Influence of concentration gradients of salts on the behaviour of four plant parasitic nematodes

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SUMMARY

The movement of juveniles of one population of *Meloidogyne javanica* and of two populations of *M. incognita* was studied in salt gradients created in agar by Hoagland solutions between 0.13×10^{-3} and 13×10^{-3} M/l. The capacity to migrate toward the region having the lower salt concentration was common within these three populations.

The behaviour of juveniles of *Heterodera oryzae* and of *Scutellonema cavenessi* (randomly selected regardless of age, sex or stage) was compared with the movement of juveniles of *M. javanica* in concentration gradients created in agar by KNO_3, NaCl, $Ca(NO_3)_2$ and $MgSO_4$ solutions at 0.031×10^{-3}, 1.25×10^{-3} and 5×10^{-3} M/l. With juveniles of *M. javanica* a significant repulsion was observed with the four salts at concentrations of 5×10^{-3} and 1.25×10^{-3} M/l. With juveniles of *H. oryzae* only NaCl was repellent. With *S. cavenessi,* the four salts failed to exhibit a repulsion.

It would appear logical to assume that the capacity to migrate toward the region having the lower salt concentration, in a concentration gradient of salt is common within *M. javanica* and *M. incognita* populations ; however, it appears that this capacity is not common within all plant parasitic nematodes.

RÉSUMÉ

Orientation du déplacement de quatre nématodes phytoparasites dans des gradients de concentration de sels minéraux

La capacité de migrer vers la région ayant la plus faible concentration en sels est commune aux juvéniles d'une population de *M. javanica* et de deux populations de *M. incognita* lorsqu'ils sont placés dans des gradients créés dans une colonne de gélose par des solutions de Hoagland ayant une concentration saline totale comprise entre $0{,}26 \times 10^{-3}$ et 13×10^{-3} M/l.

Les comportements des juvéniles de *M. javanica,* des juvéniles de *H. oryzae* et de *S. cavenessi* (sans distinction d'âge, de sexe ou de stade) ont été comparés dans des gradients créés dans une colonne de gélose par des solutions de KNO_3, NaCl, $Ca(NO_3)_2$ et $MgSO_4$ avec des concentrations de $0{,}031 \times 10^{-3}$; $1{,}25 \times 10^{-3}$ et 5×10^{-3} M/l.

Avec les juvéniles de *M. javanica,* une répulsion significative est observée avec les quatre sels à des concentrations de 5×10^{-3} et $1{,}25 \times 10^{-3}$ M/l. Avec les juvéniles de *H. oryzae,* seul NaCl est répulsif. Avec *S. cavenessi* aucun des quatre sels ne provoque de répulsion.

S'il semble logique de supposer que la capacité de migrer vers la région ayant la plus faible concentration, dans un gradient de sels minéraux, est commune aux populations de *M. javanica* et de *M. incognita,* il apparaît que cette faculté n'est pas générale à tous les nématodes phytoparasites.

All juveniles of *Meloidogyne javanica* (Treub, 1885), Chitwood, 1949, derived from a clone established from a single egg mass, placed in gradients created in agar by salt solutions between 0.125×10^{-2} and 0.5×10^{-2} M/1, moved preferentially toward the region having the lower salt concentration (Prot, 1978 a). Examination of tracks of these juveniles in salt gradients indicated that the repulsion from salts was the result of a real orientation of their movements (Prot, 1978 b). In order to determine if this capacity of orientation in salt gradients was specific to this clone or whether it occured frequently among the *Meloidogyne* and the other genera found in West Africa, movement of one field population of *M. javanica*, two populations of *Meloidogyne incognita* (Kofoid & White, 1919), Chitwood, 1959, *Heterodera oryzae* Luc & Berdon, 1961 and *Scutellonema cavenessi* Sher, 1963 was studied in salt gradients.

The movement of the three populations of *Meloidogyne* was studied in concentration gradients created in agar by Hoagland solution between 1.3×10^{-4} M/l and 1.3×10^{-2} M/l. The behaviour of juveniles of *H. oryzae* and of S. *cavenessi* was compared with the movement of juveniles of *M. javanica* in concentration gradients created in agar by KNO_3, NaCl, $Ca(NO_3)_2$ and $MgSO_4$ solutions at 0.031×10^{-2}, 0.125×10^{-2} and 0.5×10^{-2} M /l.

Materials and Methods

The origins of nematodes used in these experiments were as follows :

M. javanica: roots of kenaf (*Hibiscus cannabinus* L.) collected from the field. (FAO Station, Cambérène, Sénégal).

M. incognita, population 1 : maintained in the glasshouse on a local cultivar of sweet potato (*Ipomoea batatas* Poir.).

M. incognita, population 2 : roots of tomato (*Lycopersicon esculentum* L. cv. Roma) collected from the field (BUD-Sénégal Exploitation).

H. oryzae: collected from the type locality in Ivory Coast and maintained in the glasshouse on rice (*Oryza sativa* L. cv. Morobérékan).

S. cavenessi: soil around roots of groundnut (*Arachis hypogea* L.) collected from the field. (Station ISRA, Bambey, Sénégal).

Only individuals not more than 48 h in age were used, except for *S. cavenessi* in which individuals were randomly selected regardless of age, sex or stage.

All necessary manipulations were made under sterile conditions ; all glassware, salt solutions, demineralized water and agar were autoclaved (120 °C for 20 min).

The composition of Hoagland solution used in these experiments is given below :

$Ca(NO_3)_2$	5×10^{-3} M/l	H_3BO_3	1.69 mg/l
KNO_3	5×10^{-3} M/l	$MnSO_4, H_2O$	1.68 mg/l
$MgSO_4$	2×10^{-3} M/l	$ZnSO_4, H_2O$	0.0876 mg/l
KH_2PO_4	1×10^{-3} M/l	$(NH_4)6Mo_7O_{24}, 4H_2O$	0.0368 mg/l
		$CuSO_4$, $5H_2O$	0.0788 mg/l

Effect of Hoagland solution on the movement of juveniles of two populations of *M. incognita* and the population of *M. javanica* was studied in U-tubes as previously described (Prot, 1978 *a*) with an agar column of 1.2 cm in length in the central part of the U-tube. An undetermined number of juveniles was introduced into the agar 3 h after the salt solution (1.5 cm^3) was placed in one branch of the U-tube, whereas 1.5 cm^3 of demineralized water was placed in the other. Twenty-four hours later the juveniles were recovered from the two ends of the tube. The experiments were made in the dark at 28 °C and were repeated sixty times with *M. javanica* and the first population of *M. incognita* and forty times with the second population of *M. incognita* for the various concentrations of Hoagland solution between 1.3×10^{-4} M/l and 1.3×10^{-2} M/l.

Effects of KNO_3, NaCl, Ca $(NO_3)_2$ and $MgSO_4$ on the movement of juveniles of *M. javanica* and *H. oryzae* and of female, male and juveniles of S. *cavenessi* were studied, in the same U-tubes as previously described but with an agar column 5 cm in length, with three concentrations of salt (0.031×10^{-2}, 0.125×10^{-2} and 0.5×10^{-2} M/l). Ten nematodes were introduced singly into the center of the agar 36 h after the introduction of 1.5 cm^3 of the salt solution in one branch of the U-tube (1.5 cm^3 of demineralized water were placed in the other). Twenty-four hours later the nematodes were recovered from the two ends of the tube. The experiments were made in the dark

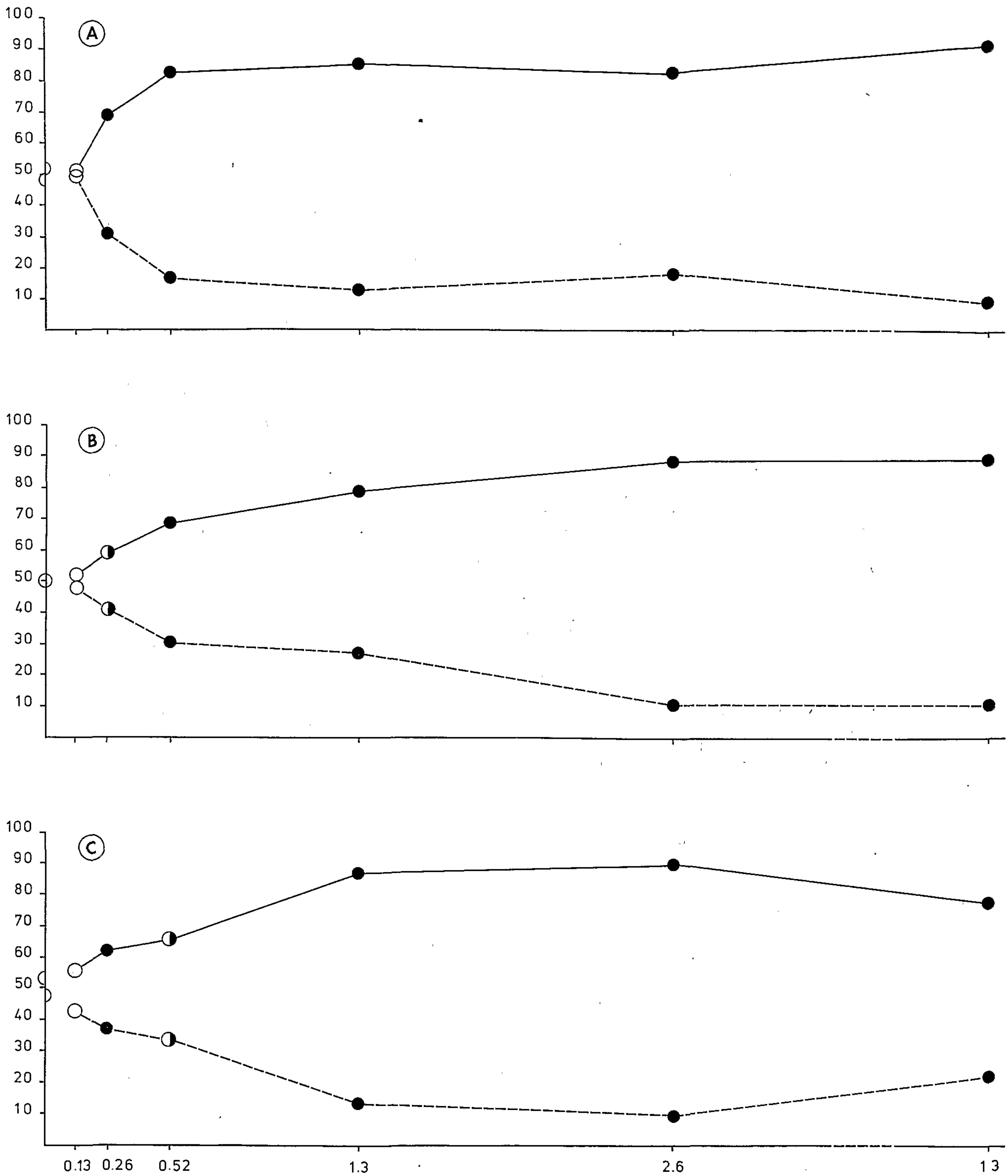

Fig. 1 : Influence of Hoagland solution on the movement of juveniles of *Meloidogyne javanica* (A), *M. incognita* population 1 (B) and *M. incognita* population 2 (C). Abscissa : concentration of the salt solution as $M \times 10^{-3}$ M/l. Ordinate : mean percentages of juveniles recovered in demineralized water (solid line) and in salt solution (dotted line). Black circles indicate a significant difference (sign test at a probability of 0.01) between the numbers recovered in the two branches of the tube ; black and white circles indicate a significant difference at a probability of 0.05 with the same test. White circles indicate that the figures were not significant.

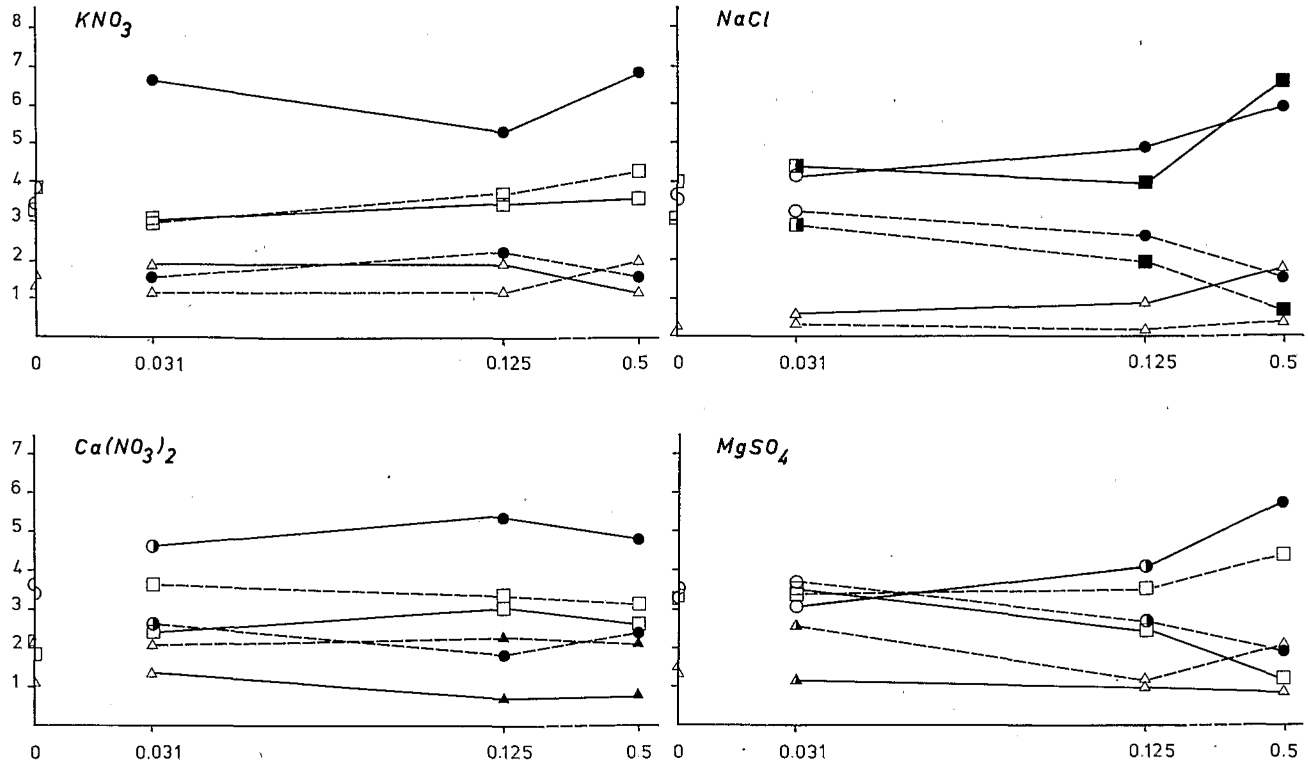

Fig. 2 : Influence of KNO_3, NaCl, $Ca(NO_3)_2$ and $MgSO_4$ on the movement of juveniles of *Meloidogyne javanica* (circles), *Heterodera oryzae* (squares) and undetermined stages of *Scutellonema cavenessi* (triangles). Abscissa : concentration of the salt solution as $M \times 10^{-2}$ M/l. Ordinate : numbers of nematodes recovered in demineralized water (solid line) and in salt solution (dotted line). Black symbols indicate a significant difference (Wilcoxon test at a probability of 0.01) between the numbers recovered in the two branches of the tube ; black and white symbols indicate a significant difference at a probability of 0.05 with the same test. White symbols indicate that the numbers were not significant.

at 28 °C. Twenty replications were used for all nematodes and concentrations of the four salts tested.

Results

Figure 1 shows the mean percentages of juveniles found in Hoagland solution and in the demineralized water after 24 h of migration. Results were the same for the three populations of *Meloidogyne* tested (A : *M. javanica*, B : *M. incognita* population 1 ; C : *M. incognita* population 2). In effect, juveniles of these populations moved preferentially toward the region having the lower salt concentration. The observed repulsion was significant in gradients created in agar by Hoagland solutions between 0.26×10^{-3} and 13×10^{-3} M/l.

Figure 2 summarizes the results of the effects of KNO_3, NaCl, Ca $(NO_3)_2$ and $MgSO_4$ at three concentrations on the movements of *M. javanica*, *H. oryzae* and *S. cavenessi*. With juveniles of *M. javanica* a significant repulsion was observed with the four salts at concentrations of 0.5×10^{-2} and 0.125×10^{-2} M/l. With juveniles of *H. oryzae* only NaCl repelled juveniles ; this repulsion was not observed with KNO_3, Ca $(NO_3)_2$ or $MgSO_4$. The four salts failed to exhibit a repulsion on *S. cavenessi* ; an attraction was observed with Ca $(NO_3)_2$ at concentrations of 0.125×10^{-2} and 0.5×10^{-2} M/l.

A mean number of 7.4 juveniles of *M. javanica* and 6.6 juveniles of *H. oryzae* of the 10 introduced per tube were recovered from the test solutions and demineralized water; however a mean of only 2.6 was observed with *S. cavenessi*.

Discussion

On the basis of these results it is concluded that the capacity to migrate toward the region having the lower salt concentration in concentration gradients of salts is common within populations of *M. javanica* and *M. incognita* from Senegal. It would appear logical to assume that the same ability exists within *M. javanica* and *M. incognita* populations from other geographical areas as well ; however from this study it appears that this capacity is not common within all plant parasitic nematodes. In effect, *H. oryzae* and *S. cavenessi* behaved differently from juveniles of *Meloidogyne*. Among four mineral salts tested KNO_3, $Ca(NO_3)_2$, $MgSO_4$ and NaCl, only NaCl exhibited a repulsive effect on the juveniles of *H. oryzae* and no repulsion was observed on *S. cavenessi* with any salt tested.

The sensitivity of the juveniles of *Meloidogyne* is very great, i.e. they moved preferentially towards the region having the lower salt concentration in a gradient created in agar by Hoagland solution between 0.26×10^{-3} and 13×10^{-3} M/l. These concentrations are close to those found in the soil (Adams, 1974).

It is possible that mineral salts can influence the behaviour of juveniles of *Meloidogyne* in the soil when a concentration gradient exists. At least two factors can induce concentration gradients of mineral salts : a moisture gradient and plant roots. Indeed, in soils containing adequate quantities of mineral salts an increase of moisture can dilute the soil solution and plant roots can affect the distribution of ions in the soil because of absorption of nutrients and water and because their physical presence affects soil bulk density.

Plant roots absorb water, which contains inorganic ions, causing a flow of water from the soil to the root surface. The concentration of an ion in the soil near the root may increase, remain the same or decrease depending on the balance between the rate of supply to the root by mass-flow and the rate of absorption into the root. When a concentration gradient normal to the root is established the ions diffuse.

When mass-flow and diffusion exceed uptake rate, an accumulation occurs in the soil near the root. When uptake rate exceeds mass-flow and diffusion, a depletion occurs in the soil near the root.

Mineral salt depletion near the roots has been demonstrated by a number of authors, for example : a depletion of calcium around lupine roots (Barber, 1974) and a depletion of rubidium about corn roots (Barber, Walker & Vasey, 1963). In the same way, concentration gradients of mineral salts created by root uptake have been measured in the soil over a distance of 1.2 cm perpendicular to roots with the lower concentration at the root surface. Thus, with onion seedlings such concentration gradients of phosphorus (Bagshaw, Vaidyanathan & Nye, 1972), chloride (Dunham & Nye, 1974), potassium and phosphate (Dunham & Nye, 1976) were observed.

In these cases, the ability of the juveniles of *M. javanica* and *M. incognita* to move preferentially toward the region having the lower salt concentration could partly explain root attraction for these nematodes.

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