PLANT VIRUSES AND VIRUS DISEASES

T. E. T. BOND

Disease in plants, as in animals, can be due to a variety of causes and in tea there are as many types of disease as in most other plants. A primary distinction is between those which are due chiefly to faulty environment and are *non-infectious* and those which are associated with the presence of a parasitic fungus or other organism and which are to a greater or less degree *infectious* or *contagious*. Of the former group, the "bitten-off" disease of tea seedlings is a good example. This disease, as Gadd [3] has shown, is due to adverse soil conditions, usually either excessive alkalinity or waterlogging. The actual destruction of the taproot to produce the "bitten-off" symptom is doubtless the work of micro-organisms which are favoured by these conditions in the soil, but, nevertheless, we are justified in regarding the disease as of primarily physiological origin. It is not infectious, and affected seedlings, if removed in time and transplanted to well-drained acid soil, will produce new roots and will grow into perfectly healthy plants. The various nutritional deficiency diseases, some of which were dealt with in the previous volume of *The Tea Quarterly* [1] are an even better illustration of this type of disorder, where the cure may consist in a simple application of fertiliser containing the deficient element. The *infectious* diseases are well represented in tea and most planters are only too familiar with at least some of them—*Poria* root disease, *Cercosporella* leaf spot, eelworm, and *"Phloem necrosis"* are representative examples. In all of these, infectiousness implies an infective agency which can pass from one plant to another and the science of plant pathology had its origin in the study of these disease-producing agencies. Usually, they proved to be distinct and easily recognisable organisms having a separate existence and life-cycle of their own but obtaining their food materials directly or indirectly from the infected host plant. The great majority of them were found to be parasitic fungi, but many other groups of living organisms (of which eelworms and bacteria are the most important) are known to be capable of producing disease in higher plants. In all of these instances the disease is diagnosed not so much from its external symptoms as from the occurrence of the causal organism within or upon the tissue of the host. But, beginning from the end of last century, and to an ever-increasing extent during recent years, a new class of infectious disease has emerged for which no visible causal organism can be detected, even under the highest powers of the microscope. That is not to say that these diseases are less conspicuous in their effects, or are less easy to recognise from their external symptoms than any others: in fact, the reverse is very often the case and their symptoms can be of a quite startling nature. This new type of disease is the *virus disease* and before attempting any further definition of a *virus* it will be well to have a clearer idea of the diseases themselves.

THE PLANT VIRUS DISEASES

Accepting the position that virus diseases are infectious disorders for which no visible causal agency exists, it follows that for their recognition we are dependent, in the first instance at least, solely upon the symptoms produced in the host plant. There are two principal categories of plant virus diseases — the "mosaic" group, and the "yellows" group.

The first group, of the "mosaic" diseases, is so named because the most characteristic symptom produced on the various hosts is a mottling or mosaic of pale green, yellow, or white areas intermingled with areas of normal, darker green colour. Many of the "variegated" foliage plants commonly grown in mid-and low-country gardens show symptoms of this kind and in some cases, but not all, the variegation may be due to a virus disease. In other plants the mottling is much less pronounced, but may be accompanied by varying degrees of distortion of the foliage. In Ceylon, "mosaic" diseases are commonly seen up-country on ornamental tobaccos (*Nicotiana* spp.) and on French and Kidney beans.

The "yellows" group is perhaps rather less easy to define as a whole than the "mosaic" group. As the name implies, the whole plant may be yellowed, or *chlorotic*, but this is not universally the case. Distortion, or at least modification of the plant's normal habit and appearance, occurs quite commonly, and many of the names of individual diseases refer to this as the most typical symptom. Thus, in Ceylon, the "bunchy-top" of plantains belongs to this group and, in India, the "spike" disease of sandalwood. Another common symptom is a rolling or curling of the leaves of affected plants: thus we have the potato "leaf-roll" disease and "curly-top" of sugar beet. In all probability, *"Phloem necrosis"* of tea in Ceylon will be found to belong to the "yellows" group of virus diseases, and it will be remembered that one of the most characteristic external symptoms of *"Phloem necrosis"* is the dwarfed appearance of the shoots, with the leaves curled backwards and the stems prominently zigzagged. Another Ceylon disease of this type is the tobacco leaf-curl, and there is evidence [4] that the same virus is also responsible for the striking symptoms commonly seen on the goat-weed *Ageratum conyzoides*, ("Pum-pillu," Tamil). Here there is little distortion but a "yellow vein-banding," a clear yellow network following the fine veins. At first sight, this might be mistaken for an intense form of mosaic disease, but the restriction of the symptoms to the immediate vicinity of the veins is a characteristic of the yellows group of diseases.

Despite the occasional resemblance of their symptoms, the "mosaic" and the "yellows" groups of viruses appear to be, biologically speaking, fairly distinct. The two groups, for instance, show important differences in their natural mode of transmission and spread and these will be referred to later. Other types of virus diseases exist which afford a whole range of symptoms — severe distortion, modification of floral into vegetative organs. "witches brooms," etc. — and which show various modes of transmission. They need not be given any separate consideration here.

The economic importance of plant virus diseases would be difficult to estimate: all kinds of crops are affected in all countries of the world and the number of such virus diseases appears to be steadily increasing. Probably, nearly 150 distinct plant viruses have been described. The best illustration of the importance of virus diseases in one of the world's basic food crops is that of the potato industry. All growers at home are aware that potato varieties quickly degenerate if grown for more than a year or two from home-saved "seed" and that for good results Scotch "seed" potatoes must be imported each year. This degeneration is due entirely to a whole complex of virus diseases affecting the potato which, under ordinary conditions of cultivation, cause an increasing loss of vigour in the affected stocks. These diseases, upon the relative freedom from which in upland, wind-swept districts, the Scotch "seed" potato trade depends, have been estimated to cause a loss to the industry as a whole of some £2,000,000 annually.

THE VIRUS: LIVING OR NON-LIVING

So far, the virus itself has been considered only as an invisible agency of disease — rather a negative qualification which is, in fact, all that can be said of a great many viruses: they are recognised as such only because all the other possibilities have been eliminated. Many other viruses, however, particularly those causing diseases of the "mosaic" type, are a great deal better known and, in fact, the study of such viruses has proved to be of great theoretical interest in the discussion of such fundamental issues as the distinction between living and non-living matter

Viruses are studied in *extracts* from infected plant tissues, that is, in the juice or sap expressed from diseased plants, which can be submitted to various methods of purification and chemical treatment. The earliest, and in some ways most fundamental discovery concerning the nature of the virus extract, was that it could be passed through a bacteria-proof filter and still remain infective, hence the term "filterable viruses" which is still frequently used for the group as a whole. While some workers have held that the virus was actually a liquid substance, it is now realised that it is essentially "particulate" in nature, and further experiments have enabled the size and shape of the virus particles to be estimated with a fair degree of accuracy. Some of the particles appear to be spherical, others are rod-shaped. The term "filterable" is now known to be relative only and it appears that some virus particles can be held back by special filters and are consequently not so very different in size from the smallest known bacteria after all. On the other hand, the smallest known virus particles are so incredibly small that they are of the same order of magnitude as the largest known *single molecules*. Bacteria are small enough, but what are we to make of these?

Another striking property of certain virus extracts is their resistance to antiseptics and other forms of chemical treatment, and to heat. The virus will remain infective, in many cases, long after any ordinary bacterium would be killed. Similarly, certain viruses can withstand long periods of drying, and storage under various conditions. One thing that no virus appears capable of doing away from the host plant is to multiply, whether in the crude virus extract or in specially prepared food media known to permit the growth of bacteria or other micro-organisms. Many viruses will stand very high degrees of dilution — for example, an extract of the tobacco mosaic virus is still infective at a dilution of one in a million — yet even so it can be demonstrated that no multiplication of the virus particles has occurred to counteract the effect of the dilution.

In all these ways we have seen the virus in the virus extract — that is away from the host plant — behaving much more like an inert but powerful chemical substance than a living micro-organism. Yet within the host plant, there is a strong suggestion that the virus is actually alive. For if a drop of highly diluted virus extract (perhaps so dilute that it contains only a few individual virus particles) is used to inoculate a healthy seedling plant of a susceptible variety, that plant will begin to show symptoms of infection within some few days and may eventually grow into a large plant which is diseased in all its parts. From this plant others may be infected, and the process may be continued indefinitely, and yet from any plant in the series a virus extract may be obtained of full strength and identical in its properties with the original, undiluted extract. Here is incontrovertible evidence of multiplication, and of multiplication true to type, in a manner which all our experience leads us to regard as characteristic of living matter.

The evidence in favour of the chemical nature of viruses was finally confirmed by the recent discovery that some viruses at least could be obtained in a pure state and actually crystallised. These virus crystals have been shown to be of a complex protein nature and and they exhibit all the properties of the virus: there is now no question but that the virus is really obtainable in this chemically pure form. We are still faced with the question, how does this virus protein reproduce itself so efficiently within the living cells of the host. The answer, if we knew it, would take us a long step forward into that fascinating region of cell physiology which holds the secret of the nature and origin of life itself.

HOW VIRUS DISEASES ARE SPREAD

From what has already been stated of the properties of certain virus extracts — particularly their resistance to ageing and dilution —, and of the rapid multiplication of viruses in their hosts, it will not be surprising to find that virus diseases can be extremely infectious, probably more so than most other types of disease. One of the simplest ways in which virus diseases are spread is by mechanical contact. This applies especially to the "mosaic" group from which infective virus extracts are most easily obtainable. Mechanical contact does not necessarily imply a direct transfer of the virus from a diseased plant to an adjacent healthy one although this frequently happens when the plants are growing so that their branches or leaves are touching. More frequently the contact is effected indirectly, as for instance by the transfer of infected sap on the hands or on knives or similar implements. A better description of this type of transfer would be "sap inoculation." If tea were ever to become subject to a virus disease of this kind, the disease would quickly be spread by pluckers and pruners and it is fortunate that, so far as we can tell at present, *"Phloem necrosis"* is not capable of transmission by sap inoculation. The extreme case with which certain mosaic diseases can be transmitted by sap inoculation is illustrated in a striking manner by some diseases which affect both tobacco and tomato plants. These viruses are so resistant to heat and desiccation that they can survive the curing process of the tobacco leaf and remain infective in the manufactured product. Thus, the viruses can be obtained from most brands of cigarette, and, in commercial tomato cultivation, stringent precautions are necessary to prevent outbreaks of disease through contact with smokers. In American tobacco fields, mosaic diseases can similarly be spread from chewing tobacco.

Perhaps the most certain way of transmitting a virus disease from a diseased to a healthy plant is by grafting, although this is scarcely likely to happen as a regular occurrence in nature. As an experimental method it is invaluable, since it applies to all types of virus disease including some for which no other means of transmission has yet been found. This is the position at present with regard to *"Phloem necrosis"* for example. The graft normally consists of two components, the *stock* which is growing on its own roots, and the *scion* which is grafted on to it. Either component may be diseased, the other, healthy at the time of grafting, becomes diseased later as the virus passes across the graft union.

In nature, the commonest means of spread of virus diseases is through the agency of insects. The insects concerned are said to be *vectors* of the virus or viruses. The insect vectors do not appear to be themselves diseased; they seem to act merely as passive carriers of the virus within their bodies. The position is not quite analogous to the transmission of malaria by the Anopheles mosquito, because in the latter case there is an undoubted organism, the malarial parasite, which is known to pass through certain stages of its normal life-cycle in the body of the insect. With the virus there seems to be some kind of biological relationship with the insect, but of this very little is known. The chief, almost the only, insects which transmit plant viruses are the sucking bugs of which the aphids — "green-" and "black-fly" — are the best known examples. Some of these insects will transmit a single virus only, others are known to transmit a dozen or more. The insect feeds on plant sap which it obtains by means of its delicate, syringe-like proboscis. This is inserted into the plant tissues and saliva is pumped down it : the saliva mingles with the sap and the resultant mixture is then sucked up again into the body. Thus the feeding mechanism of the insect allows it to obtain the virus from infected sap, and to inoculate it again into the tissues together with the saliva. Factors which contribute to the efficiency of insect transmission, especially in the case of "yellows" viruses which are not readily transmitted by mechanical means, are that the virus never comes into contact with the air ; it is obtained with the minimum of injury to the plant and is again introduced into the plant in direct contact with the living cells. On the other hand for the insect to transmit a virus from a diseased to healthy plant, it has to feed for a certain minimum period in each case and usually a so-called "incubation" period has to elapse before the insect becomes infective. The motility of the insect, and its feeding habits generally, will obviously be of prime importance in determining the extent to which it will spread the infection. One interesting feature of the vector relationship is that in some cases the insect appears to differentiate between two viruses, both of which are associated with a given disease. Thus, it may happen that both the viruses are transmissible by sap inoculation but that only one of them is transmitted by the insect, so that different symptoms will be produced on healthy plants according to the method of transmission, and the virus complex can thus be resolved. Many other varied and complicated situations arise in working with insect vectors of virus diseases and there are many problems still to be solved.

PREVENTION AND CONTROL

Virus diseases are usually not transmitted through the seed (the common bean mosaic is an exception to this) so with annual crops that are raised from seed one can at least start with healthy stock. Many of the most important virus diseases, however, occur in crops that are propagated vegetatively or that are perennial, occupying the same ground for many years at a time. The potato is an example of a perennial crop which is raised anew each year : consequently, the use of Scotch seed potatoes as healthy stock is reasonably effective provided that this stock is renewed every year or so. Where the crop remains in the ground for many years at a time, as it does in the case of all woody plants like fruit trees and tea, the use of clean stock to start with would confer very little benefit unless protective measures could be relied on in the later stages. In practice, this is very rarely the case, since complete control of insect vectors would be necessary and this is impossible for plants in the field. It follows that virus diseases are of great importance for perennial crops and especially for those that are raised by vegetative means. Once the source of planting material becomes infected, virus will be present in all the progeny raised from cuttings, offshoots, budding, etc. and the crop as a whole may be faced with virtual extinction. Examples of the spread of virus diseases in this way are to be found in the growing of raspberries and of strawberries at the present time.

Fortunately, however, the control of virus diseases has not proved to be quite so impossible as might be concluded from the preceding paragraph. Most crops which are subject to virus diseases have been found to show considerable differences in susceptibility among their several varieties. These differences are revealed in various ways. For instance, many plants will react differently to virus infection according to the climate in which they are grown and it is quite common for the symptoms of virus disease to be much less noticeable at relatively higher temperatures. Thus, infection may take place and produce severe symptoms in the spring and the disease may almost disappear in the summer, to become more severe again in the autumn. With *"Phloem necrosis"* of tea, for example, it is possible that the relative severity of the disease in the Nuwara Eliya and Kandapola districts compared with, for instance, Talawakelle, may be due largely to the higher altitude and lower average temperature of the former. Again, it happens in some crop varieties that the relative or complete absence of symptoms of infection by a virus may become permanent, so that in the latter case it would normally be impossible to tell whether a plant was infected or not. Such plants, which contain a virus without showing any symptoms of disease, are known as "carriers" and with *"Phloem necrosis"* it has recently been shown[2] that the apparent absence of the disease in *high jat* supplies may, in fact, be due to this cause. In tea, the use of "carrier" varieties will quite likely prove to be of value in eliminating the effects of *"Phloem necrosis,"* but it is easy to see that there are dangers inherent in this procedure and, in some crops, "carriers" are regarded with suspicion as hidden sources of infection to other healthy plants.

Finally, the plant's reaction to the virus may take the form of an inability to become infected at all, and this condition, which is described as resistance or *immunity*, is the most desirable of all. We do not know yet whether it exists in tea, but in crops as diverse as sugar-cane, strawberries, and cotton; resistant varieties have been obtained by selection or by breeding and a very effective measure of control of virus diseases has become possible. It seems likely that the best hope for the future lies in work of this sort, which is being actively pursued in many countries.

CONCLUSION:

In the foregoing, a selection only of the many interesting aspects of the study of plant virus diseases has been possible. The importance of these diseases for world agriculture needs no emphasis and for the study of biology as a whole they are also of great theoretical significance. No review of the plant viruses would be complete without mention of their relationship to the viruses which affect man and animals: small-pox, influenza, rabies, foot-and-mouth-disease — all of these and many others are caused by viruses which have various features in common with those affecting plants. Those who wish for further information on the subject of virus diseases as a whole should read Smith's[5] recent book — "The Virus: Life's Enemy": the title is probably not without justification.

REFERENCES

(1). Bond, T. E. T.—Deficiency diseases and the role of the "minor elements" in plant life—*The Tea Quarterly* Vol. 16, pp. 9-15 (1943).

(2). ——— Annual Report for the Year 1942—*Tea Research Institute Bulletin* No. 24 pp 25-27 (1943)

(3). Gadd, C. H. —"Bitten-off" disease of tea seedlings—*The Tea Quarterly* Vol. 13, pp. 54-58 (1940)

(4) ——— and Loos, C. A.—A virus disease of *Ageratum conyzoides* and Tobacco—*The Tropical Agriculturist* Vol. 96, pp. 255-264 (1941)

(5) Smith, K. M.—"The Virus: Life's Enemy" Cambridge (1940)