STRONG REPELLENCY OF THE ROOT KNOT NEMATODE, *Meloidogyne incognita* BY SPECIFIC INORGANIC IONS

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Abstract—Simple inorganic salts of the ions K⁺, NH₄⁺, Cs⁺, NO₃⁻, and Cl⁻ are strongly repellent to infective second-stage larvae of the root knot nematode, *Meloidogyne incognita*. Some of these salts are known to be beneficial to plant growth. The results suggest a new means of plant protection.

Key Words—Chemotaxis, repellents, *Meloidogyne incognita*, root knot nematode, inorganic ions, salts, gradients.

INTRODUCTION

Attraction of parasitic nematodes by plant roots may be the key to host recognition, specificity, and subsequent infection (Dusenbery, 1987; Steiner, 1925). On the other hand, environmentally tolerable repellents offer an alternative to pesticides in the protection of plants from these root parasites.

We have recently described a quantitative bioassay for the attraction and repulsion of plant parasitic nematodes by chemical substances (Castro et al., 1989). The method was illustrated with infective second-stage larvae of *Meloidogyne incognita* (MiJ₂) and chemotactic fractions isolated from cucumber roots. In the course of this work we have found that fertilized cucumber seedlings contained an additional repellent fraction. Examination of the fertilizer constituents prompted a scan of the response of MiJ₂ to a variety of inorganic salts. In this work we describe the surprisingly strong repellency of simple inorganic ions to this parasite. Our results suggest that salts beneficial to plant nutrition,
suitably applied, may be used to shield roots from nematode attack. They suggest a new means of plant protection.

METHODS AND MATERIALS

$^{14}$C-Labeled glycerine, glycine, and sodium acetate were commercial samples (New England Nuclear, Dupont) and were used without purification. Specific activities were UL labeled glycerine 9.1 mCi/mmol, [1-$^{14}$C]glycine 4.5 mCi/mmol, and [1-$^{14}$C]sodium acetate 1.8 mCi/mmol. HFeEDTA $\cdot$ 2H$_2$O was prepared by literature procedures (Garcia Basalotte et al., 1986) and recrystallized twice from water-acetone before use. Monosodium and potassium salts were obtained from it by reaction with stoichiometric amounts of the corresponding carbonates. The resulting monohydrate complexes were recrystallized from water-acetone. All other salts used in this work were purchased as analytical reagent-grade substances.

Bioassay. The essence of our bioassay is to effectively restrict nematode movement to one dimension. Narrow 0.8% agarose tracks are employed. Precise gradients of test substance can be established, and the population of nematodes along the track can be monitored with time. The test substance is placed at the end of the track at fixed concentration. After a suitable time ($\Delta T$), the nematodes (100-150 larvae) are inoculated into the center of the track and the population distribution is allowed to ‘‘develop.’’ Employing the average number of animals in each 0.5-cm section of track from replicate runs ($N$), and the distance of that section from the center ($D$), we have defined an attractant ($A$) repellent ($R$) ratio at time $t$ as:

$$(A/R)_t = \frac{\sum (N)(D)_{\text{toward}}}{\sum (N)(D)_{\text{away}}}$$

The sum $(N)(D)$ is the distribution of the population weighted for the distance it has moved towards or away from a given substance in that time ($t$). The details of this procedure have been presented (Castro et al., 1989).

Gradients. The salts KNO$_3$, NH$_4$NO$_3$, and NaOAc and the water-soluble substances glycerine and glycine all establish the same gradient under the same conditions of time and concentration. For example, the 22-hr, 10$^{-2}$ M gradient for KNO$_3$ shown in Figure 1 is identical for all of these substances. Consequently, we assume the gradients for the other salts in this report are the same in this matrix. The concentration of radiolabeled substances in each 0.5-cm section of track was determined by liquid scintillation counting as previously described (Castro et al., 1989). Nitrate ion was determined spectrophotometrically by reduction to nitrite, diazotization of sulfanilamide, and coupling with N-(1-naphthyl)ethylenediamine (Chow and Johnstone, 1962). The intensity of