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Proceedings of a meeting of the Society held on Thursday, 19th January, 1922, at 8.15 p.m., at 11, Chandos Street, Cavendish Square, W. 1, preceded by a Demonstration at 7.45 p.m. on "Some Stages in the Development of *Trypanosoma lewisi* of the Rat, by Dr. A. C. STEVENSON.

Sir JAMES CANTLIE, K.B.E., F.R.C.S. (*President*), in the Chair.

SOME RELATIONS BETWEEN VEGETABLE AND
HUMAN PATHOLOGY.

BY

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The differences in the organisation of the higher plants and animals are such—especially in the absence of true circulatory, nervous and digestive systems in plants—as to preclude any comparison between the great mass of the disorders from which the two kingdoms suffer. But since, in their ultimate structure, they are composed of the same cellular units, and since they are exposed to the attacks of similar parasites, it would be strange if there were no points of contact between vegetable and animal pathologists.

Vegetable pathology is a younger science than that of man and the higher animals, and there are, as yet, no handbooks that give in any detail a comparative survey of the pathological processes in plants.

But, I think, there are grounds for hoping that the older science may yet gain something from the study of certain disease phenomena in plants. The directions in which this may be sought are in certain branches of cellular pathology; in the diseases involving changes in physiological processes, such as altered metabolism; in certain classes of parasitic and virus diseases; and in the effect of environmental conditions on the interaction of host and parasite.

I propose to give only a very few instances of disease processes in plants that have some analogy to those in the animal kingdom, but without attempting to define their exact bearing on particular aspects of human pathology. While they are not specially confined to the tropics they are prevalent there, and my own actual experience has been almost entirely tropical.

The first is tumour formation in plants, as exhibited by the well-known crown gall tumour caused by *Bacterium tumefaciens*. I have recently had an opportunity of going over with Dr. ERWIN F. SMITH, of Washington, the work on this disease on which he has been engaged for the last eighteen years. It is regarded by plant pathologists, the world over, as one of the masterpieces of scientific work in their field, and has raised Dr. SMITH to the position of a recognised leader amongst them.

The tumours produced by this organism on a multitude of plants are usually hyperplasia caused by the repeated division of the cells to produce a solid small-celled tumour tissue without cavities. The cells are often markedly embryonic in character, and divide mitotically and amitotically. Secondary tumours frequently develop, often at a considerable distance and deep-seated in the tissues. They are not caused by migration of the bacteria across normal tissues, but by a definite outgrowth from the primary tumour in the form of a tumour-strand of infected cells of marked characters, and easily recognised in section, though often only a few cells thick. Such strands have been traced for ten inches in length. The secondary tumours have the characters of the primary, so that if a leaf bears tumours from one on the stem, their structure will be a stem structure. Secondary tumours can also be produced by grafting a part of a primary tumour on a suitable part of the plant. Growth of the tumour is unlimited and devoid of polarity, and the destructive action on the plant is usually merely the result of pressure and crushing, though necrosis permitting secondary infection is common.

The structure of the tumour varies according to the tissue primarily infected. Most of them are of the ordinary ground or conjunctive tissue cells, *e.g.*, in the cortex or pith, and are the nearest approach that one could expect to get in plants to Sarcomata. There is no epithelium in plants, and the epidermal tissues are thin, usually a single layer, and hard to inoculate without involving the underlying ground tissue. But a few cases have been described in which the tumour appears to be composed mainly of epidermal elements, including hairs (the plant hair being an outgrowth from a single epidermal cell), and these may be compared with Epitheliomata. The most interesting type, however, is Embryomata. These were first detected as recently as 1916. They are composed of a jumbled mass of young shoots and roots of usually incomplete structure, having only bits of organs, but sometimes forming more or less complete dwarf shoots. The secondary tumours developed from these have usually the same embryonic teratoma structure.

The organism which causes crown gall is a cell parasite of a highly developed type, in which the host cell is not destroyed but is stimulated to increased activity. There are many such cases in plant parasites. To the best of its ability the parasite aims at preserving the life of the host cell for its own needs. That it over-stimulates the cells, causing death by crushing and necrosis, may be regarded, more or less, as an accident.

Bacterium tumefaciens can be isolated and cultivated so that its morphology and characters are well known. It is rare in the tissues, at least in an active state; in one case it was estimated that there were only 200 living bacteria per cubic centimetre of the tumour. But perhaps the most significant thing that I learned from Dr. SMITH was that, contrary to his earlier belief, he is not now certain that he has ever recognised the organism in the tissue. What he previously saw he now thinks were only cell inclusions or chondriomes. Thus we have a parasite capable of causing a tumour having all the characters of a malignant growth; the organism has been isolated, grown in pure culture, and new tumours produced at will with it, but it is not with certainty to be recognised in the tissues of the tumour.

More recently Dr. SMITH has been able to produce experimentally small tumours of limited growth, having the characters of incipient crown galls, by injecting in suitable parts of plants certain of the diffusible products of the metabolism of *Bacterium tumefaciens*, including ammonia, amines,

organic acids, etc., and he considers that the tumour is ordinarily caused by the continual production of small quantities of these excreted products by the parasite.

I am aware that the path of cancer research is strewn with the wrecks of parasitic theories; but the recent work on plant cancers, the experimental production of embryonic teratomata, the extreme difficulty of detecting the parasite in the cells (it is certainly *in*, not between the cells), and the evidence that it acts by the production of repeated small quantities of an excreted stimulus, makes it easy to understand the view held by Dr. SMITH, and other vegetable pathologists, that cancer will ultimately be found to be due to some similar cause.

A second direction in which disease phenomena in plants may throw light on certain animal diseases is found amongst the obscure group of diseases which the vegetable pathologists are calling "virus" or "contagium" diseases. These are diseases in which etiological studies suggest the presence of parasitic agent, but in which, with the means at our disposal, no parasite can be detected, and filtration through the Berkefeld filter does not cause a reduction in virulence.

The number of such diseases in plants is large, but many of them have only recently been recognised. They have all certain characters in common, but they fall into several groups, the most important of which is that known as the "mosaic" group, because its most obvious symptom is a variegated appearance of light and dark green patches on the leaves. So rapidly have new cases of mosaic been recorded in the last few years that one is tempted to say that every cultivated plant has at least one such disease. Cross-inoculations show that while allied plants (as, for instance, potato and tomato) may suffer from the same mosaic, the mosaics of distantly related plants are usually distinct.

These diseases are constitutional, the virus reaching all parts of the plant in three to four days after inoculation in the mosaic of cucumber. The incubation period before the first symptoms appear in this mosaic varies from four to fifteen days, and in potato and cabbage mosaic from twenty to thirty days. In sugar-cane mosaic the virus is restricted in its distribution, and may not reach every shoot. Many tissues and organs, though containing the virus, do not react to it in any way that has been hitherto detected. Also "carriers" have been found, *i.e.*, plants that show no symptoms after inoculation, yet contain infective virus in a

transmissible form. Actual tissue reactions (cell modifications) are often caused in the leaves and fruit. Necrosis of the light areas in the leaves also occurs not uncommonly in the older stages. The other symptoms are due to deep-seated alterations in the physiological activities of cells and organs, revealing themselves in altered metabolism and abnormalities of growth (dwarfing, crinkling or folding of flat surfaces, shortening of axes, and, at times, proliferation). The disease is not usually fatal in annual plants, though death from weakness may occur. The reduction in vigour may be very great or slight. Mosaic and allied diseases of potato in England may cause a reduction of over 50 per cent. in the yield, and in Bermuda losses up to 75 per cent. have been recorded. On the other hand, I recently saw fields of sugar-cane in Louisiana with 100 per cent. infection from mosaic, but was informed by the local plant pathologist, EDGERTON, that the loss of yield was slight, though the disease has been present for seven years, and in other localities, *e.g.*, Hawaii, has been known to reduce the sugar yield by 60 per cent. The so-called deterioration of many cultivated plants, such as potatoes, leading to the constant demand for new varieties, has recently been recognised to be, in many cases, the result of the gradual accumulation of some one or other of these virus diseases.

From the standpoint of comparative pathology, what is important is the knowledge that has already been gained as to the methods of transmission and certain other characteristics of these virus diseases.

So far as is known all these diseases are insect borne. There are other methods by which infection can be produced, but the normal method in nature is by insects, chiefly sucking insects, especially aphids. In no case has transmission by mere contact been demonstrated. In one case, curly top of beet, no other method of infection than through the insect (a leaf hopper, *Eutettix*) has been discovered, and in this case the insect has to be kept for a period of twenty-four to forty-eight hours after feeding on a diseased plant before it can inoculate a healthy one, after which it can remain infective for 100 days. This suggests that a part of the life-cycle of the parasite is passed in the insect. In most cases, however, the virus can be obtained by extracting the juice (cell sap) of a diseased plant, and can be successfully inoculated on a healthy plant by injection into any part of the plant (tobacco mosaic) or into certain parts only (growing point in sugar-cane mosaic, any part but the root or flower

in cucumber). It can also be transmitted often by crushing a diseased leaf on a healthy one, so as to injure both tissues, or even (cucumber mosaic) by bringing the broken hairs of healthy and diseased leaves into contact. In such cases the virus is evidently in each individual cell. In spinach blight the crushed bodies of aphids that have fed on diseased plants are infective. In this disease, too, the offspring of infected aphids are infective occasionally. In certain cases the disease is carried in the seed borne by diseased plants, but in a good many this does not occur. There is much evidence that the virus travels in the plant through that part of the vascular system, the phloem, in which the elaborated sap is conveyed. This system does not reach to the seed, the plant embryo feeding entirely by osmosis. It is most interesting to note that recent studies show that certain aphids, including some that are known to carry mosaic, feed by inserting their very fine and flexible probosces into the cells of the phloem, avoiding other cells as a rule.

One other method of transmission must be mentioned. Even in the most refractory cases, as beet curly top, infection can be secured by grafting a diseased on to a healthy plant, provided organic union is obtained.

After extraction from the plant, the life of the virus varies from only twenty-four hours (sometimes in cucumber mosaic) to a year or more (tobacco mosaic if dried). It is killed by a temperature usually of between 70°C. and 80°C., but can withstand exposure to very low temperatures. In some cases the ordinary antiseptics kill it, but the tobacco mosaic is resistant to most weak antiseptics. The concentration in the sap is such that a needle drop can be diluted to 1,000 without any loss of virulence, though virulence is reduced by a dilution to 10,000 (tobacco and cucumber). Filtration through porous filters depends on the size of the pore. The cucumber mosaic virus filters through the Berkefeld but not through the Chamberland; tobacco mosaic filters through both, but is retained by powdered talc.

Though there are still a few vegetable pathologists who prefer the enzymic or toxic hypotheses to the parasitic explanation of these diseases, most agree that their study has made it necessary to assume that they are caused by ultramicroscopic living organisms. The mere fact that they have not been successfully grown *in vitro* has no weight with the vegetable pathologist who is familiar with whole groups of parasitic fungi,

such as the rusts, not one of which has ever been grown apart from its host plant. It is as certain as anything can be that no visible organism is present in extracted cell sap that is known to be highly infective. This does not preclude the possibility that the organism may have a visible stage, and indeed quite recently two independent workers have claimed to have discovered, in cells of sugar-cane and maize suffering from mosaic, bodies which in one case may perhaps be suggestive of a parasite not unlike a stage in *Rickettsia*.

Another set of problems, of no less interest to the vegetable than to the animal pathologist, is the geographical and seasonal distribution of certain diseases.

The experimental study of the relations of plants to their environment is naturally much easier than with human beings. In our botanic gardens we habitually grow tropical plants with success, being able to imitate their natural conditions. It is only recently, however, that plant pathologists have made use of their opportunities to study the influences of such climatic factors as humidity and temperature on plant disease. The results have been remarkable. It has been found possible to correlate the geographical distribution of a considerable group of plant diseases with the temperature and humidity reactions of the parasite and its host.

In the simplest cases a single factor, such as the degree of relative humidity of the air, acts by limiting the development of the parasite. One of the best examples of this is a disease of the rice plant which I studied in India. It was strictly confined to a large rice area at the head of the Bay of Bengal, and while the main crop from July to December was often totally destroyed, the secondary crop in the spring entirely escaped, though it was shown by artificial inoculations to be equally susceptible. I found that the organism could neither move nor feed nor reproduce at an air humidity of below 90°, while, though it is actively mobile, it is unable to feed or reproduce in water. To be able to multiply and attack the rice plant it therefore requires moderately prolonged periods when the air humidity is over 90°. This it only gets in certain limited areas in India during the second half of the year; and at a time when the disease was causing considerable alarm, it was possible to state that the bulk of the rice-growing area was not threatened.

The meteorologic factor may act by affecting the receptivity of the host-plant. Thus in the vine mildew, infection is dependent on the

condition of the stomata, through which the parasite enters. If these remain semi-closed, as in young leaves or in dry conditions, infection is impossible. Infection has been found to fail if the moisture of the soil in which the vine is growing is below 15 per cent., unless the air humidity is above 80 per cent. If the soil moisture is above 20 per cent., the stomata open widely enough to permit infection at air humidities above 40 per cent.

But it would seem that in many cases the action of the meteorological factors on both the parasite and the host in their mutual relations—on what may be, for convenience, termed the host-parasite complex—has to be taken into account. In the onion smut, for instance, it is possible to get 100 per cent., or no successful infections, according to the soil and air temperatures used. The onion seed will germinate at soil temperatures from 10°C. to 30°C. but the best top growth is got with the soil temperature of 20°C. to 25°C. Infection by the smut fungus can be got freely from 10°C. to 25°C. but is slight at 27°C. At a soil and air temperature of 25°C. a great many plants will outgrow the infection and recover, but at an air temperature even as high as 30°C. to 33°C., infection can be got if the soil temperature is kept below 25°C. In outdoor plantings the disease was completely inhibited when the mean soil temperature reached 29°C., and increased progressively with lower temperatures. The relation is believed to act chiefly through the host-plant, as successful incipient infection can be got at the higher temperatures, but the subsequent development of the fungus is inhibited. The geographical distribution of the disease in the United States, where it is absent from large areas in which onions are sown in the warm season, has been completely explained by this work.

Recently it has been shown that the severe outbreaks of epidemic malaria in the Punjab are governed by a set of temperature and humidity factors affecting both the malarial parasite and the insect host. With a warm-blooded animal like man the temperature factor is probably of slight importance, except in so far as it acts on the parasite while exposed to the air, as in the external air-passages. But in insect-borne diseases, and in the life of the parasite outside its host, these studies have a certain interest, and the recent discovery that so small a difference as 5 per cent. relative humidity or 3°C. or 4°C. temperature can cause a difference of 100 per cent. in infection has come as a surprise to many plant pathologists.

In these few instances, selected from widely different fields, I have tried to show that the problems which face the vegetable pathologist are often akin to those of human pathology. Where there is any evidence that disease processes are fundamentally alike in plants and animals, I venture to think that they are worth careful examination by pathologists generally. Their study in plants has many obvious advantages in the ease of experimentation and manipulation.

Vegetable pathology is under a great obligation to the older science, from which many of its methods and principles are borrowed, and there could be no more fitting way of repaying this debt than by the discovery that the study of plant diseases may throw light on some of the more obscure problems of human pathology.

DISCUSSION.

Dr. A. BALFOUR: I think we must all welcome this paper very much. It is rather out of the usual run of papers which are given here, but it is the more interesting and instructive on that very account.

There are two matters to which I would like to make brief reference. The first, which is only indirectly associated with Dr. BUTLER's paper, is one which I know Sir PATRICK MANSON has had in his mind for many years, though I think he has never written about it. He is not here, but I do not think he will object to my mentioning the subject. It is the question, Why do certain plants produce alkaloids? Many of these are very poisonous substances; some are distinctly disagreeable to the taste. Sir PATRICK asked me whether I thought these alkaloids were developed for the protection of the plants, for instance, against insect attacks. In cinchona bark there are, of course, quinine and other bitter alkaloids, and it would be interesting to know if those particular plants are subject to insect-borne diseases.*

The other point has a definite relation to the subject of the paper, namely, the production of gum by plants—a matter of great importance to those living in the Sudan, where gum acacia is one of the staple products. This production of gum is a diseased condition common to certain species of acacia tree, when they occur in dry regions and do not receive sufficient moisture. When the plants are well supplied with water this

* The interesting question of the function of alkaloids in plants is discussed by ANNETT in his "Investigations on Indian Opium," No. 2, in the *Memoirs of the Department of Agriculture in India*, Vol. VI., No. 2, Sept., 1921.—A. B.

gum is not produced. In Australia, the work of GREIG SMITH showed that bacterial infection, in the case of two species of acacia, was responsible for the production of gum from the sugars of the sap. Later, RUHLAND isolated from diseased cherry trees an organism, now called *Bacillus spongivans*, which he cultivated, and which he found produced slime and gums.

Certain of the Ambrosia beetles have been incriminated by ZIMMERMAN as a cause of gum production, owing to the injury produced by their boring habits. In the Sudan, EDIE isolated a bi-polar staining organism from two species of acacia, and he cultivated this in a liquid medium containing levulose, glycerine, asparagin and potassium citrate; when so cultivated the organism produced slime and, eventually, gum. These are all very interesting observations, because it was definitely shown that gum is only formed when the tree is damaged, and a point of inoculation provided for the causative organism. EDIE proved that flies were operative, probably by conveying the bacterium in the gum from one tree to another. It is also possible that ants may be effective. I think this gum production may be a protective action on the part of the plant, and may possibly, in some cases, be analogous to the scab which forms over a wound and, to a certain extent, protects the latter from insects, and from invasion by micro-organisms.

Dr. P. H. MANSON-BAHR: We have evidence of *Bacillus tumefaciens* causing tumours in plants, yet the bacilli could not demonstrate in the tissues. This is a well-marked exception to the second of KOCH's postulates. I think this should give us a lead in our further study of human pathology. In such a disease as sprue it may be possible for an intestinal bacillus to be responsible for the pathological condition, yet that bacillus, though it may be isolated by culture, may not be demonstrable in sections of the gut itself.

Dr. T. P. BEDDOES: I would like to ask to what extent there has been any treatment. When it comes to management of woodlands, certainly an improvement can be made. Take, for instance, one of the most destructive diseases—the larch disease. Instead of growing the larch unprotected in the open, if it is grown among other trees, such as the oak, then it grows in better condition. By management, woodlands can be very much improved in the matter of protection from diseases.

Another question of interest is as to how far lime and manure affect treatment. There is nothing more interesting than to consider the use of lime. It was first introduced by the Romans, and, for many years, England was the centre of the lime trade, the substance being exported to Holland and the Lowlands. Then the industry went out for many years, and at about the middle of the eighteenth century in it came again, and certainly with very great improvement. Thus I am prompted to ask to what extent management—*i.e.*, the promotion of nourishment and similar factors—lessens the injurious conditions which are being discussed.

Dr. J. C. G. LEDINGHAM commented, as a welcome sign of the times, on the increasing evidence of helpful co-operation in problems of common interest between pathologists and bacteriologists in all spheres of microbiology.

The following also took part in the discussion:—Dr. C. F. HARFORD, Mr. A. D. COTTON, Mr. A. L. SHEATHER, Sir PERCY BASSETT-SMITH, and Dr. C. M. WENYON.

The PRESIDENT: We sincerely thank you, Dr. BUTLER, for having taken us over this new ground. I think plant pathology will find a place in the ordinary medical curriculum in the Tropical School, though this new subject may be thought by some to be making confusion worse confounded.

We are very much indebted to Dr. BUTLER and the various gentlemen who have spoken and brought up points of interest; and if each contributes his particular knowledge to the discussion going forward, we shall all learn a great deal.

Dr. E. J. BUTLER: Recently some work has been done in France on the group of so-called alkaloid producing plants, so as to study such substances as nicotine and atropine. In some cases, such as the aconite, it was shown they were definitely resistant to parasites if in concentration. Nicotine was not of sufficiently high concentration in the plant to have an effect on the organism. Certainly, many plants do successfully resist infection, because they have alkaloid in them, though whether the action is definitely protective is more doubtful.

GREIG SMITH has shown gum production to be due to bacteria in a certain group of cases, but, in the commonest kind in this country, a parasite has not been detected; it is apparently a gummy degeneration of the cell wall, and the conditions which bring it out are a rapid alteration in the degree of humidity of the surrounding air. There is a sugar form to which Dr. GREIG SMITH has drawn attention. So far as I know these gums are not protective, and I am speaking of the acacias.

So far as I am aware, no work has been done in the way of conferring protection. In a few cases it has been said that products have conferred immunity to plants, but on careful examination every supposed instance has broken down.

I believe the transmission experiments to which I have referred have been done only on plants of the same species. I think Dr. SMITH has worked that out, but I cannot say definitely.

Replying to Dr. MANSON-BAHR, in artificial inoculation there is no difficulty in recovering the organism from the plant, though it cannot be seen there. Dr. SMITH has repeatedly cultivated the organism after the disease has been produced, though he could not see it while it was in the tissues.

In answer to Dr. BEDDOES, much work has been done on the control of these diseases, but it would take too long now to speak of it in any detail. The tree disease has been shown to be associated with attacks of certain large insects, and if you can keep the wounds down which are so produced, you improve the trees very much. Larch disease is said to occur more on certain soils than on others, and that is so with other groups of plants. There are plants which suffer from having excessive lime, and others which suffer from an insufficiency.

A paper was read by Dr. W. SALISBURY SHARPE, M.R.C.P., F.R.C.S., on "Diseases of the Ear in Relation to Life in the Tropics." This has been published in the *Journal of Tropical Medicine and Hygiene*, February 15th, 1922.