ROTHAMSTED RESEARCH

Report for 1949

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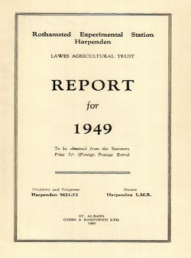

Plant Pathology Department

F. C. Bawden

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PLANT PATHOLOGY DEPARTMENT

By F. C. Bawden

Dr. F. M. L. Sheffield who has held the post of cytologist in the department since 1929 left in October to join the staff of the East African Agricultural and Forestry Organization. Dr. F. M. Roberts was seconded in November to work for a year with the Clove Research Scheme in Zanzibar.

During the spring, the field station for research on sugar beet diseases was transferred from Hackthorn to more convenient and larger premises on an airfield at Dunholme Holt.

Dr. R. Hull attended two conferences, one in Belgium in February and the other in France in July, organized by the International Institute for Sugar Beet Research; Mr. F. C. Bawden attended the Fifth International Congress for Comparative Pathology, at Istanbul in May; Mr. H. L. Nixon attended the meeting of the Electron Microscope Society in Holland in July.

In March Mr. F. C. Bawden visited Zanzibar as a member of the mission sent by the Colonial Office to report on a proposed scheme for the control of sudden death disease of cloves.

Dr. P. H. Gregory was awarded the D.Sc. degree of London University.

Viruses and Virus Diseases

Laboratory and glasshouse work

The performance of the electron microscope was much improved by fitting new lenses and eliminating faults in the electrical system and its resolving power for favourable specimens is now about 2 mμ. It has been mainly used to study viruses but such varied subjects as insect cuticle, fungus spores and clay minerals have also been examined. Using the shadow-cast technique, the sizes and shapes of several viruses and virus strains were determined, some by the direct examination of clarified infective sap and others in purified preparations. Tobacco mosaic, potato X, potato Y, potato paracrinkle, tobacco etch, henbane mosaic, cucumber mosaic and cabbage blackring viruses all have rod-shaped particles of constant widths either about 10 mμ or about 15 mμ but variable lengths. Tomato bushy stunt, tobacco ringspot, a newly discovered virus from broad bean and the various tobacco necrosis viruses studied have spherical particles. Infective preparations of the Rothamsted culture of tobacco necrosis virus nave particles of two sizes, whereas crystalline, largely non-infective preparations contain only the smaller particles; the relationship between these two lots of particles is obscure. No specified particles were identified in the sap from plants infected with potato leaf roll, tomato spotted wilt, cauliflower mosaic, beet yellows and beet mosaic viruses, presumably because these occur too diluted or are similar in size and shape to normal host constituents. No virus particles could be detected in the contents of stomachs of infective aphids. The changes in properties of potato virus X during purification are correlated with the aggrega-

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tion of separate particles into extensive rope and net-like structures (101).

A previously undescribed virus found causing much damage to a crop of broad beans in the midlands showed some unusual properties. Sap from infected plants contains large quantities of a specific antigen not present in healthy beans, giving a precipitin titre of about 1/500, approximately the same dilution end-point at which sap also ceases to cause infections when inoculated to healthy plants. Although highly resistant to heat, sap still being infective after 10 minutes at 90°C., infectivity is lost in a fortnight at room temperature. All plant viruses previously studied are precipitated by one-third saturated ammonium sulphate solution, but the bean virus is not precipitated until the salt concentration reaches three-quarters saturation. It has been isolated in the form of a ribose nucleo-protein, which has not so far been made to crystallise.

A virus that appears to be related to tobacco ringspot, not previously known to occur in the British Isles, was obtained from a mottled syringa (*Philadelphus* sp.).

The loss of precipitating power of antisera to viruses and other antigens that occur during the early stages of heat denaturation is partially reversible. When the heated sera are incubated with trypsin the complexes, which were formed by antibodies uniting with other serum proteins, are disrupted and the antibodies liberated in a form in which they again precipitate their antigens.

With the development of a suitable method for applying statistical tests for significance to counts of local lesions, the effect of dilution on the numbers of lesions produced by viruses could be tested for compatibility with contrasting hypotheses. Experimental results fitted the hypothesis that variable numbers of virus particles are needed to produce a lesion at different sites and were incompatible with the previously held idea that lesions are produced by chance encounter between single virus particles and uniformly susceptible sites (100).

Considerable work was done with potato leaf roll. Infected tubers were freed from the virus when maintained at 37·5°C. in a humid atmosphere for periods of 20 days (99). *Datura stramonium* is more readily infected than potato by aphids; symptoms develop more rapidly and are more easily identified with certainty in the current season. Using *D. stramonium* as a test plant a series of experiments was started to gain information on the factors that influence the transmission of leaf roll virus by *Myzus persicæ*. Other viruses whose transmission by aphids was studied were pea mosaic, pea enation mosaic (96), cauliflower mosaic, cabbage blackring, sugar beet yellows and sugar beet mosaic. With some of those, more infections were caused by infective aphids if their feeding on healthy plants was repeatedly interrupted than if they were allowed to feed undisturbed. In an attempt to gain information on the feeding habits and movement of aphids, experiments have been started using plants containing radio-active phosphorus.

Experiments on the effects of host-plant nutrition on the susceptibility to infection and on the multiplication of certain viruses were concluded (88, 89). Reducing light intensity, increasing humidity and the amount of water supplied to host plants before inoculation increased their susceptibility to infection with

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several different viruses. Whether these effects reflect changes increasing the likelihood of injuries occurring at the time of inoculation, or changes in the constitution of cells that facilitate the establishment of virus particles, is uncertain. Such treatments appear to have little effect on the concentration of virus reached in infected plants. No evidence was obtained to support the claim of French workers that spraying infected plants with growth hormones greatly reduces virus multiplication. Sap from *Datura stramonium* plants infected with potato virus X, and from tobacco plants infected with viruses X and Y, which were severely deformed as a result of spraying with 2:4-dichlorphenoxy acetic acid or with 2-methyl-4-chlorophenoxy acetic acid, gave the same precipitin titres with virus antisera as sap from unsprayed plants; sap from sprayed and unsprayed plants also seemed equally infective and produced similar symptoms when inoculated to healthy plants.

Further work was done in attempts to find the nature of the cause of the yellowing disease that occurs in a Breeders' line of sugar beet, Family 41 (103). Seed from each of 20 single plants produced yellowed plants, but the extent and time of appearance of yellowing varied with different plants. Seed has been saved from the crosses green × green, green × yellowed and yellowed × yellowed, to see what influence the appearance of parent plants has on that of the offspring. Seed was also saved from Kleinwanzleben E plants that had been colonized with aphids from yellowed family 41 plants, to see whether the condition is transmitted through the seed of other lines of beet. Only 3 of 50 Klein E plants colonized developed symptoms, suggesting a much smaller rate of transmission than is usual with beet yellows virus.

Beta patellaris, *B. maritima*, a cross between the two, and two samples of polyploid beet, selected at Cambridge as possibly resistant or tolerant to yellows, were all susceptible and were infected by aphids as readily as commercial beet. There was some evidence that the type of symptoms, particularly whether or not there was etching of the veins or leaf necrosis, depended on the variety.

Evidence was also found for the occurrence of beet mosaic virus in strains of different virulence, one of which seems to cause the "silver-leaf" disease prevalent in some varieties of red beet. The same isolates of mosaic virus, however, may cause different symptoms in different varieties of red beet.

Sap from beet leaves contains material that inhibits the infectivity of beet mosaic, tobacco mosaic and various other viruses. It is destroyed by heating to 80°C., is non-dialysable and is absorbed on to activated charcoal. Its nature has not been determined but it can be precipitated and concentrated by the treatments previously used to isolate a virus-inhibiting glycoprotein from *Phytolacca decandra*.

In previous reports the underground spread of potato virus X from infected to healthy plants has been described and to gain further information on the manner whereby this occurs experiments have been made on the infection of plants through their roots. Roots of tomato seedlings became infected when they were directly inoculated with tomato bushy stunt, tobacco mosaic and potato X viruses, or when these viruses were added to soil or culture solutions in which plants were growing. Sometimes the viruses remained

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localised and multiplied only around their initial entry points, sometimes they invaded the whole root system but did not enter the aerial parts, and sometimes they produced a full systemic invasion of roots and shoots. When the roots of potato plants were inoculated with virus X, tubers set were often infected though the virus rarely spread into the haulm and caused symptoms.

FIELD WORK

There was the earliest and most severe attack of sugar beet yellows in 1949 we have yet experienced; on most plots all the plants were infected soon after singling and this unusually early and high incidence of the disease meant abandoning experiments designed to test the effect of varying plant population, and of introducing scattered sources of infection, on the incidence and spread of yellows. Beet mosaic was much less prevalent, and with this, as with yellows in previous years, reducing the density of the stand increased the percentage of plants that became infected. Contrary to general belief, uneven spacing had no effect on the incidence; the same percentage of plants became infected in plots carrying the same total of plants per unit area whether they were uniformly distributed or spaced irregularly with variable gaps.

Despite the dry summer, the irrigation of sugar beet had no effect on the aphid infestation or on the incidence of yellows and mosaic. Overhead watering did not wash the aphids off the plants and the peak aphid population was similar on irrigated and unirrigated plots. Early in the season more virus-infected plants were counted on the irrigated plots, but later there was no difference, and the effect is probably attributable to a delay in the development of symptoms in the unwatered plants.

The value of raising sugar beet stecklings in districts isolated from sources of yellows virus was again demonstrated. Four lots of stecklings from the north had less than 1 per cent. plants infected; four from Wales had 1, 1, 5 and 6 per cent. respectively, and one from Gloucestershire had 6 per cent., whereas four lots from the eastern counties all had more than 85 per cent. plants infected with yellows; the last also had considerably more plants with mosaic and downy mildew. Sowing under a cover crop of barley also reduced the incidence of yellows; seed sown under barley in April 1948 gave stecklings 11 per cent. of which had yellows compared with 87 per cent. and 100 per cent. for a July and September sowing without cover.

Spraying with some of the new systemic insecticides gave useful results in reducing the prevalence of yellows in the seed bed. Stecklings were sprayed from one to three times in the autumn of 1948; in June 1949 85 per cent. of the unsprayed plants were infected whereas those sprayed three times with E 605 had 10 per cent., with Pestox III 23 per cent. and HETP 41 per cent.; octachlor and nicotine were much less beneficial. With all the insecticides, the first spraying was least beneficial and the last the most. Experiments with insecticides on the root crop in 1949, like those on plant population, gave no useful results as yellows was too prevalent on the plots before the treatments had time to operate.

In an attempt to find lines of beet resistant or tolerant to yellows, more than 700 different lots were grown on a plot surrounded

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by infected seed plants. The lines which had the largest proportion of healthy looking plants and showed least yellowing have been selected for further study.

In potatoes, as in sugar beet, there was an unusually early and extensive spread of viruses. The populations of *Myzus persicæ* on the early spring hosts were higher than for many years and large numbers migrated to potatoes in late May. There were also many winged *Macrosiphum euphorbiæ*, *Aphis rhamni* and *A. fabæ*, but the large numbers of predators and parasites that soon developed prevented the production of a large infestation of apterous aphids. Overhead irrigation greatly increased plant size and yield of tubers, but did not affect aphid populations, which were uniformly small. The aphids present were uniformly distributed per area of land and not per plant, so that plants spaced three feet apart carried twice as many as those spaced 18 in. and four times as many on those spaced 9 in. DDT, E 605 and Pestox III sprayed at weekly intervals controlled aphids but their effects on the spread of viruses will not be known until 1950.

Records on the health of plants grown from tubers saved from experiments in 1948 showed that leaf roll spread as readily to potatoes surrounded by sticky barriers, which prevented the passage of wingless aphids, as to those not so protected, suggesting that spread is largely occasioned by winged forms. Virus Y also infected nearly as many protected as unprotected plants. Experiments on the effects of roguing again showed that the removal of diseased plants in mid-June was too late to prevent most of the spread of viruses, except for potato virus Y in Derbyshire. The experiment on effects of planting date and manurial treatment showed that viruses spread equally in plots planted at different dates, though the later ones had fewer aphids. Plots with phosphate had significantly more aphids, but the only effect of manuring on spread of virus diseases was, as previously found, that most plants became infected on plots that received dung.

An analysis of the results of experiments made in different parts of England and Wales between 1941 and 1947 showed that the spread of leaf roll is correlated with the number of winged *Myzus persicæ* caught on sticky traps throughout the season. Spread of virus Y is also correlated with these trap catches, but to a lesser extent (93).

Mycology

A wind tunnel designed for use in studying various problems in plant pathology such as the dispersal and deposition of fungus spores and protectant dusts, and aphid flight, was completed and put into operation. It is designed to work at wind speeds from 10 to 0·1 metres per second. So far tests have been made mainly on the deposition of Lycopodium spores on sticky cylinders of various diameters from 0·2 to 2·0 cm. At wind speeds from 1 to 10 M/s, the efficiency of the traps increases as the cylinder diameter is reduced and the wind speed is increased. This had been predicted by workers on aerodynamics, but has not previously been studied quantitatively. The large increase in efficiency at low speeds for very narrow cylinders that has been found, however, seems to be unpredicted.

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Tests on the efficiency of spore traps in the open give similar results to those in the wind tunnel; they also show that there is a greater deposit of spores per unit area on vertical than on horizontal surfaces and that the traps are more efficient with the large spores of Lycopodium (30μ diam.) than with the small spores of Lycoperdon (4μ diam.).

A routine spore trap operated continuously in the open showed that there were not the heavy showers of either powdery mildew or *Alternaria* sp. that occurred in 1948.

Studies were continued on the effects of cultural treatments on wheat with and without eyespot (*Cercosporella herpotrichoides*). With plants grown in pots out of doors, eyespot reduced yield by 23 per cent. when ammonium sulphate was given and by 41 per cent. when it was not. Under glass, it reduced yield by 26 per cent. when nitrate was applied and by 90 per cent. when it was not. Applying nitrogen in March and April increased yields of both infected and uninfected wheat more than applications in May. In the field, spring spraying with sulphuric acid increased yields by 4·4 cwt. of grain per acre when applied to plots receiving ammonium sulphate compared with 1·9 cwt. with those that did not.

Depth to which the fungus penetrates leaf sheaths was used to measure the relative susceptibilities of 12 cereals to eyespot. There were slight differences between the seven wheat varieties tested, but all were more susceptible than oats, and rye was less than oats. *Triticale*, a wheat rye cross, was intermediate between the two parents. Whereas infection caused an average loss of 50 per cent. of grain yield in the wheat varieties, rye and oats suffered no loss.

A culture of *C. herpotrichoides* isolated from a crop of oats in Bedfordshire that was severely attacked, was as pathogenic for wheat as cultures derived from wheat, but it was more pathogenic for oats. *Lolium italicum* and *L. perenne* and *Dactilis glomerata* were infected with *Cercosporella herpotrichoides* isolated from wheat; the fungus was re-isolated from each of the grasses and found to be still pathogenic for wheat.

The survival of resting spores of *Plasmodiophora brassicæ* under various crops was again tested, but the previously reported effects of brassicæ in reducing survival were not again obtained. Studies on the relation between spore concentration in the soil, the number of root-hair infections per plant, and the proportion of plants that develop clubs, shows that clubbing can result from a few infected root hairs, and it seems that a club may develop from a single infection.

Some preliminary experiments on factors affecting the production of rhizomorphs by *Armillaria mellea* confirmed the part played by genetical factors but also indicated that the carbon/nitrogen ratio in the medium is important. Rhizomorphs were formed most abundantly with a moderate carbo-hydrate supply and little nitrogen.

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