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Nematology Department

B. G. Peters

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NEMATOLOGY DEPARTMENT

B. G. PETERS

T. Goodey died suddenly from a heart attack on the 7th July, a few weeks before his projected visit to Australia to advise the government there on nematological research and training. He was the most distinguished nematologist this country has ever had, and his death has been a great blow, not only professionally to nematology and to this department, but also personally to all members of the staff and to his many friends elsewhere. At the time of his death he was engaged in a study of species of *Anguina* from grasses and in preparing (with J. B. Goodey) a new edition of his text-book *Plant Parasitic Nematodes*.

B. G. Peters spent the three months April to June at the Connecticut Agricultural Experiment Station, New Haven, to advise them on nematological problems. He is most grateful to all the staff there with whom he came into contact for their extreme kindness. During this time he was generously given opportunities of visiting Cornell, Beltsville, and the two golden nematode laboratories on Long Island. In July he visited nematological laboratories at Ottawa, Salt Lake City, Berkeley, Modesto, Riverside and Seal Beach.

In July M. T. Franklin paid short visits to Wageningen and Lyngby, on her way to attend the 14th International Congress of Zoology at Copenhagen. Mr. N. Hague (Imperial College, London) and Mr. T. Mabbott (Department of Agriculture for Scotland) spent two weeks in the department acquiring experimental techniques.

MATERIAL AND METHODS

The department continues to receive much eelworm material for identification, most of this work being done by M. T. Franklin and J. B. Goodey. Populations of various species of *Meloidogyne* are building up well in the heated glasshouse beds, this material being used for morphological studies and for comparison with unidentified material sent in. (M. T. Franklin.)

The standard hatching test, for estimating the population of living larvae of *Heterodera rostochiensis*, involves soaking cysts in potato root diffusate for several weeks and counting the larvae which hatch from their egg-shells and emerge from the cysts. Great variability in the numbers of eggs within a cyst entails very high replication (several hundreds) in the cysts used. A new technique is under test wherein eggs are first removed from cysts and then treated with enzymes before being exposed to root diffusate; the object is to attain equal precision with the expenditure of fewer cysts and a much shorter time. (D. W. Fenwick.) Two other techniques, developed elsewhere, are being tested. An

Two other techniques, developed elsewhere, are being tested. An established alternative to hatching *Heterodera* larvae was to grow indicator plants in infested soil, stain the root systems and count larvae within the roots; it now seems that this can be greatly improved by cutting up the roots in a homogenizer. Both hatching and the growing of indicators have been necessitated by the virtual

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impossibility of deciding visually when a given larva, coiled up within its egg-shell, was living or dead. In the past much time has been wasted in seeking *intra vitam* stains which would differentiate these. It now seems likely that the fluorescence colours of larvae stained with acridine orange and examined by ultra-violet light will reliably distinguish living (green) from dead (red) larvae, at least under certain conditions. These two techniques have applications outside the *Heterodera* field. (C. C. Doncaster.) Pre-soaking cysts in weak solutions of several ionizing and non-

Pre-soaking cysts in weak solutions of several ionizing and nonionizing detergents, in order to facilitate the subsequent rapid diffusion of root-diffusate, has not proved satisfactory. (B. G. Peters.)

HOST-PARASITE RELATIONSHIPS

Under this general heading are grouped studies on life-histories, resistance and pathogenicity.

Life-histories

The life history of *Heterodera cruciferae* has been investigated by growing cabbage seedlings in heavily infested soil for two days, transplanting to clean sand and using nutrient solutions, and removing two to four plants daily for fixing and staining, over a period of five weeks. In this way all stages up to mature adults have been produced, each stage dated to within two days. (C. C. Doncaster.)

produced, each stage dated to within two days. (C. C. Doncaster.) By sowing barley in November (1952) in soil infested with *Heterodera major*, and staining roots at intervals, it has been shown that there is a considerable invasion of roots during the winter months, whether the pots of barley were grown in a glasshouse or plunged outside. There had been doubts whether winter invasion by this species occurred. (A. R. Forster.)

The host ranges of several species of *Meloiodogyne* are being investigated. (M. T. Franklin.)

Resistance

Stringent pot tests and three field trials of several oat varieties for resistance to *Ditylenchus dipsaci*, in conjunction with the National Institute of Agricultural Botany, have shown that most varieties are susceptible. Of spring oats only Milford (S.225) is resistant; two or three winter varieties show promise, and work on these is continuing. In co-operation with the N.A.A.S., one field trial of the lucerne race of *D. dipsaci* has shown two lucerne varieties to be susceptible, and two others, along with most other legumes, to be resistant. A second field trial involving the carrot race of *D. dipsaci* is under observation. (J. B. Goodey.)

Not since 1935 has any extensive survey been made of the varietal susceptibility of potatoes to *D. destructor*. This year twenty-four commercial varieties have been tested both in pots (at up to 200,000 eelworms per pot) and in a randomized plot experiment in the Fens. No variety showed any resistance. The mushroom race of this species seems, in this country, to be almost entirely confined to mushrooms. Experiments on the biology of this race are in progress. (J. B. Goodey.)

Heterodera rostochiensis in potatoes may yet be controlled by

breeding into commercial varieties the resistance factor found in Solanum tuberosum andigenum from South America. Tubers of S. t. andigenum have been grown in infested soil to discover what type of resistance is involved. Counts are not yet available, but it is already clear that larvae of *H. rostochiensis* penetrate the roots of this plant but mostly fail to attain maturity. (C. C. Doncaster.)

this plant but mostly fail to attain maturity. (C. C. Doncaster.) Resistance to *Heterodera major* infestation is being investigated in twenty-one varieties of barley and seventeen varieties of spring oats, in co-operation with the National Institute of Agricultural Botany and the N.A.A.S. (J. J. Hesling.)

Pathogenicity

The dynamic equilibrium which exists between a plant host and its worm parasite can be upset, under certain conditions, in the direction of plant disease, with consequent loss of yield. This situation is difficult to investigate. In *Heterodera rostochiensis* it is being approached from opposite ends. First, and more fundamentally, the biological complexity of soil as an ecological habitat could be side-tracked if bacteriologically sterile larvae could be made to invade sterile roots. Tomato roots have been successfully grown by sterile tissue-culture methods, but so far it has not been possible to produce viable sterile larvae. (D. W. Fenwick.) The end result may be disease, revealed by various symptoms. A start has been made on testing symptom-assessment in attacked potatoes with a view to finding sensitive symptoms amenable to scoring and statistical analysis. These symptoms are largely those of drought, and the past season was too wet to yield many helpful results. (B. G. Peters and A. R. Forster.)

The study of several populations of supposed Aphelenchoides parietinus has been started. This species is common in decaying plant material, and the problem is whether, and under what conditions, it can become pathogenic. (M. T. Franklin.)

Two other situations should be mentioned in which the rôle of eelworms in causing manifest disease is doubtful. First, the investigation of *Hoplolaimus uniformis* in Sitka spruce, in collaboration with the Chemistry Department, is being pursued, and secondly, further advice has been given to Mr. A. R. Bull of the West African Institute of Oil-palm Research on the eelworms associated with seedling oil palms suffering from a disease called "Blast", of doubtful aetiology. (J. B. Goodey.)

EELWORM POPULATION CHANGES

Heterodera rostochiensis

By arrangement with the Kirton Experimental Husbandry Farm (N.A.A.S.) annual samples are being taken from an eightcourse and a five-course rotation experiment there. These rotations have been laid down by the N.A.A.S. to reduce the eelworm population, and we are following the course of its fall. At Rothamsted one complete cycle of the three-course rotation experiment has now been completed : this year's counts are not yet available. (C. C. Doncaster.)

Eelworm strains from eight localities have been used in a pot experiment to compare the multiplication rates of the strains when

cysts of differing size were used as inocula. The new cysts produced, also graded by size, and of known age, are available for future use. An experiment to test the effect on multiplication-rate of wintering cysts outside and in the laboratory, prior to using them as inocula for potatoes planted in March and May, showed that the method of wintering had no effect upon multiplication rate, but the Mayplanted potatoes produced more of the largest cysts, and also more tubers, than those planted in March. (J. J. Hesling.)

Heterodera major

By the kindness of Mr. P. R. J. McMorland, annual samples are being taken from his fields, near Winchester, to follow changes in the cereal root eelworm population under various rotations. At Stoke-on-Tern (Salop), with the valued co-operation of Mr. H. C. F. Newton, a micro-plot experiment has been laid down to study the rate of increase of H. major over several years, in the presence of both autumn- and spring-sown oats, wheat, barley and rye. (J. J. Hesling.)

Heterodera cruciferae

In a small plot on which crucifers are grown each year to find the maximum achievable level of population under field conditions, the density of eggs and larvae per g. of air-dried soil has risen from 12 to 64. (C. C. Doncaster.)

ROOT DIFFUSATE PROBLEMS

Heterodera rostochiensis

Reference was made last year to the tripartite scheme for research into the chemical nature of potato (or tomato) root diffusate; this year we have continued to monitor the charcoal extraction carried out at Reading and to bio-assay numerous fractions produced by Cambridge. Results are now available on the breakdown of potato-root diffusate in sand, gravel, peat and clay; breakdown is most rapid in sand and gravel, and least rapid in peat. It is difficult to interpret the results, partly owing to the varying water capacities of these four materials. Partial soil sterilization neutralizes root diffusate for a few weeks, after which breakdown appears to be slower than in unsterilized soil. Moreover, the rate of breakdown is increased following successive applications of diffusate. So far as they go, these results suggest bacterial breakdown, whereas previous *in vitro* tests had shown that bacterial filtration did not materially delay breakdown. Further investigations are proceeding. (D. W. Fenwick and E. Widdowson.)

Root diffusates from sixteen species of Gramineae, including five cereals, collected after two and three weeks of growth, failed to stimulate hatching from *Heterodera rostochiensis* cysts. Stimulation had been observed earlier, in some of these grasses, by both M. J. Triffitt and M. T. Franklin, and accordingly the present experiment was carried out twice, with the same negative result. A preliminary comparison of diffusates from *Solanum andigenum*, potato, and *S. demissum* showed strong stimulation from all three and roughly in that order ("roughly", because no account was taken of differential root size or growth rate). Thus, there is no *a priori*

reason why S. and igenum \times S. tuberosum crosses, bred for resistance to H. rostochiensis, should not also produce active diffusates, which would give them a valuable trap-cropping effect. This would be a biological alternative to the manufacture and application of artificial hatching stimulants. (J. J. Hesling.)

Heterodera cruciferae

A suggested inhibiting effect of white mustard diffusate on the hatching of *Heterodera cruciferae* larvae has not been confirmed. Mixtures of mustard diffusate with those of sprouts, swedes and rapekale gave larger hatches than any of the latter without mustard, though mustard alone was less active than sprouts alone. In the latter test some cysts were found with an external sub-crystalline layer and some with mineralized contents; in a few cases the whole cyst was solid. This occurred mainly in cysts exposed to sprouts diffusate, and not at all in those from soil leachings. The mineralized cysts are being examined spectrographically by H. H. Le Riche (Soil Survey); preliminary results are anomalous in suggesting both calcification and silicification. (C. C. Doncaster.)

CHEMICAL CONTROL METHODS

Pot tests

Annual pot tests of nematicides in glazed pots holding 20 lb. of soil infested with *Heterodera rostochiensis* have continued. Potatoes are subsequently grown in the pots to detect phytotoxic or stimulating effects, and to show how far the eelworm population can recover after a crop.

Analysis of the 1952 results showed that six compounds based on 8-hydroxyquinoline gave disappointing results even at 10 g. per pot. Two which were water-soluble gave no kill, and in the other four, which were formulated with diacetone alcohol, any lethal effect was masked by that of the diacetone alcohol. Two of these four containing iodine had phytotoxic effects, as measured by length of potato haulms and yield of tubers.

A number of soil-fumigant tests can be briefly summarized as follows :

Probit Relationships. Using seven dosages in two-fold steps (1, 2, 4, 8, etc.), probit lines were fitted for D-D mixture and Chlorobromo-propene. The fit was reasonably good up to above 90 per cent kill; at higher dosages there were more survivors than would be expected, suggesting that a few cysts in the soil are protected from the fumigant.

Vertical Diffusion. Using D-D to fumigate soil in wooden boxes made up from 2-inch vertical sections (so that the soil could subsequently be analysed in 2-inch layers) and lined with aluminium foil, it was found that the kill was negligibly small in the top and bottom sections exposed to ventilation. Sealing by sprinkling water on the soil surface or by covering it with a piece of aluminium foil gave only a slight improvement.

Soil Moisture. A dosage of D-D giving a 90 per cent kill in normal soil gave no detectable kill at all in air-dried soil. Additional moisture did not significantly change the kill from 90 per cent.

Field test

The co-operative experiment with Shell Chemicals Ltd. and the West Norfolk Farmers' Manure Co., to test the effects on a potato root eelworm population of single and annually-repeated D-D injections at various stages in a five-course rotation, is now in its third year. In last year's report was recorded the non-significant kill shown by the soil samples taken in spring 1952, following the first injections of autumn 1951. It is of interest to record that samples taken in autumn 1952, immediately before the second injections and twelve months after the first, showed a significant average kill of 41 per cent. The samples of spring 1953 show reasonable kills in plots injected twice, and in those injected in the first year only, but poor kills in plots injected in the second year only. It will be interesting if the latter give an improved result in the samples of autumn 1953. The whole situation suggests the novel idea that a delay of six months in taking soil samples after injection is insufficient to give a true picture of the kill. (B. G. Peters.)