Effect of Root (wilt) diseases on microsporogenesis in coconut (cocos nucifera L.)

By

M. C. NAMBIAR AND T. O. PRASANNAKUMARI

Central Coconut Research Station, Kasaragod

INTRODUCTION

It has been known for many years that diseases caused by virus and other pathogens may induce deviations from the normal cytological processes found in plants. Though the literature concerning the nature and behaviour of plant viruses is now very large, very few reports have as yet appeared in connection with the possible effects of virus upon nuclear behaviour. Kostoff (1933) found mosaic virus infestation in tobacco causing abnormalities in all stages of meiosis leading to the abortion of pollen. Caldwell (1952) reported complete breakdown of the normal meiotic processes in tomato plants infected with aspermy virus. Swaminathan *et al.* (1959) described the cytological effects of infection with mosaic and leaf roll viruses on microsporogenesis and seed fertility in three varieties of *Capsicum annum* L. and discussed the results in relation to the role of infection in plant evolution and rarity of virus transmission through seed. Thomas Varkey and T. A. Davis (1960) reported higher percentage of dummy pollen in coconut palms infected with Root (wilt) disease and a positive correlation between the percentage of dummy pollen and stage of infection. The present paper records the indication from preliminary investigation on the effect of Root (wilt) disease of *Cocos nucifera* on the formation of microspores of the palm.

MATERIALS AND METHOD

A few diseased palms of Central Coconut Research Station, Kayangulam, which were infected mainly by the Root (wilt) disease were used in this study. For the study of meiosis, flower buds at proper stages were fixed in Carnoy's fluid and brought over to Kasaragod for cytological investigations. The fixed materials were stored in a refrigerator. The anthers were smeared in a drop of aceto-

carmine on a clear slide and analysis were carried out in most cases in temporary preparations. Pollen sterility was estimated only by stainability in aceto-carmine. The data collected on the cytological behaviour of the diseased palms were compared with those of the West Coast Tall palms of Central Coconut Research Station, Kasaragod.

OBSERVATIONS

Root (wilt) disease is one of the most important diseases of coconut areas of Kerala State affecting about a lakh of acres. The characteristic symptoms of the disease are general wilting of the leaves, flaccidity and ribbing of leaflets and necrosis of leaf tips. In the advanced stage, the crown gets very much reduced in size, ceases to produce flower bunches and finally succumbs to the disease. Recovery of diseased palms has not been observed so far (Menon and Pandalai, 1958). A comparison of meiotic behaviour of a few diseased palms and two healthy palms are listed below.

Meiosis was regular in the healthy West Coast Tall palms. There were 16 bivalents at diakinesis and metaphase I (Fig. I) and anaphase separation was normal. The bivalents were found associated with the nucleolus during diplotene and diakinesis. On an average the chiasma frequency per bivalent was found to be 1.84 (Table I). Some irregularities were, however, observed in the infected palms. There was slight reduction in chiasma frequency at metaphase I in the infected plants and frequency of rod bivalents were comparatively more (Table I). Partial to complete desynapsis (Fig. 2) was also observed at metaphase I. Irregularities at anaphase and subsequent stages were very few. One inversion bridge and a fragment, two cells with sticky bridges, and few cells with laggards were the only irregularities observed (Table II). At sporad stages few monads and diads (Figs. 3 and 4) resulting from the formation of restitution nuclei, also occurred in addition to regular tetrads (Table III).

DISCUSSION

The problems of transmission of virus by seed or otherwise have been of interest for virus workers and is clearly of practical as well as of theoretical importance. The rarity of their transmission from one plant generation to another through reproductive cells is an interesting limitation to the spread of virus in nature and several views have been advanced to explain the prevention of transmission of viruses through seed. According to Caldwell (1952) tomato plants infected with aspermy virus may occasionally contain embryo sacs and pollen grains which are free of viruses and these are the only ones which undergo fertilization. Gametes infected with viruses are non-functional. Swaminathan *et al.* working on the virus disease of *Capsicum annum* suggested the presence of inhibitory factor in capsicum seeds and the possibility of this virus inactivation mechanism present in capsicum operating at an early stage in the development of the embryo, being the reason for the comparative rarity

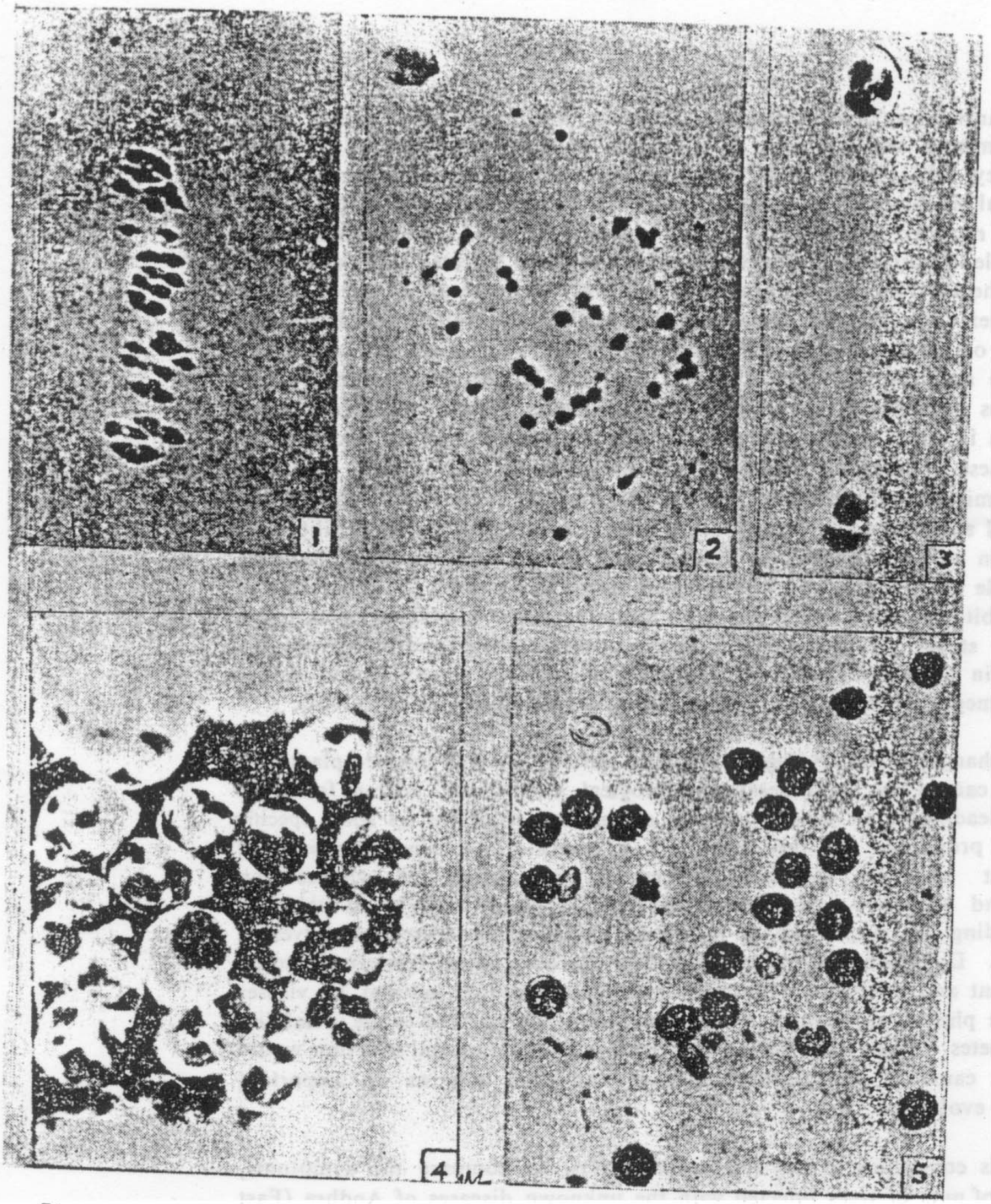

Fig. 1 Metaphase in a healthy coconut palm showing 16 bivalents. Figs. 2 to 5 Meiosis in Root (wilt) infected palm. Fig. 2 Metaphase I — Partial disynapsis. Fig. 3 Sporads showing a dyad in addition to normal tetrad. Fig. 4 Sporads showing monads in addition to normal tetrad. Fig. 5 Normal and sterile pollen in infected plants.

of seed transmission of virus disease. There are, however, several viruses which are transmitted through seed, e. g., virus causing false stripe in barley (Mc Kinney, 1951), bean mosaic virus and lettuce mosaic virus (Couch, 1955). Mechanical transmission of certain stone fruit viruses from *prunus* pollen has also been reported by Ehlers, C. G. and Moore, J. D. 1957. Hence, it may not be plausible to postulate that all the gametes which are infected with the viruses, abort. The aberrant meiotic behaviour may only be the result of infection and not an index of the presence of the virus in the cell. Our preliminary observation recorded on few diseased palms especially the cytological aberrations at metaphase and sporad stage, though very low in frequency, resemble the aberrations regarded as typical of cell divisions found under pathogenic conditions in other crops. Though further detailed studies are necessary to confirm these findings, the significance of these observations in plant evolution and transmission of viruses cannot be overlooked. The low frequency of cytological aberration and comparatively high percentage of pollen fertility and seed set in palms infected with Root (wilt) disease may therefore indicate, (a) possible transmission of virus through reproductive cells, (b) possibility of some inhibitory factor, as suggested by Swaminathan *et al.*, (1959) operating in the early stages of development of reproductive cells and/or (c) possible variation in the amount of virus or absence of virus in some cells and tissues including mega and microspore mother cells as suggested by Caldwell (1952).

The changes in the cytological processes noticed in virus infected plants are probably caused by the general physiological disturbance arising from the infection leading to an upset in normal enzymatic activity or by some specific chemical properties of the virus or by both. An understanding of the effect of the individual chemical components of viruses on mitosis and on micro and megasporogenesis in plants will be of value in understanding the mechanism of the effects produced by viruses themselves on such cells. Darlington (1944) has discussed the correlation between heredity, development and infection. The many cytological changes induced by viruses may cause phenotypic changes if such aberrations do not lead to the inviability of the gametes. Thus, infection besides being a force in natural selection would also be a cause of spontaneous mutation and thus acts as an important factor in evolution.

In this connection it may also be of interest to compare the symptoms, especially of nuts of trees infected with the unknown diseases of Andhra (East Godavari) with that of Root (wilt) disease in Kerala. The nuts of the trees infected with this unknown disease are shrivelled and atrophied and in some cases show longitudinal cracks and gumming at those cracks. In such nuts the kernel is not formed (Menon and Pandalai, 1958). The effects of Root (wilt) disease on floral parts is not so severe. The aetiology of this unknown disease is not known and is being investigated and if it turns out to be viral in nature the effects of the virus of unknown disease of Andhra are apparently different from

those of Root (wilt) disease of Kerala. The absence of the kernel in the nut and other symptoms of the infected trees of Andhra point to the possibility of the infection of the gametes and their consequent degeneration. The virus in this case may probably prevent the formation of normal tetrads in micro and megasporogenesis and thereby prevent the transmission of disease through reproductive cells. A detailed investigation of these and similar diseases of the coconut palms may, therefore, be necessary to elucidate this interesting and important problem of transmission of virus diseases in *Cocos nucifera*.

SUMMARY

Investigations of the effect of Root (wilt) disease on the microsporogenesis in *Cocos nucifera* have shown very few irregularities at meiosis. These irregularities, however, recall the aberrations regarded as typical of cell divisions found under pathogenic conditions. The results of investigations are discussed in relation to rarity of transmission of virus through seed and the role of infection in relation to plant evolution.

ACKNOWLEDGEMENT

The authors gratefully acknowledge their indebtedness to Dr. K. P. V. Menon, Ex-Director, Central Coconut Research Station, Kayangulam for the constructive criticism and valuable guidance at every stage of the work, to Dr. K. M. Pandalai, Director, Central Coconut Research Station, Kasaragod for his keen interest in the study and to Dr. M. S. Swaminathan, Head of the Division of Botany, Indian Agricultural Research Institute, New Delhi, for his valuable advice on the manuscript. Our thanks are also due to Kumari Sumathikutty, Sri Jayarajan and Sri Kunjan of Central Coconut Research Station, Kayangulam for their kind help in collecting and fixing the material required for study.

REFERENCES

1. Caldwell, J. 1952. Some effect of plant virus on nuclear division. *Ann. Appl. Biol.*, 39: 98-102.

2. Couch, H. B. 1955. Studies on seed transmission of lettuce mosaic virus. *Phytopathology*, 45: 63-70.

3. Darlington, C. D. 1944. Heredity, development and infection. *Nature*, 154: 164-169.

4. Ehlers, C. G. and Moore, J. D. 1957. Mechanical transmission of certain stone fruit viruses from *Prunus* pollen. *Phytopathology*, 47: 519-20.

5. Kostoff, D. 1933. A contribution to the sterility and irregularities in the meiotic processes caused by virus diseases *Genetics*, 15: 103-114.

6. McKinney, H. H. 1951. A seed borne virus causing false stripe in barley. *Phytopathology*, 41: 563-564.

7. Menon, K. P. V. and Pandalai, K. M. 1958. *The Coconut Palm - A Monograph*. Indian Central Coconut Committee, Ernakulam.

8. Swaminathan, M. S., Ninan, T. and Magoon, M. L. 1959. Effect of virus infection on microsporogenesis and seed fertility in Capsicum. *Genetica*, XXX: 63-69.

9. Thomas Varkey and T. A. Davis. 1960. Studies on coconut pollen with reference to the leaf and root (wilt) disease. *Indian Coconut J.*, Vol. XIV. No. 1, Oct.-Dec. 1960.

Received in December, 1963.

TABLE I

Chiasma frequency in healthy and diseased palms

Material	Stage	No. of cells studied	Mean association per cell		Mean No. of xta per nucleus	Mean No. of xta per bivalent	Remarks
			Bivalent	Univalent			
Healthy palm (of C.C.R.S., Kasaragod)	1. Metaphase	20	16	...	29.45	1.84	
	2. ,,	20	16		29.35	1.83	
Diseased palm (of C.C.R.S., Kayangulam)							
VII/399		9	15.1	0.90	25.66	1.7	
VII/375		9	13.66	2.34	22.99	1.67	
III/188		8	15.4	0.60	25.1	1.63	
VII/280		9	13.2	2.80	20.6	1.55	

TABLE II

Irregularities at Anaphase I and later stages

Material	Stages	No. of PMCs studied	Normal PMCs	No. of cells with			
				Inversion bridge	Sticky bridges	Laggards	Micronuclei
Healthy palm	A I	20	20				
(i)	A II	20	20				
	TI & TII	40	40				
(ii)	A I	20	19			1	
	A II	20	20				
	TI & TII	40	40				
Diseased palm VII/399	A I	50	48	—	1	1	
	A II	64	64				
	TI & TII	127	127				
VII/375	A I	50	49		1		
	A II	68	68				
	TI & TII	103	103				
III/188	A I	43	40	1	—	2	
	A II	74	74				
	TI & TII	171	171				
VII/280	A I	70	66			4	
	A II	52	52				
	TI & TII	145	145				

TABLE III

Abnormalities at sporad stage

Material	No. of sporads studied	Monads	Diads	Tetrads	Pentads	% of normal sporads	% of fertile pollen
Healthy palm (i)	142			142		100	96
(ii)	137			135		98.6	94
Diseased palm							
VII/399	329	4	—	324	1	98.2	92
VII/375	347	4	1	341	1	98.2	93
III/188	299	5	—	296	—	98.7	94.9
VII/280	224	5	2	217	—	97.0	92.9