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# **Plant Pathology Department**

## F. C. Bawden

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## DEPARTMENT OF PLANT PATHOLOGY

#### By F. C. BAWDEN

STAFF CHANGES

Dr. J. Henderson Smith retired from the post of Head of the Department on September 30th, 1940, and was succeeded by Mr. F. C. Bawden. The post of Virus Physiologist was then merged into that of Biochemist to the Station and filled by Mr. N. W. Pirie. In October, 1940 Dr. P. H. Gregory and Mr. J. P. Doncaster were appointed as Research Officers of the Agricultural Research Council and attached to the Department to study the factors affecting the spread of potato virus diseases in field crops. Mr. B. Kassanis, who entered the Department in October, 1938 with a research scholarship, was taken on the staff in March, 1941. Dr. A. Kleczkowski has worked in the Department since February, 1940, first with a grant from the Rockefeller Foundation and since 1942 as a Beit Memorial Research Fellow. From 1940 to 1943 Dr. F. M. Roberts worked on Verticillium wilt of tomatoes with a special grant from the Empire Cotton Growing Corporation and in 1943 was appointed to the staff for work on potato virus diseases. Dr. E. M. Crook came to the Station with a research grant from the Agricultural Research Council in 1942, and in 1944 was appointed to the staff of the Department to work on the electron microscope. Miss W. M. Ritchie (Mrs. Dion), who came in October, 1940 to replace Miss B. I. M. Mitchell for work on the insect transmission of virus diseases, later obtained a research fellowship to work on Cercosporella herpotrichoides from 1941 to 1943. Miss J. Smith was appointed in 1941 to assist with the work on the insect transmission of virus diseases. In 1942 Miss J. Moore was given a research grant by the Agricultural Research Council to study the factors responsible for wastage in stored potatoes and in October, 1944 was appointed to the Staff to work with Miss M. D. Glynne.

Others who have worked in the Department for periods of three months or longer are :----

Mr. W. Billet, Mr. I. D. Blair, Mr. S. P. Capoor, Mr. A. Christoff, Mr. J. J. Eshuis, Mrs. L. Kapica, Mr. B. Kvicala, Mr. P. Manil, Mr. T. S. Sadasivan, Mr. C. I. Shen, Miss J. Thurston, Miss E. M. Turner and Mr. A. G. Walker.

In 1939 Miss F. M. L. Sheffield was awarded the D.Sc. degree of London University and Miss M. D. Glynne the D.Sc. degree of the University of Wales in 1943. Ph.D. degrees of London University were obtained by Mr. I. D. Blair, Mr. S. P. Capoor, Mr. J. J. Eshuis, Mr. B. Kassanis, Dr. A. Kleczkowski, Miss B. I. M. Mitchell, Miss J. Moore, Miss W. M. Ritchie, Mr. T. S. Sadasivan, Mr. C. I. Shen, Miss E. M. Turner and Mr. A. G. Walker.

In addition to carrying out the research work summarised below, individual members of the staff have served on committees of the Agricultural Research Council, the Ministry of Agriculture, the Colonial Office and the Medical Research Council, as well as of various scientific societies. They have undertaken special pieces

of research for the Agricultural Research Council, the Ministry of Agriculture and the Ministry of Supply, and have prepared reports or papers for the Agricultural Research Council, the Agricultural Improvement Council, the Ministry of Information and the British Council. In February, 1945 at the invitation of the Portuguese Ministry of Agriculture Mr. F. C. Bawden visited Portugal under the auspices of the British Council, where he gave lectures at Lisbon, Oporto and Coimbra.

A large amount of diagnostic work has been done at the request of Provincial Advisory Officers and others.

#### VIRUSES AND VIRUS DISEASES

Before the war the work of the Department on viruses dealt exclusively with such problems as their relationships with their insect vectors, the cytology of infected plants, the purification of the viruses and the study of their chemical, physical and serological properties. All these lines of work have been continued since 1939 as far as possible, but in addition much attention has been paid to field work, especially with potatoes and sugar beet, in attempts to elucidate the factors affecting the rate of spread of virus diseases in these crops and, if possible, to design control measures. In conjunction with this field work, much of our glasshouse accommodation was also given over to experiments on the virus diseases of these crops.

#### LABORATORY WORK

During the period under review considerable attention has been paid to studying the factors affecting the liberation of viruses from infected plant tissues. In the past all work has been done on the virus contained in the sap, and the fibrous residues have been assumed to be virus-free, as no further virus is obtained by extracting them with water or buffer. These residues, however, may contain more virus than the sap, but it is held in an insoluble form. It can be released by fine grinding or, better, by incubation with certain proteolytic enzymes (34, 35, 36). Work on different kinds of extract from plants suffering from tobacco mosaic has provided further evidence showing how readily tobacco-mosaic virus aggregates. By differential ultra-centrifugation the virus can be separated into fractions differing widely from one another in infectivity and in their physical and serological properties. Preparations of tobacco-mosaic virus have been made that show little or no anisotropy of flow and sediment slowly in the centrifuge ; these are unstable and pass into forms that are anisotropic and sediment rapidly. Electron micrographs show that this change is accompanied by a great increase in the average length of the virus particles.

Electron micrographs have been made of a number of different plant viruses; these have confirmed many earlier deductions about the sizes and shapes of virus particles. Much research, however, has been needed on the machine itself. Because of their small size and power of stopping electrons, plant viruses are difficult subjects for study with the electron microscope and considerable attention has therefore been given to devising methods which give

better resolution (38). In addition to viruses, electron micrographs have been made of clay minerals, lung residues, bacteria and specially prepared sections of biological materials.

Tobacco necrosis has been found to be a disease that is caused by a number of serologically unrelated viruses. Several of these have been isolated in the form of crystalline nucleoproteins. Most crystallise when precipitated slowly with ammonium sulphate in the cold, but one fails to crystallise with this treatment. This virus, however, readily crystallises when ultra-centrifuged or when kept in concentrated salt-free solutions. It has smaller particles than any virus previously studied, and is also unusual in that many treatments destroy infectivity without affecting its serological activity (17, 31, 35).

The inactivating effects of a wide range of different treatments have been studied (29, 30, 33, 52, 58). Some treatments readily destroy the infectivity of all the viruses used while leaving their general physical and serological properties unaffected. Other treatments do this with tomato bushy-stunt and tobacco-necrosis viruses, but not with tobacco-mosaic or potato-virus X, although with all treatments infectivity is the first property to be affected. Inactivation of tobacco-mosaic virus and potato-virus X is usually accompanied by the separation of nucleic acid from the protein, but inactivation of bushy-stunt virus is not.

The serological reactions of anisotropic viruses such as tobaccomosaic virus resemble in every way those of bacterial flagellartype antigens, whereas those of viruses with spherical particles resemble those of somatic antigens. New interpretations of the different behaviour of these two types of antigen have been advanced as a result of the serological studies made on the virus preparations. Viruses and antibodies have been shown to form complexes with other proteins, and it has been possible to change normal precipitating antigens into non-precipitating forms (23, 24, 25, 26, 55, 56, 57, 59, 60, 61). With one and the same antiserum, preparations of tobacco-mosaic virus have been shown to behave like flagellar or somatic antigens, depending on whether or not they show anisotropy of flow and contain elongated particles, Clearly the different behaviours depend on the antigens and not as previously thought, on properties of the antibodies (36).

Antisera were prepared that reacted specifically with the sap of plants suffering from sugar-beet yellows. This is the first time that serological methods have been applied to a virus not transmitted mechanically, and it allowed the properties of the virus to be studied (63). Attempts to apply the method to strawberry viruses failed, possibly because of difficulties of extracting the viruses from infected plants (27).

Cytological studies were made of the inclusion bodies formed by several different viruses. Previously undescribed forms of cytoplasmic inclusions were found in plants infected with various strains of tobacco-mosaic virus (53). Tobacco-etch viruses were found to produce crystalline inclusions in the nuclei of infected cells, the first time this phenomenon has been found in virus diseases of plants though in animals it is common, and the properties of these inclusions were studied (47, 20, 67). Inclusions were found

for the first time in plants infected with potato-virus Y, and the form of those produced by virus X was found to vary with different strains and different hosts (37). In addition to the study of inclusions, cytological investigations were made of the characteristic blotches formed by Arran-Pilot potatoes (69) and of the phloem • necrosis found in leaf-roll potatoes (70).

#### GLASSHOUSE WORK

The relationships between the following viruses and their vectors were studied :---potato-virus Y, cucumber-virus 1, hyo-scyamus-virus 3, tobacco-etch viruses, lettuce-mosaic virus, sugarbeet-mosaic virus, dandelion-mosaic virus and sugar-beet-yellows virus (18, 49, 51, 64, 71, 72, 73, 74, 75). Except for the last two, all behaved similarly; the efficiency of all vectors of the first six was much increased by a preliminary period without feeding, followed by a feeding period on the infected plant of only a few minutes. These viruses do not persist long in the vectors, though aphides prevented from feeding sometimes remain infective for 12 hours. Dandelion-yellow-mosaic virus also does not persist in the aphis, but the efficiency of its vectors is not increased by preliminary fasting, as they need to feed on the infected plants for some hours before they become infective. Sugar-beet-yellows virus behaves quite differently. The efficiency of its vectors increases with increasing time spent on the infected plant. There is no response to a preliminary period without feeding, and the aphides remain infective longer when feeding continuously than when deprived of food : vectors can remain infective for as long as three days after feeding on a diseased plant.

Experiments were also made on the insect transmission of potato-leaf roll, pea mosaic, pea-enation mosaic, soya-bean mosaic and of strains of viruses found attacking brassicae. A new aphis, provisionally named *Myzus ascalonicus* (the Shallot Aphis), was described and its ability to transmit a number of different viruses compared with that of *Myzus persicae*, which the new aphid superficially resembles (40).

Work on the viruses causing necrotic diseases of the potato showed that potato-virus B is a strain of virus X and that virus C is a strain of virus Y (37). Virus C, however, was not transmitted by any of the aphides that transmit virus Y. The variation in symptoms shown by plants in the field infected with virus Y was found to be a result of the occurrence of this virus in a number of strains : in Majestic potatoes the symptoms caused by these vary from a barely perceptible mosaic with no necroses in the first year of infection to a severe form of leaf-drop streak. Different potato varieties were shown to vary in their susceptibility to potato-virus Y, and a glasshouse test using aphides was designed for comparing varietal susceptibility (23): this gave reproducible results, comparable with those obtained in field trials, and may prove useful in breeding for resistance.

A new type of interaction between viruses was described in plants infected simultaneously with tobacco-etch viruses and potatovirus Y. Severe-etch virus protects plants against virus Y, but plants infected with virus Y are susceptible to severe-etch virus. Severe-etch virus apparently suppresses virus Y and replaces it even in tissues where it was well established (20, 21). Further evidence was obtained that the degree of protection afforded to a plant by one strain of a virus against infection with another is proportional to the concentration of the first strain in the inoculated tissues (65).

A number of viruses which are transmitted by insects are not transmitted mechanically, or are transmitted only when special methods of inoculation are employed. Studies of the factors affecting infection by mechanical inoculation suggest three different explanations of this phenomenon; the extracts of some host plants are such that viruses are inactivated (27): the virus content of extracts of plants infected with some viruses is below the dilution end point (48, 51): some viruses may need to enter deep-seated tissues before they cause infection (51, 64). The susceptibility of plants to infection can be varied by altering the physiology of the host (45), and the effects of nutrition of the host on susceptibility are now being studied.

#### FIELD WORK

This has been the main new line of work on virus diseases and has been intensively pursued since 1940. Annual surveys of insects and virus diseases of commercial crops of potatoes and sugar beet have been made, together with many different types of experiment to determine the factors influencing the rate of spread of viruses.

In collaboration with Provincial Advisory Mycologists and others, experiments on the same general plan have been conducted in different parts of the country. Plots of healthy potatoes containing a standard amount of infected plants were laid out, and samples were lifted at fortnightly intervals. In this way much information has been gained on the normal growth of the potato plant (41, 41a) as well as on the extent of spread of potato leaf roll and potato-virus Y in different districts and on the date at which spread occurs. Spread is mainly over small distances, so that a few infected plants within a crop are much more important than badly affected stocks at a distance. This suggested the importance of self-sets as sources of virus diseases : in many districts these have been found to survive in large numbers for some years. The health of such self-sets seems to depend on the health of the crop from which they came, for when growing in crops such as cereals, isolated potato plants do not seem to contract virus diseases at all rapidly.

After the severe winters of 1939-40 and 1940-41, there was little spread of potato-virus diseases, but after the mild winters of 1941-42 and 1942-43, when Myzus persicae overwintered on brassica and other crops, there was a much greater spread. No close correlation was found between the total aphis infestation and the amount of spread of virus diseases. The work has shown the great importance of M. persicae as a vector and the results so far suggest that most of the spread may be caused by the winged migrants rather than by the wingless individuals that make up most of the infestation. Much of the spread in the south and east of England occurs early in the season, which no doubt explains the relative failure in experiments to control the diseases by such measures as roguing, early lifting and fumigation with nicotine.

Much of the field work on sugar beet has been carried out in collaboration with Dr. R. Hull of the Midland Agricultural College. The results of field observations have closely paralleled those with potato crops and again suggest the importance of M. persicae as a vector. In 1940, '41, '42, there was very little virus yellows in the root crop until late in the season, and there were virtually no M. persicae found. This aphis, however, overwintered on stecklings in 1942 and in 1943 was observed in large numbers on the root crops. In this year there was a severe outbreak of virus yellows in the districts where seed crops were grown, but it was slight elsewhere. There was some correlation between the health of root crops and the infestation of M. persicae, but much closer correlation between health and the number of winged M. persicae caught on the sticky aphis traps.

In 1944 there was an early and widespread epidemic of yellows, extending to districts remote from those growing seed. Although there was a severe infestation of Aphis fabae, which may have helped to spread the disease, this was not of primary importance, for extent of the disease was correlated with the incidence of M. *persicae*, though the infestation of this was much smaller than that of A. fabae. In 1945 there was again an infestation of M. *persicae* and where this was heavy, especially in the vicinity of seed crops, there was again a severe attack of yellows (43, 44).

Field trials have shown the importance of early sowing to offset the loss by yellows (72), and methods have been devised to distinguish yellows caused by virus infection from chlorosis produced by other causes (42, 63).

#### MYCOLOGY

During the period under review, most attention was given to plant diseases caused by soil-borne fungi. The majority of rootinfecting fungi are highly specialised parasites, and their life history is divided into two sharply contrasting phases (i) an active parasitic phase on the underground parts of the host plant, during the growing season of a susceptible crop (ii) a non-parasitic phase, as saprophytic mycelium in infected plant residues, or as resting spores or sclerotia, during which the numbers of the plant parasite are progressively reduced. Although most time was given to the study of such specialised root-infecting fungi, as responsible for the most important soil-borne diseases of field crops, work was also done with certain less specialised saprophytic fungi, in which parasitism of crop plants was incidental to their life as regular soil saprophytes.

In continuation of work on the parasitic activity of Ophiobolusgraminis, the loss in yield of wheat due to the take-all disease was shown to be aggravated by deficiencies of N, P or K in a sandculture experiment (85). The survival of this fungus in infected wheat stubble buried in the soil was found to depend chiefly upon supply of nitrogen from the surrounding soil adequate to maintain continued mycelial development in the infected tissues. This supply of nitrogen was reduced, and the life of O. graminis curtailed, by

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incorporation of nitrogen-poor but easily decomposable organic materials with the soil-or by the growing of non-susceptible plants (84, 87). These conclusions are being tested in a field experiment in progress at the Woburn Experimental Station, on light land favourable for the development of take-all. During the war interest in take-all centred upon the likelihood of its development in wheat and barley crops taken after ploughed-up grassland. A test was therefore made of the longevity of O. graminis on the roots of a number of common pasture and weed grasses; timothy (Phleum pratense) was the only species which could be considered a noncarrier, though the rye grasses (Lolium spp.) did far less damage as take-all carriers than did the common bent grasses (Agrostis spp.) (86). During the war, losses from take-all in wheat and barley crops were greatest in 1943: a survey of this disease in the Southern Advisory Province emphasised the danger of weed-grass hosts nullifying the effect of crop rotations otherwise adequate for control of this disease (81). Although in England oats are immune to takeall, and can be used in the rotation for reducing carry-over of O. graminis, in Wales this crop is susceptible. This was shown to be due to the common occurrence in Wales of a distinct variety of the take-all fungus, Ophiobolus graminis Sacc. var. Avenae, E. M. Turner, which has been described and named (104). The oatattacking variety was also found to be widely distributed in Scotland, where the second consecutive crop of oats after grass may suffer severely from take-all (88).

Another fungus causing root and foot rot of cereals is *Fusarium* culmorum. This disease is sometimes seed-borne, and the seedling blight is likely to be serious only when seed is sown without any fungicidal treatment: it was found to be most severe under soil conditions unfavourable to the cereal seedling, such as low soil moisture, acidity, or shortage of plant nutrients, especially nitrogen (103). Similarly, foot rot of older plants is uncommon except in unduly wet or dry soils, or in acid soils. In contrast to 0. graminis, F. culmorum was shown to be a common soil saprophyte, and an important coloniser of healthy wheat stubble when ploughed under the soil (101). Penicillium spp. were found to be of equal importance with F. culmorum as colonisers of the wheat stubble : their relative preponderance on the isolation plates depended upon the method of surface sterilisation employed (105).

Another common soil fungus, *Rhizoctonia solani*, was also shown to exist as a free-living saprophyte in the soil (80). Its growth on Rossi-Cholodny slides was most rapid at a relatively low soilmoisture content, and was favoured by good soil aeration. Although the fungus was found capable of growing indefinitely through untreated fallow soil as a free-living saprophyte, its activity was reduced by addition of certain organic materials to the soil, apparently through the competition of other organisms better fitted to decompose such material.

In contrast to these saprophytic root-infecting fungi is the Myxomycete *Plasmodiophora brassicae*, which seems unable to live as a saprophyte, and survives in the soil in between susceptible cruciferous crops in the form of resting spores. The effect of soil conditions upon infection of cabbage seedlings was studied by the infected-root-hair count method: soil conditions, such as high alkalinity and low moisture, which were well known to be unfavourable to the development of clubroot in pots and in the field, also gave low infected-root-hair counts (102). This technique is now being used in a study of the survival of the resting spores of *P*. *brassicae*. The survival of another type of organ, the multicellular sclerotium, is being studied in the fungus *Helicobasidium pur-pureum*, responsible for violet root rot of sugar beet and other crops.

In 1939 we were asked by the Empire Cotton Growing Corporation to undertake a general inquiry into wilt disease caused by *Verticillium* spp., because of the increase of this trouble in cotton fields in Uganda and elsewhere. The investigation was carried out with tomato plants grown under glass. Development of wilt was favoured by soil of high available – nitrogen content, and by high light intensity and other conditions making for rapid carbon assimilation by the host plant. Evidence was obtained in favour of the hypothesis that an infected plant does not become infectious to its neighbours until it becomes moribund, and the wilt fungus emerges from the vascular cylinder into the cortex of the host (99, 100).

In addition to the investigations on root-infecting fungi, work was carried out on several other fungi which, though not root-infecting, are at least partially soil-borne. In 1943, we co-operated in the survey of wastage in potato clamps carried out by Drs. A. R. Wilson and A. E. W. Boyd, of the Midland Agricultural College. This co-operation was at first confined to the making of fungus isolations from various types of potato-tuber rots, but when a rot caused by Fusarium avenaceum was found to be quite prevalent, though hitherto unreported from this country, comparative experiments on rotting by this fungus and by the dry-rot organism, Fusarium caeruleum, were started. Whereas optimum temperature for rotting by F. caeruleum was found to be 15°C., that for F. avenaceum was  $20^{\circ}-25^{\circ}$ C. The fact that F. avenaceum is less common as a cause of potato wastage than is F. caeruleum was attributed not only to its relative inactivity at temperatures below 15°C., but also to its requirement of a high atmospheric humidity for development of rotting (98).

The fungus Cercosporella herpotrichoides, causing the eyespot disease of cereals, is partially soil-borne, inasmuch as the fungus is spread by spores formed in damp weather on infected stubble, halfburied or left lying on the surface of the soil by faulty ploughing. The disease does not affect the roots, but only the stem, eyespot lesions being formed most commonly just above soil level. As the fungus is dispersed by spores, its spread from a few initial foci of infection in a young crop is more rapid than the subterranean mycelial spread of the take-all fungus, *Ophiobolus graminis*, which passes only slowly from root to root. A crop rotation long enough to control take-all is therefore quite inadequate to control the more rapidly spreading eyespot disease. Evidence on the distribution of eyespot suggests, however, that the spores of the fungus are spread by rain splash rather than by air currents, as dispersal from initial foci of infection seems to occur only within the crop, and does not usually involve

other crops on adjacent fields. In agreement with this is the decline in prevalence of the disease with less frequent occurrence of susceptible cereal crops in the rotation, as shown in extensive surveys of this disease made in 1941 and 1944 (90, 94). Certain varieties of wheat with a shorter straw, such as Wilma and Vilmorin 27, are somewhat resistant to the eyespot disease : outstanding in this respect is the very short-strawed variety Desprez 80. The apparent resistance of these varieties may be due in part to the lower relative humidity of the air near soil level under such short-strawed varieties. Much evidence has been accumulated that any measure which reduces relative humidity of the air around the tiller bases tends to decrease infection by the eyespot fungus. Such measures are late sowing of winter wheat, reduced rate of drilling, wide spacing of rows, shallow drilling, and attention where necessary to soil drainage. The checking of forward growth in the spring by feeding-off with sheep, mowing, or spraying with sulphuric acid, is also sometimes beneficial in checking infection (91-93). Application of nitrogenous fertilisers increases tillering, and so tends to increase the incidence of evespot : the increased incidence of disease is more than offset, however, by the increase in yield of grain from infected as well as from healthy plants (96). A new disease resembling eyespot has been described, and the causal fungus identified as Corticium (Rhizoctonia) solani. This disease, which has been named "sharp eyespot," is not of great economic importance, as it seldom affects more than 1 per cent. of the straw in any crop, and does not seem to increase with the frequency of susceptible crops in the rotation, as does true eyespot (75).

Trials of the effect of certain dyestuffs on spore germination in C. herpotrichoides led to more comprehensive tests of these dyestuffs as inhibitors of spore germination in Fusarium culmorum, which proved a more satisfactory test organism. This work was carried out in collaboration with the Department of Insecticides and Fungicides (82).

#### PUBLICATIONS

#### VIRUS DISEASES

#### BOOKS

- 1. BAWDEN, F. C. 1939. Plant viruses and virus diseases. First edition, pp. 272 XVI, Chronica Botanica Co., Leiden, Holland.
- 2. BAWDEN, F. C. 1943. Plant viruses and virus diseases. Second edition, pp. 294 XIV, Chronica Botanica Co., Waltham, Mass., U.S.A.

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- 3. BAWDEN, F.C. 1939. Some recent work on plant viruses. Emp. J. Exp. Agric., 7, 1-10.
- 4. BAWDEN, F. C. 1940. The sizes of plant viruses. Chron. Bot., 6, 13-14.
- 5. BAWDEN, F. C. 1940. Some properties of plant viruses. Chem. Ind., 788-89.

- 6. BAWDEN, F. C. 1941. Virus diseases of the dahlia. The Dahlia Year Book, 20-26.
- 7. BAWDEN, F. C. 1942. Potato virus diseases. Nature 150, 476-77.
- 8. BAWDEN, F. C. 1942. Crystallography and plant viruses. Nature 149, 321-22.
- 9. BAWDEN, F. C. 1945. The nature of viruses. Sci. Month., 60, 48-50.
- 10. BAWDEN, F. C. 1945. The potato and its ailments. School Sci. Rev., No. 101, 62-75.
- 11. BAWDEN, F. C. 1945. Plant viruses and virus diseases. Nature, 155, 156-60.
- 12. BAWDEN, F. C. and N. W. PIRIE. 1938. Plant viruses I. Serological, chemical and physico-chemical properties. Tabulae Biologicae, 16, 355-71.
- PIRIE, N. W. 1945. Physical and chemical properties of tomato bushy stunt virus and the strains of tobacco mosaic virus. Adv. Enzymology, 5, 1-29.
- 14. SHEFFIELD, F. M. L. 1944. The nucleolus. Nature, 153, 687-88.
- 15. SHEFFIELD, F. M. L. 1944. The electron microscope and its use in biological research. School Sci. Rev., No. 96, 201-9.
- 16. SMITH, J. HENDERSON. 1938. Some recent developments in virus research. Ann. Appl. Biol., 25, 227-43.

#### SCIENTIFIC PAPERS

17. BAWDEN, F. C. 1941. The serological reactions of viruses causing tobacco necrosis. Brit. J. Exp. Path., 22, 59-70.

A method is described for the purification of tobacco necrosis viruses that leads to little or no loss of infectivity. Evidence is presented showing that tobacco necrosis is a disease that can be caused by a number of serologically unrelated viruses. All the viruses used failed to crystall se, and it is shown that the virus culture from which Pirie *et al.* in 1938 obtained crystals was a mixed one. It is suggested that the serologically distinct tobacco necrosis viruses have particles of different sizes, and that they may differ in stability. The behaviour of the tobacco necrosis viruses on heating, ageing or treating with dilute alkali differs from that of potato virus "X," for the former are rendered non-infective without being denatured, and they remain fully active antigenically.

18. BAWDEN, F. C. 1941. Problems in breeding for disease resistance. Chron. Bot., 6, 247-8.

A note on the value of breeding for hypersensitivity.

19. BAWDEN, F. C. 1943. Some properties of the potato viruses. Ann. Appl. Biol., 30, 82-83.

An abstract of a paper given at a symposium on potato virus diseases.

• 20. BAWDEN, F. C. and KASSANIS, B. 1941. Some properties of tobacco etch viruses. Ann. Appl. Biol., 28, 107-18.

The symptoms of severe and mild etch are described; plants suffering from either contain both intranuclear and cytoplasmic inclusions. Fewer and larger crystals are formed in the nuclei of plants with mild etch. Plants infected as seedlings with S.E.V. develop malformed leaves, in which the cytoplasmic inclusions crystallise to give rise to birefringent needles. M.E.V. protects plants against infection with S.E.V.; the two are serologically related, for antisera prepared against either reacted with S.E.V. Precipitation was not obtained with M.E.V., presumably because the virus content of sap is too small.

Although not serologically related to potato virus Y or Hyoscyamus virus 3,

S.E.V. has similar properties in vitro and is transmitted in the same way. The interactions of these three viruses in the plant suggests that they are related. Plants infected with either of the other viruses are not protected against S.E.V., and those infected with P.V.Y. are susceptible to HY.V.3. Plants infected with S.E.V., however, are protected against the other viruses, and those infected with HY.V.3 are protected against P.V.Y. S.E.V. is able to suppress these two viruses when healthy plants are infected with a mixed inoculum, and to supplant them in tissues in which they are already established. Similarly, HY.V.3 can suppress and supplant P.V.Y. Possible interpretations are given for these results. S.E.V. has asymmetrical particles, for concentrated preparations showed anisotropy of flow.

# 21. BAWDEN, F. C. and KASSANIS, B. 1945. The suppression of one plant virus by another. Ann. Appl. Biol., 32, 52-57.

Severe etch virus prevents the multiplication of potato virus Y and Hyoscyamus virus 3 and replaces them even in plants in which they are established. Mild etch virus reduces the concentration of potato virus Y but does not suppress it completely. Cucumber virus 1 multiplies normally in mixed infections with any of the three other insect-transmitted viruses. Possible implications of these results on the mechanism of virus multiplication are discussed; it is suggested that these viruses inactivate in cell sap at approximately the same rate as they denature *in vitro*.

No differences were found between the stability of antibodies to viruses with different properties.

#### 22. BAWDEN, F. C. and KASSANIS, B. 1946. Varietal differences in susceptibility to potato virus Y. Ann. Appl. Biol., 33, 46-50.

In addition to giving different kinds of symptoms when infected with potato virus Y, individual potato varieties also differ in their susceptibility to infection, in the concentration of virus attained in their sap, and in their efficiency as sources of virus for aphides. Their relative susceptibility in the open when exposed to equal chances of infection is correlated with the ease with which they become infected when colonised with infective aphides, and can be assessed from tests made under glass. Methods for making such tests are described; these need few tubers and give reproducible results. It is considered that they could be applied in studying the inheritance of this type of resistance and to test the behaviour of new seedlings. The American variety Katahdin was the most resistant of those tested, but there were significant differences between commerical British varieties.

In the open, all varieties were equally colonised by aphides and resistance to infection with virus Y was not correlated with resistance to leaf roll.

# 23. BAWDEN, F. C. and KLECZKOWSKI, A. 1941. Non-precipitating protein antigens. Nature, 148, 593-94.

When tomato bushy stunt virus or serum globulin is heated with albumin the products cease to precipitate with antiserum to the virus or globulin, though they still fix complement. Such products are still antigenic and when injected into rabbits produce antisera that precipitate the virus or globulin respectively.

# 24. BAWDEN, F. C. and KLECZKOWSKI, A. 1941. Some properties of complexes formed when antigens are heated in the presence of serologically unspecific proteins. Brit. J. Exp. Path., 22, 208-19.

When tomato bushy stunt virus, human serum globulin and albumin are heated at temperatures around  $80^{\circ}$  C. in the absence of serologically unspecific proteins, they are still able to precipitate with their specific antisera. If heated in the presence of serologically unspecific serum albumin, however; they produce complexes behaving like non-precipitating haptens. Human globulin, and albumin behave in this way after being heated for 5 minutes at  $100^{\circ}$  C., but bushy stunt virus does not combine with antibody after heating for 10 minutes at  $95^{\circ}$  C.

The presence of albumin in solutions of tobacco mosaic virus heated at pH 7.0 reduces the rate of inactivation, but this flagellar type antigen still flocculates with its antiserum. This virus loses its ability to combine with antibody after 10 minutes at 90° C.

Heating either virus in the presence of albumin prevents the separation of

precipitates of denatured protein, and that combination between the viruses and albumin occurs is shown by the fact that the viruses can then be removed by precipitation with antiserum to the albumin.

#### 25. BAWDEN, F. C. and KLECZKOWSKI, A. 1942. The antigenicity of nonprecipitating complexes. Brit. J. Exp. Path., 23, 169-78.

Bushy stunt virus and human serum globulin remain antigenic when changed into non-precipitating complexes by heating with rabbit albumin in suitable conditions. The injection of such complexes into rabbits produces antisera apparently identical in all their serological reactions with those produced against the normal virus or globulin. Solutions of the non-precipitating antigens fix complement with antisera as strongly as solutions of the normal precipitating antigens, showing that complement fixation does not depend on the formation of insoluble material. Sufficient heating destroys most of the original antigenicity of bushy stunt virus and globulin, but there is no evidence that heating creates any new specificity.

# 26. BAWDEN, F. C. and KLECSZKOWSKI, A. 1942. The effects of heat on the serological reactions of antisera. Brit. J. Exp. Path. 23, 178-88.

The effect of heat on serological reactions of antisera to O-type antigens (bushy stunt virus and human serum globulin) and to an H-type antigen (tobacco mosaic virus) were studied.

The complement-fixation power of all antisera, and the specific neutralization of infectivity by virus antisera, were equally effected by heat. Tobacco mosaic virus antisera lost all their serological reactions almost simultaneously, but with O-type antigens less heating was needed to destroy precipitability than to destroy other serological reactions.

These results are explained by the formation of complexes between antibodies and other serum proteins during heating. Mixed complexes, formed with albumin, combine with antigens but are unable to fix complement, neutralize infectivity or cause precipitation; they inhibit the precipitation of O-type antigens by other antibodies, but do not interfere with complement fixation. Antisera partly denatured by heat contain mixtures of antibodies in different states; a small proportion of mixed complexes prevents the precipitation of O-type antigens, but has little effect on complement fixation or on the precipitation of H-type antigens.

#### BAWDEN, F. C. and KLECZKOWSKI, A. 1945. Protein precipitation and virus inactivation by extracts of strawberry plants. J. Pom. Hort. Sci., 21, 2-7.

Specific antisera could not be produced against extracts of virus-infected strawberry plants, possibly because of properties of the host plant. No soluble protein could be extracted from fruit, leaves, runners and roots; and aqueous extracts from all organs reduced the infectivity of tobacco mosaic virus. Except those from the fruit, all extracts contained much tannin and they precipitated serum proteins. The possible effect of this on failure to transmit strawberry viruses by mechanical inoculation is discussed.

#### 28. BAWDEN, F. C. and PIRIE, N. W. 1939. The purification of insecttransmitted plant viruses. Brit. J. Exp. Path., 20, 322-29.

Liquid crystalline preparations of nucleoproteins have been made from the sap of tobacco plants infected with the two insect-transmitted viruses, potato virus "Y" and Hyoscyamus virus 3. The yields are from 0.5 to 3.0 mg. per litre of infective sap. These proteins give specific precipitates, of the flagellar type, with antisera when diluted to  $1\cdot10^6$ . They are infective, but the methods used in the purification processes seem to destroy the infectivity of much of the virus without destroying its serological activity. The chemical and physical properties of preparations of these viruses closely resemble those of potato virus "X." Some properties of a lipoid-containing contaminant are described.

29. BAWDEN, F. C. and PIRIE, N. W. 1940. The inactivation of some plant viruses by urea. Biochem. J., 34, 1258-77.

The literature on the effects of urea on proteins, tissues, bacteria and viruses is reviewed. The four viruses, tobacco mosaic, potato "X," tomato bushy stunt and tobacco necrosis, are irreversibly denatured by urea. The denaturation is accompanied by loss of infectivity and serological activity. For each

virus there is a critical concentration of urea below which there is no irreversible effect on infectivity. This concentration is smallest for potato virus "X" and greatest for tomato bushy stunt virus. The rate of inactivation is greatly increased by the presence of alkali. The rate of inactivation is minimum at about 20°C., and is much increased by cooling to 10°. The inactivation of purified tobacco mosaic virus by urea proceeds only slightly more slowly than that of virus in crude infective sap. The inactivation of the nucleic acid and protein, but the inactivation of bushy stunt and tobacco necrosis viruses is not. Changes in the absorption spectra that accompany inactivation are described.

30. BAWDEN, F. C. and PIRIE, N. W. 1940. The effects of alkali and some simple organic substances on three plant viruses. Biochem. J., 34, 1278-92.

The effects of alkali and of 15 simple organic substances on tobacco mosaic virus and tomato bushy stunt virus are described. Some experiments with potato virus "X" are also included. Bushy stunt virus is the most resistant to denaturation and potato virus "X" the least. The effects of alkali on tobacco mosaic virus are complex; gentle treatment may increase infectivity, slightly more severe treatment causes loss of infectivity but not loss of serological activity, and more severe treatment causes loss of all characteristic properties. With bushy stunt virus inactivation without loss of serological activity occurs over a wider pH range, and crystalline non-infective preparations can be made from alkali-treated material. Apparently similar crystalline and non-infective preparations can be isolated from expressed sap allowed to age for some months. In the presence of alkali, sodium dodecyl sulphate readily destroys all the viruses, separating the nucleic acid from the proteins. With the exception of nicotine and arginine, which form with tobacco mosaic reversible, fibrous precipitates, all the substances we have tested at concentrations below 4M inactivate the viruses in neutral solution. Dilute solutions of these agents are often precipitants whereas concentrated ones dissolve the products of denaturation. Inactivation of tobacco mosaic virus and potato virus "X" is usually accompanied by the separation of the nucleic acid from the protein, but inactivation of bushy stunt virus is not.

31. BAWDEN, F. C. and PIRIE, N. W. 1942. A preliminary description of preparations of some of the viruses causing tobacco necrosis. Brit. J. Exp. Path., 23, 314-28.

The purification of six separate cultures of viruses causing tobacco necrosis is described; these cultures have been labelled Potato, Princeton, Tobacco VI, Rothamsted, Tobacco I and Tobacco II. The last two are probably identical, but the remainder differ in their properties, although they produce identical symptoms in tobacco and bean. The first three share antigens but are serologically unrelated to the others, suggesting that the disease can be caused by different viruses, each of which may occur in a number of strains. On precipitation with ammonium sulphate the products from four cultures have behaved systematically: Rothamsted gives an amorphous precipitate; Princeton shows anisotropy of flow but gives no recognisable crystals; potato crystallises as thin lozenge-shaped plates; and tobacco VI as hexagonal prisms. Tobacco I and II, for unknown reasons, crystallise in a variety of different forms. The two commonest are dodecahedra and bipyramids, but thin round laminae and elaborately twinned structures also occur. The materials isolated from all the cultures of tobacco necrosis seem to be essentially nucleo-proteins.

### 32. BAWDEN, F. C. and PIRIE, N. W. 1943. Methods for the purification of tomato bushy stunt and tobacco mosaic viruses. Biochem. J., 37, 66-70.

Methods, requiring only low-speed centrifuges, are described for the purification of tomato bushy stunt and tobacco mosaic viruses.

These preparation appear to contain virus that is weight for weight as infective as that in clarified sap. There is evidence, however, that the tobacco mosaic virus particles have undergone some aggregation.

33. BAWDEN, F. C. and PIRIE, N. W. 1943. The inactivation of tomato bushy stunt virus by heating and freezing. Biochem. J., 37, 70-79.

Tomato bushy stunt virus loses its infectivity when heated insufficiently to cause denaturation and loss of serological activity. The temperature coefficient for the loss of infectivity is small and for loss of serological activity is large. The amount of heating needed for denaturation varies with the pH.

No differences have been found between the chemical and physical properties of non-infective, but serologically active, material and those of fully active preparations.

The rate of inactivation by freezing is increased by increases in the concentration of the virus, in the duration of freezing and in the acidity of the fluid. The virus is protected from inactivation by salts and some other substances. The efficiency of different salts depends on the salt: ice: water eutectic temperature.

In general, loss of infectivity is accompanied by the separation of precipitate and loss of serological activity, but in some conditions freezing destroys infectivity without altering serological activity.

34. BAWDEN, F. C. and PIRIE, N. W. 1944. The liberation of virus, together with materials that inhibit its precipitation with antiserum, from the solid leaf residues of tomato plants suffering from bushy stunt. Brit. J. Exp. Path., 25, 68-80.

After the sap has been expressed from minced tomato leaves infected with tomato bushy stunt virus, the solid residues contain approximately as much virus as the sap. This virus is most effectively liberated by incubating the residues with a commercial trypsin preparation and then passing them through a roller mill; some virus is liberated by either treatment alone. Incubation with "trypsin" greatly increases the amount of virus liberated by milling, whereas extended milling reduces the amount liberated by "trypsin." Purified preparations of virus from sap and from the solid residues have

Purified preparations of virus from sap and from the solid residues have similar properties. Extracts of milled fibre contain some virus combined with chromoprotein to form a non-precipitating antigen; such extracts do not precipitate with virus antiserum until the chromoprotein has been removed. Non-precipitating complexes of virus and chromoprotein can be formed by milling fibre of uninfected plants to which purified virus is added.

Extracts of fibre from healthy and infected leaves, which has been incubated with "trypsin," contain material that inhibits the precipitation of bushy stunt virus by its antiserum.

35. BAWDEN, F. C. and PIRIE, N. W. 1945. Further studies on the purification and properties of a virus causing tobacco necrosis. Brit. J. Exp. Path., 26, 277-285.

A nucleoprotein that is not present in the leaves of healthy plants has been isolated from bean and tobacco leaves infected with the Rothamsted culture of tobacco necrosis virus. This has not crystallised when precipitated with salt, but it crystallises slowly from concentrated salt-free solutions or during sedimentation by ultra-centrifugation. It has a sedimentation constant of 49S, smaller than that of other preparations of plant viruses previously studied. The Rothamsted culture of tobacco necrosis virus readily loses infectivity and the relationship between the crystallisable protein and the virus is uncertain. It is most likely that much of the protein is a non-infective derivative of the virus having many physical, chemical and serological properties in common with it.

36. BAWDEN, F. C. and PIRIE, N. W. 1945. The separation and properties of tobacco mosaic virus in different states of aggregation. Brit. J. Exp. Path., 26, 294-315.

It is shown that tobacco mosaic virus aggregates when exposed to constituents of sap and to many other agents. A method is described for extracting virus from infected leaves which greatly minimises aggregation. The virus of such extracts is inhomogeneous and can be separated by differential ultracentrifuging into fractions with widely different properties. The most slowly sedimenting fractions contain much material other than virus nucleoprotein ; the virus in them shows no anisotropy of flow, has serological behaviour resembling that of somatic antigens and only small infectivity. The most rapidly sedimenting fractions contain little except virus nucleoprotein, show anisotropy of flow and have serological behaviour characteristic of flagellar type antigens.

All the fractions are unstable and readily pass into forms that sediment rapidly, show intense anisotropy of flow and have a serological behaviour characteristic of flagellar antigens. In most fractions this change is accom-

panied by the destruction of material other than virus nucleoprotein. It is suggested that the primary virus particle is small and not greatly elongated, and that it occurs in the plant combined with extraneous materials, the removal of which sets free groups capable of combining with one another. Reasons for the variation in infectivity of different fractions are discussed, but no definite conclusions reached.

#### 37. BAWDEN, F. C. and SHEFFIELD, F. M. L. 1944. The relationships of some viruses causing necrotic diseases of the potato. Ann. Appl. Biol., 31, 33-40.

Potato virus B, and some other viruses with reactions in potato varieties different from any previously described, are strains of virus X. All produce intracellular inclusions which vary with different hosts and virus strains. Except with virus B, the inclusions are larger and more frequent in potato than in tobacco or tomato. All give systemic infection when inoculated to tobacco, tomato and potato varieties in which they are carried or cause mosaic symptoms; some give systemic infection when inoculated to varieties in which they cause top-necrosis, whereas others give only local lesions.

Potato virus C is a strain of virus Y : in tobacco and a few potato varieties both produce similar symptoms, but in those varieties in which Y causes leaf-drop streak, C causes top-necrosis. C causes systemic infection when inoculated to tobacco and to potato varieties in which it causes mosaic symptoms, but not when inoculated to potato varieties in which it causes topnecrosis. Virus C was not transmitted by *M. persicae*. Viruses C and Y produce a few small intracellular inclusions in potato and tobacco.

Virus A is not related to Y or X: no inclusions were found in plants infected with A alone.

38. CROOK, E. M., SHEFFIELD, F. M. L. and CHILTON, L. V. 1945. Photographic plates for use in the R.C.A. electron microscope. Photographic J. B, 85, 6-12.

Characteristic curves of a range of Ilford plates to electrons of energies from 15 up to 60 kV have been determined in the R.C.A. Type-B electron microscope by applying to the plates an intensity scale of exposures based upon changes in net beam current. In all cases the curves show a rise of gamma with electron energy. This effect, which is linear over the lower part of the kV range, is attributed to electron absorption by the emulsion layer. It confirms similar findings by von Borries for Agfa and Perutz plates. The dependance of image contrast-on electron energy is briefly discussed in relation to image-formation by (a) differential absorption, (b) differential scattering. With electrons of 45kV energy, the plates studied exhibited a range of speeds of only 13: 1 at a density of 2 as compared with a range of speeds to white light of 100: 1. Speed comparisons from one kilovoltage to another were made possible by adjusting the magnification to give a standard brightness on the fluorescent screen of the instrument at a standard beam current. Some of the plates studied showed signs of a maximum sensitivity to electrons at the higher end of the kV-range, similar to that found by previous workers.

#### 39. DONCASTER, J. P. 1943. VI. The life history of Aphis (Doralis) rhamni B.d.F. in eastern England. Ann. Appl. Biol., 30, 101-104.

The successive stages in the life cycle of *Aphis rhamni* are described in detail. The climatic and other factors which influence migration, and the growth and development of infestations on potatoes, are discussed. Though known to be a vector of Potato Virus Y, *A. rhamni* does not appear to be responsible for the spread of this virus in the field.

40. DONCASTER, J. P. and KASSANIS, B. 1946. The Shallot Aphis, Myzus ascalonicus Doncaster and its behaviour as a vector of plant viruses. Ann. Appl. Biol., 33, 66-69.

A new species of aphis, Myzus ascalonicus Doncaster, is briefly described, and compared with Myzus persicae Sulz., which it resembles superficially. It has been found on shallots in storage and on onions and other species of plants both in glasshouses and in the open between October and June. Its summer habits and hosts are unknown. In comparative virus transmission tests with Myzus persicae it was found that Myzus ascalonicus transmits dandelion yellow mosaic virus, which is not transmitted by M. persicae; and also cucumber virus 1, hyoscyamus virus III and sugar beet yellows virus all of which are transmitted by M. persicae. M. ascalonicus does not transmit the viruses of potato Y, severe etch, lettuce mosaic and sugar beet mosaic which are transmitted by M. persicae.

# 41. GREGORY, P. H. 1943. VII. The spread of potato virus diseases in the field. Ann. Appl. Biol., 30, 104.

No correlation is found between total aphis population and amount of spread of virus diseases. Much of the spread occurs early in the summer. The chances of healthy plants becoming infected fall off rapidly with increasing distance from diseased plants.

#### 41a. GREGORY, P. H. 1944. Early planting for increased potato yields. Agriculture, 50, 557-559.

A close correlation is shown between the date of planting of Majestic potatoes and the yield obtained at lifting. Ten days delay in planting decreased yield by as much as  $3\frac{1}{2}$  tons per acre.

## 42. HALE, J. B., WATSON, M. A. and HULL, R. 1946. Some causes of chlorosis and necrosis of sugar beet foliage. Ann. Appl. Biol., 33, 13-28.

The symptoms and characteristics of two virus and one fungus disease and four nutritional disorders of sugar beet which cause chlorosis and necrosis of the foliage are described. The causes of the diseases and methods of distinguishing between them have been investigated by analytical, pathological and field experimental methods.

Experiments in which diagnosis was confirmed by serological and spectrochemical methods show that the two often easily confused diseases, sugar beet yellows virus and magnesium deficiency, can be visually distinguished.

Sugar beet Yellows virus reduces the potassium but slightly increases the magnesium content of the leaves.

Magnesium deficiency symptoms are associated with a low magnesium content of the foliage, but may be induced by salt applications without greatly affecting the magnesium analysis. "Potash" deficiency symptoms are often, but not necessarily, associated

"Potash" deficiency symptoms are often, but not necessarily, associated with a low potassium analysis and may actually be caused solely by a deficiency of sodium. In the field symptoms are induced by sulphate of ammonia and phosphate applications and may be prevented in some cases by the application of either salt or muriate of potash, in others by salt only. Some inter-changeability of the functions of potassium and sodium in the

plant is suggested.

Manganese deficiency symptoms are associated with a low manganese content of the leaves, which can be readily increased by spraying or injection with manganese sulphate solution, but a high concentration of manganese in the foliage, such as sometimes occurs naturally on acid soils, has a toxic effect.

43. HULL, R. and WATSON, M. A. 1945. Virus yellows in sugar-beet root and seed crops. Kirton Agric. J. No. 10, 45-48.

Symptoms of sugar-beet yellows are described and possible control measures are discussed, such as isolating steckling beds from the root crop and aphis control by use of insecticides.

#### 44. HULL, R. and WATSON, M. A. 1945. Virus yellows of sugar beet. Agriculture, 52, 66-70.

Losses of up to 50 per cent. can occur because of yellows, if infection occurs early. The disease is usually more serious in districts where seed crops are grown and the overwintering stecklings seem one of the main sources of virus for the annual root crop. The most important vector is *Myzus persicae*.

45. KALMUS, H. and KASSANIS, B. 1944. Reduction by carbon dioxide of susceptibility of beans to tobacco necrosis viruses. Nature 154, 641-42.

Exposure to an atmosphere containing carbon dioxide reduced the number of local lesions produced by tobacco necrosis viruses, whether plants were inoculated immediately before or immediately after exposure. The effect is temporary; 4 hours after exposure plants regain their full susceptibility.

# 46. KALMUS, H. and KASSANIS, B. 1945. The use of abrasives in the transmission of plant viruses. Ann. Appl. Biol., 32, 230-34.

The effect of different abrasives on the transmissibility of several plant viruses was tested. Celite and animal charcoal were as effective as carborundum in increasing the number of lesions produced by a given inoculum; 400-mesh carborundum was the most effective among the different sizes which were tested; this gave a result equivalent to increasing the virus content a hundred times. Some preparations of carborundum and charcoal reduced infectivity.

Uninjured plants resisted infectivity when virus solutions were sprayed over them. Leaves previously rubbed without abrasives developed only few lesions whereas leaves rubbed with abrasives developed large numbers. Three hours after rubbing with abrasives, leaves had regained their resistance to sprayed virus solutions.

The effects of rubbing leaves with abrasives are described and their significance discussed.

#### 47. KASSANIS, B. 1939. Intracellular inclusions in virus infected plants. Ann. Appl. Biol., 26, 705-09.

Two kinds of intracellular inclusions in solanaceous plants infected with severe etch virus are described. One occurs in the cytoplasm and is similar to the X-bodies found in many other plant virus diseases. The other occurs only in the nuclei. These intranuclear inclusions appear to be crystalline, have the form of thin rectangular plates, and resemble the inclusions described in the polyhedral disease of silkworms more than any other type of previously recognised virus-inclusion.

#### 48. KASSANIS, B. 1941. Transmission of tobacco etch viruses by aphides. Ann. Appl. Biol., 28, 238-43.

Severe virus is transmitted by Myzus persicae, M. circumflexus, Aphis rhamni, A. fabae and M. gei. Although the content of mild etch virus in sap is much less than that of S.E.V., both are transmitted to the same extent by M. persicae. The percentage of infection using single aphides is greatest with aphides that are starved for 4 hrs. or more and then fed on the source of infection for 2 mins. Continuous feeding on healthy plants or diseased plants greatly reduces the efficiency of the vector. The length of time for which aphides remain infective is also increased from 15 minutes to a few hours if the aphides starve instead of feed; it is also greatly increased in starved insects if they are kept at a low temperature. Provided the feeding time on each test plant is small, one aphis may infect up to four plants.

## 49. KASSANIS, B. 1942. Transmission of potato virus by Aphis Rhamni (Boyer). Ann. Appl. Biol., 29, 95.

It is shown that in glasshouse experiments *Aphis rhammi* is as efficient a vector as *Myzus persicae* in transmitting potato virus Y. The ability of both aphides to transmit is increased by preliminary starving.

#### 50. KASSANIS, B. 1943. Neutralisation of some plant viruses by rabbit sera. Brit. J. Exp. Path., 24, 152-59.

The unspecific neutralisation of plant viruses by normal and heterologous sera was so large that the additional specific effect of homologous antisera was small in comparison. This specific neutralisation could be used for demonstrating serological relationships only if sera of the same age and stored similarly were compared; the relationships indicated were the same as those indicated by precipitin tests.

Unless sera were kept frozen their unspecific neutralising power fell rapidly on storing. All heterologous antisera reduced infectivity more than normal sera, stored comparably.

Precipitating antibodies did not appear to be responsible for neutralisation. No correlation was found between precipitin titre and neutralising power, and removal of precipitins did not affect neutralisation power. Only quantitative differences were found in behaviour between homologous and other sera ; the infectivity of all virus-serum mixtures was regained by dilution. 51. KASSANIS, B. 1944. A virus attacking lettuce and dandelion, Nature, 154, 16.

A description of a new virus, dandelion yellow mosaic, transmitted from dandelion to lettuce by Myzus ornatus but not by M. persicae. The relationships between the virus and its vector seem different from any previously described.

### 52. KASSANIS, B. and KLECZKOWSKI, A. 1944. The effect of formaldehyde and mercuric chloride on tobacco mosaic virus. Biochem. J. 38, 20-24.

(1) Tobacco mosaic virus was inactivated by 2 per cent. formaldehyde at all pH values between 3 and 7.5; the rate of inactivation was min mal at pH 3.5. Inactivation could be stopped at any stage by dilution or dialysis, but there was no evidence that inactivated virus regained infectivity by these treatments.

(2) Loss of infectivity caused by formaldehyde does not depend on changes in groups giving the Van Slyke test for amino-nitrogen, for preparations inactivated by formaldehyde treatment at pH 3.0 give the same value as control virus. Formaldehyde treatment at pH near 7.0 leads to a greater fall in the Folin pH 8.0 colour value than treatment at pH 3.0. There is no real correlation between the decrease of infectivity and of the colour value.

(3) Mercuric chloride in sufficient concentrations acts as an inhibitor of infectivity. At pH values greater than  $6 \cdot 0$  it causes loss of infectivity and serological activity. Dilution, acidification or addition of certain salts prevent inactivation or interrupt its progress at any stage, but there is no evidence that any of these treatments can reverse it.

#### 53. KASSANIS, B. and SHEFFIELD, F. M. L. 1941. Variations in the cytoplasmic inclusions induced by three strains of tobacco mosaic virus. Ann. Appl. Biol., 28, 360-67.

According to previous accounts tobacco mosaic virus regularly induced striate material and amoeba-like inclusions and occasionally raphides in the host cells; enation mosaic virus gave striate material and amoeba-like Xbodies; whilst aucuba mosaic virus induced either striate material or a large amorphous inclusion which later gave rise to striate material. A spike-like body recorded in early descriptions of aucuba mosaic disease had not been seen for some years. In 1940, a variety of new forms were induced by all three strains. These new forms were mostly fibrous. The spike-like body reappeared, spindle-shaped bodies, masses of short needle-like fibres and extremely long coiled fibrous forms occurred. New amorphous forms were also found. All these arose either directly or from pre-existing inclusions of the previously recorded types. Variation in the inclusions produced is not due to mutation of the virus. The type of inclusion is to some slight extent determined by the host plant but seems to be largely controlled by the amount of light and heat available to the host.

54. KLECZKOWSKI, A. 1941. Quantitative studies on the serological reactions of some plant viruses and of a pea nodule bacterium (Rhizobium leguminosarum). Brit. J. Exp. Path., 22, 44-58.

Quantitative investigations of serological reactions of tobacco mosaic, aucuba mosaic, and bushy stunt viruses and a strain of pea nodule bacteria, with their homologous antisera, and of cross-reaction between tobacco mosaic and aucuba mosaic viruses, were made.

Antibody-antigen ratios in the precipitate formed at equivalence point by these plant viruses with their homologous antisera occupy an intermediate position between ratios for bacterial agglutination and for precipitation of smaller antigens like ovalbumin or blood serum proteins.

An aucuba mosaic virus antiserum contained antibodies reacting with aucuba mosaic virus but not with tobacco mosaic virus, in addition to antibodies reacting with both, whereas all antibodies in a tobacco mosaic virus antiserum reacted with both viruses.

With the same amount of antibody maximum precipitate with the rodshaped tobacco mosaic and aucuba mosaic viruses is much greater than with the spherical (or almost spherical) bushy stunt virus, and is formed in much greater antigen excess.

Qualitative differences between strong and weak tobacco mosaic virus antisera were found.

### KLECZKOWSKI, A. 1941. The formation of protein complexes in heated solutions of rabbit serum proteins. Brit. J. Exp. Path., 22, 188-92. 55.

The effects of heating normal rabbit serum albumin and euglobulin fractions separately and together are described. When a mixture of the two fractions is heated a product is formed with properties different from those of either fraction heated separately. This product is a complex formed by the two fractions uniting as they undergo denaturation.

#### KLECZKOWSKI, A. 1941. Effect of heat on flocculating antibodies of rabbit antisera. Brit. J. Exp. Path., 22, 192-208. 56.

Experiments on the effect of heat on rabbit antisera to the following antigens have been made : human serum albumin and globulin, a strain of pea nodule bacteria (*Rhizobium leguminosarum*); and purified preparations of the following plant viruses-tobacco mosaic, potato "X," tomato bushy stunt and tobacco necrosis.

Antisera to the rod-shaped viruses (tobacco mosaic and potato "X") behave like those to flagellar type antigens, whereas antisera to the other antigens named behave like those to somatic type antigens, much more heating being needed to destroy the flocculating power of the former. How-ever, euglobulin fractions (containing antibodies) of all the antisera behave similarly, and they require more heat to destroy their flocculating power than do the original antisera.

Flocculating antibodies undergo at least two changes during heating. Complexes composed of antibody particles and of particles of other unspecific proteins present in the solutions are first formed. Antibodies changed in this way can still combine specifically with antigens, but the result of this com-bination depends on the quantity and quality of unspecific proteins present in the solution during heating. Complexes formed when antibodies are heated in the presence of euglobulin fraction of the antiserum flocculate their antigens. Complexes formed when antibodies are heated in the presence of other serum fractions, notably albumin, cannot flocculate their antigens, although they combine with them ; this combination interferes with the flocculating action of antibodies that are unchanged. The degree of this interference depends on the type of antigen, being large with antigens of type "O" and small with those of type "H." This fact, and not a difference in heat stability, explains the differences in the behaviour of heated antisera to the two types of antigen.

The second change of antibodies during heating corresponds with a further stage of denaturation and is shown by the loss of ability to combine with antigen.

### KLECZKOWSKI, A. 1943. The effect of salts on the formation of protein complexes during heat denaturation. Biochem. J., 37, 30-36. 57.

The formation of complexes between different proteins, undergoing heat denaturation together, is greatly influenced by the presence of salts. In the absence of salts only mixtures containing water-soluble serum globulin formed any detectable amount of complexes.

The efficiency of different salts in promoting the complex formation, expressed as the reciprocal of the lowest effective concentration, follows Hardy's law. Ions of higher valency are much more effective than those of lower valency. On the alkaline side of the isoelectric point of the proteins, the efficiency of salts is determined by the valency of the cations, and on the acid side by the valency of anions.

#### 58. KLECZKOWSKI, A. 1944. Combination of potato virus X and tobaccomosaic virus with pepsin and trypsin. Biochem. J., 38, 160-67.

(1) Pepsin combines with potato virus X and casein, which are substrates for its proceelytic activity, but not with tobacco mosaic virus, which is not a substrate.

(2) Tobacco mosaic virus denatured by heat is readily hydrolyzed by pepsin and combines with pepsin almost to the same extent as potato virus X.

(3) Invertase does not combine with potato virus X, with tobacco mosaic

(4) More trypsin combines with tobacco mosaic virus, which is not a substrate for its proteolytic activity, than with potato virus X, which is a substrate. The combination of trypsin with tobacco mosaic virus could account for the reversible inhibition of infectivity of the virus by trypsin.

(5) Combination between trypsin and tobacco mosaic virus protects trypsin from spontaneous inactivation at pH 7.0.

(6) Trypsin and invertase adsorbed on charcoal can be set free by casein ; invertase can also be extracted by tobacco mosaic virus, but not by sucrose.

# 59. KLECZKOWSKI, A. 1945. The reaction of products of initial stages of peptic proteolysis of human and horse serum albumin with antisera to the original albumins. Brit. J. Exp. Path., 26, 24-33.

Peptic digestion of human or horse albumin in suitable conditions eventually leads to loss of their ability to react in any detectable way with antibodies to the original albumins. Before this stage is reached some products of peptic proteolysis of the albumins are still able to combine specifically with the antibodies. These products differ from the original albumins in that their precipitability by the antisera to the original albumins is greatly reduced. They do not differ from the original albumins in their diffusion rates. Precipitability of the products by the antisera can be restored by heating solutions of the products in the presence of salt.

# 60. KLECZKOWSKI, A. 1945. Conversion of non-precipitating and inhibiting protein complexes into forms again precipitable by the antisera to the original proteins. Brit. J. Exp. Path., 26, 33-41.

When tomato bushy stunt virus is heated together with a serum albumin the virus can combine with the albumin to form a complex that does not precipitate with the antiserum to the virus. The virus, which is resistant to peptic proteolysis, can be recovered in the precipitable form by peptic hydrolysis of the complex. Non-precipitating complexes similarly formed between human and horse serum albumin can, in suitable conditions, be split by means of pepsin into forms again precipitable specifically by antisera, although both components of the complex are susceptible to peptic proteolysis.

## 61. KLECZKOWSKI, A. 1945. Specific precipitation of one protein by antiserum to another. Brit. J. Exp. Path., 26, 41-49.

Products of interaction between human and horse albumin formed during heat denaturation of mixtures of the two, could be precipitated by the antisera to either albumin. The product formed in mixtures, where human albumin was in excess, combined with antibodies to horse albumin, but did not precipitate with them. It precipitated with antibodies to human albumin. The converse of this was true when horse albumin was in excess over human albumin in heated mixtures.

Products of interaction formed between antibodies to bushy stunt virus and human or horse serum albumin, formed when mixtures were heated, could combine, but not precipitate, with the virus, although they could be specifically precipitated with antiserum to the albumin.

#### 62. KLECZKOWSKI, A. and THORNTON, H. G. 1944. A serological study of root nodule bacteria from pea and clover inoculation groups. J. Bact., 48, 661-72.

Twenty-nine strains of nodule bacteria derived from plants of the pea inoculation group, and 161 strains derived from clover nodules, were tested for agglutination with antisera against six strains of nodule bacteria, four derived from clover and two from pea.

No antigen or set of antigens of either O or H type was found to be common to the whole group. Some strains reacted with none of the antisera, some with only one, and others with several antisera, but none with all of them.

Strains belonging to both inoculation groups were found to give agglutination with five of the antisera; one antiserum reacted only with a small number of strains all from clover.

Neither ability to cross-inoculate between clover and pea host plants, nor effectiveness in fixing nitrogen within the host plant, nor susceptibility to a bacteriophage was necessarily associated with the presence or absence of any one antigen or group of antigens. Indeed, an effective strain and a very ineffective variant derived from it were found to be serologically identical.

There was, however, a partial correlation between effectiveness and the H antigenic constitution.

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63. KLECZKOWSKI, A. and WATSON, M. A. 1944. Serological studies on sugar beet yellows virus. Ann. Appl. Biol., 31, 116-20.

Specific antisera have been prepared against the sap expressed from beet plants infected with beet yellows virus.

The antigen is unstable. In the sap it is destroyed by keeping for 2 to 3 days at room temperature or by heating for 10 minutes at 52° C. It is unaffected by pH changes between 5 and 9. In detached leaves at room temperature it remains unchanged for at least 6 days, whereas the ability of aphides to transmit from these leaves fell considerably in 4 days.

The antigen can be reversibly precipitated by ammonium sulphate or sedimented by high-speed centrifugation. However, all the attempts to isolate it from other sap constituents or to concentrate it have failed.

The precipitin reaction is of value for diagnosis, and works successfully with crude sap.

### 64. ROBERTS, F. M. 1940. Studies on the feeding methods and penetration rates of Myzus persicae Sulz., Myzus circumflexus Buckt., and Macrosiphum gei Koch. Ann. Appl. Biol., 27, 348-58.

(1) The feeding habits and penetration rates of three aphides Myzus persicae, M. circumflexus, and Macrosiphum gei on tobacco, and of Myzus persicae on sugar beet were investigated in relation to their transmission of Hyoscyamus virus III, potato virus Y, cucumber virus I, and sugar beet yellows virus.

(2) Neither *M. persicae* or *M. circumflexus* were found to reach the phloem after 5 minutes feeding on tobacco. Very few *M. persicae* penetrated the phloem in 15 minutes on tobacco or sugar beet. Even after 24 hours a few aphides do not penetrate the phloem but feed on non-vascular tissue.

(3) Myzus persicae and Macrosiphum gei were found to penetrate by the intracellular method in more than 50 per cent. of the slides examined, and Myzus circumflexus in less than 50 per cent. Macrosiphum gei showed a higher percentage of phloem penetrations in 1-24 hr. than the other aphides.

(4) There were no visible toxic effects on the part of the host plants to the insects' saliva with either tobacco or sugar beet.

(5) A possible correlation between the behaviour of Myzus persicae and M. circumflexus with cucumber virus I and localisation of this virus in the leaf.

(6) Increased infections with increased feeding times on both infected and healthy plants, in the transmission of sugar-beet yellows virus by its vector M. persicae is discussed in relation to phloem feeding.

# 65. SADASIVAN, T. S. 1940. A quantitative study of the interaction of viruses in plants. Ann. Appl. Biol., 27, 359-67.

When the saps of healthy plants are mixed with potato virus " $X^s$ " or aucuba mosaic virus *in vitro* there is an inhibition of the lesion production on *N. tabacum* and *N. sylvestris* leaves. Saps containing unrelated viruses also reduce the infectivity to the same extent as healthy saps. However, saps containing strains of related virus, have a greater and specific inhibitory action. Experiments were performed to show that this specific reduction produced by related strains of viruses is due to the viruses themselves and not other metabolic products present in the saps.

Further study by inoculating related strains of viruses *in vivo* has shown that the local lesions of one strain are inhibited by another when the latter systemically infects hosts, and that the efficiency depends on the concentration of the systemically infecting viruses in such hosts. The degree of inhibition of aucuba mosaic virus is directly proportional to the number of active units of tobacco mosaic virus present in the leaf tissues at the time of reinoculation.

# 66. SHEFFIELD, F. M. L. 1939. Some effects of plant virus diseases on the cells of their hosts. J. Roy. Microsc. Soc., 59, 149-61.

Some virus diseases cause no apparent abnormalities in the cells of the host plant. Others induce the production of inclusion bodies. These are in some cases amorphous, in others crystalline. The different types of inclusion are described and compared. The nature of intracellular inclusions is discussed briefly in the new light thrown by the recent work on the isolation of plant viruses.

# 67. SHEFFIELD, F. M. L. 1941. The cytoplasmic and nuclear inclusions associated with severe etch virus. J. Roy. Microsc. Soc., 61, 30-45.

Severe etch virus induces two types of intracellular inclusion.

The cytoplasmic inclusions are amorphous. Chemically they consist of mixtures of proteins with fats and lipoids. They are formed by the aggregation of particles which appear in the streaming cytoplasm.. They may contain some birefringent particles and may give rise to small needle-like bodies. They can be pricked or divided into portions with a microneedle. They contain the virus, but this is also present in other parts of the cell. They are numerous and occur in most tissues of the plant.

The intranuclear inclusions give protein reactions and are more stable than the cytoplasmic inclusions. They take the form of thin rectangular plates, and as many as 30 may be found in a single nucleus. They can be isolated but can be broken or dissolved only with difficulty. Almost every nucleus contains them and they have been found in almost all tissues. They occur in the seed but have not been found in the young embryo. The virus is not transmitted through the seed.

The two types of inclusion are briefly discussed and compared with other inclusions occurring in diseased and healthy tissues.

68. SHEFFIELD, F. M. L. 1942. Presence of virus in the primordial meristem. Ann. Appl. Biol., 29, 16-17.

It was thought that the absence of intracellular inclusions from the apical meristem of infected plants might be due to failure of the virus to penetrate there. However, if this tissue is dissected out from shoots or roots of plants with tobacco or aucuba mosaic, infections can be obtained from it.

## 69. SHEFFIELD, F. M. L. 1942. The "blotches" on leaves of Arran Pilot potatoes. Ann. Appl. Biol., 29, 341-45.

About flowering time a greyish green blotch appears on some of the leaves of Arran Pilot potato plants. It is due to necrosis of the epidermis, followed by cell division in the palisade tissue resulting in the formation of several layers of small, thin-walled colourless cells. This proliferation may occur on the top only or on both sides of the leaf. The new tissue partially masks the green colour of the plastids in the cells nearer the centre of the leaf. After a few weeks the central tissue dies. The blotching, which is almost certainly of genetic origin, is discussed in comparison with other plant effects which resemble it in one or other of its characters.

## 70. SHEFFIELD, F. M. L. 1943. Value of phloem necrosis in the diagnosis of potato leaf-roll. Ann. Appl. Biol., 30, 131-36.

In plants infected with leaf-roll virus a type of phloem obliteration and necrosis occurs which is distinct from any abnormality produced by other pathogens or arising from physiological causes. The necrosis occurs in the primary phloem only of the bicollateral bundles. The affected tissue reacts with phloroglucinol in HC1. It was present in all of 179 plants of 33 varieties showing secondary leaf-roll which were examined and was not found in any of 83 healthy plants of 20 varieties. The amount of necrosis varies in different plants. If the disease is severe, necrosis may extend to almost all parts of the plant except the stolon, tubers and roots. If the infection is mild it may be confined to a very few strands in two or three nodes near the base of the main stem. Phloem necrosis can always be found before leaf rolling is apparent.

In primary leaf-roll, slight necrosis can be found in the stem near the bases of the lowest rolled leaves and sometimes in the petioles.

A technique is suggested for the use of this symptom in diagnosis.

71. WATSON, M. A. 1940. Studies on the transmission of sugar beet yellows virus by the aphis, Myzus persicae (Sulz). Proc. Roy. Soc., B, 128, 535-52.

The efficiency of the vector Myzus persicae in transmitting sugar beet yellows virus increased greatly with increasing feeding time on the infected plants. Infection was produced on a succession of healthy plants for 1, 2, or 3 days depending on the length of the infection feeding time. The infectivity of the vectors increased with increasing feeding time on the healthy plants undergoing infection, and decreased with increasing feeding time on healthy

plants prior to those on which the infection trial was made. There was no clearly defined "incubation period" of the virus in the vector, below which no insect could cause infection, but there was variation in the time between cessation of infection feeding of the aphis and the initiation of infection in the healthy plants.

The relations of this virus with its vector differ from those of the viruses already described as *non-persistent* (Watson and Roberts, 1939). For the latter viruses infectivity is lost by *M. persicae* soon after cessation of infection feeding; after fasting the vectors become optimally infective almost immediately on penetrating infected tissues of the leaf. Their infectivity decreases with increasing feeding time on the infected plants, and increases only slightly with increasing feeding time on the healthy plants.

The behaviour of sugar beet yellows virus is compared with that of curlytop virus of sugar beet, in which infectivity also persists for an indefinite period in the vector and increases with increasing feeding time on infected and healthy plants.

#### 72. WATSON, M. A. 1942. Sugar beet yellows virus. A preliminary account of experiments and observations on its effect in the field. Ann. Appl. Biol., 29, 358-65.

The symptoms of sugar beet yellows virus in the field vary with different meteorological and cultural conditions. Illustrations of typical symptoms are given, and some account of their appearance and distribution in the field. Infection greatly reduces the yields of roots and sugar. In experiments early infection on late sown beet caused a loss of 67 per cent. of the root, and 71 per cent. of the sugar yield. The loss decreased with later infection and earlier sowing date. The main source of infection and means by which the virus is carried over from year to year appears to be the sugar beet seed crop. Proximity of the seed crop to the root crop determines the number of viruliferous migrant aphides which enter the root crop at the initial infestation. Subsequent spread of virus in the root crop is determined by the rate of reproduction and of movement of the apterous aphides. Reproduction is more rapid on late than on early sown beet, and seems to be increased by poor nutrition of the plants.

73. WATSON, M. A. 1946. The transmission of beet mosaic and beet yellows viruses by aphides; a comparative study of a non-persistent and a persistent virus having host plants and vectors in common. Proc. Roy. Soc., B, 133, 200-219.

The behaviour of beet mosaic and beet yellows viruses in relation to their insect vectors was compared, using Myzus persicae (Sulz.) as the vector and sugar beet as the common host plant. Beet mosaic virus proved to be one of the "non-persistent" group, resembling Hyocyamus virus 3, and potato virus Y, etc. Its vectors were optimally infective when allowed to feed for only 2 or 3 minutes on the source of infection after a period of fasting. Feeding time on the healthy plant had only a small and limited effect on infectivity, in some conditions of infectivity of the aphides. The vectors lost infectivity rapidly when feeding and slowly when fasting. When optimally infective they could infect several healthy plants in succession but only during the first half-hour after cessation of infectivity for 20 hours.

Vectors of beet yellows virus were optimally infective after about 6 hours infection feeding and 3 hours feeding on the healthy plants. They could infect in much shorter time, but their infectivity increased up to about these limits. The efficiency of the vectors was not affected by a period of fasting given before infection feeding. The period of delay in production of optimum infectivity during feeding on the healthy plants was independent of the infection feeding time. Vectors of beet yellows virus lost infectivity more rapidly when fasting than when feeding on healthy plants. There was no increase of infectivity during the first few hours of fasting as there was during feeding, and the curve relating loss of infectivity to fasting time was similar to that obtained with beet mosaic virus.

Aphis fabae (Scop.) behaved similarly to *M. persicae* with both viruses. Differential fasting and feeding treatments were used to separate the viruses when they infected the same plant, and the method could be used for separating complexes which occur in nature and which include non-sap transmissible viruses such as beet yellows virus. 74. WATSON, M. A. and ROBERTS, F. M. 1939. A comparative study of the transmission of Hyoscyamus virus 3, potato virus Y and cucumber virus 1 by the vectors Myzus persicae (Sulz.), M. circumflexus (Buckton), and Macrosiphum gei (Koch). Proc. Roy. Soc., B, 127, 543-76.

Three strains of *Hyoscyamus* virus 3, two of cucumber virus 1, and potato virus Y, were tested for their transmissibility by the aphides, *Myzus persicae*, *M. circumflexus*, and *Macrosiphum gei*. The efficiency of the vectors in transmitting all the viruses increased with increasing time of fasting before feeding on the infected plants. Their efficiency decreased as the time of feeding on the infected plants increased. The most probable explanation of these effects is that the viruses are inactivated by some substance produced by the aphides when feeding.

The most successful vector on the whole was *Myzus persicae*, and the least successful was *Macrosiphum gei*, but the relative efficiency of the vectors varied with the different viruses, indicating that their degree of success depended upon several interacting factors. The most important of these factors appeared to be:

- (a) The concentration of virus in the host plant.
- (b) The localisation of virus in the host plant.
- (c) The capacity of the vector for inactivating the virus.

The viruses which have thus been shown to be similar in their insect-virus relationships, are also similar in their physical properties, and there are many other aphid transmitted viruses which resemble them in this respect. It is suggested that such viruses may form a natural group, with the same type of vector-virus relationship. This relationship appears to be a complex one, and it is unlikely that the viruses are mechanically transmitted.

# 75. WATSON, M. A. and ROBERTS, F. M. 1940. Evidence against the hypothesis that certain plant viruses are transmitted mechanically by aphides. Ann. Appl. Biol., 27, 227-33.

Individuals of M. persicae, when given an adequate fasting period followed by 2 minutes feeding on the infected plants, can transmit Hyoscyamus 3, potato Y, and severe-etch viruses from tobacco to a number of successive healthy plants. The number of plants infected seems to be that on which the insects can be given 2 minute feeding periods, within the time for which they would normally retain their infectivity if prevented from feeding at all. This is considerably longer than the time for which they would remain infective when feeding on a healthy plant. Rapid loss of infectivity of the vectors, especially when feeding, is a characteristic property of the type of virus used in these experiments which we have defined as "non-persistent" (Watson & Roberts, 1939). The present results confirm our previous hypothesis that loss of infectivity is not due to cleansing of the stylets when feeding, or to normal deterioration of the virus in the insects when fasting. The viruses appear to be inactivated by some substance produced by the aphides after they have been feeding for about 2 minutes. Persistent viruses, which are retained by their vectors for indefinite periods, are not affected by this substance, the vectors being able to acquire lasting infectivity by prolonged feeding upon infected plants, even when the virus available in the leaves is highly localised or very limited in amount. Most of the major differences between the results of transmission experiments with the two types of viruses could be accounted for by differences in the effect of the inactivating substance produced by the vectors. It is therefore unnecessary to postulate fundamentally different mechanisms of transmission between the two types.

#### MYCOLOGY

#### BOOK

76. GARRETT, S. D. 1944. Root disease fungi. The Chronica Botanica Co., Waltham, Mass., U.S.A., p. 77.

#### REVIEWS

77. GARRETT, S. D. 1939. Soil-borne fungi and the control of root disease. Imp. Bur. Soil Sci. Tech. Comm., 38, p. 54.

78. GARRETT, S. D. 1942. The take-all disease of cereals. Imp. Bur. Soil Sci. Tech. Comm., 41, p. 40.

#### 79. GARRETT, S. D. 1945. The root-infecting fungi. Endeavour 4, 104-7.

#### SCIENTIFIC PAPERS

80. BLAIR, I. D. 1943. Behaviour of the fungus Rhizoctonia solani Kühn in the soil. Ann. Appl. Biol., 30, 118-27.

Rhizoctonia solani was found able to grow as a saprophyte through natural unsterilised soil. Its rate of growth under different soil conditions in glass tumblers was studied by the Rossi-Cholodny soil-plate method. Growth was most rapid at the lowest soil-moisture content tested, viz.: 30 per cent. saturation, and was accelerated by forced aeration of the soil. The maximum distance to which mycelial growth could be supported on the food reserves of the agar inoculum alone was some 5 cm., as shown by extent of growth through tubes of moist sand, but in 23 days the fungus grew 21-24 cm. through tubes of soil. Removal of the agar disk 2 days after inoculation of the tubes reduced growth through sand by more than half, but through soil by only a small proportion. In soil, Rhizoctonia was able to cause 100 per cent. damping-off of radish seedlings planted at a radial distance of 9 cm. The depressing effect of additions of 1 per cent. ground-wheat straw or dried grass to the soil upon growth of the fungus was attributed to (a) the negligible cellulose-decomposing ability of Rhizoctonia ; (b) nitrogen starvation of the mycelium, through rapid util.sation of the available soil nitrogen by the cellulose-decomposing micro-organisms multiplying upon the fresh organic material ; (c) fungistatic action on Rhizoctonia of the respiratory carbon dioxide produced by the cellulose-decomposing

#### 81. BUDDIN, W. and GARRETT, S. D. 1944. Take-all of cereals in 1943. Agriculture 51, 108-10.

Quantitative methods used for this take-all survey in the Southern Advisory Province are described, and examples given of the effect of susceptible grass weeds acting as carriers of the take-all fungus.

#### DION, W. M. and LORD, K. A. 1944. A comparison of the toxicity of certain dyestuffs to the conidia of Fusarium culmorum. Ann. Appl. Biol., 31, 221-31.

The toxicity of a number of dyestuffs to the spores of *Fusarium culmorum* and *Cercosporella herpotrichoides* was determined by the slide-germination technique. No attempt was made to distinguish between fungistatic and fungicidal activity.

The toxicity of basic dyestuffs was unaffected by the acid radicle associated with the dye base.

The high toxicity to *Fusarium culmorum* of malachite green dye base was reduced weight for weight and mole for mole by substitution of ethyl, propyl, or butyl groups for methyl groups.

The reduction of malachite green to malachite green *leuco* base removed toxicity.

The substitution of amino groups and alkylated amino groups in benzene nuclei of triphenyl methane increased toxicity, whereas acid groups reduced toxicity. Sulphonation and carboxylation reduced toxicity to vanishing point.

Alkylation of amino groups increased, but alkylation of benzene nuclei did not affect toxicity appreciably. When the central carbon atom of the triphenyl methane dyestuffs was

When the central carbon atom of the triphenyl methane dyestuffs was replaced by nitrogen (e.g., Bindschedler's green) the diphenyl ammonium compounds were less toxic than the corresponding triphenyl methane compounds.

The prevention of rotation of the aminated benzene rings by bridging, in the O-position to central atom, with O or N, and so obtaining a planar molecule, only slightly affected toxicity.

Certain acid dyes stimulated fungal growth.

The toxicity of the basic dyestuffs seems to depend not on one specific part but on the molecule as a whole, and within certain limits the structure may be varied without pronounced changes in toxicity.

#### 83. GARRETT, S. D. 1940. Utilisation of nitrogen by Ophiobolus graminis. Nature, 145, 108.

A pure culture of *Ophiobolus graminis*, growing on sterilised wheat straw, was found to utilise nitrate nitrogen.

## GARRETT, S. D. 1940. Soil conditions and the take-all disease of wheat. V. Further experiments on the survival of Ophiobolus gramin's in infected wheat stubble buried in the soil. Ann. Appl. Biol., 27, 199-204.

Further experiments are reported on the survival of *O. graminis* in infected wheat straw buried in soil. The longevity of the fungus appears to be closely related to the available nitrogen content of the soil. Microscopical observation has shown that the characteristic dark mycelium of *Ophiobolus* slowly continues to develop in the artificially infected straw after this has been buried in the soil. This development of dark mycelium is greater, and also more sustained, in soils well supplied with nitrogen. Additional nitrogen is now considered directly to benefit *Ophiobolus*, by enabling the fungus to assimilate more of the undecomposed carbohydrate reserves of the straw. Conditions such as high aeration favouring general microbiological activity in the soil hasten the disappearance of *Ophiobolus*, probably by promoting a more rapid consumption of the available food material by the fungus itself and by associated micro-organisms; the supply of nitrogen normally limits both the amount of available food material and the survival of *Ophiobolus*.

#### 85. GARRETT, S. D. 1941. Soil conditions and the take-all disease of wheat. VI. The effect of plant nutrition upon disease resistance. Ann. Appl. Biol., 28, 14-18.

Red Marvel spring wheat plants were grown singly in sand culture in 7 in. glass flower pots under conditions of full nutrient supply, and under deficiencies of nitrogen, phosphate and potash and of all three together, respectively. After one month's growth, the plants were inoculated with *Ophiobolus* by the insertion of two pieces of infected wheat straw into the sand on each side of and just below the crown.

Satisfactory root infection occurred in every plant inoculated, but was lightest in the nitrogen-deficient plants, whilst the potash-deficient plants exhibited a rather more intense root infection than those of any other series. Percentage infection of the stem-bases was lowest in the full nutrient and in the nitrogen-deficient plants, and highest in the series deficient in all three nutrients.

In the uninoculated control plants, a significant depression in grain yield was produced only by the deficiency of phosphate; in the inoculated plants, however, deficiency in any one of the three plant nutrients significantly reduced grain yield. Infection significantly reduced yield in every series except that receiving full nutrients; the percentage reduction was greatest in the phosphate-deficient plants.

## GARRETT, S. D. 1941. Soil conditions and the take-all disease of wheat. VII. Survival of Ophiobolus graminis on the roots of different grasses. Ann. Appl. Biol., 28, 325-32.

Sixteen species of grasses were inoculated with Ophiobolus graminis, and their roots examined under the binocular dissecting microscope for runner hyphae and discoloured disease lesions. Whilst some species were obviously susceptible and others showed few signs of root infection, there were yet other species which were difficult to classify either as susceptible or as resistant. The effectiveness of these 16 grasses as perpetuators of Ophiobolus was therefore directly tested, as follows. The seed was sown in contact with a minimal amount of inoculum in boxes of a light-textured soil; 2 months after planting, the grass tops were cut off, and the sods inverted in the boxes. The degree of survival of Ophiobolus in the inverted sods of the different grasses was determined at approximately monthly intervals by the planting of test wheat seedlings. Whilst all 16 species propagated Ophiobolus to some extent, as compared with a negligible survival in fallow soil and under clover, there were notable differences in the longevity of the fungus under different grasses. The resistance of Phleum pratense, reported by previous investigators, was confirmed, and seeds mixtures employing this grass and Avena elatior in place of Lolium spp. were suggested for use on heavily-infected land.

#### 87. GARRETT, S. D. 1944. Soil conditions and the take-all disease of wheat. VIII. Further experiments on the survival of Ophiobolus graminis in infected wheat stubble. Ann. Appl. Biol., 31, 186-91.

Assimilable nitrogen in various forms prolonged the life of *Ophiobolus* graminis in infected wheat straw, whether added directly to the straw or to the surrounding soil. When the infected straws were buried in washed quartz sand, 0.5 g. nitrogen per 100 g. air-dry straw was the optimum dressing for longevity of *Ophiobolus*. Addition of sodium phosphate did not significantly increase longevity.

Nitrogen is considered to prolong the life of *Ophiobolus* by enabling the mycelium to form new branch hyphae, which can explore unexhausted parts of the substrate; it is suggested that aged mycelium dies from carbohydrate starvation, through exhaustion of the zones of enzymic erosion around the hyphae. This hypothesis is supported by the extended life of the fungus in infected straws that were shaken twice weekly in 3% dextrose solution. *Ophiobolus* was found to survive longer in infected straws buried in a

Ophiobolus was found to survive longer in infected straws buried in a fallow soil than in the same soil under oats, mustard or trefoil; this finding suggests the use of catch crops as competitors with Ophiobolus for soil nitrogen.

88. GARRETT, S. D. and DENNIS, R. W. G. 1943. Note on the occurrence of Ophiobolus graminis Sacc. var. Avenae E. M. Turner in Scotland in 1942. Trans. Brit. Mycol. Soc., 26, 146-7.

Giving ascospore measurements of O. graminis var. Avenae from 21 collections of diseased material.

89. GLYNNE, MARY D. 1939. Eyespot lodging of wheat caused by Cercosporella herpotrichoides Fron. Agric. Prog., 16.

Paper read before Conference of Advisers, December, 1938.

90. GLYNNE, MARY D. 1942. Cercosporella herpotrichoides Fron., causing eyespot of wheat in Great Britain. Ann. Appl. Biol., 29, 254-64.

Surveys of wheat in 1941 showed that the probability of general lodging in heavy crops is greatly increased when a high percentage of the straws is infected by *Cercosporella herpotrichoides*. Individual straw lodging resulting from infection occurs both in heavy and in light crops, so that the straws fall in all directions. The trouble has been known to farmers for many years under various dialect names, and when severe is estimated to cause a reduction in grain yield of about 30%. A survey of 170 randomly selected fields in sixteen counties showed increases in the frequency and severity of infection by eyespot from west to east,  $3\cdot6\%$  of the fields in North Wales showed eyespot and  $84\cdot5\%$  in the eastern counties ; the percentage area lodged increased in the same way from  $1\cdot1$  to  $16\cdot4\%$  and the percentage of the area under wheat (in 1939) from  $0\cdot17$  to  $12\cdot89\%$ . Two hundred and thirty-five fields not randomly selected, in three independent surveys, included 118 in which no wheat or barley had been grown in the preceding 4 years ; thirtyseven of these showed eyespot infection, always less than 20% straws being infected ; 115 fields which had wheat or barley recorded at least once in the preceding 4 years, included eighty-nine with eyespot, more than half having over 20% straws infected. Infection of over 20% and over 70% occurred on a few fields in which the last preceding wheat or barley crop had been taken in 1937 or 1938, but they were most frequent where wheat or barley (or one of each) had been grown in at least 2 of the preceding 4 years. The disease is likely to increase in wheat-growing districts under the more intensive wheat cultivation resulting from war-time conditions. Measures for reducing loss from eyespot, while increasing wheat acreage, are discussed.

- 91. GLYNNE, MARY D. 1942. Eyespot in wheat. Agriculture, 49, 91-94. Giving general field observations and recommendations for control.
- 92. GLYNNE, MARY D. 1944. Eyespot, Cercosporella herpotrichoides Fron., and lodging of wheat. Ann. Appl. Biol., 31, 377-8.
  - Paper read before Association of Applied Biologists.
- 93. GLYNNE, MARY D. 1945. Eyespot of wheat and barley. Adv. Leafl. Minist. Agric. Fish., 321.
  - Giving general field observations and recommendations for control.

#### 94. GLYNNE, MAEY D. 1946. Eyespot of wheat and barley in Scotland in 1944. Ann. Appl. Biol., 33, 35-39.

In August 1944, eyespot (*Cercosporella herpotrichoides* Fron.) was found in 90 out of 121 autumn-sown wheat crops distributed over 12 Scottish counties. It was abundant enough in 40 fields to be likely to harm subsequent crops, and was causing obvious loss in all crops. The disease was found in 17 out of 18 spring-sown barley crops, more than half the straws being infected in 7 of them. About 4 per cent. of the wheat inspected was lodged mostly by eyespot and about 38 per cent. of the barley, mostly non-parasitically. Eyespot increased with the frequency of wheat and barley in the two preceding years, but a few infected crops occurred on fields where no wheat or barley had been grown for many years. In Scotland, where the atmosphere is more humid, eyespot tends to increase more rapidly than in similar rotations in England; lesions are found higher up the straw and the disease is much more prevalent on spring-sown barley. The long rotations practised in Scotland prevent more extensive damage by eyespot.

#### 95. GLYNNE, MARY D. and RITCHIE, WENDY M. 1943. Sharp Eyespot of wheat caused by Corticium (Rhizoctonia) solani. Nature, 152, 161.

Although one heavily infected crop had been reported, sharp eyespot rarely affected more than 1 per cent. of the straws in the numerous wheat fields of England and Wales in which it had been found. It did not seem to increase with the frequency of wheat and barley in the rotation as did true eyespot, due to *Cercosporella herpotrichoides*.

96. GLYNNE, MARY D., DION, W. M. and WEIL, J. W. 1945. The effect of eyespot (Cercosporella herpotrichoides Fron.) on wheat and the influence of nitrogen on the disease. Ann. Appl. Biol., 32, 297-303.

The effect of eyespot throughout the season on wheat receiving different amounts of nitrogen was studied in pot experiments. Plants inoculated in December showed chlorosis of outer leaves in February. Among plants with high nitrogen, eyespot killed 11 per cent., caused straggling of 31 per cent. and whiteheads in 14 per cent. of the surviving ear-bearing straws, reduced yield of straw by 8 per cent. and of grain by 16 per cent. The loss in straw yield was due to reduction in plant number, that of grain was about half due to reduction in number of ears and half to production of lighter grains. Among plants with low nitrogen the disease killed 23 per cent. of the plants, caused straggling of 86 per cent. and whiteheads in 18 per cent. of the surviving ear-bearing straws, and reduced yield of straw by 23 per cent. and of grain by 44 per cent. The loss in straw yield was due to death of plants, that of grain was about two-thirds due to the production of fewer ears and one-third to that of lighter grains. In the high-nitrogen series the number of shoots at the time of maximum tillering was reduced by 29 per cent.; in the low-nitrogen series the disease caused reduction in height, a very uneven crop, delay in ear and anther emergence, and an increase in tail corn from 4 per cent. in the controls to 30 per cent. in the inoculated plants.

4 per cent. in the controls to 30 per cent. in the inoculated plants. All inoculated plants became infected, but those receiving high nitrogen had only 49 per cent. of the ear-bearing straws with severe lesions at harvest, while those receiving low nitrogen had 86 per cent. The larger number of tillers produced when nitrogen was applied probably enabled the less severely diseased shoots to survive and bear ears while the most severely infected died.

## 97. GREGORY, P. H. 1945. The dispersion of air-borne spores. Trans. Brit. Myc. Soc., 28, 26-72.

The deposition of air-borne fungus spores decreases with increasing distance from a source. The factors controlling the scattering are reviewed, and observed gradients of deposition are discussed.

The terminal velocities of spores depend on their dimensions and are of the order expected for smooth, spherical particles from Stoke's law. Fungus spores have been observed to fall with velocities between 0.04 and 2.5 cm. sec., and pollen grains between 1.5 and 40 cm. sec. The mean wind velocity at 10 m. is near 300 cm. sec. Attempts to calculate the dispersal of spores as the result of vertical fall under gravity and horizontal wind movement are shown to apply only to non-turbulent air movement, and therefore to be inapplicable at heights more than a few millimetres above the earth's surface. Differences in rate of spore fall are probably not major factors in dispersal. It is more appropriate to consider a spore cloud in suspension in the air in process of being diluted by eddies in the course of its transport by the wind.

In support of the concept of the spore cloud as a suspension it is shown that at heights in the atmosphere above the surface layers, the concentrations of pollen and spores decreases exponentially with increasing height in the manner that would be expected if particles falling under gravity were balanced by other particles diffused upwards by eddies. The values for eddy diffusivity, K, varying from  $1.5 \ge 10^4$  to  $3.3 \ge 10^5$ , deduced for spores and pollen, are of the order usually found in meteorological work.

While terminal velocity may play only a small part in spore dispersal, it may be more important in causing deposition of spores brought down by eddies to the boundary layer of relatively still air a few centimetres thick at the earth's surface.

The earlier theories of eddy diffusion, including that of Schmidt, are compared with recent work by Sutton who considered that the size of eddies effective in diluting a cloud, instead of remaining constant, increases with the distance travelled by the cloud. Experiments by Stepanov, in which spores were liberated from a point in the open air and trapped at various distances and in different directions, are shown to be in excellent agreement with Sutton's theory, and to lead to almost identical values for the parameters for diffusion and turbulence with those found by Sutton.

Based on Sutton's theory, equations are given for the deposition of spores at various distances from a point source, and from Stepanov's data it is concluded that in travelling across 1 sq. cm. of surface there were deposited the equivalent of the number of spores present in a layer on the axis of the cloud about half a millimetre thick. This value is expressed as a coefficient of deposition, p, and it is regarded as a parameter of considerable biological significance.

The relevance of the deposition formula is discussed. Observed gradients of air-borne plant infections originating from a point source are shown to be closely predicted by this theory. Fungi known or suspected of being splash-dispersed on the contrary show gradients incompatible with the theory. Gradients from strip sources cannot be dealt with satisfactorily at present but an approximate formula is given, and observed gradients from strip sources are found to show reasonable agreement.

The significance for plant hygiene of this interpretation of fungus spore dispersal is that, while attention should be paid to isolation, most emphasis should be placed on eliminating foci of disease within a crop.

98. MOORE, F. JOAN. 1945. A comparison of Fusarium avenaceum and Fusarium caeruleum as causes of wastage in stored potato tubers. Ann. Appl. Biol., 32, 304-09.

Fusarium avenaceum is reported for the first time as a cause of rotting of potato tubers in Britain. The progress of rotting in tubers infected with F. avenaceum has been compared with dry rot due to F. caeruleum in the laboratory, clamp and potato store. Of the four varieties, Majestic, King Edward, Doon Star and Arran Pilot, tested for susceptibility, King Edward was the most susceptible to F. avenaceum and Doon Star to F. caeruleum.

Optimum temperatures for growth on potato-dextrose agar were 20-25° C. for F. avenaceum and 20° C. for F. caeruleum; maximum temperatures were >30° and 30° C. respectively. For infection of wounded potato tubers, cardinal temperatures for F. avenaceum were similar to those on agar, but for F. caeruleum the optimum was  $15^{\circ}$  C. and the maximum  $25^{\circ}$  C. The optimum temperature for rotting tended, with both species, to be higher in the more susceptible potato varieties. At low temperatures F. caeruleum caused quicker rotting than did F. avenaceum, even though its rate of growth on agar was scarcely more than half that of the latter.

High humidity favoured rotting especially by *F. avenaceum*; *F. caeruleum* was more tolerant of relatively low humidity. Both species caused quicker rotting in the clamp than in store, even though there was no appreciable difference in mean temperature between the clamp and the store. This was attributed to the higher atmospheric humidity in the clamp.

## 99. ROBERTS, F. M. 1943. Factors influencing infection of the tomato by Verticillium albo-atrum. Ann. Appl. Biol., 30, 327-31.

Infection of tomato plants by *Verticillium albo-atrum* was encouraged by application of nitrogenous manures. Application of phosphate had no significant effect on the progress of the disease, but a deficiency of potash tended to encourage it. Steam-sterilized soil inoculated with *Verticillium* immediately after treatment produced a very high total of infected plants. When inoculation of the steamed soil by *Verticillium* was delayed for 17 days or longer after steaming, the steamed soil. Spread of *Verticillium* from the roots of an infected plant to those of neighbouring healthy plants was hastened by killing the infected plant.

#### 100. ROBERTS, F. M. 1944. Factors influencing infection of the tomato by Verticillium albo-atrum. II. Ann. Appl. Biol., **31**, 191-3.

Tomato plants receiving adequate supplies of mineral nutrient acquired great resistance to infection by *Verticillium albo-atrum* if the leaf-shoot ratio was reduced; this effect is attributed to reduction in carbohydrate content of the host. Wide variation in potash manuring did not affect susceptibility of tomatoes to *Verticillium*.

#### 101. SADASIVAN, T. S. 1939. Succession of fungi decomposing wheat straw in different soils, with special reference to Fusarium culmorum. Ann. Appl. Biol., 26, 497-508.

A study has been made of the sequence of fungi developing on wheat straw buried in four arable soils, an allotment soil, and a glasshouse compost. Both natural untreated straw, and straw autoclaved in a 2 per cent. solution of sodium nitrate were employed. Fusarium culmorum and Mucor spp. appeared to be dominant organisms in the earlier stages of straw colonization, but these were replaced by Penicillium spp. in the latter stages of decomposition. The nitrogenous treatment of the straw favoured the development of Penicillium spp. at the expense of Fusarium culmorum and Mucor spp. The pathogenicity of the Fusarium culmorum isolates to wheat seedlings was established by inoculation experiments.

The data provided by this investigation are considered sufficient to justify the inclusion of *F. culmorum* in Reinking & Manns' (1933) group of *soilinhabiting Fusaria*, or true soil fungi.

#### 102. SAMUEL, G. and GARRETT, S. D. 1945. The infected root-hair count for estimating the activity of Plasmodiophora brassicae Woron. in the soil. Ann. Appl. Biol., 32, 96-101.

A method is described for estimating the relative numbers of *Plasmodio-phora brassicae* spores germinating in different soils, by counting infected root hairs. Cabbage seedlings are grown for 1 week at  $25^{\circ}$  C. in glass tumblers of the infected soils; after washing out, the tap roots are stained in 1 per cent. aceto-carmine and a count is made of the number of root hairs containing zoosporangia of *P. brassicae*, along 2 cm. of root. It is shown that by this method it is possible to study, for example, the action of different bases in inhibiting root-hair infection; the main inhibiting factor was found to be soil alkalinity, however produced. Other factors were found to influence infection in lesser degree; thus the number of infected root hairs was reduced in soils receiving N/10 sodium and potassium chlorides in place of distilled water. Root-hair infection was also inhibited by low soil-moisture content.

### 103. SHEN, C. I. 1940. Soil conditions and the Fusarium culmorum seedling blight of wheat. Ann. Appl. Biol., 27, 323-29.

The influence of soil conditions upon the Fusarium culmorum seedling blight of wheat has been studied by means of the glass tumbler technique. Intensity of infection increased with density of the spore suspension used to inoculate the seed, but was reduced by germinating the seed for 48 hrs. or more before inoculation. Infection was most severe at a low soil moisture content and in acid soil. In nutrient sand culture, infection was highest in

the no-nutrient series and lowest in that receiving full nutrients. Nitrogen alone appeared to be as effective in reducing infection as the full nutrient solution, phosphorus was less effective, whilst potassium had no beneficial effect at all upon seedling resistance.

Outbreaks of take-all in oats have often been reported in recent years from Wales, and occasionally from Australia, Denmark and Holland, although in most parts of the world it is commonly held that oats resist the disease. Isolates of *Ophiobolus* from oats grown in Wales were indistinguishable in cultural behaviour from *O. graminis*, but they were very pathogenic to oats, which were found to be highly resistant to ordinary *O. graminis*. The histology of the infection of oat plants by the fungus from Wales and by *O. graminis* was studied in detail. It was found that there were significant differences between the two groups of isolates in the length and

septation of the ascospores, the Welsh material giving a length of  $101-117\mu$ , the English material  $79-86\mu$ . The fungus from Welsh oats is therefore regarded as a new variety

Ophiobolus graminis Sacc. var. Avenae E. M. Turner.

#### 105. WALKER, A. G. 1941. The colonization of buried wheat straw by soil fungi, with special reference to Fusarium culmorum. Ann. Appl. Biol., 28, 333-50.

A study has been made by Sadasivan's (1939) technique of the colonization by fungi of wheat straw buried in the soil. One-inch lengths of sterilized straw were buried in the experimental soils in 31-in. flower pots; after incubation at laboratory temperature (16-20° C.) for the required period the straws were washed out of the soil, surface sterilized by mercuric chloride or other sterilizing agent and plated out on acidified potato-dextrose agar (pH 5.0).

Fusarium culmorum and Penicillium spp. were numerically the most important organisms developing from the straws on the plates, at least during the first 5 months of incubation in the soil. Both groups of organisms, together with others, appeared generally to be present in the decomposing straw, but the method of surface sterilization employed apparently decided which organism produced a colony on the isolation plate.

Fusarium culmorum, a fungus of a vigorous and rapid habit of growth, showed low resistance to the action of the more severe sterilizing agents, such as mercuric chloride and silver nitrate, but developed better after surface sterilization of the straws with calcium hypochlorite, a mild sterilizing agent, and best of all after a mere washing in sterile water. Penicillium spp. were apparently crowded out by the more vigorous growth of Fusarium culmorum after these mild treatments of the straws ; on the other hand, they were very tolerant of the more severe surface sterilizing agents, mercuric chloride and silver nitrate, and after the longer period of treatment were often the only organisms developing on the plates.

The pathogenicity to wheat seedlings of the isolates of F. culmorum obtained from decomposing wheat straw was shown to be comparable with that of isolates of the same fungus secured from diseased cereal plants.

TURNER, E. M. 1940. Ophiobolus graminis Sacc. var. Avenae var. N., as the cause of take-all or whiteheads of oats in Wales. Trans. Brit. Mycol. Soc., 24, 269-81. 104.