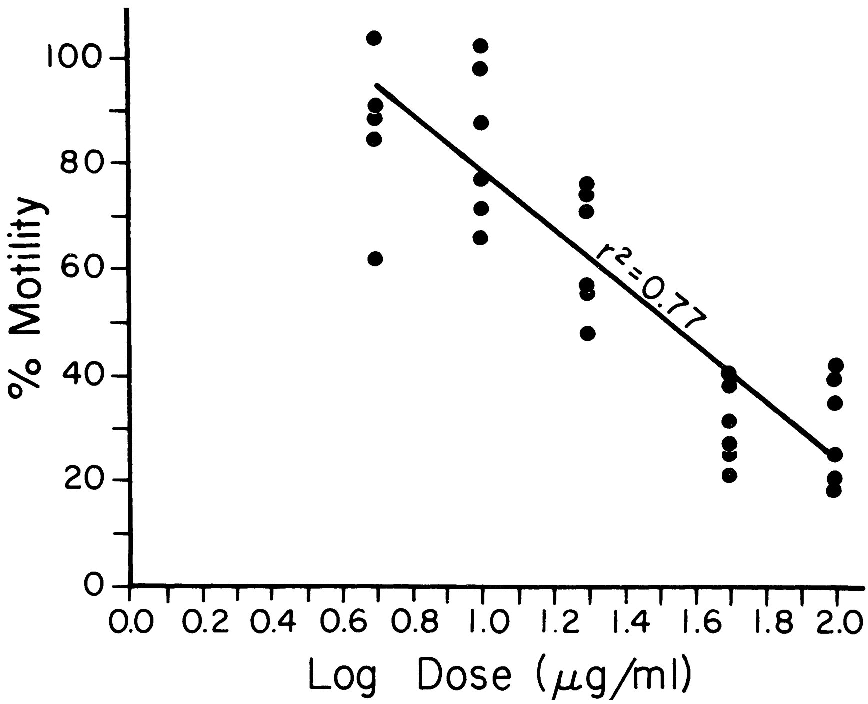

Fig. 2. Dosage-response relationship between nicotine and motility of *Meloidogyne incognita* second-stage juveniles.

selectively to mannose residues on the cuticle surface surrounding the cephalic sensilla of many nematodes, and ricin also may act in the same way. If this were the case, the capping of the receptors of ricin would result in a modification of chemotatic behavior (18). Ricin also is known to inhibit protein synthesis in vitro in mammalian systems, but whether this occurs in nematodes is not known (13). It is possible that amphids or other natural openings may provide means of entry for ricin and that inhibition of protein synthesis in neuronal cells may indirectly influence motility (i.e. cellular enzymes necessary for processing motility signals would be inhibited).

While further work on the mode of action is needed, it would appear that the comparatively high EC_{50} values may not render the compound useful as a nematicide. Ricin, however, may be one of the chemicals in castor bean meal that adversely affects plant-parasitic nematodes.

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