

directe du bromure de méthyle sur les nématodes celui-ci éliminant totalement du sol les espèces endoparasites, et presque complètement les espèces ectoparasites (Tabl. 1).

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REFERENCES

- ANON. (1978). S. Afr. Sugar Ass. Exp. Stat., *Ann. Rep.* 1977-1978 : 21.
 ANON. (1979). S. Afr. Sugar Ass. Exp. Stat., *Ann. Rep.* 1978-1979 : 13.
 ANON. (1980a). S. Afr. Sugar Ass. Exp. Stat., *Ann. Rep.* 1979-1980 : 17.

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ANON. (1980b). Sugar cane diseases in South Africa. *S. Afr. Sugar Assoc. Bull.*, 9 (rev.) : 5.

CADET, P. & MERNY, G. (1978). Premiers essais de traitements chimiques contre les nématodes parasites de la canne à sucre en Haute-Volta, *Revue Nématol.*, 1 : 53-62.

CADET, P., QUÉNÉHERVÉ, P. & MERNY, G. (1982). Pathogenic action of nematodes on irrigated sugarcane. *Revue Nématol.*, 5 : 205-210.

CADET, P. & SPAULL, V. W. (1985). Comparison of the relationship between nematodes and sugarcane in South and West Africa : Plant cane. *Revue Nématol.*, 8 : 131-142.

HITCHCOCK, B. (1979). New chemicals for nematocidal control in cane. *Cane Grow. quart. Bull.*, 42 : 62-65.

MOBERLY, P. K. & CLOWES, M. J. S. (1981). Trials with nematicides registered for use on sugarcane in South Africa. *Proc. S. Afr. Sugar Technol. Assoc.*, 1 : 7.

PICARD, D. (1968). Étude sommaire du type de loi de distribution de certains paramètres racinaires. *Cah. ORSTOM, Sér. Biol.*, 5 : 3-13.

CHITIN IS PRESENT IN GELATINOUS MATRIX OF *MELOIDOGYNE*⁽¹⁾

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Chitin has been found "only in the (egg) shell and in no other parts of adult or larval nematodes" (Bird, 1976). The chemical composition of the gelatinous matrix — studied only in *Meloidogyne* — is reported to consist of an acid mucopolysaccharide-tanned protein complex and a number of enzymes (Bird & Rogers, 1965). In the present study we employed a relatively novel and sensitive technique to find out whether chitin is present in the gelatinous matrix (G.M.) and egg shell (E.S.) of three different plant-parasitic nematodes.

Chitin, a polymer of N-acetylglucosamine, has been known to be insoluble in strong alkali solutions, while undergoing hydrolysis in strong mineral acids, even at room temperature (Muzzarelli, 1977). It can be detected by its specific lectin, wheat germ agglutinin (WGA), although this lectin is also specific for terminal glycoprotein-bound sialic acid units (Roth, 1978). E.S. and G.M. of *Meloidogyne javanica*, *Tylenchulus semipenetrans* and

Meloidoderita kirjanovae were dissected and separated from infected tomato, citrus and mint roots, respectively, as described by Bird and McClure (1976). The labelled lectins: fluorescein isothiocyanate FITC — WGA, rhodamine-limulus polyphenus (from horseshoe crab, specific for sialic acid, glucuronic acid and phosphoryl choline analogs; Roche & Nonsigny, 1974) — were reacted with the G.M. and E.S. by the labelling technique described for neural crest cells (Sieber-Blum & Cohen, 1978). The specificity of the observed lectin absorption and the fluorescence microscopy observations were evaluated as described by Sieber-Blum and Cohen (1978). E.S. of *T. semipenetrans* and *M. kirjanovae* were highly fluoresced by FITC-labelled WGA (Fig. 1), as were E.S. and G.M. of *M. javanica* (Fig. 2), thereby confirming earlier observations with this nematode (Spiegel & Cohn, 1982). However, this lectin did not reveal any fluorescence with the G.M. of

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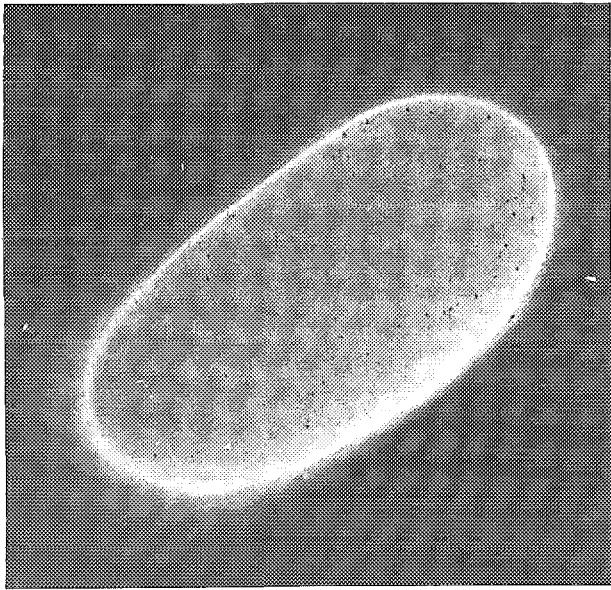


Fig. 1. Binding of fluorescent lectin (wheat germ agglutinin) to egg shells of *Tylenchulus semipenetrans* ($\times 1,250$).

M. kirjanovae and *T. semipenetrans*. Preincubation of the WGA lectin with its respective haptenic sugar, N-acetyl-D-glucosamine, prevented its absorption by the E.S. of *T. semipenetrans*, whereas incubation with sugars other than the correct hapten inhibitor did not interfere with binding of the lectin. Overnight pretreatment of *M. javanica* E.S. and G.M. with 10 mg/ml chitinase (from *Streptomyces griseus*, Sigma), followed by labelling with WGA-FITC, revealed a significant reduction of the fluorescence in both E.S. and G.M., compared to the untreated ones. In addition, E.S. and G.M. of *M. javanica* were preincubated for 90 mn at room temperature with concentrated sulphuric acid (H_2SO_4 , pH — 0.3), 1 N glacial acetic acid (CH_3COOH , pH — 2.9) for 30 mn at 100° , or with 10 N sodium hydroxide (NaOH , pH — 14.0) for 90 mn. They were then thoroughly washed with PBS (pH — 7.2) and labelled with WGA-FITC. Most of the H_2SO_4 — treated E.S. and G.M. was dissolved and the remainder showed no labelling. In the CH_3COOH -treatment, almost all the G.M. was dissolved and the remaining eggs were barely fluorescinated. However, NaOH -treated material was not damaged and revealed strong labelling with WGA-FITC. The rhodamine-limulus poly-

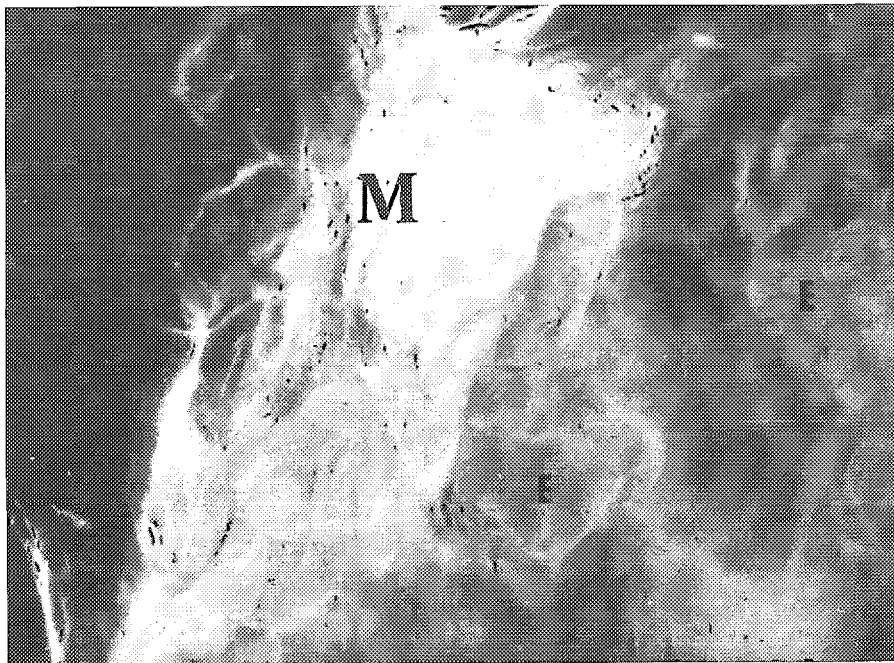


Fig. 2. Binding of fluorescent lectin (wheat germ agglutinin) to the gelatinous matrix (M) and egg shells (E) of *Meloidogyne javanica* ($\times 200$).

phemus lectin did not reveal any fluorescence with the G.M. or with the E.S. of the three aforementioned nematodes. This finding ruled out the possibility that the described WGA binding patterns were the result of sialic acid presence.

Thus, while chitin was detected in E.S. of all three nematode species, it was only found in G.M. of *M. javanica*. This difference in the G.M. composition of three different phytonematodes may perhaps be better understood in the light of the known differences in G.M. formation among the three species. In *M. javanica*, G.M. is produced by specific rectal glands and secreted through the anus, whereas in *T. semipenetrans* it is produced on the excretory system and exuded via the excretory pore (Maggenti, 1962). G.M. formation in *M. kirjanovae* has not been described in detail, but our observations indicate that it emerges from the vulval slit, as in *Heterodera cruciferae* (Mackintosh, 1960).

REFERENCES

- BIRD, A. F. (1976). The development and organization of skeletal structures in nematodes. In: Croll, N. A. (Ed.). *The Organization of Nematodes*. London, Academic Press 107-137.
- BIRD, A. F. & McCLURE, M. A. (1976). The tylenchid (Nemato-) egg shell : structure, composition and permeability. *Parasitology*, 72 : 19-28.
- BIRD, A. F. & ROGERS, G. G. (1965). Ultrastructural and histochemical studies of the cells producing the gelatinous matrix in *Meloidogyne*. *Nematologica*, 11 : 231-238.
- MACKINTOSH, G. MACD. (1960). The morphology of the brassica root eelworm, *Heterodera cruciferae* Franklin, 1945. *Nematologica*, 5 : 158-165.
- MAGGENTI, A. R. (1962). The production of the gelatinous matrix and its taxonomic significance in *Tylenchulus* (Nematoda : Tylenchulinae). *Proc. helminth. Soc. Wash.*, 29 : 139-144.
- MUZZARELLI, R. A. (1977). *Chitin*. New York, Pergamon Press.
- ROCHE, A. C. & NONSIGNY, M. (1974). Purification and properties of limulin : a lectin (agglutinin) from hemolymph of *Limulus polyphemus*. *Biochem. biophys. Acta*, 371 : 242-254.
- ROTH, J. (1978). *The lectins : molecular probes in cell biology and membrane research*. Iena, Gustav Fischer Verlag, 186 p.
- SIEBER-BLUM, M. & COHEN, A. M. (1978). Lectin binding to neural crest cells. *J. Cell. Biol.*, 76 : 628-638.
- SPIEGEL, Y. & COHN, E. (1982). Lectin binding to *Meloidogyne javanica* eggs. *J. Nematol.*, 14 : 406-407.

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REMARQUES SUR LE GENRE *LAIMAPHELENCHUS* FUCHS, 1937

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Deux travaux récents (Baujard, 1981; Hirling, 1982) sont consacrés au genre *Laimaphelenchus*. Baujard (1981) synonymise les genres *Laimaphelenchus* Fuchs, 1937 et *Ruidosaphelenchus* Laumon & Carle, 1971, considère les espèces *Laimaphelenchus moro* Fuchs, 1937 et *Laimaphelenchus ulmi* Khan, 1960 comme synonymes mineurs de *Laimaphelenchus penardi* (Steiner, 1914) Filipjev & Schuurmans Stekhoven, 1941, et décrit une nouvelle espèce, *L. pini*. Hirling (1982) considère le genre *Ruidosaphelenchus* comme valide et décrit trois nouvelles espèces, *Laimaphelenchus montanus*, *Laimaphelenchus silvaticus* et *Laimaphelenchus*

parvus, sans désigner ni les types ni les localités-types.

Hirling (1982) reconnaît que la seule différence séparant les genres *Laimaphelenchus* et *Ruidosaphelenchus* réside dans la morphologie des spicules. Nous avons signalé (Baujard, 1981) que la même situation existe dans le genre *Bursaphelenchus*; et d'autre part Giblin et Kaya (1983) considèrent le genre *Huntaphelenchoides* Nickle, 1970 comme synonyme mineur du genre *Bursaphelenchus* arguant du fait que la seule différence entre ces deux genres, à savoir la morphologie des spicules, n'est pas suffisante pour justifier l'existence de deux genres différents.

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