

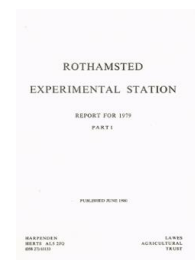
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## Report for 1979 - Part 1

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### Nematology Department

**A. R. Stone**

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## NEMATODOLOGY DEPARTMENT

A. R. STONE

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### Introduction

Dr. F. G. W. Jones retired in November after 24 years as Department Head; under his inspiration and sympathetic leadership the Department has attained a high international standing.

The Department is now host to the Overseas Development Administration's (ODA) Nematology Liaison Officer and his staff. This welcome addition will increase our involvement with tropical plant-parasitic nematodes. We now receive ODA support for work on the taxonomy of root-knot nematodes, enabling a return to active research in the department on this important group, and a Potato Marketing Board (PMB) grant to fund research on intracellular parasites of cyst nematodes. This year our report takes a different format with emphasis on taxonomy and morphology, although work has also continued, as in recent years, on aspects of plant-parasitic nematode biology, crop damage, and control by chemicals, resistant crops and biological agents. In future reports particular subjects will be chosen for special consideration. The report is shorter and inclusion of substantial quantities of data from experiments, which will appear in published papers, has been avoided.



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### Morphology and taxonomy

Plant and soil nematodes show relatively little morphological diversity. Groups of very similar species are common among plant-parasitic nematodes, especially among the sedentary forms with limited host ranges, and may be a product of co-evolution of parasites and hosts (*Rothamsted Report for 1978*, Part 1, 180–182). Among root ectoparasites with more catholic host ranges morphological distinctions between species are sometimes as slight. We seek to improve recognition of such species by supplementing classical morphology with hybridisation studies and electrophoresis of proteins. Ultra-structure and fine morphology may throw light on inter-relationships between taxa at the generic and higher levels as for instance in groups of cyst nematodes (*Rothamsted Report for 1972*, Part 1, 157–159; *for 1974*, Part 1, 175–176; and *for 1975*, Part 1, 192).

**Hybridisation studies.** Application of the biological species concept is the ultimate test of specific status for amphimictic animals. If barriers to gene flow due to genetic incompatibility, or other mechanisms, occur between populations, then they must be considered distinct species.

Of the members of the *Globodera* species complex parasitising Solanaceae, *G. pallida* and *G. rostochiensis* produce few cysts containing viable eggs when crossed and the F<sub>1</sub> juveniles are of low viability (*Rothamsted Report for 1969*, Part 1, 185; and *for 1973*, Part 1, 153–154). This demonstration of a barrier to gene flow supported their recognition as distinct species. In contrast, when reciprocal crosses using the same technique were made between *G. solanacearum*, *G. virginiae* and the unnamed Mexican cyst-nematode a larger percentage of females developed into cysts with embryonated eggs containing F<sub>1</sub> juveniles. The cysts so produced were inoculated on to tomato plants and F<sub>1</sub> juveniles released developed to cysts containing F<sub>2</sub> embryonated eggs. When these cysts were in turn inoculated on to tomato plants the multiplication rate from the F<sub>1</sub> to F<sub>2</sub> generation (F<sub>2</sub> cysts with embryonated F<sub>3</sub> eggs/F<sub>1</sub> cysts with F<sub>2</sub> embryonated eggs) of the hybrids was  $\times 1.6$ – $9.6$  that of selfed lines  $\times 2.5$ – $2.8$ . *G. solanacearum*, *G. virginiae* and the Mexican cyst-nematode have similar morphology; further *G. solanacearum* and *G. virginiae* are both described from Virginia and have a common host range so that populations are likely to be sympatric. This, and the apparent absence of barriers to gene flow between the species suggest they are conspecific. (Stone, Rowe and Payne)

It has been suggested recently that *Pratylenchus fallax* and *P. penetrans* are conspecific. We have studied the reproductive compatibility of populations of these two root endoparasitic species using excised maize root and lucerne callus tissue cultures. Intraspecific crosses showed that breeding readily occurs under these conditions but interspecific crosses gave few reproductive cultures and progeny were few and probably F<sub>1</sub> generation only. There is evidently incompatibility between these species and their separate specific status is endorsed. (Perry, Plowright and Webb)

**Electrophoresis of stem nematode races.** Races of the stem nematode, *Ditylenchus dipsaci*, are presently distinguishable only by differences in host range, except for the 'giant race', a tetraploid with larger body size. Chemical characterisation would help identification of races of this important endoparasite. Electrophorograms of oat race cultured on oats and on field beans are identical except for the presence in the former of a faint band, migrating to about 2 mm from the 'origin'. This small difference is probably due to protein from the host plant. Patterns obtained from giant race *D. dipsaci* cultured on beans show differences from the oat race patterns almost certainly due to differences in nematode proteins. The band referred to above is lacking. In addition an intense band, migrating to about 6.5 mm in the oat race patterns is absent in the giant race, but a strong



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band in giant race samples migrating to about 7.5 mm is absent in oat race. Distribution and numbers of bands in the middle regions of the gels also vary slightly between giant and oat races, and the giant race pattern has a distinct band at about 35 mm that is absent in the oat race. The difference in protein complement observed between oat and giant races is as great as that between some closely related nematode species. (Greet)

**Ultrastructure of nematode sperm.** Sperm characters of the various nematode groups so far studied may be a useful indication of affinities between groups. We have previously described sperm structure and development in species of Heteroderidae and Meloidogynidae (Secernentea, Tylenchida, Tylenchina), Aphelenchoididae (Tylenchida, Aphelenchina) and Rhabditidae (Secernentea, Rhabditida, Rhabditina) and have now studied the structure of sperms of the longidorid species *Longidorus macrosoma* and *Xiphinema index* (Adenophorea, Dorylaimida, Dorylaimina). Like other nematode sperm those of longidorids are amoeboid and in these two species the whole sperm surface gives rise to very many pseudopodia and filopodia, more even than in the tylenchine sperm. Sperm in the seminal vesicle of *Longidorus* are about 10  $\mu\text{m}$  across, spherical to slightly ellipsoidal, whereas those of *Xiphinema* are more elongate, about 10  $\mu\text{m}$  long and 5  $\mu\text{m}$  wide. Their outer membrane is lined with parallel arrays of microtubules of width similar to those in *Meloidogyne* sperm. The central nucleus consists of a network of chromatin strands, coarse in *Xiphinema*, finer in *Longidorus*, in an ellipsoidal capsule formed by the fusion of mitochondria. The mitochondria are all orientated along the long axis of the enveloping structure. A junctional complex is formed where individual mitochondria adhere to one another and a layer of electron-dense granular material surrounds the whole mitochondrial complex. The only other inclusions in the sperm cytoplasm are bundles of fibrillar material, seen in other nematode sperm, and the centrioles. The organelles, called 'membrane specialisations', found in aphelenchoid and rhabditid sperm and in other orders, are absent from longidorid and tylenchine sperm.

Only very few representatives of each nematode group have been studied so far but there is no reason to believe that they will prove to be untypical. Dissimilarities between the Aphelenchina and the Tylenchina in sperm structure together with the cytology of the anterior alimentary tract, stylet structure and protractor musculature, suggest that these two groups may be less closely related than is now thought. Sperm of Tylenchina have more attributes in common with those of the Dorylaimida examined than with the Aphelenchina sperm, a provoking observation. (Shepherd and Clark)

**Head fine morphology of Rotylenchinae and Tylenchorhynchidae.** Examination with the scanning electron microscope of the head region of spiral nematodes (Rotylenchinae) from Britain showed that five *Helicotylenchus* spp. have a perioral disc surrounded by a first head annule which varies within and between species in the amount it is divided. Amphid apertures are generally distinct, in a lateral position at the edge of the perioral disc. In *H. pseudorobustus* the perioral disc is elongated in the dorso-ventral plane and the first head annule undivided. In *H. vulgaris*, *H. exallus*, *H. digonicus* and *H. canadensis* the perioral disc is round to oval and elongated laterally, if at all. The first head annule is often grooved, indicating the lateral sectors; subventral and subdorsal sectors are sometimes also clearly demarcated but variation within all five species is such that even the lateral sectors are not always clearly indicated. By contrast, in *Rotylenchus robustus*, *R. fallorobustus*, *R. goodeyi* and *R. pumilus* the first head annule is clearly divided into six sectors, and *R. robustus* has additional dorsal and ventral divisions. Cephalic and labial papillae are generally not visible in any of the above species except for *R. fallorobustus* and *R. pumilus* where six pits (opening of labial papillae) are present immediately around the oral aperture.



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Within Tylenchorhynchidae (stunt nematodes) 'face' views are very variable between and within genera. In *Amplimerlinius* (*A. icarus* and *A. socialis*) the face lacks any distinct division and the widely spaced amphid apertures tend to be displaced dorsally. The face pattern of *Nagelus obscurus* is similar but somewhat elongated laterally. In *Merlinius microdorus* and *M. nanus* the lateral sectors are marked by distinct grooves radiating from the amphid apertures but the subventral and subdorsal regions are indicated only by slight depressions. *Tylenchorhynchus lamelliferus* has a rectangular perioral plate surrounded by an undivided first head annule. In *T. dubius* and *T. judithae* the perioral plate is divided into subdorsal and subventral sectors and the first two or three head annules converge on the amphid apertures; these two species have six distinct raised papillae immediately around the oral aperture. *T. maximus* and *Scutylenchus quadrifer* have a distinct hexaradiate pattern with prominent lateral sectors. *Geocenamus longus* has a prominent perioral disc with the whole head region divided into six sectors by deep grooves. (Hooper and Cham)

**Morphology of potato cyst-nematodes.** Following demonstrations that an isolate of *G. pallida* (Cadishead) has a lower optimum temperature for multiplication than an isolate of *G. rostochiensis* (Feltwell) (*Rothamsted Report for 1976*, Part 1, 203; and *for 1977*, Part 1, 177) the effect of temperature during development on characters used in separating these morphologically similar species was investigated.

Cyst size varied considerably with temperature over the range 12 to 22°C, and was largest at the optimum temperature for multiplication of each species. Curves for the size against temperature are best described by cubic equations. Cyst length/breadth ratio, a value sometimes used in taxonomy, did not vary significantly. The distance between anus and vulval basin, and the number of cuticular ridges crossing the anus-vulval basin axis were related to cyst size and values for the two species were almost identical at 22°C. Although of importance in the separation of *G. pallida* from *G. rostochiensis* these last characters are of limited use because of overlap in their ranges and it is apparent that specimens cultured in the glasshouse, where temperatures are typically 20–25°C, will be most difficult to separate. Granek's ratio (anus to vulval basin distance/vulval basin length) also varied significantly with temperature but in a linear manner and of all cyst characters most effectively distinguished the two species. Second-stage juvenile length was temperature dependent, with that of *G. rostochiensis* juveniles increasing significantly over the range 12 to 22° and that of *G. pallida* juveniles decreasing significantly. At 22°C body length was ineffective as a differential character. Stylet length of *G. rostochiensis* was independent of temperature, that of *G. pallida* was just significantly dependent at  $P=0.05$ . Stylet length has the greatest utility of all juvenile and cyst characters. (Stone and Parrott)

**Morphometric formulae.** In studying how plant-parasitic nematodes feed we often need to know volumes and sectional areas of the lumina in different regions of the anterior alimentary tract, and how these quantities change during activity. For example, the volume of sap drawn from the host cell into the oesophageal feeding pump is important in studies of host-parasite relations. Accordingly a series of simple formulae is being developed that gives, among other quantities, the lumen volume of a tri-radiate feeding pump at any degree of opening. The pump lining plates can be a curved shape (as in many tylenchids) or elongate (e.g. *Longidorus* spp.) with straight, circular or elliptical ends. An example is the formula for volume in a pump with semi-elliptical lining segments:

$$1.33 L r^2(3 \cos \theta \sin \theta + \sqrt{3} \sin^2 \theta)$$



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where  $L=0.5$  lumen length,  $r$ =radius of fully-closed pump lining where it is greatest (i.e. as seen in an equatorial section) and  $\theta$ =half the internal angle of the points of the three-pointed star represented by a transverse section of the lining at a given degree of opening. (Seymour, with Dr. G. M. Jarman, Zoology Department, University of Bristol)

**Cyst nematodes of uncultivated land.** Cysts occurred in 45% of samples from soil around the roots of grasses and in 75% from around dicotyledons growing in nature reserves in Middlesex, Hertfordshire, Bedfordshire and Huntingdonshire. Most of those from dicotyledonous plants are in the *Heterodera goettingiana* group and only a much smaller proportion in the *H. schachtii* group, an unexpected result because the latter has the greater host-range. The high incidence of cyst nematodes in pockets of uncultivated land in an otherwise intensively cultivated region emphasises the possibility of poorly known taxa intruding into agricultural samples. Of samples from eight sand dune and salt marsh sites on the coast of southern and eastern England and South Wales 39% contained cyst nematodes, chiefly the previously recorded but undescribed species on marram grass (*Ammophila arenaria*) and sea lavender (*Limonium vulgare*). These two species are being described. (Hill and Stone)

**New records.** During the routine examination of soil samples the following were recorded for the first time from Britain: *Geocenamus longus* from Scotland, *Merlinius joctus* from Cornwall and *Tylenchorhynchus microphasmis* from Bedfordshire. (Hooper and Cham)

A cyst nematode received from Egypt, parasitising *Trifolium alexandrinum*, is *Heterodera daverti* reported previously only from western Europe on *T. repens*. The centre of diversity for the clovers is in the eastern Mediterranean and the presence of this nematode in that region is another example of a cyst nematode occurring in the locality where its host group is centred, indicating that co-evolution of host and parasite has taken place. (Stone, Bennison and Burrows)

### Cyst nematode biology

**Hatching of cyst nematodes.** Atkinson and Ballantyne (*Annals of Applied Biology* (1979) 93, 191-198) suggested that hatching of *Globodera rostochiensis* might be a calcium-mediated process. In support of the hypothesis, they reported that the inhibitors of calcium transport systems in the cell, lanthanum chloride and ruthenium red, inhibited the hatching of *G. rostochiensis*. We also find that a 4 mM solution of lanthanum chloride in potato root exudate inhibits hatching by about 50%. However, the same concentration of lanthanum chloride in distilled water has a similar hatching activity. Apparently lanthanum chloride does not fully inhibit hatching activity. 4 mM ruthenium red added to potato root exudate and then removed from the solution, destroyed its hatching activity. The failure of solutions of potato root exudate containing ruthenium red to cause hatching of *G. rostochiensis* may therefore be due to inactivation of the hatching factor in the exudate rather than interference with the hatching mechanism in the egg. In other experiments substantial hatching of *G. rostochiensis* was obtained with potato root exudate solutions from which calcium ions had been removed and which contained up to 12 mM of the chelating agent, 1,2-di(2-aminoethoxyl)-ethane-NNN'N'-tetra-acetic acid. We conclude that there is, at present, no direct evidence that calcium has an essential part in the hatching mechanism of *G. rostochiensis*. (Clarke and Hennessy)

Some eggs in cysts of *G. rostochiensis* from a field soil hatched in water, others only in potato root exudate. Multiplication of nematodes hatching in water produced a second



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generation with a water hatch twice as great as that of nematodes selected for hatch in exudate. This suggests that the ability to hatch in water rather than exudate may be inherited in a simple manner. (Evans)

Dormancy in potato cyst-nematodes is reported to begin at a time when flowering occurs in the potato crop and tubers are initiated, becoming the primary sink for nutrients. Cysts of *G. rostochiensis* were picked off the roots of three potato cultivars maturing in early, mid and late season. The plants had been inoculated with juveniles 8 weeks previously. Tuber formation was altered by growing the plants in 8.5 or 16 h days, and spraying them with gibberellic acid or lidocaine hydrochloride (a lipid anti-oxidant which delays ageing). Percentage hatch was greatest from cysts grown on the earliest maturing cultivar and least from cysts grown on that maturing last. Treatments which delayed tuber formation in all cultivars (gibberellic acid or lidocaine hydrochloride) also increased hatching from cysts in all cases. Short day growing conditions increased hatching from cysts collected from the early and late maturing cultivars. Physiological condition of the potato plant can apparently regulate onset of dormancy of the eggs produced by the nematodes feeding on the plant. A large hatch of eggs produced early in the season might, in the wild, permit an immediate second generation while increased dormancy in late season eggs would facilitate overwintering. (Evans)

Failure to induce a high rate of hatch may be an aspect of resistance to potato cyst-nematodes. This is not so for resistant *andigena* potato hybrids but past reports for *S. vernei* × *S. tuberosum* are conflicting, because earlier workers took no account of root weights of the hybrids. Numbers of juveniles hatching over 6 weeks in root exudate from two *Solanum vernei* × *S. tuberosum* hybrid lines, one resistant to *G. rostochiensis* and *G. pallida*, the other susceptible, were adjusted to the hatch g<sup>-1</sup> fresh root weight. Exudate from the resistant clone was significantly less effective in hatching *G. rostochiensis* (Ro1, Ro2) and *G. pallida* (Pa1, Pa3) ( $P=0.001-0.05$ ). There was no significant difference between the numbers of juveniles hatched by exudate from the susceptible ex *vernei* clone and a susceptible *S. tuberosum* (Arran Banner) control. (Turner and Stone)

**Pathotypes.** Resistance in *S. vernei* has been held to be of the horizontal or field type (i.e. effective against all pathotypes) but a *S. vernei* × *S. tuberosum* hybrid may depress multiplication of different potato cyst-nematode isolates to markedly different extents (*Rothamsted Report for 1978*, Part 1, 179–80). In *G. pallida* isolates the incidence of individuals developing on such hybrids may be increased by repeated multiplication on the same ex *vernei* resistant clone. By the third generation some isolates of *G. pallida* pathotypes Pa1, Pa2 and Pa3 showed up to a 150% increase in virulence compared with the initial generation (e.g. from 25 to 80% of the numbers of cysts of the same isolate developing on a susceptible control) but other isolates of the same pathotypes showed no increase. Resistance in *S. vernei* may be based on an array of major genes and the efficiency of the resistance against a nematode isolate then presumably depends on the frequency of corresponding virulence genes in the isolate. Where these occur frequently in the initial isolate they may be selected rapidly in the type of experiment described. Further evidence comes from an examination of resistance in the wild *S. vernei* parents of the hybrids. When tested against pathotypes resistance was effective at a high level against them all (0–5% of the multiplication rate on a susceptible control). Evidently some genes for resistance have segregated out during the breeding programme. The utility of potato cultivars with resistance derived from *S. vernei* may much depend upon the frequency with which they are grown and the amount of heterozygosity in nematode field populations. (Turner and Stone)

An isolate of *G. pallida* from the Woburn farm has been much used in experimental work during the past decade. Originally classified as pathotype E in the U.K. scheme, it



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has now been tested on the full range of differential hosts for *G. pallida* pathotypes and is Pa3 in the pathotype identification scheme now used widely in Europe. (Stone and Payne)

### Nematicides

#### Potato cyst-nematodes

**Methods of incorporating nematicides in soil.** Aldicarb ('Temik') or oxamyl ('Vydate') control potato cyst-nematode well when about 5 kg a.i. ha<sup>-1</sup> is thoroughly incorporated in the top 12–15 cm of the soil. This can be done by spreading the granules on the soil surface in spring, just before planting and rotavating the top 15 cm of the soil. Harrowing in the granules leaves two-thirds or more of them in the top 5 cm of the soil and results in less control in most soils. Rotavation is slow, mixes weathered with unweathered soil and may harm soil structure. The 'Vertical Band-Roterra' (VBR) technique (*Rothamsted Report for 1977*, Part 1, 183), which overcomes these difficulties, was further assessed in silt loam at Benwick, Cambs. Before King Edward potatoes were planted, oxamyl as 10% granules or in aqueous solution was blown into vertical bands in the top 12–15 cm of the soil and mixed laterally with the soil by a rotary harrow (Lely 'Roterra') travelling at 1 mph and 220 or 300 rpm. This was compared with spreading the granules on the surface and incorporating them by rotavation. Immediately after treatment the distribution of oxamyl was determined in successive 5 cm depth fractions of the soil down to 20 cm. The two methods of incorporating nematicide into soil were equally effective and the slower rotor speed of the Roterra was as effective as the faster rotor speed. We conclude that the VBR technique incorporates non-fumigant nematicide granules or sprays into seedbeds safely, efficiently and with the advantage of no detriment to soil structure.

**Cultivar responses.** At the Arthur Rickwood Experimental Husbandry Farm in 1978 we showed that Maris Piper and Cara potatoes, which are resistant to *Globodera rostochiensis*, responded less to nematode control by oxamyl than the susceptible cultivar Pentland Crown. In 1979 we studied the reactions of these and five other potato cultivars to *G. rostochiensis* in lightly and heavily infested peaty loam soils which had or had not been treated with oxamyl. In the heavily infested soil, yield response to oxamyl varied greatly between cultivars. Record, Desirée and Pentland Dell grew poorly in heavily infested soil and senesced early but grew and yielded well in soil treated with oxamyl. King Edward and Croft grew better in untreated soil but also responded to soil treatment with oxamyl. Because of their tolerance yield increases of Maris Piper and especially Cara in treated soil were less, as in 1978. Also, in pots, the roots of these two cultivars contained only a third or less of the juveniles recovered from the roots of the susceptible cultivars. In the lightly infested soil all varieties grew well but yields of Pentland Dell and especially Croft, were significantly increased by oxamyl, which also controlled the nematode well. Nematode increase was greater on King Edward than on other susceptible cultivars. (Whitehead, Tite, Bromilow, Fraser, French, Mewton and Nichols, with Mr. L. Short, ADAS)

**Stem nematode.** The reactions of different field crops to an 'oat race' of stem nematode, *Ditylenchus dipsaci*, and the effect of aldicarb in the seed furrows on crop growth and yield and on nematode increase were studied in heavily infested soil at Rothamsted. The nematodes invaded field beans, peas, maize, onions and oats (cv. Manod) but did not invade lucerne, sugar beet, wheat or oats (cv. Maris Tabard). Maize and onion were very sensitive to attack. The nematodes increased greatly in field beans and Manod oats. The susceptibility of Manod oats is surprising, as this variety is known for resistance to



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stem nematode. Aldicarb in the seed furrows greatly increased yields of field beans, oats, wheat, maize, onion and lucerne. Yield responses in lucerne, wheat and Maris Tabard oats could not be ascribed to control of stem nematode. Aldicarb reduced nematode increase in Manod oats but not in field beans, which apparently supported more multiplication of the nematode from a population initially reduced by nematicide. Nematode numbers declined in soil under weeds or bare fallow. (Whitehead, Tite, Fraser, French and Mewton)

### Pathogens of nematodes

**Fungal parasites of cyst nematodes.** The presence of such fungi in field soils can be determined only by examination of female nematodes for mycelia or spores, or, in some cases, by inoculating a culture medium with infected eggs. Two methods were used to measure the amount of the fungus *Nematophthora gynophila* present in soil infested with *Heterodera avenae* collected from 35 fields in Berkshire, Buckinghamshire, Oxfordshire and Hampshire. Female nematodes from roots of oats cv. Milford, growing in soil from each site in plastic pots (diam. 9 cm), or in observation plates (Crump & Kerry, *Nematologica* (1977) 23, 398–402) were examined for the presence of a mycelium or spores. Pots were sampled once; females in the observation plates were examined over a number of weeks. Overall 86% of the soils contained *N. gynophila*. In pots an average of 52 females per root system was recovered of which 11% became infected, whereas in the plates, only four females per root system could be seen, but 47% eventually became infected. In soils where nematode densities were low, often too few females could be examined on the roots in observation chambers for the fungus to be detected. There was no correlation in the rates of fungal parasitism in the two containers and there is a need for a quantitative method of estimating fungal spore numbers in soil. (Kerry, Crump and Mullen, with Mr. L. E. W. Stone, ADAS, Reading)

Pure cultures of fungi were obtained from about 100 eggs of *H. avenae* from Butt Close, Woburn, by culturing on corn meal agar containing 30  $\mu\text{g}$  aureomycin  $\text{ml}^{-1}$ . *Verticillium chlamydosporium* was isolated from about 71% of eggs but 15% contained general soil species: *Cylindrocarpon destructans*, *Phoma* sp., *Penicillium* sp., *Chryso-sporium* sp., *Hormodendron* sp. and *Phialophora* sp. Such soil saprophytes may attack only dead or moribund eggs. The remaining fungi (about 15%) failed to produce spores on corn meal or potato dextrose agar and were not identified. (Kerry and Mullen with Salt, Plant Pathology Department)

**Cereal cyst-nematode population changes in the field.** Studies of population changes of *H. avenae* on susceptible crops and fungal parasitism of nematode females and eggs were made at three sites at Butt Close field, Woburn, Beds., Pylon field, Tidworth, Hants. and Watership Down, Sydmonton, Berks. All sites had been monocultured with cereals for at least 4 years and contained precropping *H. avenae* populations of 4 eggs  $\text{g}^{-1}$  soil at Woburn, and 22 and 10 eggs  $\text{g}^{-1}$  soil at Tidworth and Sydmonton respectively. Adult females were sampled at weekly intervals from June until harvest in August and examined for fungal parasites. *N. gynophila* was present at all sites but *V. chlamydosporium* was found particularly at Tidworth and Sydmonton, in large numbers of females and eggs. This fungus previously considered a parasite of nematode eggs, was recovered from virgin females before any eggs had been produced. The conditions which favour infection of the female are unknown. At Woburn despite irrigation, *N. gynophila* was not sufficiently active to prevent nematode numbers increasing: possibly the nematode density was too low for the fungus to spread rapidly. Nematode numbers increased at Woburn ( $\times 4.3$ ) on oats but declined at Tidworth ( $\times 0.3$ ) and Sydmonton ( $\times 0.4$ ) on barley. (Kerry, Crump and Mullen)



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**Micro-organisms associated with cyst nematodes.** Studies on the ultrastructure and biology of the micro-organisms thought to parasitise the potato cyst-nematode, *G. rostochiensis*, and the pea cyst-nematode, *H. goettingiana*, have shown that they have the properties of the members of the order Rickettsiales. The micro-organisms exist intracellularly and the only extracellular forms observed, e.g. in the intestinal lumen of the nematodes, appeared to be degenerating and dying. The cell wall and plasma membrane of the micro-organism are trilaminar and look identical to those of most rickettsias. Their cell wall differs from that of gram-negative bacteria in that it does not possess a 'dense intermediate' or 'middle R layer' between the cell wall and the plasma membrane. The micro-organisms are susceptible to penicillin, indicating that their cell wall contains muramic acid, reported to be present in rickettsial cell walls. Oxytetracycline hydrochloride also inhibits growth and reproduction of the nematode micro-organisms. Strands of electron-dense material, probably corresponding to DNA, are seen in ultra-thin sections of micro-organisms, along with small electron-dense particles recognisable as ribosomes. The micro-organisms appear to reproduce by binary fission, also typical of rickettsias. Rickettsias are classified according to life cycle and pathogenicity to their hosts, most of which are arthropods. Because the forms in cyst nematodes are not transmitted to other organisms by their hosts, they would be placed in the tribe *Wolbachiae* of the Rickettsiales and because they are thought to be pathogenic to their hosts, in the genus *Rickettsiella*. However, *Rickettsiella* has a complex reproductive cycle not observed in the micro-organisms from cyst nematodes, which thus differ from all other members of the order Rickettsiales. (Walsh)

### Feeding in plant-parasitic nematodes

***Ditylenchus dipsaci*.** Cinémicrographic film analyses of the oesophageal feeding pump (*Rothamsted Report for 1978*, Part 1, 184) have been extended. The anchorage mechanism of the radial pump muscles when they open the pump lining seems to involve elastic and semirigid properties of the membrane surrounding the pump, as well as locking together of the expanded muscles themselves. In passive feeding the pump does not operate; food flows down a pressure gradient from the plant cell to the nematode. But at rest the pump lining appears closed and was not seen to open when food began flowing. This, together with measurements of transverse sections, suggests that enough lumen stays open in the 'closed' pump for flow to occur. However, part of the pump (the oesophageal lining just behind the pump chamber) acts as an outlet valve and is seen to open widely as food starts to flow. It has been assumed that all the radial muscle elements in the pump that open the pump lining contract at once but we have seen 'twitches' in the bulb musculature that are apparently all-or-none contractions of a single muscle element or small group of elements. At onset of pumping the pump periphery is pulled in irregularly and the lining may jerk about within the pump. Apparently the vigour of pumping is regulated by number of elements working, starting with only a few when passive inflow has just ended and suction required is minimal. (Seymour and Doncaster)

***Longidorus caespiticola*.** A simple conceptual model of feeding-pump action in *L. caespiticola* based on earlier, mostly qualitative, observations, did not describe pumping adequately. The investigations of Towle and Doncaster (*Nematologica* (1978) **28**, 277-285) have been extended and an elaborated but still largely non-mathematical pictorial model has been developed which incorporates timing and extent of all observed pump behaviour. It accurately depicts known details of pump action and may predict those not yet observed. (Seymour and Doncaster)



## ROTHAMSTED REPORT FOR 1979, PART 1

### Miscellaneous studies

**Damage to sugar beet by *Helicotylenchus vulgaris*.** The spiral nematode *H. vulgaris* was first associated with poor growth of sugar-beet seedlings in 1976 (*Rothamsted Report for 1976*, Part 1, 61–62). Although several cases were recorded that year, the problem has not recurred commonly and trials in subsequent years showed moderate (1977) or negligible damage (1978) (*Rothamsted Reports for 1977*, Part 1, 187–188; and *for 1978*, Part 1, 171–172, respectively). The possibility that the problem had been associated with the unusually warm summer of 1976 was investigated in pots with soil temperatures of 7°, 10° or 15°C. (Soil temperatures at 10 cm approximated 15° in May 1976.) Comparisons during the first 2 months of growth of sugar beet in soil infested with 2000 *H. vulgaris* 1<sup>-1</sup> and in uninfested controls, indicated that the nematodes slowed growth at all temperatures and that the effect was significantly greater at the higher temperatures. This effect, together with unusual stress conditions imposed on the crop that season, may account for the appearance of damage in 1976. (Spaull)

**Egg-shell permeability and hatching of the pig roundworm.** The hatching mechanism of the pig roundworm, *Ascaris suum*, has been found to resemble that of *Globodera rostochiensis* in that changes in permeability of the egg membrane precede hatch, with loss of trehalose from the egg fluid in both cases. The two nematodes are only distantly related but both have eggs in which the juvenile remains quiescent until a specific external stimulus occurs. (Clarke and Perry)

**Behaviour of bovine lung-worm.** Infective third stage juveniles of *Dictyocaulus viviparus* can be dispersed from cattle faeces to clean pasture by the agency of fungi, *Pilobolus* spp. (Robinson, *Nature, London* (1962) **193**, 353–354).

*Dictyocaulus*-infested (but *Pilobolus*-free) calf dung, supplied by Messrs. Glaxo Ltd. (Dictol Unit), was mixed with fresh bullock dung in December and stored in Petri dishes at 15°. Six to 9 days later third stage *D. viviparus* larvae were swarming on the dung surface and climbed about 10% of the fruiting bodies of *Pilobolus* developing on the bullock dung. Many of these nematodes avoided being trapped in drops low down on the fungus by climbing up only one side of the stipe. They could thus reach the base of the sporangium where at least 50 nematodes could accumulate. When the sporangium dehisced, exposing spores and their gelatinous matrix, the nematodes burrowed into the spore mass, sometimes displacing spores. The matrix appears to be hygroscopic, while the sporangium wall is un-wettable, and nematodes enclosed in the sporangium undoubtedly avoid desiccation for longer periods than individuals exposed on the surface. At abscission, when the sporangium is explosively discharged, the nematodes are carried inside it for up to 3 m. Few free-living nematodes of other genera swarming on the dung surface climbed sporangiophores of *Pilobolus*. (Doncaster)

### General cinématography

Two films were produced and accepted by the British Universities Film Council for distribution (publication) and for hire by the Higher Education Film Library: *The Stem Nematode: a Pest in Agriculture* (Doncaster and Hooper) and *Aphid-trapping Potato Plants*. The film of stem nematode shows symptoms of attack on several crops commonly grown in temperate regions, how the nematode can survive desiccation, be distributed on seed, and persist on common weed hosts. With high-power cinémicrography and explanatory animated diagrams the film shows how the nematode feeds in host tissues and how the tissues react to the nematodes' feeding. *Aphid-trapping Potato Plants* illustrates the role of sticky glandular hairs on the foliage of *Solanum berthaultii* × *S. tuberosum* hybrids



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produced by R. W. Gibson (Plant Pathology Department) in trapping insects and mites and thus limiting the spread of virus diseases. (Doncaster with Gibson, Plant Pathology Department)

### Staff and Visitors

J. Bridge joined the Department as the ODA's Nematology Liaison Officer, together with S. Page and Sunniva Jordan. Susan Walker was appointed to a research post funded by ODA and J. A. Walsh to one funded by PMB. Susan Turner was awarded a grant from the Perry Foundation to complete her student training and Amanda Hill continued as a NERC student. A. R. Stone visited the USSR Academy of Sciences Zoological Institute and the All Union Institute for Plant Protection, Leningrad, under the auspices of the Royal Society and the USSR Academy of Sciences. F. G. W. Jones was invited to the Research Institute for Vegetable Crops, Skierniewice, Poland. A. G. Whitehead visited nematological laboratories at Antibes and Rennes under the Anglo-French Collaboration Agreement on Agricultural Research. B. R. Kerry visited nematological laboratories at the University of California, Riverside, and Cornell University and attended, with J. A. Walsh, the 18th Meeting of the Society of Nematologists in Salt Lake City and the IXth International Congress of Plant Protection, in Washington, the latter also attended by F. G. W. Jones. B. R. Kerry and D. H. Crump attended an International Organisation for Biological Control Working Group on Soil Pests in Wageningen. D. J. Hooper gave invited lectures at the European Science Foundation Workshop on Virus Vector Nematodes, in Wageningen. K. Evans returned from a 1 year visit to Cornell University, under auspices of the USDA, ARC and the host university. Department members contributed to the Royal Society soirées in May and June, the Association of Applied Biologists/Federation of British Plant Pathologists meeting on Screening Techniques in Plant Breeding, Edinburgh, in July, the 75th Anniversary Meeting of the Association of Applied Biologists, Reading, in September and the Federation of British Plant Pathologists Meeting on Methods in Plant Pathology, Leeds, in December. *The Stem Nematode: A Pest in Agriculture* was accepted by the Journées Internationales du Film Scientifique 1979, Paris. J. A. Walsh received the Society of Nematologists Best Student Paper Award for 1979.

Dr. N. Vovlas (Bari, Italy), Mr. J. Philis (Cyprus) and Dr. M. Sarwar (India) worked in the department for extended periods.

A meeting on Lighting Techniques in Cinémicro- and macrophotography was held in the department in collaboration with the British Universities Film Council.

### Publications

#### GENERAL PAPERS

- 1 (BABATOLA, J. O.) & BRIDGE, J. (1980) Feeding behaviour and histopathology of *Hirschmanniella oryzae*, *H. imamuri*, and *H. spinicaudata* on rice. *Journal of Nematology* **12**, 48-53.
- 2 CLARKE, A. J. & PERRY, R. N. (1979) The hatching mechanism of *Ascaris suum*. *Parasitology* **79**, xlvii-xlviii.
- 3 HOOPER, D. J. (1979) Nematode vectors of plant viruses—anatomy, taxonomy and biology. *Nordisk Jordbrugsforskning* **60**, 763-765.
- 4 HOOPER, D. J. (1979) Distribution of Longidorid and Trichodorid nematodes in the British Isles. *Nordisk Jordbrugsforskning* **60**, 766.



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- 5 JONES, F. G. W. (1980) Some advances in plant nematology. *Annals of Applied Biology* **94**, 335–347.
- 6 JONES, F. G. W. (1980) The problems of race-specificity in plant resistance breeding. *Proceedings 1979 British Crop Protection Conference, Brighton—Pests and Diseases* **3**, 741–752.
- 7 WHITEHEAD, A. G. (1978) Chemical control of nematodes and other soil-borne pests of field crops. In: *Maximising yields of crops. Proceedings of a Symposium organised jointly by the Agricultural Development and Advisory Service and the Agricultural Research Council, 17–19 January 1978*. London: HMSO, pp. 70–76.
- 8 WHITEHEAD, A. G. (1980) Principles of efficient nematicide use. *Proceedings 1979 British Crop Protection Conference, Brighton—Pests and Diseases* **3**, 709–716.

### RESEARCH PAPERS

- 9 CLARKE, A. J. & PERRY, R. N. (1980) Egg-shell permeability and the hatching of *Ascaris suum*. *Parasitology* **80**, 447–456.
- 10 KERRY, B. R. & CRUMP, D. H. (1980) Two fungi parasitic on females of cyst-nematodes (*Heterodera* spp.). *Transactions of the British Mycological Society* **74**, 119–125.
- 11 (KLAR, A. E.) & FRANCO, J. (1979) Effects of cyst-nematodes on the water relations of two potato varieties. *Turrialba* **29**, 41–44
- 12 MCEWEN, J., COCKBAIN, A. J., FLETCHER, K. E., SALT, G. A., WALL, C., WHITEHEAD, A. G. & YEOMAN, D. P. (1979) The effects of aldicarb, triazophos and benomyl plus zineb on the incidence of pests and pathogens and on the yields and nitrogen uptakes of leafless peas (*Pisum sativum* L.). *Journal of Agricultural Science, Cambridge* **93**, 687–692.
- 13 PAGE, S. L. J., BRIDGE, J., (COX, P. & RAHMAN, L.) (1979) Root and soil plant parasitic nematodes of deep water rice areas in Bangladesh. *International Rice Research Newsletter* **4**, 10–11.
- 14 PERRY, R. N., CLARKE, A. J. & HENNESSY, J. (1980) The influence of osmotic pressure on the hatching of *Heterodera schachtii*. *Revue de Nematologie* **3**, 17–24.
- 15 SPAULL, A. M. (1980) Effect of the cereal cyst nematode, *Heterodera avenae*, on the early growth of oats, wheat and barley. *Plant Pathology* **28**, 138–142.
- 16 STONE, A. R. (1979) Co-evolution of nematodes and plants. *Symbolae Botanicae Upsaliensis* **22**, 46–61.
- 17 STONE, A. R., (FULLER, J. M. & HOWARD, H. W.) (1980) Pathotype Pa2 of *Globodera pallida* present in the United Kingdom. *Plant Pathology* (1979) **28**, 134–137.
- 18 STONE, A. R. & VALENZUELA, A. (1979) Pathotypes of *Globodera rostochiensis* (Woll.) present in the United Kingdom. *Nematologica* **25**, 374–375.
- 19 WALSH, J. A., (LEE, D. L.) & SHEPHERD, A. M. (1979) Intracellular micro-organisms parasitizing cyst-nematodes and their pathogenic effect on the potato cyst-nematode, *Globodera rostochiensis*. *Parasitology* **79** xlvii–xlviii.
- 20 WHITEHEAD, A. G., TITE, D. J., FINCH, P. H., FRASER, J. E. & FRENCH, E. M. (1979) Chemical control of beet cyst-nematode, *Heterodera schachtii*, in some peaty loam soils. *Annals of Applied Biology* **92**, 73–79.
- 21 WHITEHEAD, A. G., BROMILOW, R. H., TITE, D. J., FINCH, P. H., FRASER, J. E. & FRENCH, E. M. (1979) Incorporation of granular nematicides in soil to control pea cyst-nematode, *Heterodera goettingiana*. *Annals of Applied Biology* **92**, 81–91.



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- 22 WHITEHEAD, A. G., FRASER, J. E. & FRENCH, E. M. (1979) Control of potato cyst-nematode, *Globodera pallida*, on tomatoes grown under glass, by applying steam or chemical nematicides to the soil. *Annals of Applied Biology* **92**, 275-278.
- 23 WHITEHEAD, A. G., TITE, D. J., FRASER, J. E. & FRENCH, E. M. (1979) Control of stem nematode, *Ditylenchus dipsaci*, in onions, by granular nematicides applied to the soil. *Annals of Applied Biology* **93**, 213-220.
- 24 WILLIAMS, T. D. & BEANE, J. (1979) Temperature and root exudates on the cereal cyst-nematode *Heterodera avenae*. *Nematologica* **25**, 397-405.
- 25 YEATES, G. W. (1979) Nine new Dorylaimida (Nematoda) from the New Zealand region. *Nematologica* **25**, 419-438.
- 26 YEATES, G. W. (1979) Soil nematodes in terrestrial ecosystems. *Journal of Nematology* **11**, 213-229.
- 27 YEATES, G. W. (1979) Oesophageal length in *Dorylaimellus* (Nematoda: Dorylaimida) poorly correlated with body length. *Nematologica* **25**, 275-278.

## FILMS

- 28 DONCASTER, C. C. & HOOPER, D. J. (1979) *The Stem Nematode: A Pest in Agriculture*. London: British Universities Film Council. HEFL No. 3HE 410.
- 29 DONCASTER, C. C. (1979) *Aphid-trapping Potato Plants*. A research project of Dr. R. W. Gibson. London: British Universities Film Council. HEFL No. HE 411.