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ROOT (WILT) DISEASE - RESUME OF WORK DONE SINCE 1964

by

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SUMMARY

The root (wilt) disease of coconuts in Kerala has been spreading and now infects about 250 thousand hectares of coconut plantations. It is more intense in poorly aerated land. Flaccidity of leaflets is the primary symptom of the disease, while yellowing, necrosis, nut fall etc. complete the syndrome. The disease is probably caused by a virus-like pathogen which is soil borne and transmitted by an insect as well as mechanically. Mode of spread in nature is not fully understood. The cowpea indicator host has been very erratic and seems to be highly sensitive to prevailing weather. Water stagnation or high water table seems to play an important role in the initiation and spread of the disease. There is a gross derangement in the physiological processes of infected palms. Eradication of diseased trees to check the further spread of the disease in new areas, as well as to re-establish healthy coconut gardens in diseased areas, is being experimented.

INTRODUCTION

The nature of the root (wilt) disease of Kerala continues to be shrouded with mystery despite investigations in concerned fields resulting in a large volume of available information. The disease has been infecting healthy coconut palms and seedlings within the diseased tract and spreading into new areas lying outside the outposts of disease as well. It now engulfs an area covering nearly 250 thousand hectares of coconut plantations spread fully over four districts and spreading into two more in the State of Kerala. The spread is not continuous, hence pockets of healthy coconut gardens occur throughout the diseased area. It is more frequent and more severe in trees growing in lowlying high water table areas. Of special interest are the reclaimed 'Kari' (peaty acidic) soils which though low lying and subject to waterlogging are practically free from the disease except in very old reclamations. Severe yellowing of leaves due to poor drainage in large areas of coconut gardens subject to waterlogging which also results in poor yields is often mistaken by the cultivators for root (wilt) disease. The leafrot disease cuased by fungi which often superimposes itself on root (wilt) infected trees presents a complex syndrome and makes the infected trees look horrid and crippled and adds to their deterioration and to the panic of the coconut cultivator. While the trees infected with root (wilt) disease alone decline slowly and are not generally killed, those infected with both the diseases deteriorate much faster and are sometimes killed.

SYMPTOMS

The characteristic symptom of the disease is a slow wilting of the foliage. Foliar symptoms of flaccidity and ribbing of leaflets, abnormal bending and breaking of petiole, yellowing of outerwhorls of leaves and tip and marginal necrosis of leaflets are common associated symptoms in infected palms. A study to establish corelations in foliar symptoms to work out the characteristic diagnostic symptoms of the root (wilt) disease has been undertaken. The data (Table 1) supports that flaccidity of leaflets is the primary characteristic symptom. Necrosis rarely occurs independent of flaccidity while occurrence of yellowing free of flaccidity is more frequent and appears as a delayed feature independent of root (wilt) disease. The low incidence of yellowing of leaves in younger palms and the occurrence of yellowing in more than 7.0 per cent of the adult palms in the absence of other symptoms is perhaps suggestive of a condition associated more with nutritional factors than of a disease symptom. Necrosis of distal ends of spadix has also been observed to occur in 18.0 to 25.0 per cent of diseased trees varying with intensity of the disease. The number of palms exhibiting complete disease syndrome was lower in laterite soils as compared to that in sandy loam and reclaimed clayey soils (Table 2). In tests of transmission under controlled conditions flaccidity of leaflets was discernable in infected seedlings further confirming that flaccidity is the primary diagnostic symptom of the disease. The infected seedlings also exhibited some yellowing of foliage but no necrosis. Root rot was established in infected seedlings alone after *Rhizoctonia solani* was introduced in the soil thus confirming that root rot is secondary.

SPREAD OF THE DISEASE

How the root (wilt) disease of coconut plam spreads in nature is not fully understood although it has been proved that banana lace wing bug (Stephanitis typicus) is a vector and can transmit the disease to healthy trees experimentally. The disease is also soil borne and spreads through the soil as well. The disease is found to occur in coconut plantations in all soil types but the rate of spread was observed to vary with the soil type. Moreover a high water table favoured its development. Preliminary studies were started in 1964 to study the pattern and rate of spread of the disease under different soil conditions. The spread of the disease was linear and slow in the direction of a pond perhaps influenced by flow of subsoil water in the plot with sandy soil during the first two years. In the third year, however, the disease spread was irregular and rapid when the land was partially inundated. The spread was irregular and rapid in the plot with alluvial loam soil also following flooding. On the other hand, the disease did not spread at all during the three year period, in the laterite soil with low water table. The spread of the disease was, thus, rapid in areas with high water table, suggesting active spread through the soil, water perhaps aiding the spread of the disease.

CAUSAL AGENT

(i) Nature of pathogen:

The cause of the disease has been believed to be a virus (Nagaraj *et. al.*, 1956) although no direct evidence is yet available. Holmes (1965) suggested that spirochaetes and sporozoa may cause the disease although no records of these organisms associated with diseased palms and suspected them of causing the root (wilt) condition by plugging the conducting vessels of coconut stem. Similar ciliate organisms were since reported from healthy coconut trees as well and could not be consistently observed in diseased palms; further investigations were not persued (Per. comm., 1968).

It is proved that the unknown agent of root (wilt) disease is sap, insect and soil transmissible. The pathogen has been detected from roots, leaflets, flowers, and parts of the nut from diseased tree by inoculation on cowpea seedlings and is considered systemic. Tests of filterability and electron microscopy with the pathogen were suggested (Holmes, 1965). Brande's dip method for preparing electron microscopic grids was adopted using sap from cut leaflets and nut water. No conclusive evidence regarding the presence of any specific particules in diseased material has yet been obtained although presence of some spherical bodies has been observed in the electronmicrographs in one test.

(ii) Test of pathogenicity:

Earlier investigations have already proved that the disease was transmitted to adult palms in the field by mechanical inoculation using infective S. typicus adults and by root connections (Nagaraj et. al. 1956). The disease was also transmitted successfully to 4 year old seedlings grown under insect proof screen house (Shanta, et. al., 1964).

TRANSMISSION STUDIES

(i) Insect transmission:

Adults of Stephanitis typicus have already been reported to transmit the disease. The insects acquire infectivity in two hours and retain it for twenty-two hours. The 4th and 5th instar nymphs of the insect also transmitted the disease to cowpea seedlings after 24 hours acquisition and 48 hours infection feeding.

(ii) Seed transmission:

The husk, kernel, water and embryo from nuts collected from diseased trees in varicus stages of development, were infective on cowpea seedlings. Seednuts collected from diseased palms were sown in sterilised soil. Three months later when the nuts germinated the sprouting embryo, haustorium, kernel and nut water were tested for infectivity on cowpea seedlings. All parts of the germinated nut gave positive reaction indicating presence of the infective principle. Further tests to confirm whether the disease was transmitted to the progeny through the seednut have been undertaken.

(iii) Soil transmission:

Root samples of a large number of diseased palms in different age groups and with varying intensities of disease were examined under microscope for zoospores of Olpidium or such lower fungi adopting Teakle's method. Root washings of 50 palms were centrifuged at 2000 rpm. and the pellet was tested for infectivity on cowpea seedlings both by mechanical abrasion as well as by adding it to the soil. Cowpea seedlings were also raised at the base of diseased palms in the field and in wooden boxes with sterile soil irrigated by water suspension of diseased soils. Seedlings with malformed leaves were examined. No Olpidium like fungi were observed. Roots of the weeds commonly present and susceptible to Olpidium sp. e.g. Cleome viscosa and C. monophila from diseased gardens were examined. No Olpidium sp. were present.

Weischer (1967) studied the possible role of plant parasitic nematodes in root (wilt) disease. Soil samples were taken from different places representing seven soil types in diseased and healthy areas from the rhizosphere of healthy, slightly

diseased and severely diseased trees (Table-3). Nematodes were present both in healthy and diseased areas. No relation between the number of individuals or sps. and the disease was found. There was no significant difference in the nematode fauna of healthy and early and advanced stages of diseased trees. Population density and number of species differed in certain degrees with soil type. The low population density of nematodes, their wide-spread occurrence and the general distribution pattern of the disease indicated that the plant parasitic nematodes can be excluded from being considered as primary cause of root (wilt) disease. The nematodes can however be considered as disease incitants by permitting entry of pathogens through injured roots. Xiphinema was present in both healthy and diseased trees, Longidorus was found in diseased areas or very near the border between the two areas. Host-parasite relationships between coconut palms and nematodes and the role of Xiphinema and Longidorus as vectors of the pathogen need to be studied.

SOIL INFECTIVITY

The root (wilt) disease pathogen is soil borne. In sandy loam soil samples from six metres away from the base of a diseased tree were still found infective. Wet and air-dried diseased soil samples were still infective after storage for 40 days at 28-30°C. as against 9-12 days reported earlier. Soil samples from a diseased coconut garden from which all the trees were removed, continue to exhibit infectivity after 28 months, indicating the persisitence of infectivity for unusually long periods in the soil under natural conditions.

INDICATOR HOST

Holmes et. al. (1965) observed some peculiar soil reactions on cowpea seedlings by mixing samples from diseased soils and reported that either the infectivity of soils was destroyed or suppressed due to slight changes in the soil condition or the cowpea inoculation test was erratic. A large number of cowpea varieties were tested for selecting a better consistent indicator host for root (wilt) pathogen but var. 'New Era' continued to show the best results. Further tests confirmed that susceptibility of cowpea was variable and was perhaps governed by temp-humidity factor. The cowpea test had been unusually erratic since 1965 (Table 4). Factors favouring infectivity of cowpea seedlings to the root (wilt) disease pathogen are under study. Inoculation of cowpea seedlings by infective insects proved to be more effective as compared to mechanical abrasion.

Nutritional and Biochemical Investigations

(i) Soil survey:

It was reported earlier that nutrient status of soils from healthy and diseased areas essentially had no differences except that the diseased soils were poorer in calcium, available potash, total exchangeable bases and low in pH (Varghese, et. al., 1959). As a result of a second reconnaissance survey of healthy and diseased areas, Varghese (1966) reported that the diseased tracts of the four major soil types under study (sandy, loamy, clayey and laterite) contained greater amounts of moisture, organic matter and nitrogen than the corresponding healthy areas. How the moisture retaining capacity of these soils orientate soil conditions favourable for action of pathogenic organisms for initiation and spread of the disease is not known. Excess nitrogen may favour susceptiability of trees to infection. Perhaps combined effect of excess soil moisuture and nitrogen may set in soil and physiological conditions conducive to disease incidence. Available potash was deficient in most diseased soils. Potash deficiency is known to affect parasitism, water relationships and weakens root system eventually leading to symptoms of wilt.

Lime and magnesium contents were uniformly poor and variations in pH values were of a low order in all soils. Faulty nutrient ratios particularly K_2O/MgO, K_2O/CaO, and N/K_2O were obtained suggesting impaired nutrient balance as reported earlier (Varghese, et. al., 1959).

High incidence of disease occurred in reclaimed clayey soils but it was practically absent from 'Kari' soils though both are waterlogged. Kari soils are also fairly rich in nutrient content, but are highly acidic. Perhaps that is largely responsible for keeping them free from infection.

(ii) Effect of waterlogging:

Poor soil aeration due to waterlogging or high water table has a profound influence on the development and intensity of the disease. Waterlogging results in lower uptake of nutrients and consequent foliar yellowing and stunted growth of coconut palms. Provision of drainage channels, application of river sand and manuring cured the foliar yellowing and significantly improved the growth and yield of coconuts. Under pot culture conditions, river sand proved to be a better medium for growth of seedlings under waterlogged conditions than sandy loam soil.

(iii) Tissue analysis:

Further studies on the amino acids in coconut palm tissues have revealed that the trend of accumulation of free amino acids noticed in leaves is evident in coconut water and kernel also. Tissues of West Coast tall variety contained higher quantities of amino acids than dwarf orange, dwarf green and T x D cross.

(iv) Nutritional studies:

Both healthy and root (wilt) infected palms often develop yellowing of outer whorls of leaves. Yellowing characteristically commences from tips and margins of leaflets. Leaf analysis showed magnesium deficiency. A set of healthy and diseased palms showing foliar yellowing were given monthly foliar spray with 2 per cent Magnesium sulphate solution. At the end of 18 treatments, yellowing was completely cured in 16 per cent of the treated palms. Sixty per cent of the palms exhibited reduction in yellowing. The results indicated that the yellowing associated with root (wilt) disease symptoms may be largely due to deficiency of Magnesium.

Physiological Investigations

Water relations of the diseased palm comprising of the rate of uptake of water by root tissue, and the trunk, turgidity of leaves and rate of transpiration were studied. Earlier studies had shown that the rate of transpiration of the leaves of diseased palms was higher than that of the healthy ones. A study showed that the rate of uptake of water by the root tissue in diseased palms was 35 per cent less than that in healthy tissue. Anatomical studies of the root tissue further showed internal browning and disintegration of vascular tissue in 60 per cent of the roots of diseased palms and 33 per cent of the roots of apparently healed palms growing in diseased gardens, against no such disintegration in the roots of healthy palms (Indira, 1968). A study on the rate of uptake of water in the stem just above the level of the bole revealed that in the diseased trees, the uptake is 5-30 per cent less than that in the healthy ones. These results suggest the slow development of an internal water deficit in the palms affected by root (wilt) disease.

Nitrogen, Phosphorus and carbohydrate metabolism of diseased palms was studied. Gross derangement in the path of nitrogen resulting in considerable increase in the non-protein nitrogen content which is concomitant with a sharp decrease in the water soluble nitrogen and protein nitrogen fractions was observed in the diseased leaf tissue. Considerable reduction in C/N ration of both leaves and roots of diseased palms was also indicated. A sharp decrease in the total carbohydrate content of diseased leaf tissue further supports derangement in the metabolism of nitrogen. There is urgent need to re-examine the nitrogen nutrition of diseased palms.

METHODS OF CONTROL

(i) Inhibition of soil infectivity:

Holmes et. al. (1965) treated infective coconut soils with ash, calcium hydroxide, proteolytic enzymes and planted them with cowpea in a preliminary experiment to determine inhibition of infectivity due to soil treatment. The results proved promising. In an earlier experiment PCNB (Penta-chloro-nitro-benzene) 2.5-5.0 gm/kg. of soil destroyed infectivity of soils. Calcium hydroxide, copper sulphate, mercuric chloride, tannic acid, formaldehyde, methylene blue, malachite green and gentian violet did not give any conclusive results. Large scale applications have not yet been tried. Growing annual crops which might hasten inhibition of soil infectivity is also undertaken.

(ii) Eradication of diseased trees.

Holmes (1965) suggested experimenting eradication of diseased trees as a control. He suggested that: Attempts be made first in healthy areas at the borders of the disease to check its further spread in new areas; the soil must be constantly tested and replanted with coconut seedlings only when it has lost infectivity; seedlings raised from seednuts from healthy coconut palms in the diseased tracts may be tried. Any new case of diseased palm must be immediately eradicated.

Another set of eradication experiments may be undertaken in highly diseased areas by removing all palms from a garden; replanting may be undertaken only when the soil is tested to be non-infective; the soil may be replanted with such crops which are immune to the root (wilt) disease: the population of the insect vector (S.typicus) may be kept low by scheduled insecticide application. A small scale experiment was started in February, 1966. All trees were removed from a one-acre plot at the research station farm. The soil was deep ploughed and has been planted with Tapioca (Cassava) twice. The soil is still infective after 28 months.

(iii) Disease resistance:

The progenies from apparently healthy trees of the West Coast tall variety in the diseased areas did now show any resistance and behaved similar to the "quality" seedlings from healthy areas. Malayan yellow, orange and green dwarfs and green semi-tall varieties have been planted for testing resistance against the disease.

DISCUSSION

The possibility of a virus-like pathogen being the causal agent of the disease has been surmised (Menon, 1963; Lal 1966 and Holmes et. al. 1965) particularly because the disease is systemic, is transmitted mechanically, has an insect vector and is soil-borne, thus possessing some of the important characteristics of virus

diseases of plants. If the nature of the pathogen is finally confirmed, by direct evidence, to be a virus, it is going to be a peculiar plant virus which has not been recorded so far.

The derangements in the physiological functions and consequent responses in diseased palms leading to internal water deficit and improper utilisation of nutrients perhaps leads to the appearance of primary symptoms of flaccidity and wilt. The role of soil factors in development of the disease complex is evident from the fact that in transmission trials under controlled conditions, the primary symptom of the disease alone was produced while the field tests exhibited all the symptoms of the syndrome.

The significant role of high water table and waterlogged soil conditions in the incidence, intensity and spread of the disease requires further elucidation. The series of chemical and biological changes ensuing such unfavourable conditions probably act as conditioning factors in pathogenesis. No lower fungi seem to be involved in soil transmission but the role of nematodes, particularly the vector species needs to be determined.

The reasons for the sudden erratic behaviour of cowpea seedlings to the root (wilt) disease pathogen during the past four years are not understood. This has greatly been responsible for limiting the progress of investigations in various fields.

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Table 1. Foliar condition in relation to age of palms.

Age of palms	% Healthy	F	Y	N	F+Y+N	No.of palms observed
3-9 years	29.1	47.1	1.8	1.6	15.0	861
10-15 years	44.4	2.9	8.5	0.2	46.7	768
30 years and above	17.6	1.3	7.6	0.1	74.5	1592

Table 2. Foliar symptoms in relation to soil type.

Soil type	% Healthy	% exhibiting Flaccidity	Yellowing	Necrosis	F+Y+N	No. of palms observed
Sandy loam	17.6	1.3	7.6	0.1	74.5	1592
Loamy to clayey	28.2	15.3	4.0	—	55.1	915
Reclaimed clayey	2.6	8.9	—	—	88.5	2000
Lateritic	16.5	23.0	2.5	6.0	50.4	1375

Table 3. Plant parasitic nematodes from the rhizosphere of coconut palms in Kerala, South India

	DISEASED AREA						HEALTHY AREA		
	Soil type						Soil type		
	Sandy	Loamy sand	Laterite	Alluvial	Black	Clay	Sandy loam	Laterite	Clay
	No. samples analysed						No. samples analysed		
	18	12	9	13	8	3	1	2	1
Xiphinema	+	+	+		+	+	+	+	+
Longidorus	+	+		+			+		
Tylenchorhynchus	+		+	+	+	+	+	+	+
Meloidogyne	+		+	+	+		+		
Hoplolaimus	+	+	+	+		+			
Helicotylenchus	+		+	+	+	+	+		+
Dolichodorus		+							
Pratylenchus	+	+	+	+					
Radopholus		+							
Rotylenchulus			+	+			+	+	
Criconema				+					
Hemicycliophora	+			+					
Hemicriconemoides				+			+		
Criconemoides	+	+	+	+	+				
Paratylenchus					+				

Table 4. No. of days showing positive cowpea reaction in the different months of the year from 1965 - '68.

Months / Year	1965	1966	1967	1968
January	Ex-	-/19	4/22	-/26
February	peri- ment	6/22	-/21	3/23
March	start- ed from	8/25	6/23	8/23
April	July.	5/22	2/23	-/22
May		4/24	10/26	5/26
June		7/24	7/25	3/24
July	4/25	4/24	4/25	
August	-/24	9/15	-/22	
Sept ember	9/22	-/23	-/18	
October	7/21	6/23	2/22	
November	6/25	10/24	-/16	
December	7/25	-/25	1/24	

Nominator expresses the number of days of positive reaction and the denominator, the total number of days in a month on which the text was made.