ROTHAMSTED RESEARCH

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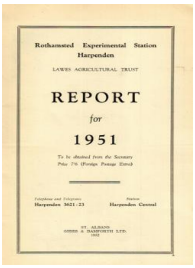
Full Table of Content



Nematology Department

T. Goodey

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NEMATOLOGY DEPARTMENT

By T. Goodey

General

From September 3rd-14th an International Nematology Course and Symposia were held at Rothamsted Experimental Station. They were the first of their kind to be arranged for the subject of Nematology in this country, and were sponsored by the Food and Agriculture Organization of the United Nations. The instructional course, held in the laboratories of the department, September 3rd-10th, was attended by scientists from the following countries: Belgium (1), Denmark (2), Egypt (1), Eire (1), France (1), Haiti (1), Indonesia (1), Israel (1), Italy (1), Netherlands (2), South Africa (1), Sweden (2), Switzerland (2), Uganda (1).

The members of the scientific staff gave lectures and organized practical work on nematological techniques and methods. In this work, too, we had the benefit of the help of Mr. Gerald Thorne from the United States. After the course symposia were held in the Small Public Hall, Harpenden, September 11th-14th, at which most of the members of the staff contributed one or more papers on problems in plant nematology. These meetings were attended by those who had taken the instructional course and by additional scientists from some of the countries already mentioned, as well as by several British specialists. Two days were devoted to the discussion of problems connected with the potato root eelworm, *Heterodera rostochiensis*; one day was spent on a visit to Cambridge and the surrounding country to see eelworm-infested crops; and a final day was devoted to further discussion of problems connected with the stem eelworm, *Ditylenchus dipsaci*; the potato tuber eelworm, *D. destructor*; the leaf and bud eelworms, *Aphelenchoides* spp.; and the root lesion nematodes, *Pratylenchus* spp. In the making and carrying out of the necessary arrangements for the course and the symposia much valuable help was given by Mr. W. C. Moore and his staff of the Ministry of Agriculture and Fisheries Plant Pathology Laboratory, Milton Road, Harpenden. A report on the course together with the papers given at the symposia and the ensuing discussion has been prepared and was issued by F.A.O. in January 1952.

During the year four temporary workers have spent periods of a month or more in the department acquiring nematological technique. They were Miss Daphne Bishop (Imperial College of Science and Technology), Mr. Wahib Doss Hanna (Egypt), Miss F. van der Tuin (Netherlands) and Dr. J. Gallego (Spain). Mr. J. J. Hesling is at present holding an Agricultural Research Council Studentship in the department.

The Ph.D. degree of the University of London was awarded to J. Basil Goodey in July.

During a short stay in Italy Dr. T. Goodey was able to spend a few days with Prof. G. Goidanich and his staff at the Laboratory of Experimental Plant Pathology, Bologna, and to help in the diagnosis of certain eelworm infestations of hemp, wheat and various garden crops.

Research carried out by the members of the staff falls, as in

https://doi.org/10.23637/ERADOC-1-73

previous years, into two main divisions: (1) problems connected with plant infestation by eelworms belonging to the families Tylenchidae and Aphelenchidae and soil nematodes generally (Dr. T. Goodey, Dr. Mary T. Franklin and Dr. J. Basil Goodey); (2) problems connected with root-infesting nematodes belonging to the family Heteroderidae (Dr. B. G. Peters, Mr. D. W. Fenwick and Mr. C. C. Doncaster).

Tylenchidae and Aphelenchidae

Tylenchidae

Dr. T. Goodey continued investigations on the stem eelworm, *Ditylenchus dipsaci*, infesting oats and teasel. A number of spring oat varieties were tested, both by a laboratory method and on an experimental plot, for susceptibility to attack. It was found that the variety S.225, raised by Professor E. T. Jones of Aberystwyth, showed good resistance. This and a few other spring oat varieties which also showed signs of resistance will be tested further. Field observations made at a farm in east Hertfordshire, where susceptible oats commonly fail from stem eelworm attack, and where S.225 was being grown, confirmed the resistance of this variety, as no affected plants could be found.

Mr. M. V. Tracey, of the Biochemistry Department, using infested and uninfested oat seedlings supplied from some of the foregoing tests, found that the enzyme cellulase occurs in greater amount in infested than in healthy seedlings of the same age. He also found cellulase in the nematodes themselves.

Oats of the very susceptible variety S.147 when grown on the teasel eelworm plot were attacked by *D. dipsaci* with the production of typical "tulip root" symptoms. From this and from observations made more than 80 years ago by Kühn in Germany, who found that the stem eelworm from teasel would infest rye, it may be inferred that it is probably one and the same biological race of the parasite which will infest teasel, rye and oats.

Some small-scale tests have been made with the stem eelworm attacking hemp in order to determine the range of crop hosts which this race is capable of infesting. It is hoped by this means to afford some help to Italian plant pathologists who have asked for assistance with this problem.

Dr. J. Basil Goodey reports that work has continued chiefly on the problems connected with the parasitic species of *Ditylenchus*, the stem eelworm, *D. dipsaci*, and the tuber rot eelworm, *D. destructor*. There are various biologic races of *D. dipsaci* found in this country, some of which occur naturally in common weeds of pastures and arable land. Races are found, for instance, on *Plantago lanceolata* and *Hypochoeris radicata* growing on waste land, and experiments have been started this year with these and some other races to try to understand more of the biology of *D. dipsaci* generally and the relationship of these races to those known to cause serious damage to agricultural crops.

Field beans, *Vicia faba*, are commonly attacked by the oat race of stem eelworm when severe damage to the stem base usually results. There is also a so-called giant race of *D. dipsaci* which attacks *V. faba*. This occurs in Portugal and has also been recorded

https://doi.org/10.23637/ERADOC-1-73

in this country. Experiments carried out during the last few years have shown that the infestation of *V. faba* by the giant race is seed-borne to the extent of 30 per cent, while the oat race is not transmitted by seed, although occasionally, on badly attacked plants, pods are affected and eelworms have been found, in one or two instances only, closely associated with the seeds. The red clover race of stem eelworm may also attack beans at the stem base, but observations and experiments indicate that the eelworms cannot maintain themselves on this plant and die out after a time.

The stem eelworm on *Plantago lanceolata* has been found to be seedborne to a small extent. A new weed host of the oat race of *D. dipsaci* has also been discovered.

A species of *Ditylenchus* has been known for a number of years to be the causative agent of a serious disease of mushrooms. The mushroom beds begin to crop quite well and then suddenly fail. Examination shows that the mycelium has been more or less destroyed, and there is often a characteristic smell produced, reminiscent of acetylene. It has recently been shown in Holland that the eelworm responsible for this damage is *D. destructor*. Work on this problem has been carried out in this department during the past year and observations which have been made on numerous samples of compost from diseased mushroom beds have confirmed the identification of the eelworm. Experiments attempting the transfer of the eelworms from compost to potatoes and mint have all failed so far. The work is continuing. In this connection it may be mentioned that M. V. Tracey of the Biochemistry Department has shown that chitinase is present in *D. destructor* infesting mushroom compost.

Studies have continued on various aspects of the host range of *D. destructor*; in particular the influence of the host plant on the parasite. Considerable variation in size, body ratios and growth of gonads occurs; variation which is so striking as to possibly warrant the view that more than one species of *Ditylenchus* may be involved. This view is, however, untenable since a study of the anatomy of the eelworms and evidence from host transfer experiments shows that all the eelworms are *D. destructor*.

Observations have continued on *Hoplolaimus uniformis* and a species of *Trichodorus* associated with the roots of Sitka spruce, *Picea sitchensis*. The numbers of eelworms present actually attacking or in close proximity to the roots are small and variable, and it is too early to be able to say what effect, if any, they have on the young seedlings. This work has been done in co-operation with the Chemistry Department. Further work is contemplated designed to investigate the relationships between these eelworms and Sitka seedlings. Recent work in Florida, U.S.A., has shown that a species of *Trichodorus* is responsible for a stubby-rooted condition of a wide range of vegetable crops. It is interesting, therefore, that *Trichodorus*, which has hitherto been considered a free-living nematode, has been encountered attacking roots of Sitka in this country.

During the year help was sought by the Cameroons Development Corporation in the identification of eelworms associated with a root disease of *Elaeis guinensis*, the oil palm. A species of *Hoplolaimus*, new to science, was discovered which has several remarkable features

https://doi.org/10.23637/ERADOC-1-73

as an eelworm. It is closely related to *H. coronatus*, which is known as a root parasite in the United States. Only comparatively few eelworms were associated with each root system, and they were found attacking the short lateral roots, causing considerable damage to the cortex. It appears unlikely that the eelworms themselves are responsible for all the disease symptoms shown by the seedlings, but more probable that the lesions they cause are the ports through which fungi or other pathogens enter and set up more serious damage.

Aphelenchidae

Dr. Mary T. Franklin reports that observations have continued on black-currant bud disease caused by *Aphelenchoides ribes*. Blackcurrant cuttings treated in November with warm water for killing the eelworms in the buds, and subsequently planted out of doors, failed to root. This was largely because the treatment caused premature expansion of the buds which were then destroyed by frost. Further examination of weeds growing amongst infested bushes have shown many of them to be carriers of the eelworm.

Investigations of the effect on blackcurrant eelworm of spraying infested bushes with Parathion have been made in co-operation with Mr. Brown of the National Agricultural Advisory Service, Cambridge. The results will not be available before next season, but do not appear to be very promising.

The nematode found last year attacking Caucasian scabious, and at first thought to be *Aphelenchoides parietinus*, has now been described as a new species, *Aphelenchoides blastophthorus* (="bud destroyer"). It has been found in scabious from 13 nurseries in various parts of England and one in Scotland. So far it has only been found in *Scabiosa caucasica* var. Clive Greaves. Seedlings of other varieties have been grown and inoculated to find out whether they are susceptible.

As *A. blastophthorus* was at first confused with *A. parietinus*, further attempts have been made to obtain *A. parietinus* from its original habitat in the lichen, *Xanthoria parietina*, at Broadmoor, Berks. The nematode was not described originally in sufficient detail and only the female was found by Bastian in 1865. It is thought that nematodes identified as *A. parietinus* and described sometimes as plant parasites and sometimes as saprophytes may belong to different species and may not all be identifiable with true *parietinus*. It is therefore necessary to have a detailed description of the true *A. parietinus* in order to clarify the situation. So far no males of the species have been found, and it is now thought that, at least in this habitat, the nematode may be parthenogenetic. It is being cultured on agar with a fungus in the hope that males may be produced. During the examination of lichens other interesting nematodes have been found, though they are probably of no practical importance.

HETERODERIDAE

Dr. B. G. Peters reports that the pot experiments to test the effects on *Heterodera rostochiensis* of various edaphic factors (sand, peat, compost and artificials) reached its second year in 1950. Analysis of the data showed that the density of eelworm larvae per gramme of soil (1/g) had decreased, after a 35-fold increase in the

https://doi.org/10.23637/ERADOC-1-73

first season. Positive effects were found for peat, compost and artificials, with no significant interactions. This index of larval density is the product of cyst density (c/g) and the mean number of larvae per cyst (1/c) ; the latter criterion showed no treatment effects in the first season, but in the second there was a positive compost effect with three negative interactions suggesting that peat interferes with the stimulating effects of the other factors.

The indication that the larval density had already reached saturation, and the lack of significance in plant criteria (height of haulms and weight of crop), led to the decision to discontinue the experiment. In its place, a new experiment has been set up in which eelworm infestation has been made a factor (i.e. it is omitted from half the pots) and has been introduced at the much lower density of 0·01 cysts per gramme of soil. This has run through its first season, but counts have not yet been made.

Pot tests of nematicides to control potato root eelworm are carried out each year. Counting of cysts and larvae, and analysis, of the 1950 samples have shown kills above 95 per cent for methallyl chloride at 16 ml. per 20 lb. soil and for the following at either 4 or 16 ml. : D-D mixture, ethylene dibromide, and bis-chloroethyl ether. D-D appears to be slightly more effective when formulated as an emulsion in water (poured over the soil as a drench) than when injected without water. A commercial preparation of phenols was not very effective against this particular eelworm. Phytotoxic effects were marked from ethylene dibromide and slight from bis-chloroethyl ether, but the potato plants appeared to be stimulated by methallyl chloride and D-D at the lower rate (4 ml. per 20 lb.).

During the current season similar tests have been carried out using D-D, bromochloropropylene, iodine, chlorophenols, chloro-isothiocyanates, chloronitrobenzenes, Parathion, and ammoniacal gas liquor. Counts are not yet available.

Analysis of the 1950 data on the vertical migration of *H. rosto-chiensis* larvae through soil (in boxes composed of 2-inch layers) shows that more than 80 per cent of the new cysts produced remain within two inches of the original inoculum, whether that was placed at the top or at the bottom of the box. Migration (up or down) of six inches or more was achieved by about 1 per cent of larvae. Half the boxes were under glass and half outside in the shade. In the latter, multiplication of cysts was twice as high from top inoculation as from bottom ; in the boxes under glass there was a very slight multiplication from top inoculation, but bottom inoculation gave a multiplication almost as great as top inoculation in the shade. This probably indicates that insolation under glass is lethal to larvae in the upper layers of soil, but those in the lower layers benefit from the higher glasshouse temperatures.

The sectional boxes just mentioned have been used during the current season filled with infested soil and each planted with a potato. The boxes have been dismantled, in pairs, at four different times during the season in order to follow changes in larval population at various depths. Counts are not yet available.

Among minor activities may be mentioned the examination of six samples of potato soil from Norway. No potato eelworm was found. Norway is now the only country in north-western Europe

https://doi.org/10.23637/ERADOC-1-73

from which the parasite has not been recorded. In growing a potato in a glass-sided box (to study migration of eelworm from a 1-inch layer of infested soil, sandwiched between uninfested layers), males of *H. rostochiensis* have been seen on two occasions: first in June, 63 days after planting, and secondly in September, 43 days after a second planting in fresh soil. The long time elapsing before parasites were seen is probably due to the cool spring.

Mr. Fenwick co-operated in a joint experiment with the West Norfolk Farmers' Co-operative and Shell Chemicals Ltd., in which D-D mixture was injected annually for three years into field plots infested with *H. rostochiensis*. Potatoes were grown each year. The experiment finished in the autumn of 1950, but the final counts were not available in time for the last report. Briefly, the results show that on a silt soil at Moulton D-D led to an increased yield of tubers sufficient to have paid for the treatment, without any cumulative build-up of the eelworm population. On a black fen soil at Prickwillow D-D led to a significant net increase in the eelworm population with no economic increase in yield. It seems evident that the type of soil largely controls the efficacy of this fumigant.

During the current year the same co-operators have started a new experiment near Wisbech, in which D-D is being injected at various stages in a five-course rotation and is to be repeated on some plots for 2-5 years. This year each of the 30 plots has been sampled, potato yields have been recorded, and the first injections have been made. It is planned to sample the plots for eelworm population changes each autumn, and again for assessment of nematicidal effect each spring.

Mr. D. W. Fenwick reports that experiments on the bio-assay of root diffusate are now completed. Disappointing results from experiments with synthetic chemicals as standards for comparison led to the investigation of purely biological phenomena. Three sets of relationships have been established. If a sample of diffusate be progressively diluted in a logarithmic series the activity falls off in a linear manner so that it is reasonable to assume that the activity of each dilution is proportional to the logarithm of its concentration. If cysts from different stocks are exposed to such a series of dilutions, the linear regression lines obtained when extrapolated to a level of hatch equivalent to that in water all give a similar dilution value indicating the existence of a threshold of concentration below which any diffusate is inactive. If similar dilutions of different diffusates act on cysts from a single stock the curves for each diffusate are parallel to one another, making it possible to express the activity of one in terms of that of any other. The threshold value of dilution furnishes a convenient reference point for any diffusate from which its original value can be calculated if tothe threshold concentration is ascribed a value of one arbitrary unit of concentration. It is proposed to express concentration logarithmically and the term L.A. is proposed; the L.A. of any diffusate being defined as the logarithm of its concentration in arbitrary units. It is an arithmetic measure of its activity in terms of larval emergence. The work is in process of being written up for publication.

The above technique has rendered possible the conduct of experiments on the factors influencing the production of root diffusate by

https://doi.org/10.23637/ERADOC-1-73

plants. An experiment on the relationship of age and diffusate production using Arran Banner potatoes shows that maximum production of diffusate occurs at an age of 2-3 weeks. Preliminary examination of the data indicates that this was the period of maximum growth of the experimental plants and it seems reasonably safe to say that production is positively correlated with the rate of growth of roots. Rate of growth is apparently far more important than root mass. Experiments on the influence of potato variety on diffusate production indicate that the maximum varietal differences are shown very early on in the plant's life ; later the differences become less and less significant. It is considered that these differences in the early stages are a reflection of increased rate of growth in the first few weeks and thus confirm the results of the experiment on root diffusate production and age. Investigations have been carried out on the effect of infestation with *H. rostochiensis* on root diffusate production and although not complete there is reason for believing that the plant response to infestation is the production of an increased quantity of diffusate. This is considered to be further confirmation of the correlation between root diffusate production and growth rate since the normal reaction of plants to infestation is the production of new roots to replace infected ones.

To expedite the setting up of hatching tests, experiments in conjunction with Miss Reid have been carried out on the errors attendant on weighing equal-sized batches of cysts instead of counting. Using a capillary micro-balance the increased error is so small that it can be ignored and the method is now in standard use. It has been written for publication.

A series of experiments, in conjunction with Professor Todd of Cambridge and Professor Stoughton of Reading, sponsored by the Agricultural Research Council, have been commenced to elucidate the activity of the stimulatory factor in tomato root diffusate. Apart from routine testing of samples and fractions, work has been conducted on the effect of different adsorption rates with animal charcoal on residual activity of diffusate and interesting results have been obtained. Experiments are also in progress on the temperature stability of different fractions.

Tests on the break-down of potato root diffusate have been continuing but as yet they are not sufficiently advanced to give definite results.

Studies on population changes during the growth of potatoes have been continued. A trial was carried out in which ten varieties of potatoes were first grown in infested and then transplanted to clean soil. It was found that the maximum invasion occurred in the case of early varieties. The cysts produced had fewer larvae, however, although the reduction in larvae per cyst was not sufficient to offset the increased invasion. A further experiment was carried out to investigate the gross reproduction rate of eelworm on the same ten varieties at three levels of infestation but results of this experiment are not yet available.

Co-operation with Dr. H. C. Gough of the Cambridge National Agricultural Advisory Service on long-term changes in eelworm population during crop rotations on different soils is still continuing.

https://doi.org/10.23637/ERADOC-1-73

Mr. C. C. Doncaster reports that a six-year plot experiment has been started in order to investigate the effect on a long-standing population of *Heterodera rostochiensis* of four different three-course crop rotations, each one including potatoes. The numbers of *Heterodera* cysts are to be estimated in soil samples from each plot once each year and the yields of potatoes will also be recorded.

Investigations on the susceptibility of *Solanum nigrum* to attack by *Heterodera rostochiensis* have shown that though larvae of the eelworm will readily penetrate the roots, they are generally unable to develop. Occasionally, seven weeks or so after invasion, abnormal and degenerating larvae are to be found corresponding to a stage which is normally reached in potato roots about ten days after invasion.

Solanum nigrum suffers more serious root necrosis than do tomatoes or potatoes.

It was found possible to infest *Solanum nigrum* and tomatoes with *Heterodera rostochiensis* in two days and the plants were transferred to clean soil and lifted at subsequent intervals. Roots were collected for histological examinations of infestations of known age.

A comparison has been made between *Solanum nigrum* and tomatoes growing in soil with four levels of *Heterodera* in inoculation ranging from zero to 180 larvae per g. of soil. Plants were lifted after one week, two weeks, four weeks and eight weeks.

Solanum nigrum appeared to show a progressive decline in its rate of growth as the eelworm infestation increased. The effect on tomato was more complex: there appeared to be a stimulating effect at low rates of infestation but at high rates an inhibition of growth occurred even more serious than that suffered by *Solanum nigrum*.

The activity of the root diffusates of the two plants appeared to be generally comparable. In *Solanum nigrum* during the first and second week before growth is noticeably retarded there appears to be a decline in root diffusate production; its activity being about 50 per cent of that of the uninfested controls, but at eight weeks, when growth is seriously checked by eelworm invasion, root diffusate is as active from infested plants as from uninfested ones.

Tomato, in its early stages of growth, produces root diffusate of greater activity as the infestation increases until at 180 larvae per g. of soil there is about a 50 per cent increase in activity. At the later stages, growth and root diffusate activity are both reduced by this infestation, the diffusate being about three-quarters as active as that from the controls.

The overall effect in *Solanum nigrum* is a decline in root diffusate production with increased soil until at 180 larvae per g. infestation the activity falls to about two-thirds of that obtained from control plants.

Cysts have been recovered from soil in which tomatoes and *Solanum nigrum* have been grown and the effect of the plants upon the cyst-contents is being determined.

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