FINAL REPORT
OF
RESEARCH PROJECT

No. Ent. XI (231)

P1-81/9-ICI-H10/0311

GENETICAL INVESTIGATIONS ON THE VECTORS OF COCONUT ROOT (WILT) DISEASE

JANUARY 1985 TO DECEMBER 1987

Central Plantation Crops Research Institute,

REGIONAL STATION, KAYANGULAM

KRISHNAPURAM-690 533

KERALA, INDIA.

SEPTEMBER 1988

FINAL REPORT

1. Institute code No. : Ent XI (231)
2. ICAR code No. : PI-81/9-ICI-H10/0311
3. Name and address of the Research Institute/Centre : Central Plantation Crops Research Institute, Regional Station, Kayangulam, Krishnapuram -690 533 Kerala State.
4. Project title : Genetical investigations on the vectors of coconut root(wilt) disease.
5. Name and designation of project leader : C.P.Ramachandran, Senior Research Assistant.
6. Name (s) and designation (s) of Project Associates Collaborators including project leader and work done.

Sr.No.	Name	Designation	Period	Project leader/ Associate
1.	C.P.Ramachandran	SRA	1985-'87	Project leader
2.	T.S.S.Rauther	S3	1985-'86	Associate
3.	K.V.Joseph	S2	1985-'87	"

7. Location of Research project with complete adress : Central Plantation Crops Research Institute, Regional Station, Kayangulam, Krishnapuram -690 533 Kerala State.
8. Date of start : January, 1985.
9. Date of termination : 31st December, 1987.
10. (a) Objectives.

To evolve genetical methods of control of vectors of root (wilt) disease and to study the strainal variation of vectors leading to refractoriness.

(b) Practical utility.

In case refractoriness is present in the vector population the same factor could be used in preventing the spread of the disease. Release of the non vector strains in large numbers in diseased tracts and in particular in ex border areas could gradually change the characteristics of the population and convert the same in to non-vector population. Here eradiation of the vector is not envisaged, but by genetical manipulation the character is changed without affecting the natural balance.

If inter or intra-specific hybrid sterility as observed in species of cabbage loopers, mosquitoes etc., in these, the same could be employed to supress the pest population. Sterile male release technique in Pest Control can be employed as part of an integrated pest management programme.

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11. Techinical programme.

1. Refractoriness. (Strainal Variation ie resistant and Susceptible)

a. Locating gardens in healthy tracts planted with seedling collected from diseased tracts and observations on the manifestation of disease symptoms and spread of the disease to adjoining palms/gardens(spread of the disease to palms of the same locality).

b. Protein pattern of Stephanites typica from various tracts, correlate the genetypic variation in relation to diseased tract, healthy pockets in diseased area, healthy tracts and isolated pockets of infestation.

2. Hybrid sterility studies, inter and intra-specific, of the vector.

3. Effect of radiation on sterility and use of sterile males in pest control.

4. Laboratory rearing of S. typica using alternate host plants and artificial diets.

12. Summary of Results:-

Carried out survey of Manenthody area of Wyanad Dist and Anamalai area of Coimbatore dist. At Wyanad seven gardens having fiftyfour surviving palms out of two hundred planting material(nuts/seedlings)collected from diseased tract. None of the palms showed any disease sympotme. At Anamalai, a garden planted with one hundred and five seedlings from Kayangulam was located. Fifteen samples from these seedlings and six samples from the local seedlings adjoining to thee plants were serologically tested. Seven out of the fifteen samples and one out of the six samples showed positive indication of disease. But none of the palms showed any disease symptoms even after five years of planting at the time of sample collection.

In Trivandrum Dist, Morinda tinctoria was found to harbour a large number of lace bugs. However, these lace bugs were got identified as Dalinius conchatur distant.

Acrylamide gel electrophoresis, both slab and disc were carried out. Different sample sizes, purification methods, buffer, gel cmcentration and voltage were tried to standardise technique. A sample size of 200 to 300 insects, gel concentration of 7.5 per cent or less cleaning lipids and lipe proteins with toluene and centrifugation at 18,000 rpm with addition of SDS was found to give seperation of thebands. Only summassive

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blue was available to stain and the band separations were not good.

13. Progress of work in relation to the time targeted for completion of work and reasons for non-achievement of targets, if any.

An initial survey of Mananthody and Anamalai areas of Wyanad and Coimbatore districts was carried out. At Mananthody seven gardens planted with 200 planting materials, seed nut/ seedlings were located. Out of this only 54 plants were surviving (details table I) and none of the plants showed any symptom of root(wilt) disease. At Anamalai, one garden was located, where 105 seedlings collected from Kayangulam area were planted. None of the palms showed any visual symptoms of disease. 15 samples were collected from the Kayangulam plants and six samples from adjoining garden. On serodigonostic test, 7 samples out of 15 and 1 sample out of six showed positive indication of MLO.

Morinda tinctoria a green manure plant was found to grow in large numbers in Trivandrum dist. These plants were found to harbour large numbers of lace bugs. The lace bug was got identified as Dalinius conchatus distant(Tingidae) by Dr. Mohana Sundaram Tamil Nadu, Agricultural University, Coimbatore. A few plants of Morinda were planted in the Institute campus and observed regularly for feeding by S. typica No feeding of S. typica was noted during the course of two years. The aim was to study the influence of flora of an area on the population of S. typica. Since Morinda was not an alternate host of S. typica the studies on these lines were discontinued.

A detailed study of the survey reports on root (wilt) disease as well as other information on the appearance of diseased palms in healthy tracts was made and information regarding gardens , their address etc were compiled.

Acrylamide gel electrophoresis using disc and slab was carried out to standerdise technique for protein assay of S. typica. The parameters involved were sample size, acrylamide concentration, purification, running time voltage, PH and staining technique. Insect samples 25,50,100,200 and 300 numbers, both male and female, were homogenised separately in 5 ml of 0.1 per cent Tris Hcl containing 0.01 per cent mercapto-ethanol or in 0.1 per cent phosphate buffer ph 7.4. The homogenate was centrifuged at 5000, 10,000 and 18,000 Rpm for 10 or 15 minutes to remove impurities. These samples gave

streaking on staining of the gels due to smaller particles, lipids and lipo proteins. Lipids and lipoproteins were removed by using Toluene and centrifugation at 18,000 RPM. The streaking was reduced by the above treatment. Aerylamide concentrations of 12.5, 10 and 7.5 per cent and gardient gels were tried. 7.5 per cent and gardient gels were gave some separation of the bands. Various voltages and running periods were tested and a voltage of 150 amps and a running time of two and a half hours to three hours was found to be satisfactory for disc. For slab three to three and half hours and 200 volts were found to be adequate.

The gels were stained with cummassive blue. Band clarity number and seperation were not satisfactory with this stain. One slab was stained with siluer stain and this gave 11 bands.

The work could not progress due to the following reasons. Due to paucity of funds, the chemicals required for testing lower concentrations of acrylamide, isozymes and staining methods could not be tried. Frequent power failure during working hours, lack of running water etc. affected the progress of work adversely. The power pack was highly erratic in its power supply. Due to various reasons like lab facilities etc. the actual work on gels electrophoresis was started only in the later half of 1987 only.

14. Papers published : Nil

Literature cited :

1. Black, L M. 1943

Genetic variation in the clover leaf hopper's ability to transmit pototo yellow-dwarf virus genetics, 28: 200-9.

2. Marjorie A. Hoy and John Mc Kelvey Jr.(edn) 1979 genetics in relation to insect management. Rockfeller foundation conference-March 31-April 5, 1978. Rock feller foundation.

3. Nagaraj,A.N., Black, E.M. (1962)

Hereditary variation in the ability of a leaf hopper to transmit two unrelated plant viruses, virology-6: 152-62

4. Storey,H.H. (1978) Transmission of maize streak disease-Ann. Appl. Biol. 15: 1-25.

15. Details of Field/Laboratory Note Book and their final location.

Primary Project File	:	One
Sub Project File	:	Nil
Experimental log book	:	Nil
Laboratory note book	:	One
Field note book	:	Nil

C.P. Ramachandran

16. Signature of Principal investigator

17. G.B. Pillai

Signature of Head of Division/Section/Station

18. Signature of Director

19. Prepared by C.P.Ramachandran. SRA.

Table: I. Showing Details of Planting material collected from diseased tract and planted in Mananthody area

Sr.No. of Garden	Place from where planting material collected	Type of planting material Seed nuts – Nos.	Seedling Nos.	Age at the time of observation Years.	No.surviving
1.	Kuriyanad Meenachil	100	–	10	16
2.	Ettumannure	–	25	15	13
3.	Thodupuzha	–	1	8	1
4.	Moovattupuzha	–	40	30	12
5.	Kothamangalam	–	15	9	8
6.	Travancore area	2	–	10	1
7.	Pala	5	–	30	3