

20. Aetiology of the wilt (root) disease : Investigations on its virological nature

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INTRODUCTION

NO general discussion on the wilt (root) disease of coconuts of Travancore-Cochin, at present seriously threatening the economy of millions of people, could be initiated without a survey of its historical aspect and the earlier work done on this perennial crop leading, step by step, to our present-day knowledge of its aetiology. Being, so far, restricted to the broad belt of coconut tract of Travancore-Cochin, the disease covers nearly one lakh of acres incurring annually, a loss of about 10 million.

The disease was first noticed about 80 years back from three different localities of north and central Travancore. From these scattered and independent foci of infection it started spreading, covering the coastal area as well as the hilly tracts. A slow but steady spread to new, healthy areas has been noticed, while the increased intensity of the disease has been threatening the coconut cultivation in the already affected areas.

Palms of 6 years or more are generally attacked although the most susceptible time is when the tree starts bearing, that is, at about 8-12 years. Rare infections on 3-4 year old seedlings are also met with.

Symptoms

The external symptoms of the disease manifested by the palms vary according to their individual resistance, genetical condition, constitution and environmental conditions. Nevertheless, the characteristic symptoms may be stated to be the abnormal bending of petioles and the flaccidity and ribbing of leaflets accompanied with necrosis and curling of their tips. Small chlorotic streaks on the younger leaves, which later become necrosed, are also met with while yellowing or chlorosis of the outer whorl of leaves are not uncommon. Yellowing and drying of intermittent leaves and drying of the

spathes are also met with in some cases. In bearing palms, on the initiation of the disease, abnormal shedding of buttons sets in, which later on results in reduction in yield. As the disease advances, the leaves get progressively smaller and fewer in number, the marginal and top necrosis of leaflets gets intensified, the dried portions being blown away in the wind. Thus the entire crown gets reduced in size and consequent to the abnormal nut-fall, produces only few under-sized fruits, ultimately becoming to tally barren. Accompanying these foliar symptoms there is a deterioration of the root system, the rotting and drying of the main roots and rootlets from tip backwards, increasing as the disease progresses. Consequent on the restricted output or the total absence of new roots, the absorption of water is impaired.

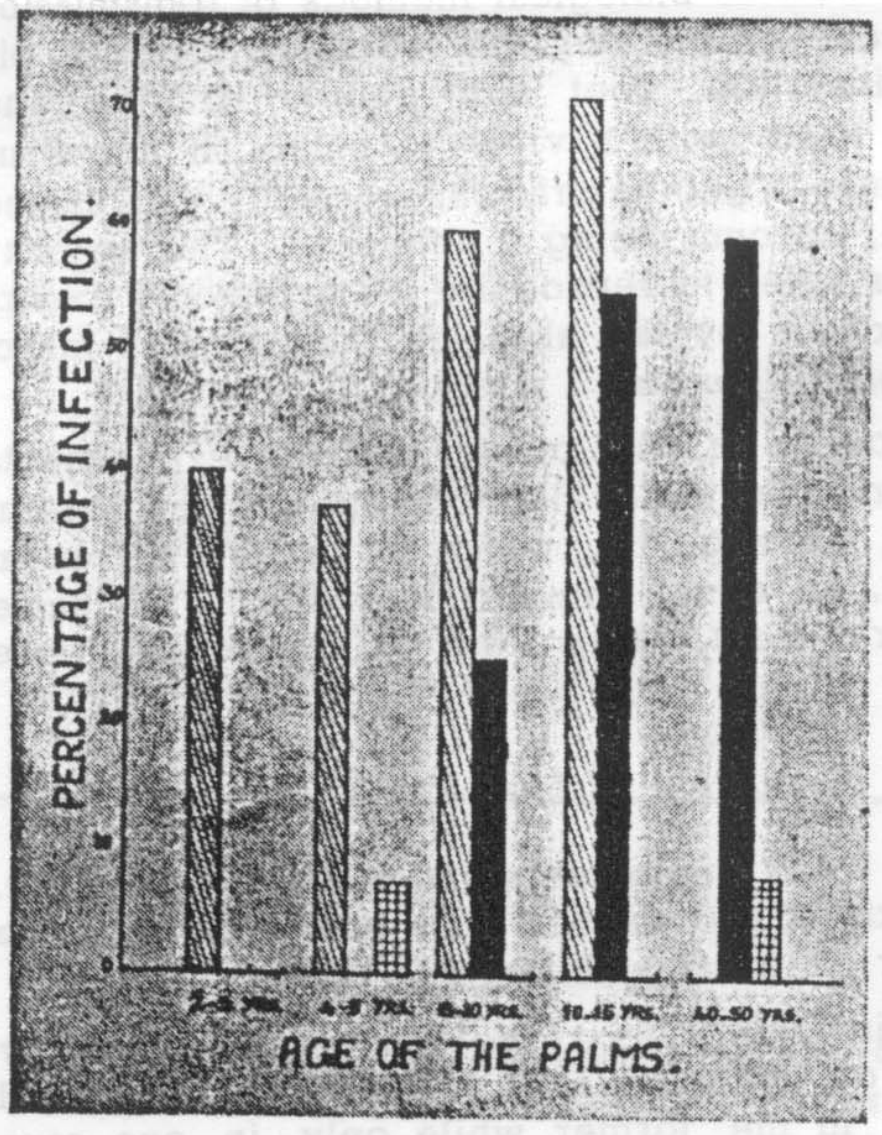
EARLY INVESTIGATIONS




The first serious study on the aetiology of the disease was started when Butler (1908) isolated *Botryodiplodia* sp. from rotted roots and considered it to be the causal organism affecting the roots under adverse conditions of drought and poor soil. Systematic investigations, however, conducted by Menon and Nair (1951) revealed the complexity of the disease caused, perhaps, by a combination of factors. They found that *Rhizoctonia solani*, *R. bataticola* and *Botryodiplodia theobromae* were invariably associated with the rotted roots and in spite of repeated inoculation trials were unable to get the symptoms reproduced on plants more than 3 years old. In view of the cosmopolitan distribution of these fungi and because of their facultative growth habit Radha and Menon (1954) supported the view of the earlier workers that these could be only secondary in nature.

Work done on the nutritional aspect of the disease revealed that no major or minor element deficiency could be a direct cause of the disease. The possibility, however, of any single or multiple deficiency or of a combination of physico-chemical factor (s) as being indirectly responsible cannot be as yet ruled out. One important finding, worthy of particular notice in this connection, is the consistently low nutrition level of soils of all types in the coconut growing tracts where this disease is prevalent. Total calcium and iron, available potassium, base exchange capacity, percentage base saturation and pH of the soil are lower in the disease affected region than in the healthy. All these factors, as one may notice, are inter-related - for instance - low calcium and potassium result in low base exchange capacity and a low percentage base saturation, which ultimately leads to acidity of soil. The normal physiological functioning of the palm, which requires large amounts of potash for a balanced nutrition,

stulsa m...
P...
...
...

Fig. 1
*Showing percentage disease incidence after
sap inoculation & insect transmission.*



-  Sap inoculated
-  Insect transmission
-  Control

is thereby impaired bringing about a predisposing environment for the attack of facultative organisms. It may be pertinent to mention in this connection that Menon and Nair (1951) reported that although no curative treatment was known for this disease, the disease could be kept in check to some extent and the infected trees made to give an economic yield. Citing phenomena like stunting of growth, the characteristic foliar symptoms, the infections and systematic nature of the disease it was suggested that the wilt disease might be of virological origin. Some of the results obtained in connection with the work on this aspect of the problem are described hereunder.

EXPERIMENTAL

Transmission

Both mechanical as well as biological methods of transmission like the inoculation of leaf extract by the abrasion method, pin prick inoculation, leaf insertion, root and petiole grafting and insect transmission were tried with a view to establish the virus nature of the disease. Failure to get the grafts successfully established was only to be expected, coconut being a monocot. The majority of the common techniques failed to reproduce the symptoms except the inoculation of leaf extract by the abrasion method and insect transmission using *Stephanitis typicus*.

It may be mentioned on the outset that the experiments herein described were carried out under field conditions at the Central Coconut Research Station, Kayangulam, which is an infected area. To obviate this drawback, an equal number of palms of the same age as the ones under treatment, were kept as check and were not artificially inoculated.

Mechanical Inoculation of Leaf Extract

Palms ranging from 3—50 years were used for this purpose. The preparation of the extract and method of inoculation were the same as described earlier (Nagaraj and Menon, 1956).

The data presented in Table 1 (Text Fig. 1) shows a consistently high percentage infection on the inoculated palms than the control ranging from 33 to 71% in the former while only in one case did the latter go up to 20%. Palms of about 6—15 years are the most susceptible both in the inoculated as well as the control series and this agrees with the general observations in the field that maturing palms are most susceptible to the disease. Although the interval between symptom expression and the first set of inoculation varied from 8—12 months, generally the disease was clearly manifested only after 12—15 months. Palms of 40—50 years seem to be

apparently resistant to the disease probably due to the low inoculum potential and more so perhaps due to the technical difficulties in inoculating leaves at such heights.

TABLE 1

Percentage disease incidence (after sap inoculation) in relation to age of the palm (12 inoculations at monthly intervals)

| Expt. No. | Treatment | Age of palm in years | No. of palms | | % disease incidence | Incubation period in months |
|-----------|------------|----------------------|--------------|----------|---------------------|-----------------------------|
| | | | Inoculated | Infected | | |
| 1. | Inoculated | 2-3 | 5 | 2 | 40 | 9-11 |
| | Control | " | 5 | — | — | — |
| 2. | Inoculated | 4-5 | 19 | 7 | 37 | 8-12 |
| | Control | " | 14 | 1 | 7 | 10-11 |
| 3. | Inoculated | 6-10 | 5 | 3 | 60 | 9-10 |
| | Control | " | 5 | — | — | — |
| 4. | Inoculated | 6-10 | 6 | 2 | 33 | 9-10 |
| | Control | " | 6 | — | — | — |
| 5. | Inoculated | 10-15 | 7 | 5 | 71 | 8-9 |
| | Control | " | 6 | — | — | — |
| 6. | Inoculated | 10-15 | 10 | 5 | 50 | 8-12 |
| | Control | " | 10 | 2 | 20 | 9-10 |
| 7. | Inoculated | 20-30 | 5 | 2 | 40 | 8-10 |
| | Control | " | 5 | — | — | — |
| 8. | Inoculated | 40-50 | 12 | — | — | — |
| | Control | " | 6 | — | — | — |

TABLE 1A

Percentage disease incidence in 2-3 year-old seedlings after fortnightly inoculations

| Method of inoculation | No. of palms | | % disease incidence | Incubation period in months |
|-----------------------|--------------|----------|---------------------|-----------------------------|
| | Inoculated | Infected | | |
| Sap inoculation | 12 | 7 | 58 | 8-9 |
| Insect transmission | 12 | 7 | 58 | 8-9 |
| Control | 12 | 3 | 25 | 6-7 |

Further trials in the field (Table 1 A) wherein 2—3 year-old seedlings were inoculated at fortnightly intervals resulted in 58% infection in the inoculated as against 25% in the control. The results indicate that while the greater frequency of inoculation supplies a greater inoculation potential and thereby increases the percentage infection in the inoculated seedlings, the incubation period of the virus remains at 8—9 months.

Insect Transmission

Preliminary investigations (Nagaraj and Menon, 1956) using a number of insects visiting the palm indicated that the banana lacewing bug, *Stephanitis typicus* was probably the insect vector responsible for transmitting the disease. The results of further trials using the same insects as vectors are presented in Table 2. About 30—40 insects were collected from the field and after 2 hours' starvation, were given a 24 hours' acquisition feeding on the tender leaf of a diseased tree. These were then fed on the tender leaves of test palms for 24 hours or till all were dead. Table 2 shows that 20 to 60% infection was obtained in the inoculated series in which the percentage infection was throughout much higher than in the control palms. Only in one case 22% of the check palms were infected, and that was at a stage when they were of the most susceptible age, that is, 10—15 years.

TABLE 2

*Percentage disease incidence (after insect transmission with *Stephanitis typicus*) in relation to age of the palm. (12 inoculations at monthly intervals)*

| Expt. No. | Treatment | Age of palm in years | No. of palms | | % disease incidence | Incubation period in months |
|-----------|------------|----------------------|--------------|----------|---------------------|-----------------------------|
| | | | Inoculated | Infected | | |
| 1. | Inoculated | 2-3 | 15 | — | — | — |
| | Control | „ | 15 | — | — | — |
| 2. | Inoculated | 4-5 | 10 | — | — | — |
| | Control | „ | 9 | — | — | — |
| 3. | Inoculated | 6-10 | 10 | 2 | 20 | 10-13 |
| | Control | „ | 9 | — | — | — |
| 4. | Inoculated | 6-10 | 8 | 2 | 25 | 10-12 |
| | Control | „ | 8 | 1 | 12 | 10 |
| 5. | Inoculated | 10-15 | 9 | 5 | 55 | 8-9 |
| | Control | „ | 8 | — | — | — |
| 6. | Inoculated | 10-15 | 10 | 4 | 40 | 8-12 |
| | Control | „ | 9 | 2 | 22 | — |
| 7. | Inoculated | 35-45 | 6 | 2 | 33 | 10-12 |
| | Control | „ | 6 | — | — | — |
| 8. | Inoculated | 40-50 | 10 | 6 | 60 | 8-10 |
| | Control | „ | 10 | — | — | — |
| 9. | Inoculated | 40-50 | 12 | 4 | 33 | 6-14 |
| | Control | „ | 12 | 1 | 8 | 12 |

TABLE 3

Disease incidence and rate of spread of wilt disease in nature during the period 1954-58

| Block No. | 1954 | | | 1955 | | | 1956 | | | 1957 | | | 1958 | | | |
|------------------------------|------|-----|--------|------|-----|--------|------|-----|--------|------|-----|--------|------|-----|-----|-----|
| | I | V | VI VII | I | V | VI VII | I | V | VI VII | I | V | VI VII | I | V | VI | |
| Total No. of trees | 416 | 351 | 458 | 397 | 406 | 330 | 452 | 388 | 395 | 326 | 427 | 386 | 314 | 413 | 313 | 410 |
| Healthy | 127 | 73 | 88 | 68 | 131 | 61 | 86 | 67 | 130 | 61 | 81 | 65 | 48 | 66 | 49 | 58 |
| Wilt | 247 | 234 | 318 | 273 | 231 | 225 | 317 | 269 | 204 | 232 | 300 | 267 | 228 | 298 | 242 | 318 |
| Yellowing with outfall | 22 | 27 | 31 | 41 | 23 | 32 | 37 | 40 | 43 | 24 | 31 | 42 | 27 | 37 | 12 | 22 |
| General chlorosis | 4 | 7 | 14 | 6 | 5 | 2 | 7 | 5 | 4 | 2 | 8 | 5 | 4 | 5 | 3 | 4 |
| Tapering disease | 16 | 10 | 7 | 9 | 16 | 10 | 6 | 7 | 14 | 7 | 7 | 7 | 7 | 7 | 7 | 8 |
| No. of trees cut due to wilt | | | | | 10 | 21 | 6 | 7 | 9 | 1 | 25 | 2 | 12 | 14 | 1 | 3 |
| % 'wilt' affected palms | 59 | 67 | 69 | 69 | 68 | 70 | 70 | 70 | 54 | 72 | 72 | 70 | 75 | 75 | 70 | 80 |

Spread of the Disease

Systematic observations were taken on the incidence of disease in a plot of nearly 32 acres at Krishnapuram, Kayangulam for a period of five years. The data (Table 3) shows that about 60—80% of the mature palms of this area, where disease was first reported nearly sixty years back, are affected by the wilt disease showing a steady increase of about 3—5% every year. When it is taken into consideration that palms showing general chlorosis and an abnormal button shedding are inter-related and may lead to the typical wilted condition, the alarming increase of the disease in this area becomes really evident. The palms in Block I, where a manurial-cum-spraying experiment was in progress, show a decrease in disease incidence revealing the influence of a balanced nutrition in checking the spread of the disease.

Observations taken on a 3.67 — acre plot of underplanted seedlings also reveal the initiation and the sporadic nature of spread of the disease within a restricted area typical of an insect transmitted virus disease. It is not meant here to draw any conclusive evidence from this data which however, does support the virus theory.

Studies on Insect Population

A weekly sampling of diurnal and nocturnal insects visiting the coconut palm was made by using fly-paper. The results of such a survey done from diseased and healthy palms over a period of 10 months are given below (Table 4). This is intended only as a preliminary work before taking up detailed investigations using wind and light traps. It is seen that *Stephanitis typicus* is the single major group which predominates over the others except ants of the diurnal visitors. It is also of some interest to note that with the exception of one pair of palms, the population of *S. typicus* was consistently higher on the diseased trees than on the healthy ones. Of the other types of insects, *Ambispha* sp. and two species of the Fulgoridae were tried as vectors of the disease in the transmission trials, which however, yielded only negative results.

DISCUSSION

Even though no clear cut evidences are forthcoming from the foregoing data, especially since the transmission trials were not carried out under rigidly controlled conditions, it lends weight to the earlier hypothesis of the virus nature of the disease. Primarily, the very fact that the disease is spreading, though slowly, into new areas

TABLE 4

Diurnal insects collection: Number of various insects collected during a 9-month period

| Block No. | Condition of palm | Stephanitis typicus | Amblyspira sp. | Earwig. | May flies | Mosquito | Lace wing fly | Spider | Beetles | Ante | Miscellaneous |
|-----------|-------------------|---------------------|----------------|---------|-----------|----------|---------------|--------|---------|------|---------------|
| I | Diseased | 30 | — | — | — | — | — | — | — | 47 | 2 |
| | Healthy | 34 | — | — | — | — | — | — | 1 | 37 | 3 |
| III | Diseased | 34 | — | — | — | — | — | 1 | — | 61 | 1 |
| | Healthy | 16 | — | — | — | — | — | 1 | 1 | 49 | — |
| IV | Diseased | 27 | — | 2 | — | — | — | 2 | — | 49 | 1 |
| | Healthy | 20 | 1 | — | — | — | — | — | — | 36 | — |
| V | Diseased | 29 | — | — | — | — | — | 1 | — | 38 | 8 |
| | Healthy | 17 | — | — | — | — | 1 | — | 1 | 41 | 1 |
| VI | Diseased | 36 | — | — | — | — | — | 2 | 1 | 36 | 1 |
| | Healthy | 24 | — | — | — | 2 | — | — | 1 | 42 | — |
| VII | Diseased | 40 | 1 | — | 2 | — | — | — | — | 52 | — |
| | Healthy | 29 | — | — | — | — | — | — | 1 | 40 | — |

points to its pathogenic nature. Secondly, this hypothesis, arrived at mainly through negative results, finds parallel cases in many a disease of importance. Thus, the cadang-cadang disease of coconuts widely prevalent in the Philippines, is suggested to be an insect-transmitted virus (Celino, 1947; Kent, 1953) since extensive epidemiological studies prove that, like the wilt (root) disease, the causal agent is neither the soil nor any agent that moves through the soil. That the wilt disease is found in soils of different types, both laterite as well as sandy, suggests that neither major nor minor element deficiencies are involved. Moreover, pot culture experiments (Physiologist, unpublished) with NPK and the more important minor elements have failed to reproduce the disease symptoms. Nevertheless, the consistently low nutrient status of those soils, especially potassium, the high acidity and water-table of the majority of soil types reveal how this predominating poor nutrition increases the disease proneness of the host.

It is possible (Park, 1928) that under unfavourable soil conditions microrhizal fungi become parasitic on their host tissues. Hence it is understandable and also probable that the facultative *Rhizoctonia*, so widely distributed in the roots and rhizosphere of healthy palms (Radha and Menon, 1954) becomes pathogenic under unfavourable conditions of waterlogging or a high water-table. Indeed, the reported ability of *Rhizoctonia* sp. to reproduce the disease symptoms under water-logged conditions on seedlings (Menon *et. al.*, 1952) tends to support this view. However, that root decay was the effect and not the cause was shown by Nagaraj and Menon (1955) who found normal root system for a number of trees at the early stage of infection. The somewhat complicated yet important disease, the 'Bunchy-top' of bananas was presenting a similar problem until Magee (1927) showed that the high percentage of rotted roots invariably associated with *Rhizoctonia* sp. was secondary to the virus attack.

It is strongly indicated that *Stephanitis typicus* may be the probable vector or one of the many that may be functioning as vectors, by the higher percentage infection of inoculated palms under field conditions. That this insect forms the single major group that visits the coconut palm is also of considerable interest in this connection. It is a matter of common observation in the field that those palms which are highly infested by this insect become diseased earlier than the others whereas Table 4 shows that with one exception, all diseased trees have a higher number of these insects visiting them than the healthy. The instance of a lacewing bug acting as a vector is rare amongst plant viruses and attempts

are made among others, to find out the host range of this insect as well as of the virus pathogen.

The phenomenon of accumulated nutrients especially nitrogen, in the leaves and tender tissues of affected palms both in the case of cadang-cadang as well as the wilt (root) disease which may be either due to the inadequate translocation or due to the impaired physiological functioning is comparable to the accumulated nutrients common in the sandal spike and potato leaf roll (Smith, 1957).

Apart from trials under insect-proof conditions to transmit the disease, it is essential, under the circumstances, to undertake a survey of all insects visiting and breeding on the palms, so as to find out the vector (s), to develop a serologic or other specific diagnostic test for identifying the disease, to find out the host range of the virus and above all, to start a long range programme of breeding for disease resistance and these items of work are under progress at the Central Coconut Research Station, Kayangulam.

SUMMARY

Results presented here, though not conclusive, support the virus hypothesis of the wilt (root) disease of coconut. A higher percentage of infection of palms inoculated by mechanical methods as well as by insect transmission under field conditions, the nature of spread of the disease and the occurrence of *Stephanitis typicus* as the single major group of insects visiting coconut palm support this view.

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