ROTHAMSTED RESEARCH

Report for 1955

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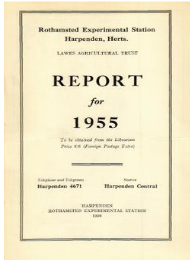
Rothamsted Experimental Station
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REPORT
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1955

Plant Pathology Department

F. C. Bawden

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PLANT PATHOLOGY DEPARTMENT

F. C. BAWDEN

The mycologists moved to the new building during the year, and some of the old laboratories were modified to make them more suitable for work on viruses. T. W. Tinsley returned from his period of secondment to the West African Cocoa Research Institute. D. H. Lapwood was appointed to replace F. T. Last, who transferred to the Department of Insecticides and Fungicides.

B. D. Harrison and J. M. Hirst gained the Ph.D. degree, and G. D. Heathcote the M.Sc. degree, of London University. F. C. Bawden was awarded the Research Medal of the Royal Agricultural Society of England.

L. Broadbent was awarded a Kellog Fellowship and spent three months visiting universities and research stations in the United States and Canada. F. C. Bawden visited West Africa in February to advise the Government of the Western Region of Nigeria on the control measures against, and research into, swollen-shoot disease of cocoa. In December he attended a meeting of a study group in Paris called " to consider how U.N.E.S.C.O. might help to promote knowledge on cell growth with a view to assisting research on cancer." R. Hull attended the Congress of the International Institute of Sugar Beet Research held in Western Germany.

VIRUSES AND VIRUS DISEASES

Strains of tobacco mosaic virus

A virus that causes a mosaic disease of legumes in Nigeria was found to be serologically related to tobacco mosaic virus: so, too, was southern sann-hemp mosaic virus from India. Although many previously described strains cause necrotic local lesions in some varieties of French bean (*Phaseolus vulgaris*), none has infected leguminous plants systemically. The viruses from Nigeria and India are not identical, but both undergo similar changes when they are transferred from leguminous to tobacco plants. When obtained from systemically infected tobacco plants, both are very similar to our type strain of tobacco mosaic virus, but when obtained from systemically infected French beans they differ strikingly from it. Not only do they cause different symptoms in many plants, but they are only remotely related serologically to the type strain, have different electrophoretic mobility and different resistance to inactivation by ultra-violet radiation. One passage through bean or tobacco is enough to produce these changes, and inoculum derived from single local lesions undergoes the same changes as the original bulk inoculum. When virus from bean is inoculated to tobacco plants kept in usual glasshouse conditions with a mean temperature of 20° C., the inoculated leaves develop necrotic local lesions; for about 5 days after infection only the bean form of the virus is recovered from such leaves, but later the tobacco form begins to appear in many. When inoculated plants are kept at 37° C., they produce no symptoms; the bean form of the virus reaches much higher quantities than at 20°, and the tobacco form is not produced. Thus, the change from one form to another depends not only on the species of plant infected but also on the environment in which the plant is growing. Whether the change from one form to another represents a reversible mutation or a phenotypic variation is uncertain. The changes in serological and physical properties induced by passage through different hosts may all arise from changes in the protein moiety of the virus while the nucleic acid remains constant.

Plants infected with all strains of tobacco mosaic contain protein that is serologically related to the virus but is not infective. Bean plants infected with the strains from Nigeria or India seem to contain unusually large quantities of such non-infective protein, for purified preparations of the specific protein from beans are, weight for weight, less than one-hundredth as infective as comparable preparations from tobacco when tested in *Nicotiana glutinosa*. (Bawden.)

The specific protein in plants infected with other strains of tobacco mosaic virus was fractionated and the part that is not sedimented at 40,000 r.p.m. found to contain from 0·1 to 0·2 per cent phosphorus instead of the 0·5 per cent present in infective virus preparations. This phosphorus, like that in preparations of the larger particles, seemed to be present as a nucleic acid. (Bawden and Pirie.)

All the components specific to infected plants have previously been thought to be serologically related to one another. Studying the serological behaviour by the gel-diffusion test, both in plates and in test-tubes with a blank layer of agar intervening between layers containing antiserum and virus preparations, suggests that this assumption is unjustified. Preparations are serologically heterogeneous, and serologically distinct components, all specific to infected plants, have been detected. The protein that usually accompanies tobacco mosaic virus when it is precipitated from sap by acid or salts, or when it is sedimented by high-speed centrifugation, seems not to be one of these antigens; after this contaminant is removed by incubation with citrate or trypsin, virus preparations still give more than one line of precipitation when tested against antisera by the gel-diffusion method.

A crystallized preparation of tomato bushy stunt virus also contained two serologically unrelated antigens that were not detected in healthy plants, but only one of these was sedimented by high-speed centrifugation. (Kleczkowski.)

The relation between carnation latent virus, potato virus S and potato paracrinkle virus

Last year we reported that carnation latent virus was serologically related to potato virus S, which occurs in stocks of many potato viruses without causing any symptoms. The strains of virus S in the varieties Arran Victory and King Edward have been studied in some detail and found to differ in several respects. Although serologically related, the two can be readily distinguished by precipitation tests with their heterologous and homologous antisera, and the one from King Edward infects tomato plants, whereas the one in Arran Victory does not. It has long been known that all stocks of King Edward contain paracrinkle virus, and our results are compatible with the idea that the aberrant strain of virus S universally present in King Edward plants is paracrinkle virus. Many experiments were made to gain evidence that King Edward plants contain two viruses, without success. Sap from King Edward plants was treated in various ways, such as heated to different temperatures and diluted to different extents, and inoculated to tomato plants, which were then tested after three weeks for virus S and paracrinkle virus. All plants whose sap precipitated specifically with antiserum to virus S also caused paracrinkle when scions from them were grafted on to Arran Victory potato plants, and no plant behaved in only one of these two ways.

Plants already infected with one strain of a virus usually resist invasion by a second. The fact that Arran Victory plants, which contain virus S, develop severe symptoms when grafted with scions from King Edward plants seems, therefore, to conflict with the idea that paracrinkle virus is a strain of virus S. However, there are examples in other viruses of strains that are only remotely related serologically failing to protect plants against each other, and this may be another. Evidence in favour of this idea was provided by experiments which showed that virus S from Arran Victory and the serologically related virus from King Edward multiplied in originally virus-free plants of the potato variety Profijt without seeming to interfere with one another.

Paracrinkle virus has been much cited in discussions on the origins of viruses and used as an example of a virus that has arisen endogenously as a component of a higher plant. Our results make the idea that it arose in King Edward quite unnecessary. They suggest that it is simply a specialized strain of a widely occurring virus, other strains of which are aphid-transmitted. None of the strains now found infecting potatoes seems to be aphid-transmitted, but they may have derived from ones that were, such as carnation latent virus now is, and lost this property, together with the ability to infect such plants as carnation, during their protracted existence in plants propagated by tubers. Evidence that another virus can gain and lose the ability to be transmitted by aphids is given below.

We have now produced plants of the King Edward potato variety that seem to be free from paracrinkle virus and from anything that precipitates specifically with antiserum to potato virus S. These seemingly virus-free lines were produced by excising the apical meristems from the shoots of potato tubers, and growing them on nutrient agar. After they had developed enough, they were first grafted to tomato plants to produce sizeable shoots, when these were struck as cuttings. Repeated tests with various antisera and by grafting to Arran Victory potato plants failed to reveal the presence of any virus. (Kassanis.)

Potato virus C and Y

Ten years ago we reported that potato virus C was serologically related to potato virus Y and suggested that the two were related strains, despite the different diseases they cause and the fact that virus C could not be transmitted by aphids. The stock of virus C we then used has since been maintained continuously by inoculation in series to *Nicotiana* sp. in the glasshouses. This year we have transmitted this virus by *Myzus persicae*, using the same techniques that failed previously. That the change in transmissibility represents a change in the virus during its existence in *Nicotiana* sp. is strongly suggested by the fact that new cultures of virus C in tobacco, obtained like the original one from potato plants of the variety Edgecote Purple kindly supplied by the Scottish Society for Plant Breeding, were not transmitted by *M. persicae*. No other change has been noted in the behaviour of virus C, and both the new and old cultures produce only necrotic local lesions when inoculated to Majestic potato plants. Transmission of the aphid-transmitted culture to Majestic and back to tobacco usually resulted in the loss of the ability to be transmitted by *M. persicae*, but a few tobacco plants infected from Majestic still contained aphid-transmitted virus.

The different abilities of virus Y and individual isolates of virus C to be transmitted by aphids seem not to be correlated with any difference in their distributions in the tissues of infected leaves. When leaves infected with any were exposed to ultra-violet radiation, the proportional drop in infectivity of the expressed sap was the same, suggesting that they were all at a higher concentration in the epidermis than in deeper tissues. The ability of *M. persicae*, when fasted and then allowed to feed for 2 minutes on infected leaves, to transmit potato virus Y from tobacco, declined in the ratio of 85 : 60 : 10 when the infected leaves were irradiated for 0, 40 and 80 seconds : the corresponding decrease in infectivity of expressed sap was about 10 : 5 : 2. (Watson.)

Barley yellow dwarf

Although virus diseases of cereals have received little attention in Britain, they may be prevalent and causing unsuspected losses. Certainly many plants in crops at Rothamsted and elsewhere show leaf symptoms in the spring suggesting that they are infected with barley yellow dwarf virus. From such plants on the Rothamsted farm, a virus was obtained that was transmitted by three species of aphids, of which *Rhopalosiphum padi* transmitted the most readily. The transmissions were made by first feeding aphids for 3 days on infected plants and then for 3 days on healthy ones. Tests were complicated by the fact that virus-free aphids cause yellow, partly necrotic lesions, but these appear only at the feeding sites, whereas symptoms of yellow dwarf occur later on the young leaves. Infected oats become characteristically red, which makes it a better experimental host than wheat or barley. In a field experiment with small plots of Plumage Archer barley, plants infected on 25 April yielded only 66 per cent as much grain as uninfected ones, and plants infected on 3 May yielded 84 per cent. After the initial reaction to infection the plants tillered well, and at harvest had the same weight of straw as uninfected ones, despite their lower grain yields. (Watson and Mulligan.)

Viruses of leguminous crops

Work was started on virus diseases of leguminous crops, and these also seem more prevalent than previously thought. Most lucerne plants taken from the Rothamsted farm were infected with a virus whose host range agrees with American descriptions for lucerne (alfalfa) mosaic virus. Its properties *in vitro* also suggest that it is a strain of this virus, and electron microscopy showed it to have spherical particles of approximately 14 mμ diameter. What appears to be a strain of yellow bean mosaic virus was isolated from nursery stocks of Gladiolus, and a similar virus is very prevalent in red clover. A severe disease found affecting a crop of French beans seems to be caused by a virus not previously recorded. (Tinsley.)

Electron microscopy

The electron microscope continued to be used for routine observations of virus preparations, but many weeks' work was lost when the microscope was refitted and housed in new quarters.

After a visit to the Research Laboratories of Associated Electrical Industries Ltd., at Aldermaston, techniques for preparing specimens were changed. The evaporating plant was modified to make it suitable for carbon evaporation, and most specimens are now mounted on carbon films. After comparative tests, platinum replaced palladium as the shadowing metal. Although difficult to evaporate, it gives far superior results, as might be expected from its higher melting point and atomic weight. In collaboration with Mr. D. E. Bradley of A.E.I., attempts were made to demonstrate structure on the surface of tobacco mosaic virus rods, using an experimental high-resolution microscope at Aldermaston. It is clear that for success in work of this kind we must use either stained or shadowed virus mounted on carbon films, and not carbon replicas, which are too thick and give too little contrast.

The new microtome has reduced section cutting to a routine little more difficult than that involved in ordinary light microscopy. Considerable trouble has been experienced from the tissue disintegrating when the embedding methacrylate polymerises, apparently because the volume changes during polymerisation. It is most troublesome with large cells having rigid cell walls, and no doubt happened previously, but with the old microtome was not distinguishable from trouble in the cutting. Possible interaction between this trouble and reagents used for fixation are being studied.

Experiments were made to see if tobacco mosaic virus in tissue could be impregnated with solutions containing heavy atoms, so that its density might be raised enough to observe virus particles easily in sections still containing the embedding plastic. Both silver nitrate and phosphotungstic acid, at pH 2, stain purified virus, but exposure to pH 2 destroyed leaf tissue, even when it was already fixed.

From our and American work, it seems that the low internal hydration of the virus makes it unlikely to be stained to high densities without using treatments that do serious damage. Tomato bushy stunt virus particles offer more promise because they are considerably hydrated and have more space into which heavy atoms might be introduced.

Sections of a tobacco hair cell containing tobacco mosaic virus particles permitted a direct estimate of the number of virus particles in the cell. A typical hair cell, with a volume of about $2{\cdot}5 \times 10^5\ \mu^3$, was shown to contain about 6×10^7 virus particles, or 240 per μ^3. This compares with a figure of 50 particles per μ^3 calculated from infectivity tests on expressed sap.

When the microscope was out of action, much time was devoted to a statistical study of the results of a series of test particle counts. This showed that contaminations were occurring in the Cascade Impactor, which collects the sprayed droplets on filmed grids. Contaminating drops are deposited in stage 3 of the impactor, and probably arise as " satellite " droplets formed on the impact of very much bigger droplets in stage 1. They are thus contaminated with material derived from a surface within the impactor, and the nature of this contamination depends upon what has recently been put through it. Once recognized, the difficulty is readily overcome by coating the inside surfaces of the impactor with a strong solution of calcium chloride; all droplets which have been in contact with the walls of the apparatus are then contaminated with crystals of calcium chloride and so recognized and omitted when counting. Nevertheless, the precision of counts is still less than that predicted theoretically. Our method will work at virus concentrations down to about 0·1 mg./l. for purified tobacco mosaic virus, and could be pushed further if the magnification of our electron microscope could be rapidly varied over a wide range, so that larger droplets might be used for dilute solutions. The method is now being used to study the way virus increases in recently infected leaves. (Nixon and Fisher.)

The control of potato virus diseases

Experiments were continued to test the effects of spraying potato crops with insecticides on the spread of leaf roll and rugose mosaic, caused by virus Y. At Rothamsted DDT emulsion was used on 0, 2, 4, 6 and 8 occasions. Aphids were few in spring, but multiplied rapidly during July, and the spray this year did not check infestations as thoroughly as previously, perhaps because the summer was unusually hot. A count during the second week of August showed 6,000 aphids per 150 leaves on unsprayed plots, compared with 2,500 on plots sprayed on 13 June and 14 July, 1,200 on plots sprayed on 13 June and 5 and 27 July, and 550 on plots sprayed on 13 and 23 June and 5, 14 and 27 July. The yields of tubers were unusually small and were unaffected by spraying.

Twenty-three other trials were made in different parts of Britain to find out for how long potato stocks can be maintained healthy enough to use as seed, when the crops are sprayed and diseased plants are removed.

To gain further information about the best time to apply sprays, plots containing 16 potato plants (12 healthy, 2 with leaf roll and 2 with rugose mosaic) were kept caged except for a single period of 3–4 weeks, and different plots were exposed at different times. The plots were sprayed with an insecticide at the end of their periods of exposure.

An attempt was made at Efford Horticultural Station to see whether the incidence of leaf roll and rugose mosaic in crops could be assessed by lifting tubers in mid-August, breaking their dormancy and immediately planting them again. It failed, for frost killed the haulms of plants growing in the open before symptoms appeared, and although rugose mosaic was usually identifiable in plants grown under glass, leaf roll was not.

Observations on plants grown from tubers saved from experiments made in 1954 showed that all four insecticides (" Systox,"

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parathion, endrin and DDT emulsion) used at Rothamsted stopped leaf roll from spreading, but they only decreased the proportion of plants that contracted rugose mosaic by about one-quarter.

At Clavering, in Essex, where "Hanane" or "Systox" were either sprayed on the foliage or dribbled on to the ridges 3 times, or parathion was sprayed on the foliage 6 times, leaf roll did not spread, whereas its incidence increased 5 times (to 26 per cent) in untreated plots. Rugose mosaic increased in all plots, by 7 times to 35 per cent in unsprayed plots and by 6 times to 28 per cent in sprayed ones, and the trial with this stock has therefore been stopped after continuing for 4 years.

At Harlow, Essex, in the third year of a trial, unsprayed plots had 27 per cent of their plants with leaf roll, plots sprayed with DDT or endrin 5·5 per cent, and plots sprayed and rogued 1·5 per cent. Leaf roll increased 7 times in plots not sprayed in 1954 and by less than one-third in sprayed plots. In all plots the incidence of rugose mosaic was less than 0·5 per cent.

A similar trial at Efford, Hampshire, gave less satisfactory control of leaf roll, perhaps because the spraying was not thorough enough. Leaf roll incidence increased from 1 to 2·5 per cent in sprayed plots and to 5·3 per cent in unsprayed ones.

Another stock at Efford and two in possible seed-growing parts of Gloucestershire, which were sprayed in 1954, as a preliminary to starting combined roguing and spraying tests in 1955, all had less than 0·5 per cent of their plants affected with leaf roll or rugose mosaic.

The samples from the trial at Sutton Bonington, where tubers were planted at various dates and part harvested 12 weeks later and part after the haulm had died, showed that viruses spread much less in 1954 than in 1953. The table shows the percentages of plants with leaf roll and rugose mosaic obtained from tubers obtained from the various plots in 1954.

Time of Harvest	Planting date in 1954					
	12 Apr.	7 May	1 June	26 June	21 July	21 Aug.
Leaf Roll						
After 12 *weeks* ...	0	3·7	4·1	8·7	5·5	0·4
After haulm died ...	18·7	10·2	7·0	7·3	6·7	7·3
Rugose mosaic						
After 12 *weeks* ...	0·4	1·3	5·4	0·7	2·5	0
After haulm died ...	3·0	7·0	7·3	3·0	3·8	0

As before, virus diseases were least prevalent in plots planted very early or late and lifted 12 weeks later. Again, seed tubers harvested after the haulm died gave plants that yielded 10 per cent more tubers than did seed harvested after 12 weeks' growth. Seed from the last two plantings yielded less than that from the others. (Broadbent and Heathcote.)

Viruses of cruciferous crops

The value of sowing strips of barley and oats around cauliflower seed-beds to decrease the incidence of mosaic was again tested at Luddington Experimental Station. Although the cereals grew poorly and were damaged by birds, and aphids were more numerous than in previous trials, there were only 2 per cent infected plants in

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protected beds 6 rows wide and 2·7 per cent in beds 12 rows wide, compared with 5 per cent in unprotected beds.

We again co-operated with the Plant Pathologists of the National Agricultural Advisory Service in their annual survey of the incidence of cauliflower mosaic, by operating 14 aphid traps in different parts of England. *M. persicae* were few until August, when they became numerous, especially in the Midlands, and they remained so during September. *B. brassicae* were very numerous during August in Bedfordshire and during September and October in Cheshire, Yorkshire and Northumberland.

Work was continued on the host range and properties of the turnip viruses. The Edinburgh and Northumberland strains of yellow mosaic virus were inactivated by heating sap for 10 minutes at temperatures between 75° and 80° C., and turnip crinkle virus between 80° and 85° C. Both viruses were transmitted from infected to healthy plants when the leaves were in contact in a wind-tunnel, through which air was flowing at 5 miles per hour.

Turnip crinkle virus has still been found only on one farm in Kincardineshire. Its host range extends beyond the *Cruciferae*; it causes necrotic local lesions in *Chenopodium amaranticolor* and *Gomphrena globosa*, and becomes systemic in *Datura stramonium*, *Reseda odorata* (Mignonette) and *Cleome spinosa*. (Broadbent and Heathcote.)

Dr. Ch. Martini, Institut für Pflanzenkrankheiten, Bonn, spent 7 months here, and studied the transmission of the turnip viruses by flea-beetles. Both viruses could be transmitted immediately by beetles that had fed for only a few minutes on infected leaves, contrary to other workers' results with turnip yellow mosaic virus. In similar conditions, beetles transmitted yellow mosaic virus more often than the crinkle virus. Beetles that fed for longer than a day on infected plants transmitted less often than those which fed for 12 or fewer hours.

Sugar-beet virus diseases

A virus obtained from wild beet, in which it seems to be common, causes discrete yellow or light-green rings in sugar beet; in old leaves the rings coalesce to give a pattern of green rings on a yellow background that is characteristic and from which the virus has provisionally been called "water mottle". It does not protect plants against infection with sugar-beet mosaic virus, but the two are transmitted by aphids in the same manner and in some conditions produce very similar symptoms. A difference is that the water-mottle virus makes the under surface of the main veins necrotic, so that the leaves bend backwards, and the necrotic lesion may spread to the petioles and eventually kill the leaves; it also gives more and better-defined local lesions in tobacco than does mosaic virus, and twice during the winter has become systemic in inoculated tobacco plants. Whether it is a distinct virus or an aberrant strain of beet mosaic virus is undetermined. (Watson.)

When transmitted to inbred lines of sugar beet, various isolates of beet yellows virus consistently produced symptoms of different severity. When titrated against virus antiserum, the sap from plants with the most severe symptoms also gave the highest precipitation end-point. Isolates that were most virulent towards

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sugar beet either failed to infect *Capsella bursa-pastoris* or caused only a mild disease, whereas isolates least virulent towards sugar beet severely stunted it and turned its older leaves purple. Sap from infected *C. bursa-pastoris* did not precipitate specifically with antiserum to yellows virus. The diseases caused by the various isolates in *Chenopodium virgatum* and *Nicotiana clevelandii* in general paralleled in severity those caused in sugar beet, but isolates that caused vein etch and yellowing in beet sometimes produced only one or other of these symptoms in *N. clevelandii*. The precipitation end-points obtained when sap from diseased *N. clevelandii* was titrated against antiserum were highest from plants that showed the most severe symptoms.

Beet plants infected with any of eight isolates that cause various degrees of yellowing and etching were protected against subsequent infection by the most virulent isolates. Not only did the characteristic symptoms caused by the virulent strains alone fail to develop, but the plants infected with the less virulent strains and then colonized 14 days later with aphids carrying virulent ones gave roots the same weight as those infected only with the less virulent strains and heavier than those from plants infected only with the virulent strains. Plants infected simultaneously by aphids carrying strains of different virulence developed symptoms intermediate in severity between those caused by plants infected with either strain alone. Yield of roots was again correlated with severity of leaf symptoms; uninfected plants in pots yielded roots averaging 19·2 g., to be compared with 4·8 g. for those infected with the most virulent strain, 12·8 g. with the less virulent strain and 8·8 g. for plants infected with both strains. In field plants of commercial beet varieties, however, there was no sign of such interference between strains, and plants infected with a virulent and avirulent strain simultaneously yielded no better than plants infected only with the virulent one. In a replicated field trial with virus strains that produce diseases of different grades of severity in glasshouse plants, loss of yield in roots was correlated with severity of leaf symptoms shown under glass. (Blencowe and Hull.)

Small field tests with 272 progenies, obtained by self-pollinating plants selected because they produced large roots although they had yellows, showed large differences in leaf type and in reaction to infection, loss of yield from infection in seedlings ranging from 35 to 66 per cent. Most of the progenies yielded less than commercial varieties, but a few yielded more, both when uninfected or infected with yellows virus, and some did so only when infected.

Larger experiments were possible with inbred progenies selected in 1953, but unfortunately the plants suffered severely from drought. However, some of the lines yielded as much sugar per acre as a commercial variety when uninfected and yielded more, and looked greener, when infected with yellows virus.

A chlorosis that developed in some seedlings raised from the inbred line H2S seems to resemble the "41 yellows" found some years ago in breeding lines in Eire. Seed from one plant produced 8 per cent and from another 20 per cent of yellowed seedlings, and seed from one cluster sometimes produced both healthy and yellow seedlings. The cause, presumably a seed-transmitted virus, was transmitted from affected to healthy seedlings by aphids. Infection

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with it did not protect plants against subsequent infection with yellows virus. The two inbred plants that gave this seed were infected with the yellows virus as seedlings and later selected in the field for propagation because they showed unusually brilliant yellowing. No seed from any other infected plants has given diseased seedlings. (Hull.)

Experiments to test how spraying with insecticides affects the incidence of yellows in the root crop were made at Cambridge, Bury St. Edmunds, Norwich and Boston. Although much of the summer seemed favourable for aphid flight and multiplication, only few *Myzus persicae* were found on the experiments, and large populations of *Aphis fabae* developed on only one.

The materials used did not kill all the aphids on the plants, possibly because of the hot weather when they were applied. A day after the third spraying at Cambridge there was a geometric mean of 90 *A. fabae* per plant on unsprayed plots, compared with 22 on those sprayed with "Systox", and 4 on those sprayed with "Metasystox". Plots sprayed with a commercial formulation of DDT in oil (a satisfactory aphicide for potato crops) had as many aphids as did the unsprayed plots. Three weeks after being sprayed with "Systox" or "Metasystox", plants had the same aphid population as unsprayed ones, and those sprayed with the DDT had higher populations.

At Bury St. Edmunds there was a geometric mean of 10 *A. fabae* per plant on the control plots in late July. Plots sprayed two weeks previously with "Systox" or "Metasystox" had 5, and plots sprayed with DDT had 70. The plots sprayed with the three materials on earlier occasions in June had the same number of aphids as the controls.

Spraying with "Systox" or "Metasystox" at the middle or end of June decreased the incidence of yellows to one-fifth, and spraying on both occasions was no more effective than spraying on either one. A spray in mid-July was effective only at Norwich. A mid-June spray of DDT emulsion decreased the incidence of yellows by two-thirds, but later sprays had no effect. At the end of August fewer than 15 per cent of the plants were infected; expected yield increase from spraying was under 4 per cent and was not detected by lifting samples from the plots.

The effects of two sprayings with "Metasystox" on yellows incidence were determined in 16 experiments arranged in co-operation with the sugar-factory agricultural staffs. Fewer than 5 per cent of plants had yellows at the end of August on the unsprayed plots of 8 experiments, and only 4 experiments had more than 10 per cent of plants with yellows. Spraying decreased the incidence of yellows; it prevented large aphid infestations from developing on most experiments and kept the plants reasonably free from aphids for 7 to 14 days.

At Dunholme, where plots were surrounded by plants artificially infested with viruliferous *Myzus persicae*, "Systox" and "Metasystox" decreased the incidence of yellows by one-third at the beginning of September, and by one-eighth in mid-October; endrin and DDT emulsion had no effect either alone or when applied with systemic insecticides.

An experiment to find the interactions between cultural factors

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and spraying with a systemic insecticide was repeated. Two dates of sowing, two levels of fertilizer dressings and two levels of plant populations were compared. No cultural treatment produced large differences in the incidence of yellows; the largest response to spraying three times with "Systox" (about 1 ton/acre of beet) was from plots with full fertilizer and high plant population, the treatments that yielded best. (Blencowe, Firth, Gates and Hull.)

Beta vulgaris spp. *maritima* grows abundantly on the eastern coast of England from the estuary of the Thames to south of Lowestoft, near Blakeney in N. Norfolk, and around the Ouse estuary near King's Lynn. A survey was started to estimate its importance as a perennial host plant of yellows virus and other pests and diseases of the commercial sugar-beet root crop. Most plants examined were infected with either yellows or beet mosaic, or both, viruses. *Peronospora schachtii* was locally abundant on wild beet, and in some areas the plants were heavily infested with larvae of the mangold fly. In two places yellows and mosaic had spread from wild beet to adjacent fields of commercial beet, but whether wild beet is generally an important virus source for beet crops in the coastal districts is still unknown. The wild beet were not colonized by aphids in the summer.

On low-lying coasts, *B. maritima* is mainly confined to the region of the equinoctial high-tide-mark, but sea flooding in January 1953 extended its distribution. Apparently seed floated inland with the flood waters, and wild beet is now established as a weed of hedgerows and waste ground. Whether it will continue in this unaccustomed habitat remains to be seen. (Blencowe.)

"Conserbeta", a proprietary powder designed to check the sprouting of clamped mangolds and fodder beets and to kill aphids, halved the growth of shoots from roots whose leaves and main growing point had been removed, but inhibited growth from untopped mangolds by only 11 per cent. Topping alone reduced sprout length; shoots from the shoulder buds on topped roots averaged 1·3 inches when the apical shoots from untopped roots were 5·0 inches. (Cornford.)

The samples of commercial sugar-beet stecklings planted at Dunholme averaged 2·4 per cent of plants with yellows; those from eastern England that were sprayed during the autumn with insecticides had 3·4 per cent, those grown under cover crops had 1·4 per cent and those from the north of England 2·1 per cent infected plants. Many of the samples grew poorly, only one-quarter of the plants surviving in some plots. Some of the loss could be attributed to downy mildew (*Peronospora schachtii*), which was commonest on stecklings from open beds in East Anglia, and to the stecklings being very small, but many large stecklings that seemed healthy when planted also failed to grow, despite favourable weather soon after planting.

In the autumn of 1955, 127 sugar-beet steckling beds had on average 0·38 per cent of plants with yellows when inspected by National Agricultural Advisory Service and British Sugar Corporation officers. Discarding 5 beds with over 2 per cent of infected plants decreased the average to 0·21 per cent. Mangold and fodder-beet steckling beds had up to 8·7 per cent of infected plants and the average for 180 beds was 0·63 per cent. (Firth and Hull.)

Plots of stecklings at Dunholme sprayed 5 or 3 times in the autumn of 1954 gave seed crops containing 6 and 10 per cent infected plants when the spray was "Metasystox", 8 and 29 per cent when "Systox", 9 and 20 when parathion, 4 and 13 when endrin and 8 and 15 when DDT.

Seedlings can be made aphicidal when seed is soaked in solutions of Hanane or N.C.7 before it is sown, but the treatment decreases germination. Watering the rows at the time of drilling affects germination less and made seedlings aphicidal for 3 weeks. (Gates.)

Mycology

Potato blight

Experiments started in 1954 to find the importance of seed tubers infected with *Phytophthora infestans* in initiating outbreaks of potato blight were continued in 1955, when more than 400 naturally or artificially infected tubers were planted, either in an unheated glasshouse or in the open. Although 11 per cent of those raised under glass produced stem lesions, showing the spread of the fungus from the tubers, none planted in the open produced any. The need for a suitable environment before such lesions are produced was further emphasized by their occurrence, despite the dry weather, in July on potato plants found growing from discarded tubers in tall grass.

Where blight occurred in the unusually warm and dry summer, it did so late and spread only slowly until late in September, when crops that had not already matured became heavily attacked. In such conditions protective sprays were not expected to increase yield; they had no effect on the yield of Majestic at Rothamsted and decreased the yield of King Edward by about 1 ton/acre, which was not statistically significant.

For five years observations on the occurrence of weather fulfilling Beaumont's criteria for forecasting outbreaks of potato blight have been made in usual Stevenson's screens and in potato crops. In wet summers there is little advantage in making measurements in the crops, but in average or dry years forecasts based on such measurements are more reliable than those made 4 feet above ground. Unfortunately there seems little hope of improving the criteria to be applied in screens 4 feet above ground, because the differences between screen and crop measurements result chiefly from such factors as soil moisture and density of crop that vary with each crop.

Apple scab

Our work with the National Agricultural Advisory Service in the Wisbech area again demonstrated the importance of ascospores as the cause of early infections of apple scab (*Venturia inaequalis*). In the course of this work it has become obvious that the extensive use of organic mercury compounds as curative sprays has created a need for accurate methods of detecting the kind of weather that permits infection. Workers in the United States consider that infection occurs when leaf surfaces remain wet for longer than a minimum time, which varies with the temperature. In 1955 we confirmed that the dew balance could be used to measure the time that apple leaves were wet, and it seems that the design of a simpler balance might allow weather favouring infection to be detected at many places and lead to greater accuracy in the timing of curative sprays.

In addition to the Wisbech area, where Bramley's Seedling is the predominant variety, a trap was operated at Sudbury, Suffolk, in an orchard of Worcester Pearmain and Laxton's Superb. Ascospores occurred three weeks earlier and were over six times as numerous at Wisbech as at Sudbury.

To estimate the number of ascospores produced in different localities by different varieties, and to test the efficacy of methods of preventing their liberation, two small wind tunnels were made to study spore liberation. These allow the total number of ascospores liberated from unit area of different samples of dead leaf to be compared. Preliminary trials with this apparatus in 1955 showed differences between the spore content of leaves from Wisbech and Sudbury orchards similar to those indicated by operating spore traps in the orchards; they also showed that, after spraying with DNOC, dead leaves liberated only 1 per cent of the ascospores liberated from unsprayed leaves. (Hirst and Stedman.)

Pea wilt and foot-rot

Serious outbreaks of pea wilt, caused by *Fusarium oxysporum* f. *pisi*, and of foot-rot, caused by *Fusarium solani* f. *pisi*, were seen in S.E. England; wilt was particularly prevalent in Essex. From N. Suffolk, a bacterium was consistently isolated from diseased peas occasionally together with *F. oxysporum*. When inoculated to the varieties Onward and Charles I in the glasshouse, either organism alone caused only a mild disease, whereas both together caused the lower leaves to wilt and collapse. The bacterium has not yet been identified.

A field experiment at Rothamsted, in which soil was artificially inoculated, showed that *F. solani* spread along pea rows more rapidly than did *F. oxysporum*, and that simultaneous inoculations with both fungi decreased the rate at which both spread. An experiment to test the effects of soil inoculum introduced with peas sown in March and late April on yields of fresh weights gave inconclusive results, although fewer seedlings emerged and the plants were shorter in the inoculated plots. These experiments are being repeated, together with similar ones on an infested field in Essex.

Further work on the variability in *F. oxysporum* f. *pisi* indicated that the formation of heterocaryons (hyphae containing nuclei from genetically different cells) may be a method whereby pathogenicity is altered. Heterocaryons formed between two mutant strains, produced from a virulent wild-type culture by exposure to ultraviolet radiation and each weakly pathogenic, were as pathogenic as the original wild type.

Cultures were derived from single spores formed by heterocaryons between physiologic races 1 and 2 of *F. oxysporum* f. *pisi* and 3 out of 10^8 of these combined properties from both races. These new combinations of properties were detected by studying the nutritional requirements of the cultures and their ability to tolerate growth products of an Actinomycete. Two cultures that combined properties from races 1 and 2 infected a variety of pea that is not attacked by either of these races and that previously has been found susceptible only to race 3. It seems that *F. oxysporum* has a system resembling the "parasexual" cycle that has been described for some other Fungi Imperfecti, and this system, which allows different nuclei occasionally to exchange genetic material, may be one factor responsible for originating races able to infect previously resistant plants.

An attempt to gain information on the mechanism whereby different pea varieties resist infection by some races of *F. oxysporum* was made by studying the effects of root exudates from different varieties on the growth in culture of the three races. Sterile water was continuously circulated over the roots of plants growing under sterile conditions, and the final solution was concentrated *in vacuo* at 40° C. Exudates from roots of varieties that resist infection by a given physiologic race inhibited the germination of spores of this race. Exudates from young roots inhibited more strongly than exudates from old ones. Concentrated exudates contained in porcelain cylinders on agar media also inhibited the mycelial growth of races which the variety of pea resists but not the growth of those which infect it. Sections through inoculated pea roots showed that races incapable of wilting a variety were checked in the outer layers of the cortex and never reached the stele. This also was most obvious at the early stages of the plant's growth.

Extracts from lesions caused by *F. solani* in stem bases suppressed the germination of spores of *F. oxysporum*, whereas extracts from normal stem bases did not. This toxic effect on *F. oxysporum* of stems infected with *F. solani* may explain the phenomenon observed in earlier experiments, that infection with both fungi causes a less severe disease than the wilt caused by *F. oxysporum* alone. (Buxton.)

Effects of ultra-violet radiation on plant pathogens

As with other cells, the damaging effects of ultra-violet radiation on spores of plant pathogenic fungi can partly be offset by visible light. Irradiated suspensions of spores of *Botrytis fabae*, the cause of chocolate spot of broad bean, or of *Uromyces fabae*, the cause of broad bean rust, produced many more infections when the spores were exposed to light before being inoculated to plants, or when the inoculated plants were exposed to light, than when they were kept in the dark. Exposing the inoculated plants to light increased infections by *B. fabae* more than did exposing the irradiated spore suspensions. When spores were irradiated similarly at different times after suspensions were rubbed on leaves, the proportion that infected increased with increasing time between inoculation and irradiation; with a dose so that spores irradiated immediately after inoculation gave 3 per cent of the infections produced by un-irradiated suspensions, those irradiated after 4 hours produced 13 per cent and those irradiated after 8 hours produced 93 per cent. These results suggest that germination increases resistance to in-activation by ultra-violet radiation and that, once the fungus has penetrated the epidermis, it is protected from the radiation.

Irradiating bean leaves increased their susceptibility to infection by *Botrytis fabae*, so that the same inoculum produced more lesions and larger ones. It had no effect on infection by *Uromyces fabae*. (Buxton and Last.)

When resting spores of *Plasmodiophora brassicae*, the cause of clubroot of crucifers, were exposed to various amounts of ultra-violet radiation and then inoculated to cabbage seedlings growing in water culture, two effects were observed: (1) loss of ability to produce zoosporangia in infected root hairs, and (2) loss of ability to infect root hairs. Doses of radiation too small to affect ability to infect root hairs left only 5 per cent of the spores able to develop to the stage of forming zoosporangia. Whether these effects can be reversed by visible light is being studied, but the study is complicated by the fact that visible light itself seems to inactivate resting spores. Exposure to direct sunlight for only short periods prevented spores from infecting, and results were inconsistent when less intense light was used. The component of sunlight that inactivates is unlikely to be in the ultra-violet region, because it passes through glass.

Spores irradiated to the extent that zoosporangia are not formed, though infections occur, are also less effective than normal spores in causing "clubs" to form. Such exposures to ultra-violet radiation decreased the weight of clubs to the same extent as did diluting the inoculum fifty-fold. Diluting to this extent decreases the number of root hairs that become infected to about the number that develop zoospores when infected with undiluted suspensions irradiated for the correct time. This suggests that infections which fail to form zoosporangia in root hairs do not develop further and confirms observations made on roots infected by irradiated spores.

Results of experiments showing that the antibiotic griseofulvin, either sprayed on the leaves of cabbages or applied to the soil in which the plants were growing, checked the development of club root are described in the report of the Insecticides and Fungicides Department. (Macfarlane.)

Cereal diseases

The weather greatly favoured the development of eyespot, *Cercosporella herpotrichoides*, in wheat sown in October, but crops sown late were checked by cold in January, and less severely infected. Take-all, *Ophiobolus graminis*, was more prevalent than usual at Rothamsted, both on autumn- and spring-sown wheat, growing where wheat or barley had been taken recently. Ergot, *Claviceps purpurea*, which very rarely occurred on wheat at Rothamsted 20 years ago, usually occurs now every year. The harvested grain from wheat on Long Hoos averaged 1 sclerotium in 400 g. Black stem rust, *Puccinia graminis*, was fairly severe on spring- but not on autumn-sown wheat.

Broadbalk was drilled unusually late and was less affected by eyespot than usual; the average incidence in 8 plots after crop was 48 per cent infected straws, 32 per cent severely; after fallow 18 per cent were infected, 8 per cent severely. Take-all was commoner than usual in sections following wheat. It was again negligible after fallow and most serious on the three plots that receive no nitrogenous fertilizer; it was less severe than in some nearby fields where wheat followed wheat, suggesting that some condition in Broadbalk is inimical to take-all. The incidence of eyespot or take-all varied from plot to plot in Section V, which was limed in 1954, but an average of all plots was the same in the lightly and heavily limed parts.

The wheat in the four- and six-course rotation experiments was sown in October, and the eyespot incidence was above average. In the four-course, 60 per cent straws were infected, 43 per cent severely, and in the six-course, 44 per cent straws were infected, 21 per cent severely.

The curious white-straw disease, caused by *Gibellina cerealis*, which is not known to occur anywhere else in Britain, was again recorded in wheat on the alternate wheat and fallow.

In the ley-arable experiment the percentage straws with eyespot lesions after arable, cut grass, grazed ley and lucerne were 43, 5, 1 and 7 respectively on Highfield, and 72, 7, 6 and 16 respectively on Fosters. About half of the infected plants had severe lesions, but the crops did not lodge, and yield was affected only on Fosters. Take-all was recorded for the first time in these experiments on 5 of the 8 plots after arable on Highfield, but on average only 4 per cent of the plants were affected. (Glynne, Salt and Slope.)

The effect on lodging in Proctor barley of sowing seed at 1, 2 and 3 bushels/acre was tested at different levels of nitrogen (0, 1½, 3 and 4½ cwt. sulphate of ammonia/acre). There was much less lodging than last year, but there was the same tendency for it to increase with increasing seed rate and nitrogen. Plots that received nitrogen averaged 3, 22 and 32 per cent of their areas lodged at harvest when sown respectively at 1, 2 and 3 bushels; plots receiving 0, 1, 2 and 3 doses of nitrogen had an average of 0, 2, 13 and 42 per cent of their areas lodged. Total grain yields, like last year's, averaged 42·9 cwt./acre for plots receiving nitrogen. Yield was not affected by seed rate; plots receiving one dose of nitrogen yielded 1·8 cwt./acre more than those with no nitrogen, 2 doses gave a slight decrease and 3 doses yielded 2·2 cwt./acre less than 1 dose. The amount of tail corn again increased with increase in seed rate and nitrogen, so that the yield of dressed grain was depressed by each additional bushel of seed; plots receiving nitrogen and sown at 3 bushels, yielded 4·8 cwt./acre less dressed grain than those sown at 1 bushel. Yield of dressed grain was again depressed by each dose of nitrogen, 3 doses yielding 7·0 cwt./acre less than 1 dose, 7·7 cwt./acre less than no nitrogen. (Glynne and Slope.)

In an experiment to measure the effects of previous crops on two varieties of wheat grown under good conditions, Cappelle sown on 15 October, after a crop of potatoes that received farmyard manure, gave 65½ cwt./acre on some plots, the highest yield yet recorded at Rothamsted. Plots receiving 3 and 6 cwt. "Nitro-Chalk", applied half in March, half in May, averaged respectively 51½ and 60 cwt./acre. Holdfast, with the same treatments, gave 41½ and 44 cwt./acre. The large response to nitrogen of Cappelle suggests that its limit of yield may not yet have been reached. When following wheat instead of potatoes, but otherwise treated similarly, Cappelle gave a mean yield for all treatments of 20½, Holdfast of 14 cwt./acre; the proportion of tail corn was greater than where they followed potatoes. The very large differences in yield and quality of grain when wheat followed different crops was associated with a much higher incidence of eyespot, take-all and weeds in the crop following wheat. The weather was exceptionally favourable for eyespot and weeds in 1954 and 1955, and both were more prevalent than is usual at Rothamsted in the second successive wheat crop. Cappelle after

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wheat had 76 per cent straws infected by eyespot, 50 per cent severely, Holdfast had 88 per cent straws infected, 74 per cent severely, supporting evidence from earlier pot experiments that Cappelle has some resistance to this disease. Take-all was more prevalent on the roots of Cappelle than of Holdfast. (Glynne and Slope.)

On light land at Woburn, winter wheat grown for a second year on the same land yielded only 14·6 cwt./acre of grain compared with 27·4 cwt. in the first year, the highest yielding plot of Cappelle giving only 28·8 cwt. compared with 50·2 cwt. in 1954. The lower yields in the second year resulted partly from a higher incidence of eyespot and take-all and partly from much increased weed infestation, *Agrostis gigantea* predominating.

Nitrogen gave large responses in both years; 6 cwt. of " Nitro-Chalk "/acre applied in March, April and May increased yield by 15·4, 22·4 and 13·9 cwt./acre respectively in 1954 and by 13·2, 13·8 and 3·3 cwt. respectively in 1955.

Eyespot affected Holdfast more than Cappelle. Its incidence was greatly increased by nitrogen, especially where applied in March; thus Holdfast given no nitrogen, and dressed in March, April and May had respectively 7, 42, 23 and 12 per cent straws infected at harvest in 1955. The crop on nitrogen-deficient plots was too sparse to favour infection by the eyespot fungus, but conditions became favourable when the luxuriance of the crop was increased by nitrogen.

Take-all was a little more prevalent than last year, 15 per cent of the straws having infected roots. There was more take-all on Cappelle (18 per cent) than on Holdfast (12 per cent), as at Rothamsted. " Nitro-Chalk " applied in April decreased the percentage infection more than where applied in March or May. (Salt.)

Potato skin spot

Attempts to produce skin spot on potato tubers with cultures of *Oospora pustulans* isolated from underground parts of growing potato plants failed, but the failure is not evidence that the isolates differ from those affecting tubers, for these also failed to cause skin spot under the conditions used.

Seedlings of both potato and tomato became infected when grown for a month in soil inoculated with an isolate from a tuber with skin spot, and such seedlings were used in preference to tubers for studying the persistence of the fungus in naturally and artificially infested soils. Soil in which potatoes had not grown for over 100 years gave no infections, but soil from an adjacent field, on which potatoes grew the previous year, gave 8 per cent infected seedlings in February and 2 per cent in March.

The underground parts of potato shoots of several varieties were examined in August at seven places where the National Institute of Agricultural Botany conducts trials of main crop potato varieties. The samples were classified in four categories according to the extent and severity of the attack by *O. pustulans* (Clean, 0; Slight, 1; Moderate, 2; Severe, 3) and the total score expressed as a percentage of the possible. The fungus was present at all places, but was commoner on some varieties and soil types than on others, although the same stocks of seed were planted at all centres. The seed tubers used for the varieties Majestic, Orion and Dr. McIntosh had skin-spot lesions and, averaging all centres, plants of these varieties had disease ratings of 60, 56 and 48 per cent respectively, whereas Ulster Supreme, the seed tubers of which were free from skin spot, averaged only 10 per cent. The average rating of these varieties at different centres ranged from 23 per cent on black fen soil to 68 per cent on clay with flints. (Hirst and Salt.)

Sugar-beet diseases

At 11 out of 12 experimental sites, seed soaked in ethyl mercury phosphate and dusted with BHC gave an average of 10 per cent more seedlings than seed dressed with "Mergamma" (organo-mercury BHC seed dressing dust). In small-scale trials, "Captan" was almost as effective as ethyl mercury phosphate. Pouring solution or suspension of fungicide along rows drilled with seed dressed with ethyl mercury phosphate gave on average 6 per cent more seedlings.

In an experiment on the "Docking disorder", nitrate of soda almost halved the proportion of fangy roots, but the yield of roots was not increased above that given by other sources of nitrogen that scarcely affected fanginess. Drenching the rows with fungicide at time of drilling also halved the proportion of fangy roots. Magnesium nitrate prevented the yellowing of the interveinal areas of the leaves. (Gates.)

When conidia of *Peronospora schachtii* were placed at intervals of a few days on the leaves of sugar-beet seedlings, all plants of inbred line N4S5 remained healthy, whereas between 88 and 98 per cent seedlings of 5 other inbred lines were killed. (Cornford.)