

NATURAL OCCURRENCE OF AN ENTOMOPATHOGENIC NEMATODE, *HETERORHABDITIS INDICUS* (POINAR, KARUNAKAR AND DAVID) FROM KERALA, INDIA.

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Abstract :- Natural occurrence of an entomopathogenic nematode, *Heterorhabditis indicus* (Poinar, Karunakar and David) from soil is reported for the first time from Kerala, India. Morphometrics of the nematode is reported. At $24\pm 1^\circ\text{C}$ the duration of life cycle was seven days on larva of *Corcyra cephalonica* Staint (Galleridae: Lepidoptera). The LC-50 for larva of *C. cephalonica* was 5.02 IJ/ larva at $24\pm 1^\circ\text{C}$. There was a direct relationship existed between the number of nematodes applied and number invading the host. The maximum multiplication of 48,000 IJ / larva was recorded when *C. cephalonica* larva was inoculated was 5 IJ/larva. Grubs of *Oryctes rhinoceros* L. (Scarabaeidae: Coleoptera), *Rhynchophorus ferrugineus* Fabr. (Curculionidae: Coleoptera) and *Leucopholis coneophora* Burm. (Scarabaeidae: Coleoptera) were recorded as hosts under laboratory condition.

Key words:- Entomopathogenic nematode, morphometrics, infective juveniles, mortality, invasion efficiency, host range, multiplication rate, *Heterorhabditis indicus*.

INTRODUCTION

Heterorhabditid and Steinernematid nematodes are potentially useful biological control agents for numerous pests (Poinar, 1979). They transmit the bacteria which are lethal to the host. The infective juveniles can be cultured either on host or on artificial media. These nematodes can penetrate and kill many economically important pests. They are relatively insensitive to agricultural chemicals eg:- insecticides, fungicides and nematicides (Hara and Kaya, 1982; Rovesti and Deseo, 1991). Because of these useful attributes numerous surveys were conducted world wide and these nematodes were recovered from numerous habitats. The purpose of this study was to investigate the occurrence of entomopathogenic nematodes to obtain locally adapted isolates for developing biocontrol programmes against pests of coconut.

MATERIALS AND METHODS

Sample collection :- A total of 95 soil samples were collected from the rhizosphere of coconut and near cowdung pits during April, 1996. Each composite sample consisting of three random subsamples collected one metre away from the bole from a 15 cm deep stratum was placed in a polythene bag and taken to the laboratory.

Nematode isolation and identification: - The rice meal moth, *Corcyra cephalonica* Staint (Galleridae: Lepidoptera) was used as bait to isolate entomopathogenic nematode. Five to ten *C. cephalonica* larvae were placed at the bottom of a glass container by packing the soil over them. The glass container was kept at $24\pm 1^\circ\text{C}$. The insect mortality was recorded one week later. The assay was repeated using fresh insect in the same soil for another one week. Dead larvae from each sample was rinsed in sterile water and incubated in modified White trap (White, 1929) to collect emerging infective

juveniles. Cadavers without emerging juveniles were dissected to detect nematodes remaining inside the insect. The infective juveniles were exposed to five fresh *C. cephalonica* larva on a filter paper in a petri dish to verify pathogenicity. Different stage of the pathogenic isolates fixed in 5% Formalin were processed to glycerine by Seinhorst method and mounted in anhydrous glycerine. Glass wool or small piece of cover slip were used to prevent the flattening of the specimens. Measurements were made using an ocular micrometer.

Larvae of *C. cephalonica* were used as host for the multiplication of nematodes. The basic in vivo productin method by Woodring and Kaya (1988) was followed for multiplication, storage and quantification of entomopathogenic nematodes.

Mortality :- Full grown larvae of *C. cephalonica* were selected for the study. The pathogenic effect of the Kayangulam isolate of *Heterorhabditis indicus* to *C. cephalonica* larvae was determined following the procedure of Dunphy and Webster (1986) at $24\pm 1^\circ\text{C}$. The inoculum levels used were 2, 5, 10, 20 and 40 infective juveniles (IJ) per larva with ten larvae per replicate and ten replicate for each level. Separate control was also maintained. The mortality was recorded 72 hours after confinement of the insect larvae in the incubated chamber. The LC 50 value was calculated by probit analysis (Finney, 1962).

Life cycle :- The life cycle of the nematode from infective juveniles to second generation second stage juvenile was studied by using *C. cephalonica* as host at $24\pm 1^\circ\text{C}$. Each insect larva was confined to a petri dish (4.5 cm) over a moist filter paper and inoculated with 40 IJ/larva. At 12 hours interval ten larvae each were dissected and examined for the developmental stages. The experiment was repeated twice and results computed from the pooled analysis.

Invasion efficiency :- The invasion efficiency of Kayangulam isolate of *H. indicus* into *C. cephalonica* larvae was studied by placing a larva in a petri dish (4.5 cm) over moist filter paper. Different levels of inoculum viz., 2, 5, 10, 20 and 40 IJ/larva were added to double layer filter paper. There were forty replication per inoculum level and petri dishes were kept at $24\pm 1^\circ\text{C}$. After the death of the larva number of nematodes that had entered inside (both live and dead) the host was counted separately. The invasion efficiency was calculated as follows.

$$\text{Invasion efficiency} = \frac{\text{No. of nematodes recovered from dissection}}{\text{No. of nematodes per treatment}} \times 100$$

(Epsky and Capinera, 1993)

Multiplication rate:- This experiment was conducted as per the method described above. Upon death the larva was placed in white trap. The infective juveniles start emerging 8-10 days after infection. After first emergence, the infective juveniles were harvested daily until the production ceased. The production of infective juveniles was determined using formula given by Woodring and Kaya (1988).

Host range:- Studies were conducted by exposing ten grubs each of *Oryctes rhinoceros* L, *Rhynchophorus ferrugineus* Fabr. and *Leucopholis coneophora* Burm. to 3000 infective juveniles in an infection chamber in the method described above and cadavers were examined 8-10 days later.

RESULTS AND DISCUSSION

Nematode isolation and identification:- Out of 95 samples collected one each from

Block IV and near the cowdung pit showed positive result to nematode infection (Table 1). The infected larva turns to reddish brown in colour.

Based on the morphometrics, nematodes isolated from two samples were identified as *Heterorhabditis indicus* Poinar *et al.*, (1992) (Table 2). The morphological features of this Kayangulam isolate resembled closely with those described by Poinar *et al.*, (1992) but for some difference. The total length of herma phroditic female was found to be greater than for the original description (2.74 - 4.57 mm as against 2.3 - 3.1 mm) which was reflected in other body dimensions also. Difference was also noticed in length of the male. The original description is based on the specimens removed from *Galleria melonella* L. (Galleridae: Lepidoptera) larvae however, the present description of the Kayangulam isolate was made based on the specimens removed from *C. cephalonica* larvae. The difference in size may be due to the host used for the study. Similarly the difference in body dimensions of second generation females of *H. bacteriophora* was also noticed by Sivakumar *et al.* (1989) when *C. cephalonica* was used as host instead of *G. melonella*. In our study, the length of the infective juveniles were 577.49 - 594.96 μm as against 479 - 573 μm . Since the measurement was taken along with second stage cuticle which adds approximately 40 μm to the length of the third stage juveniles (Poinar *et al.*, 1992). In the infective juveniles, ratio E is less than 1.03 (1.021 in present study) and ratio F is less than 0.22 (0.211 in present study), thus confirming the criteria given by Poinar *et al.*, 1992 for diagnosing *H. indicus*.

Mortality : - Perusal of data in Table 3 indicates the pathogenic effect of *H. indicus* to *C. cephalonica* larvae. Complete mortality (100 per cent) was obtained at the highest dosage level of 40 infective juveniles per larvae and the mortality rate decreased with decrease in dosage (Table 3). The cadavers assumed reddish brown colour. The LC 50 value was 5.02 infective juveniles per larva of *C. cephalonica* at $24\pm 1^\circ\text{C}$.

Life cycle :- The life cycle of *H. indicus* showed that this nematode has a heterogonic cycle with a hermaphroditic oviparous (short generation) and an amphimictic ovoviviparous (long generation). The infective juveniles which have heavily sclerotized dorsal tooth invade the host and turn to fourth stage larvae (J4) who lacks sclerotization. The hermaphroditic females were produced 48-72 hours after infection. These females lay eggs initially, but later the eggs are retained and hatch in her body (endotokia matricida). The second generation male and female appear in about 5 to 6 days. The juveniles exit from female body in 8-10 days after infection at $24\pm 1^\circ\text{C}$. The second generation females are much smaller than hermaphroditic females (1.146 mm and 3.712 mm respectively).

Invasion efficiency :- The invasion efficiency of Kayangulam isolate of *H. indicus* decreased with the increase in inoculum density. The decrease in invasion efficiency could be due to the competition among the infective juveniles to enter inside the host. There was direct relationship between the number of nematodes applied and the number invading the host (Table 4). This result coincides with the findings of Epsky and Capinera (1993) who reported that the number of nematode invading the host was directly related to the concentration applied.

Multiplication rate: - When the *C. cephalonica* larva was inoculated @ 10 infective juveniles per larva, the multiplication rate was found to be 48,000 followed by 29,575 at 5 IJ/larva (Table 5). The multiplication rate was reduced at the lowest as well as the highest inoculum level. Razak and Sivakumar (1989) studied the influence of inoculum on multiplication of *Steinernema feltiae* (DD- 136 strain) on *C. cephalonica* and it was observed that the nematode multiplication was reduced at the lowest and highest level of inoculum. Similar study conducted by Karaunakar *et al.*, (1993)

showed that the dosage of 20 IJ of *H. indicus* per fifth instar larvae of Sugarcane internode borer, *Chilosacchariphagus indicus* (Kapur) yielded significantly highest multiplication rate of 2, 10, 283.3 IJ/larva followed by 1, 99, 472.5 @ 10 IJ/larva.

Host range:- Under laboratory conditions at 24±1°C Kayangulam isolate of *H. indicus* infected grubs of *O. rhinoceros*, *R. ferrugineus* and *L. coneophora*, pests of coconut in India. *Neoaplectana carpocapsae* (DD - 136) was reported to be effective against grubs of *O. rhinoceros*, *R. ferrugineus* and *L. coneophora* (Anon, 1971). However Zelazny (1985) reported that the grubs of *O. rhinoceros* were resistant to infection by *Steinernema feltiae* (= *Neoaplectana carpocapsae*). Pardede *et al.* (1992) reported the occurrence of *Heterorhabditis* sp in diseased *O. rhinoceros* grubs and its pathogenicity. *Heterorhabditis* sp was also used for controlling *R. ferrugineus* under laboratory conditions in Egypt. (Shamseldean and Elgawad, 1994).

Since *O. rhinoceros*, *R. ferrugineus* and *L. coneophora* were recorded as hosts for the Kayangulam isolate of *H. indicus* under laboratory conditions, further studies are need to find out whether this nematode significantly suppress the pest population under field condition and if so the method of applicatin for longer persistence, multiplication and dispersal under field condition should be standardised.

Table 1: Survey of entomopathogenic nematodes

Location	No. of samples collected	No. of samples yielding EPN
CPCRI (RS)	Block I -15	Nil
Kayangulam	II - 10	Nil
	III - 10	Nil
	IV - 10	1
	V - 10	Nil
	VI- 10	Nil
	VII - 10	Nil
	IX - 15	Nil
Cowdung pit	1	1
Vazhuvadi	3	Nil
Cowdung pit	1	Nil
Total	95	2

Table 2. Morphometrics of *Heterohabditis indicus*

Character	Hermaphroditic females (n=10)	Amphimictic females (n=10)	Males (n=8)	Infective juveniles* (n=10)
Total length (mm)	3.71(2.74-4.57) (2.3-3.1)	1.147(1.073-1.249) (1.2-1.8)	0.778(0.770-0.792) (0.573-0.788)	0.586(0.577-0.595) (0.479-0.573)
Greatest width (µm)	292.80(252.00-374.40) (107-145)	74.72(66.67-81.66) (76-113)	56.84(54.52-60.32) (35-46)	20.87(18.76-22.51) (19-22)
Length of stoma (µm)	9.81(6.09-15.22) (5-8)	8.16(6.67-9.66) (4-8)	3.19(2.38-3.48) (2-4)	-
Width of stoma (µm)	11.41(6.09-15.22) (6-10)	5.29(3.33-7.24) (5-8)	5.22(3.48-5.80) (4-6)	-
Length-Head to base of pharynx (µm)	186.98(152.15-243.44) (163-179)	143.80(126.65-157.49) (120-139)	93.96(85.85-103.24) (93-109)	123.08(120.06-125.69) (109-123)
Length-Head to excretory pore (µm)	188.67(179.54-273.87) (163-187)	124.72(120.70-130.36) (118-138)	102.66(89.32-116.00) (109-138)	103.42(100.37-105.99) (88-107)
Length-Head to Nerve ring (µm)	113.35(109.55-146.06) (104-123)	110.24(106.22-113.46) (88-96)	74.24(59.16-95.12) (72-85)	87.82(85.36-92.87) (72-85)
V	46.93(42.86-51.92) (45-50)	49.36(46.67-52.16) (40-53)	-	-
Length-tail (µm)	88.59(60.86-136.94) (72-110)	46.55(39.99-53.11) (66-88)	24.65(23.20-29.00) (24-32)	98.96(97.55-103.18) (93-109)
Width at anus/cloaca(µm)	59.51(42.60-69.99) (38-51)	20.40(16.67-24.14) (22-32)	27.84(25.52-30.16) (24-32)	- (93-109)
Length of spicule (µm)(SL)	-	-	42.63(40.60-45.24) (35.48)	-
Length of gubernaculum (µm)(GL)	-	-	19.72(17.40-22.04) (18.23)	-
Anal swelling extended from body (µm)	13.31(9.13-15.22) (5-14)	3.00(2.00-4.00) (1-3)	-	-
Reflexion of testis (µm)	-	-	78.30(61.48-95.12) (35-144)	-
GL/SL	-	-	0.46(0.42-0.51) (0.40-0.60)	-
a	-	-	-	25.82(23.59-27.31) (25-27)
b	-	-	-	4.69(4.58-4.75) (4.3-4.8)
c	-	-	-	5.46(5.71-5.98) (4.5-5.6)
d	-	-	-	0.83(0.79-0.84) (0.79-0.90)
e	-	-	-	1.02(0.97-1.070) (0.83-1.03)
f	-	-	-	0.21(0.19-0.23)

*Measurements were taken with the infective juveniles with second stage cuticle. Values in italics are the original values given by Poinar *et.al.*, 1992

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Table 3 Per cent mortality due to *H.indicus* on *C. cephalonica* larvae at 24±1°C.

Inoculum level IJ/larva	Per cent mortality
2	22.5
5	50.0
10	62.5
20	82.5
40	100.0

Table 4 Effect of inoculum level on invasion efficiency of *H.indicus* on *C. cephalonica* larva.

Inoculum level (IJ/larva)	No. of nematodes inside larvae			Invasion efficiency %
	live	dead	total	
2	1.2	0.4	1.6	100.00
5	2.8	0.6	3.4	68.00
10	5.4	1.4	6.8	68.00
20	10.5	1.4	11.9	59.50
40	20.6	1.7	22.3	55.75

*Average of ten larvae dissected.

Table 5 Effect of inoculum level on multiplication of *H.indicus* on *C.cephalonica* larvae (mean of 10)

Inoculum level IJ/larva	Production of IJ/Larva
2	8300
5	29575
10	48000
20	16403
40	5000

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Coconut Pests of National Importance

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Abstract

Among the different species of insects and mites recorded on coconut palm, the major ones of national importance are the rhinoceros beetle *Oryctes rhinoceros* L., the black-headed caterpillar *Opisina arenosella* Wlk. and the red palm weevil *Rhynchophorus ferrugineus* F. The bio-ecology, damage potential and the present status of the management of these pests are highlighted in this paper.

I. Introduction

The coconut palm, *Cocos nucifera* L. is infested by a variety of insect and mite pests. Nirula (1955) reviewed the world distribution of the pests of coconut and listed 106 insects and one mite infesting the palm. Kurian *et al.* (1979) listed 547 insect and mite species infesting the coconut palm and copra and furnished information on their distribution in various countries. The present status of the management of the major ones of national importance is reviewed in this paper.

II. Rhinoceros Beetle

The rhinoceros beetle, *Oryctes rhinoceros* L., is an ubiquitous and serious pest of the coconut palm infesting the unopened fronds and spathes. In Kerala alone 35-75% of the palms were found to be infested by the beetle in varying degrees of intensity (George *et al.*, 1979). The direct damage on spathes by the beetle alone causes 5.7% loss (Ramachandran, 1961; Ramachandran *et al.*, 1963). A field control trial laid out in an extensive area of 11 sq. km at Sathamkotta, Kerala, showed an increase in yield to the tune of 5 to 6 nuts per palm per year as a result of treatment of breeding sites of the beetles with insecticide (Sahasranaman *et al.*, 1966; Sahasranaman, 1969). Integrated management had yielded encouraging results in controlling the pest: (i) extraction of beetles during the peak period of pest abundance during the monsoon season (July-September), (ii) treatment of all the possible breeding sites of

the beetle such as cattle dung, compost, dead coconut logs and other decaying organic debris in and around the plantations, (iii) the prophylactic crown treatment of the main border palms with insecticide and sand mixture during the pre- and post-monsoon periods, and (iv) maintenance of field sanitation through disposal of the decaying organic matter. The data collected from the above experimental area revealed a reduction of leaf infestation from 26.25% to 4.74%, spathe infestation from 28% to 4% and fresh incidence from 45% to 5%. It was also observed that there was quicker reduction in pest infestation in this integrated control experimental area than that obtained by the individual method of control by the chemical treatment of breeding sites only (Kurian and Pillai, 1969).

Indigenous predators like *Santalus parallelus*, *Scarites* sp., *Harpalus* sp., *Pheropsophus* sp., *Agrypnus* sp., etc. feed on the eggs and/or early stage grubs of *O. rhinoceros*. The extent of pest suppression exerted by them was quite meagre in relation to the rate of multiplication of the pest. Attempts were, therefore, made for the introduction and colonisation of the exotic reduvid predator, *Platymeris laevicollis* Dist. from Zanzibar. This reduvid bug feeds on the adult beetles and, as such, is capable of numerically reducing the pest population. It is quite amenable to laboratory rearing. The predator was mass multiplied on biological hosts such as ground roaches, red palm weevil, coconut caterpillar, etc. and released on *Oryctes* infested coconut palms at regular intervals. There was remarkable reduction in pest incidence on palms receiving predator releases regularly. The observations of experimental sample palms revealed 13.06% leaf attack, nil spathe attack and 1.0% fresh incidence on spindle, as against 59.2%, 2.5% and 37.0%, respectively, of the pre-release condition. Presence of a higher proportion of dead beetles on the crowns of pest infested palms also indicated the efficacy of the predator. The release of *P. laevicollis* was also suggested as one of the components in the integrated control of the beetle and the same had been tested at Sooranadu, Quilon district, Kerala.

The green muscardine fungus, *Metarhizium anisopliae* produces epizootics in *Oryctes* population when climatic factors like low

temperature and high relative humidity conditions prevailed. A method has been developed to mass culture this fungus on tapioca chips and rice bran, supplemented with waste fish meal extract or urea, in specially designed large aluminium vessels (Mohan and Pillai, 1982). The fungal spores cultured in the laboratory could be applied to the breeding sites of the pest.

A baculovirus disease was found to occur in the natural population of *O. rhinoceros* in Kerala. The symptoms conformed to those of the baculovirus disease of the pest recorded by Huger (1966) in Malaysia and subsequently introduced into several of the South Pacific Islands to control the pest population (Zelazny, 1973; Bedford, 1977). The visual symptoms of baculovirus disease are: the midguts of diseased beetles will be white, swollen and filled with a mucoid milky fluid in contrast with very thin, brown midgut containing very little brownish fluid in healthy beetles. Other tests such as immuno-osmophoresis, bioassay and electron microscopy also confirmed the natural incidence of baculovirus infection in the population of beetles collected from different tracts of Kerala to an extent of 54.2% (Mohan *et al.*, 1983). However, the beetles collected from Minicoy, Lakshadweep were free of baculovirus incidence and this observation opened up the possibility of introducing baculovirus to the island for biological suppression of the pest. Work on this line was initiated in May, 1983 and indications of establishment of baculovirus in the natural population of beetles and in their breeding sites were obtained. Laboratory studies carried out at CPCRI have revealed that baculovirus infection results in reduction of longevity by 45% and fecundity by 95-100% of adult beetles. Baculovirus of *Oryctes* is claimed to be one of the most successful microbial control agents employed against an insect pest (Caltagirone, 1931).

III. Black headed Caterpillar

The leaf eating caterpillar *Opisina arenosella* Wlk. (= *Nepantia serinopa* Meyr.) is serious in the coastal and backwater tracts of India. Of late, this pest has appeared in severe proportions even in the interior tracts also in many states. The caterpillars live on the lower surface of leaflets in galleries made of silken

threads reinforced with leaf scraps and excreta and feed on the chlorophyll containing parenchymatous tissues of leaflets, which results in drying up of leaves and the consequent reduction in the yield of palms.

Generally outbreaks of the pest occur during summer months. Cutting and burning of badly infested leaves, spraying the palms with suitable chemicals which will be less toxic to beneficial indigenous natural enemies - parasites and predators - and subsequent release of laboratory reared parasites are the measures recommended for the management of this pest. Trunk injection or root feeding of systemic insecticides to the infested palms was also recommended as an effective method to combat this pest (Natarajan *et al.*, 1980; Ganeswara Rao *et al.*, 1980). Injury made to the palm trunk for administering insecticides will not be healed and as such, it is likely to pave way for subsequent infection by pathogenic fungi or infestation by red palm weevil, particularly in young palms. If reinfestation of the pest occurs it would be necessary to repeat the injection treatment. Another disadvantage of the injection/root feeding of systemic insecticides to palms is that harvesting of nuts (including tender nuts) has to be done prior to chemical application. It will be rather difficult to enforce the pretreatment harvesting of all nuts in small holdings.

By the intensive colonisation of laboratory bred indigenous parasites the pest could be brought under control in six years, time in Malabar and South Kanara (Rao *et al.*, 1948) and over a period of two years in the deltaic areas of East and West Godavari (Dharmaraju, 1952) of the erstwhile Madras state. According to Joy and Joseph (1977, 1978) the most important pupal parasites of *O. arenosella* are the chalcidids, *Brachymeria nosatoi* and *B. nephantidis*. Although the eulophid parasite *Trichospilus pupivora* was being mass multiplied and released in sizable numbers for several years now, its extent of natural parasitism was very meagre in southern Kerala (less than 1%) while it was 14% in north Kerala region. Pillai and Nair (1981) also found that more than half of the pest population was suppressed by the solitary pupal parasitoids. Of the different pupal parasitoids *Brachymeria*

nosatoj possesses the major attributes of an effective biocontrol agent of the pest. It is a sturdy parasite present in nature almost throughout the year. Its natural incidence is as high as 38.8% in some tracts even during summer months, which is the peak period of pest abundance. It has got greater searching ability and could locate and parasitise host pupae remaining inside the cocoons in silken galleries. Each parasite is capable of parasitising a good number of host pupae during its long life span of more than three months. A technique has been developed for mass culturing this parasite, which was hitherto considered to be not amenable to laboratory rearing (Pillai and Nair, 1982a).

Xanthopimpla punctata is another sturdy pupal parasite which exerts considerable check on pest population in some interior tracts of Tamil Nadu. *X. nana nana* is another potential parasite which is a new record on *O. arenosella*. In some localised tracts in Kerala its intensity is quite high (Pillai and Nair, 1983). Glass chimney method is suitable for rearing *Xanthopimpla* spp. using *Opisina* and *Anadevidia* pupae as hosts.

Nagarkatti (1973) observed that one obvious defect in the present parasite rearing programme for control of the coconut caterpillar was that only those parasites which could easily be bred in the laboratory has been mass bred and released, while very little had been done with those that were difficult to breed. For example, the prepupal parasite, *Elasmus nephantidis* is capable of locating and parasitising host larvae remaining inside silken galleries and it is well adapted to thrive even during summer season. It is a monophagous species with a high degree of stage specificity. If adequate supply of prepupal caterpillars of *O. arenosella* is ensured this parasite could be mass multiplied and released in pest infested fields. The bethylid, *Parasierola nephantidis* is another sturdy larval parasite which is capable of withstanding high temperature of summer months and exerting considerable check on pest population.

Attempts on colonisation of the exotic parasites like *Spoggosia bezziana* and *Eriborus trochanteratus* were made. Eventhough indications of establishment of the released parasites were obtained

hyperparasitism by *T. pupivora* and *B. nephantidis* in the former and by *B. nephantidis* in the latter were observed under the west coast conditions. Recent attempts on colonisation of the exotic tachinid *Bessa remota* were unsuccessful.

Adults and grubs of the carabid predator, *Parena nigrolineata* feed on *O. arenoella* caterpillars. A technique has been developed for the laboratory rearing of this predator. Studies on the rate of predation by different species of spiders found associated with *Opisina* infested coconut palms revealed that *Cheiracanthium* sp., *Rhane indicus* and *Sparassus* sp. are the dominant species of spiders exerting considerable check on pest population in the field.

Proper monitoring of the pest population is essential for the effective management of this pest. Necessary techniques for the same had already been developed. The caterpillars/pupae present in 40-60% of leaflets of the lowest 20% leaves are counted and from this the estimates of pest population could be made using the sampling procedure developed by George *et al.* (1982). For assessing the intensity of natural pupal parasitism in addition to recording emergence of parasites from live pupae, a method of examination of emergence holes present in empty pupal cases was developed. This method was found to be quite useful in assessing the aggregate natural parasitism of pupa during the entire pest generation (Pillai and Nair, 1982b).

IV. Red Palm Weevil

The red palm weevil, *Rhynchophorus ferrugineus* F. generally attacks palms of the age group 5-20 years. Prevention of entry of weevil is possible by treatment of wounds on palm trunk, cut ends of leaf stalk, etc. with BHC or BHC + coal tar or by cutting the petioles of young palms at a distance of 120 cm away from the stem. Prophylactic crown treatment with insecticide and sand mixture, curative treatment of the infested palms with insecticide and trapping the floating population of weevils with tender coconut logs smeared with fresh toddy fermented with yeast granules or acetic acid or pineapple + molasses + yeast are the methods recommended

Present Status of Research on Root (Wilt) Disease of Coconut

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Abstract

The root (wilt) disease of coconut has caused an annual loss of over 340 million nuts in Kerala State and has spread to the adjoining Tamil Nadu State. The symptoms and diagnostic tests, etiology and management of the disease are highlighted. Effective quarantine measures are needed to prevent the movement of planting material from disease-prevalent to disease-free areas. Until effective control measures are developed the palms in the affected area should be rejuvenated and the disease contained within its present geographical limits.

I. Introduction

Coconut root (wilt) disease was first reported, following the great floods of 1882, in three isolated pockets about 50 km away from each other in the erstwhile State of Travancore (Butler, 1908; Varghese, 1934). Since then it has spread from the original foci of infection and now occupies a contiguous area covering eight out of the thirteen districts in Kerala. According to a survey concluded in 1972 more than 30% of the 0.5 million ha under coconut in Kerala are affected by the disease causing an annual loss of over 340 million nuts (Bavappa *et al.*, 1982). The disease is non-lethal, but debilitating and palms of all age groups are affected. Recent surveys on the distribution of the disease revealed occurrence of diseased palms in North Kerala far from the core of the diseased tract and also in the adjoining State of Tamil Nadu.

II. Symptoms

The diagnostic symptom of the disease is the characteristic bending of the leaflets termed flaccidity. Yellowing and necrosis are other associated symptoms (Radha and Lal, 1972). As early as 1908 Butler held root rot as a major symptom. Drying up of the spathe and necrosis of spikelets from tip downwards even in

unopened inflorescence was noticed in certain cases. It is worth mentioning here that necrosis of inflorescence is one of the consistent symptoms of lethal yellowing disease in the Caribbean and West Africa.

III. Diagnosis

Two diagnostic tests, one based on positive serological reaction of extracted sap (Solomon *et al.*, 1983a) and another dependent on the differential stomatal resistance (Rajagopal *et al.*, 1984), have been standardised to identify diseased palms even before visual symptoms are manifested.

The sporadic occurrence of the disease and the pattern of spread are more suggestive of the involvement of a pathogen transmitted by biological agents to be the cause of the disease than nutritional or physiological factors. The changes in the physiological parameters in the diseased palms are more indicative of a pathogen mediated host metabolism.

IV. Etiology

i. Nutritional aspects : Extensive studies carried out on the nutritional status of coconut palm in relation to the disease tend to rule out the role of any major nutrient in the disease incidence (Cecil, 1975). The effect of micronutrients and heavy metals in the disease syndrome has not been investigated.

ii. Physiological studies : Some of the malfunctions encountered are higher transpiration and respiration rates, lower photosynthetic rate and electrolyte release in diseased palms. The accumulation of reducing and non-reducing sugars in leaves as reported is suggestive of impaired translocation normally encountered as a result of infection by vascular pathogens. Accelerated phenol metabolism is yet another feature known for many pathogenic diseases (Mathew and Dwivedi, 1981).

iii. Mycological studies : Since Butler's report implicating fungi, possibly *Botryodiplodia* as the probable etiological agent, *Rhizoctonia solani*, *R. bataticola*, *Fusarium equiseti*, *Cylindrocarpon effusum* and *C. lucidum* were isolated from the

roots of wilt affected palms (Lily and Joseph, 1981). These fungi on inoculation to healthy seedlings produced rotting of roots but failed to induce the foliar symptoms characteristic of the disease. These fungi may therefore not have a primary role in the causation of the disease.

iv. **Bacteriological studies:** Srivastava *et al.* (1969) reported bacterial streaming in vascular tissues of roots of diseased palm and isolated *Pseudomonas* sp. However, in subsequent studies an unconventional phytopathogenic bacterium, *Enterobacter cloacae* was isolated from the roots. Culture filtrates of *E. cloacae* induced toxic symptoms on detached coconut leaflets and caused reversible wilting in tomato seedlings (Jayasankar *et al.*, 1981).

v. **Nematological studies:** Implication of a soil bound pathogen or a pathogen transmitted through a soil borne vector warranted nematological studies. Analysis of coconut soils revealed the presence of 35 genera including species of *Xiphinema*, *Longidorus* and *Trichodorus*, known virus vectors, and the burrowing nematode *Radopholus similis* (Mathen *et al.*, 1970). The inconsistent pattern of occurrence and the low population density of these virus transmitters in the diseased tract, when viewed with the pattern of spread of the disease, tend to rule out the role of nematodes in the transmission of the disease. Pathogenicity experiments with *R. similis* on coconut seedlings while establishing the formation of root lesions leading to extensive rotting of roots and stunting of the plants failed to induce the root (wilt) syndrome (Koshy, 1981).

An elaborate pathogenicity experiment with these suspected biological agents - fungi (*Fusarium equiseti*, *Cylindrocarpon effusum*), nematode (*R. similis*) and bacterium (*E. cloacae*), singly and in various combination, is under observation in our Institute.

vi. **Virological aspects:** A viral etiology for the disease was proposed by Nagaraj and Menon (1954) based on the systemic nature of the disease and resemblance of the disease symptoms to other known plant virus diseases. Positive transmission of the