

relatively inexpensive, but until the cellulose plastics can be manufactured cheaper the application is limited to special uses where other plastics cannot be used. There is, however, a great possibility for this type of moulding and new developments may be looked for.

The manufacture of plastics is essentially a chemical industry and has thus followed in most cases routine chemical methods of manufacture. The utilization of plastics in various other industries is more allied to engineering than chemistry. On this side more ingenuity and development along specialized lines have been shown. While the hydraulic press has been available for many years the type and construction and general layout has been radically changed to meet the needs of the plastic moulding industry. There is, however, room for more development on the lines of continuous

operations to cut out the large amount of handling that is still required. There has been very little real novelty in design of finished plastic products. The industry has grown so rapidly that plastics in most instances have only substituted some other material. The past rate of development cannot be expected to continue on these lines.

Plastics can now be obtained in so many forms and colour. Celluloid in sheet and moulded casein likewise is procurable in very beautiful colours. Synthetic resinoids are available in every conceivable type of moulding of dark, warm hues to pale colours, transparent or opaque, in sheet form or panelling, constructional, electrical and mechanical uses, while the cellulose plastics show promise of development into channels where the others cannot reach, such as for pens, pencils, small articles in colour effects quite characteristic of their own.

### Virus Diseases of Plants.

AT a Joint Session of the Sections of Agriculture, Chemistry and Botany of the Indian Science Congress, a symposium on "Virus Diseases of Plants" was held during the Science Congress Week at Patna. Dr. S. L. Ghose presided over the meeting.

The subject was discussed under the following heads:—

#### Symptoms.

Morphological. [*M. Sreenivasaya.*]

The reaction of the plant to virus invasion manifests itself in diverse morphological transformations. The plant organ that is most easily susceptible and suffers the most distinctive and perceptible change, is the leaf; next comes the other aerial parts of the plant while the root is, generally, not visibly affected in the earlier stages of the disease, although this is affected in advanced stages. Symptoms at the root, however distinct and infallible, do not afford us a ready and easy method of diagnosis. It is fortunate, therefore, that the leaves of the plant are the first to suffer and exhibit the symptoms in a very decisive manner. We are thus in a position to recognize the malady at comparatively an early stage.

The symptoms generally appear with the vegetative growth and development of the affected plants; they rarely occur in tissues that are mature before infection. "Spike" of sandal appears with the bursting of dormant buds or at the growing points, as the case may be, and the general manifestation of the disease in forests synchronises with a season when the sandal plant passes through its vegetative phase of development.

Virus diseases are usually systemic in character but in the case of the higher woody plants, like sandal, the infective principle is confined to certain

tissues and portions of the plant in the earlier stages of the disease. One-sided involvements with one-sided response to the effects of virus invasion, is not uncommon. Sandal spike and peach yellows afford striking examples of this phenomenon.

Mottling, chlorosis, necrosis, proliferation, dwarfing, curling, crinkling or corrugating, bunching, phyllody and sterility are some of the main symptoms which characterise virus-affected plants. The symptoms vary with the variety of the host affected. The leaf roll of potato, for example, which affects a large variety of potato plants, is accompanied in the case of the "President" variety by a pinkish discolouration of the leaflets. Most of the virus diseases stimulate starch production in affected tissues and this deposition lends the tissues a characteristic stiffness and brittleness.

Mottling is characteristic of the several mosaics, curling and crinkling, of the several leaf rolls and streaks, while dwarfing or stunting appears to be a general effect of all the virus infections. In some instances the "dwarfing" is so intense and general that the new flushes of growth result in "bunching" which is to be found, not only in sandal "spike" but also in the so-called "spikes" of *Vinca rosea*, *Z. oenophia*, *Dudonea viscosa* and a number of other plants.

The "reproductive activity" of the plant is more or less affected in the case of virus-infected plants. The fruits become very small as in the case of cucumber mosaic, while in yet other instances the infection causes a complete sterility as in the case of sandal spike. In some cases the flower retrogrades to the leaf stage resulting in "phyllody"; this phenomenon is to be found in sandal and *Vinca rosea* spike.

The root systems of diseased plants are stunted; whether this is due to virus infection or to the diminution of nourishment from the top which



results from a congested, starch-clogged transport system, has yet to be determined.

Finally, attention should be drawn to cases of plants which do not show the symptoms in spite of their being infected. Symptomology of such disease-masking plants is not yet developed. In view of the fact that these constitute effective sources of infection, their detection with a view to their eradication is of paramount importance. Methods for detecting these plants, with reference to sandal spike, are now being developed at the Indian Institute of Science, Bangalore.

Cytological. [M. J. Narasimhan.]

During recent years, investigations on the cytology of plants affected by virus have shown that certain changes occur in the virus-affected cells. (i) The chloroplasts in the diseased cells have been found to undergo disintegration and in some cases have been reported to be loaded with starch, as in potato leaf roll and spike disease of sandal. (ii) In the case of the potato leaf roll, the phloem cells have been found to suffer from necrosis, involving the collapse of the cell walls, and affecting the function of the sieve-tubes. Phloem necrosis has been observed only in very few cases. (iii) The claims of some investigators that they have detected flagellate organisms in the tissue of the affected plants, have not been confirmed by other investigators. (iv) Cytoplasmic inclusions associated with animal virus diseases, such as rabies, fowl-pox, etc., which have been regarded to be of diagnostic value have been observed in some of the plants affected by virus. The nature of these inclusions was discussed, illustrating them with reference to the inclusions found associated with the spike disease of sandal, a serious virus disease prevalent in South India, especially in Mysore. The analogy of the inclusions found in sandal spike, with the Bollinger bodies of fowl-pox, in regard to staining reactions was pointed out.

### Transmission.

Vector Transmission. [T. V. Subramaniam.]

It has been definitely established that insects play a very important part in the transmission and dissemination of virus diseases of plants.

The biological study of the subject has received considerable attention in foreign countries by numerous workers so that at the present day we have to record about 50 plants belonging to 27 Natural orders—some of them being of very great economic importance like sugarcane, potato, tobacco, etc.—known to be subject to virus attack of which many have been found to be transmitted by insect agency.

Among the insects noted as transmitting virus diseases in plants, insects with suctorial mouth-parts have been found to be more concerned than those with biting mouth-parts. Among the former, the family *Aphididae* occupy the foremost rank as vectors of virus diseases with 27 to their credit.

A number of salient facts, viz., the incubation period, period passed by the virus principle in the insect vector, presence of the inclusion body in the insect vectors, inheritance of the transmitting power by the insect vectors, length of time necessary for the insects to feed before they are capable of transmitting a disease, production of different symptoms when a virus is transmitted

by insects and inoculation, etc., have to be considered in this connection.

Very little work has been done in India, excepting the work of Uppal showing the close association of the mosaic of (Chillies) *Capsicum* and Thrips.

In connection with a study of sandal spike, the sandal fauna of the spiked and healthy areas has been worked out. Attempts to find out the insect vector or vectors of the disease have not been a success as yet.

Artificial Transmission. [M. Sreenivasaya.]

Sap injection or inoculation and tissue transplantation are the two methods employed for transmitting virus diseases of plants. In many cases, e.g., sandal spike, peach yellows, bunchy top of bananas, etc., sap injections have not transmitted the disease. A living support or carrier is perhaps necessary for maintaining the infectivity of the causal entity which appears to be rendered innocuous once it is freed from the live tissue. This is analogous to certain enzymes like *glycolase* which do not function when they are extracted from their associated tissues.

Tissue transplantation includes scion grafting, budding, patching and leaf insertions; it involves the insertion of a piece of infective tissue, freshly derived from a diseased plant at the susceptible region, in a manner that it will intimately fuse with the operated stock.

In transmission work, a due recognition of the susceptible region of the stock and infective tissues of the diseased plant is necessary. Ignorance of these facts have been responsible for the many failures of disease transmission cited in literature. If the infective material sap or tissue is not introduced into the susceptible region, the disease will not be transmitted. In the case of sandal spike, the disease cannot be transmitted if the infective tissue is transplanted at the root or into the wood, while successful transmission can be effected when the infective material is transplanted between the bark and the wood.

The technique of disease transmission in vogue is very crude as compared with those natural to vectors. Needle inoculation which represents the nearest approach to injection by aphids is still unsatisfactory. The natural intake of sap by the aphid is selective; laboratory preparations of sap constitute a mixture of various cell saps. The vector is further capable of injecting the infective sap into the susceptible region. The laboratory methods do not lend themselves to these requirements and sap injection in many cases are therefore unsuccessful, assuming for the moment that the role of the vector is to effect a mere mechanical but selective transfer of the infective sap.

An interesting case of tissue transplantation employed for disease transmission is that of leaf insertion in the case of sandal spike. Here is an instance of how by a judicious choice of the most infective tissue and the highly susceptible region, disease transmissions could be achieved with a very high percentage of success.

Reference may be made to root transmissions effected in the case of sandal plants where mutual parasitism is very common. A curious instance of a sandal plant which had made its connection with a grafted sandal plant getting spiked before the latter exhibited the symptoms, has been noted.



### Properties.

#### "In Vitro" Cultivation of the Virus Principle. [S. V. Desai.]

It was found that the virus of sugarcane mosaic does not act as bacteriophage on organisms associated with the pest or present in the soil. Attempts to isolate organisms from diseased tissues on special nutrient agar, resulted in the growth of transport plaque-like colonies of a pleomorphic character. Healthy tissues similarly treated did not yield any organism. Subsequent culturing on agar and in broth did not affect their morphological characters. It was not found possible to free the organisms from the bacteriophage supposed to be associated with them.

A suspension of the organism was treated with a small inoculum of the virus, and incubated for five days. The filtrate obtained after passing the culture through a sterile filter candle was used for inoculating a fresh suspension of young organisms. Several passages of the virus were thus carried out to see if the virus multiplied during the serial transfers.

Inoculation experiments for the reproduction of the disease were carried with the suspension of these serial transfers. The inoculum used contained the virus principle diluted to  $1:10^{16}$ ,  $1:10^{30}$ ,  $1:10^{44}$ , far outside the limits of infective dilutions. The inoculated plants reproduced the original symptoms of the disease showing thereby that the virus principle multiplied *in vitro* either with or at the expense of the organism.

The same type of organisms were reisolated from the plants in which the disease was produced artificially.

#### Bacteria in Relation to Virus Diseases. [N. V. Joshi.]

The failure to get uniformly successful inoculations from organisms isolated from plants affected with some of the more common forms of virus diseases was regarded as proof that the diseases were not of bacterial origin and this has deterred a systematic investigation of the diseases from the bacteriological side.

However, ideas about the life cycle and morphology of bacteria under different cultural conditions have changed and are changing. Not only have many bacteriologists established that bacteria pass through a life cycle and under different conditions may exist in different visible forms, but recently Hauduroy in 1929 and Hadley in 1931, by adopting new methods, have been able to show a filterable stage in the life-cycle development of bacteria, and Swezy and Severin (1930) have shown indications of the existence of filterable forms of bacteria in the beet-root affected with curly top disease and the leaf-hopper that transmits the curly top of beets.

In the bacteriological laboratory at Pusa the mosaic diseases of several plant hosts have been studied (1931 and 1932) and cultures of bacteria from different plants affected with mosaic have been successfully isolated. Inoculation experiments with some of these cultures have been successful in reproducing all the symptoms of the virus diseases. In studying the cultures of these bacteria, the life cycle of the organisms was tried to be followed. After filtering the cultures through I-3 candles sometimes a growth could be observed and the slides were made from the smallest granules

to the usually larger sized organisms. One of the organisms that was being examined happened to be a flagellated bacterium; on staining, the small granules were found bearing flagella in the same way as the larger sized bacteria. This is perhaps the first time that such an observation is recorded. The fact that the growth in the filtrate when inoculated on agar gives the same kind of growth as the original culture shows that there is a filterable stage in the bacteria that have been studied.

There appears to be very little difference between visible bacteria and the viruses. The dividing line between them is one of size only. It is necessary to examine the possibility of the existence of filterable forms of bacteria more thoroughly as one of the possible methods of arriving at a solution of this perplexing problem of the nature of virus diseases.

#### General. [A. V. Varadaraja Iyengar.]

Absence of easily recognizable and usually associated plant pathogens like bacteria, fungi and protozoa in virus-infected plants led to the suspicion that the diseases are caused by soil deficiency: the effect of fertilizers was therefore investigated by earlier workers in the case of peach yellows, tobacco and tomato mosaics, and sandal spike, but without any significant results.

Moisture and ash contents of diseased plants are generally lower than those of healthy, thus pointing to a poor absorption of the essential nutrients from the soil. Among the ash constituents, lime is conspicuously low in the diseased leaves of peach and sandal. This deficiency could not be traced to the soil.

Abnormal starch accumulation appears to be a general characteristic of all virus-infected plants, e.g., tobacco mosaic (Wood and Hunger), potato leaf roll (Esmarch), spinach blight (True and Hawkins), false blossom of crawberry, peach yellows and sandal spike. A detailed study of the starch content of spike leaves of sandal has shown that it increases with the progress of the disease. In early stages, however, the twigs have been found to contain more starch than leaves. Esmarch ascribes this phenomenon to defective translocation of photosynthetic products of the leaf. Neger, in his study of potato leaf roll could not correlate the degree of rolling with starch content.

Significant conclusions cannot be drawn with regard to the other carbohydrates occurring in the diseased condition.

A physico-chemical study of the tissue fluids in the case of sandal spike and curly top of sugar beets, has been carried out.

#### Nitrogen Metabolism. [Y. V. Sreenivasa Rao.]

Significant changes are brought about in the nitrogen metabolism of all virus-infected plants. In his studies on the mosaic of sugar beets, Bonequet drew attention to this aspect of the problem. Nitrites and ammonia nitrogen were found to occur in larger quantities in the diseased leaves. He concluded that the internal bacterial flora were responsible for the reduction of nitrates to nitrites and ammonia; these results found confirmation in his work on potato mosaic. A systematic study of the nitrogen distribution in the tissues of healthy and blighted spinach was carried by Jodidi, Kellog and True, who found higher



concentrations of nitrite and ammonia in the diseased condition.

In the case of sandal spike, however, a study of the nitrogen distribution has revealed (a) an increase in the water soluble nitrogen, (b) a higher concentration of the basic fraction, and (c) an increase in the amino-nitrogen. Further analysis of the basic fraction has shown an abnormal amount of histidine in the diseased condition, leading to the significant suspicion that histidine is getting decarboxylated with the production of histamine, a compound known to inhibit the growth of roots. The low percentage of arginine in the basic fraction in spiked leaves is equally noteworthy, as it explains the general suppression of reproductive activity with the progress of the disease.

#### Enzymes. [B. N. Sastri.]

Significant disturbances in the enzyme make up of plant tissues accompany the onset of virus diseases. It was believed at one time that the causal entity of virus diseases is of the nature of an enzyme, a theory which has recently been supported by the work of Vincent on tobacco mosaic.

Diastases and oxidases are the two groups of enzymes that have received wide attention; an increase in diastatic activity has been found in the diseased leaves of sandal, and *Dudonea viscosa*. High oxidase and a low catalase activity have been recorded for most of the mosaics. The curly top and curly leaf of beets also exhibit a high oxidase activity.

The disturbance in the enzyme balance has been brought about by subjecting the organism to unfavourable environmental conditions. Drought and over-watering, for example, have been found to develop abnormally high oxidase activity. Starvation of cells has been found to stimulate the development of enzymes.

High diastase activity, optimum reaction and lower moisture content of the metabolic fluids are significant factors influencing rapid synthesis of starch in the diseased leaves. The occurrence of the liquefying component of the diastase in low concentration hinders the rate of translocation of the products of photosynthesis leading to an abnormal accumulation of starch in the diseased tissues.

The high percentage of amino nitrogen present in the tissue fluids of the spike leaf, suggestive of the enzymatic degradation of the leaf proteins, points to a greater activity of proteases in the diseased tissues. As in the case of other virus-affected plants, a high oxidase activity is shown by the tissues of spiked sandal. Respiration studies show that a high oxygen intake and low carbon-dioxide output are characteristic of diseased tissues. It is, therefore, clear that the oxidation-reduction mechanism is upset in the pathological condition and this explains the occurrence of mannitol and accumulation of organic acids. The discussion centres round the concentration of respiratory pigment cytochrome. An insufficiency of this favours fermentative processes leading to

the formation of alcohols. Incomplete oxidation of sugars results in the production of free acids which accumulate in the tissues owing to the lack of sufficient quantities of lime for neutralization.

#### Control Measures. [A. V. Varadaraja Iyengar.]

Control measures consist of (1) introduction of resistant varieties, (2) removal of infection centres, and (3) elimination of the carriers of infection.

The existence of resistant varieties is known and can be evolved either by selection or hybridisation. In the case of parasitic plants like sandal the possibility of building up resistance through host plants may be indicated.

The basic control method must aim at the eradication of the sources of infection. This requires a decisive method for diagnosing the disease which, in the case of sandal, is complicated (1) by the fact that the plant under certain environmental conditions exhibits symptoms analogous to spike, and (2) by the presence of disease-masking plants. The symptomology of these two classes of sandal plants are being worked out.

In practice a complete eradication of the diseased parts of the plant necessary for the control of infection is difficult and expensive, but this is the only method adopted in the case of the diseases of the peach and the leaf roll of potato. A similar method has been adopted in the case of sandal spike but with this modification that the plant prior to its uprootal is treated with an arsenical preparation, "Atlas", which has greatly facilitated the operation.

Eradication of wild hosts of the same species has been found necessary in the case of the cucumber mosaic.

#### Discussion.

Referring to the studies of transmission by insect vector, Mr. A. V. Varadaraja Iyengar spoke on the work that is being carried out at the Indian Institute of Science, Bangalore, by Mr. Cedric Dover and his collaborators in connection with the spike disease of sandal. Elaborate work on the sandal fauna, although forming a unique contribution to South Indian Entomology, has not yet yielded any positive result by way of establishing the vectors.

Dr. Gilbert J. Fowler speaking on the work carried out at Pusa, pointed out that the danger of organisms growing through the filter candles is great unless scrupulous attention is devoted to aseptic conditions of experimentation. This factor is generally overlooked.

Mr. B. N. Sastri was of opinion that the bacterial nature of the virus principle can be disputed on the basis of its resistance to several powerful cytolytic agents such as toluene and acetone.

Prof. R. H. Dastur, referring to the physiological studies, said that all the observed results are consequential to the virus attack and inasmuch as they do not give any clue as to the cause they are only of limited value.

B. N. SASTRI.

M. SREENIVASAYA.