

## FINAL REPORT

I YEAR ( 2012—2013)

“Development of Integrated Pest Management with special emphasis on Biological Agents for the Control of the Red Palm Mite (*Raoiella indica* Hirst) on coconut in Trinidad and Tobago”

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## EXECUTIVE SUMMARY

Entry of Red Palm Mite (RPM), *Raoiella indica* into the Caribbean region in 2004 and Trinidad in 2006, reflections of impact of this Invasive Alien Species (IAS) in agriculture, lead to the Memorandum of Understanding (MOU) between Indian Government and the Government of Trinidad and Tobago and eventually, Dr. A. Sujatha, Principal Scientist (Entomology) has been deputed as an Indian Technical Economic Cooperation (ITEC) expert for one year to assist in the control of Red Palm Mite in Trinidad and Tobago. Estimation of RPM damage, identification of pest and natural enemies, development of protocol for rearing of RPM, *Amblyseius largoensis*, a natural enemy of RPM and capacity building of staff are the major objectives of the project.

The estimated RPM infestation in different locations of Trinidad through the survey was varied from 1.65 to 46.8 per cent of leaf let. RPM population count ranged from 7.0 to 11,452 per leaflet, whereas the *A. largoensis*, a natural enemy of RPM recorded was 1 to 19 per leaflet. From the survey in Tobago it was found that the percentage infestation ranged from 2.5 to 81.0 per cent of leaflet. The RPM number per leaflet ranged from 892 to 8366 and natural enemies from 0.33 to 11.0. The predatory mite *A. largoensis* (phytoseiidae-Acari) was identified as a potential indigenous natural enemy of the RPM. In addition to RPM pest, prevalence of Red Ring Disease, sucking pests, nutritional disorders, stress to the palm, soil erosion with palms at shores of the ocean, lack of awareness about coconut cultivation technologies among the farming community *etc.*, were found to be factors contributing to the decline of the coconut industry in Trinidad and Tobago.

Laboratory protocols for rearing RPM, *A. largoensis*, a natural enemy of RPM were standardized. Recommended components of Integrated Pest Management (IPM) for the RPM were implemented in the demonstration plots at Research Division, Centeno. Training of staff in the Ministry of Food Production in terms of pest diagnosis, skills for laboratory rearing of RPM and its natural enemy, *A. largoensis*, bio-control laboratory maintenance were accomplished as a part of capacity building of staff. The outcome of the project was brought in the form of publications; extension activities coupled with distribution of printed material would be helpful for creating awareness about coconut cultivation among the technical staff, agricultural officers and extension workers.

Continuity of mass culturing of *A. largoensis* in the laboratory, field efficacy studies, confirmative studies and bio-agents were recommended. Field maintenance of demonstration plots with sequential application of inputs in the long run would result in the crop improvement and suppression of RPM. A large scale rejuvenation programme of coconut crop to replace dead, senile and unproductive palms with elite germplasm was suggested for plant health improvement and increased productivity. Establishment of farm level processing units or cottage industries for value addition to the coconut products would create opportunities for youth employment and boosts the crop economy. Sharing of information was strongly emphasized to build up the knowledge and make the coconut industry more productive to sustain the ‘Tree of Life’ for the benefit of mankind.

## *ACKNOWLEDGEMENTS*

I would like to express my sincere thanks to Government of India, Director General of Agriculture for considering my candidature for one year deputation as an ITEC expert to work with the Government of Trinidad and Tobago.

My special thanks are due to His Excellency, Mr. Malay Mishra, High Commissioner of India; Dr. H. Arlington D. Chesney, Executive Director, CARDI for their efforts in negotiating and being instrumental in adumbration of the project on RPM.

I am grateful to Dr. C.V.S.K. Sarma, I.A.S., the Honourable Vice Chancellor, Dr. B. Srinivasulu, In-Charge-Of Director of Research and Registrar of my parent organization, the Dr. Y.S.R. Horticultural University, West Godavari district, Andhra Pradesh, India for the acquiescence of my deputation.

I am delightful to extend my heart-felt thanks to Dr.LiloryMcComic, former Director of the Research Division, Mrs.AudineMootoo, Director of Research Division, and Dr. Mario Fortune, Chief Technical Officer, Ministry of Food Production (MFP), Trinidad and Tobago for providing infrastructural facilities, inputs required for undertaking this project and a team of staff members for assisting me in the project work.

I express heartfelt gratitude to Dr. Janet Lawrence and Mr. Bruce Laucknar, Scientists at CARDI for their logistic role in the implementation of the project, overviewing the project technically to make it scientifically fit, complete and place the outcome of the project on records as technical information.

I am pleased to underscore names of RPM Project team with special appreciation to Ms. Teresa Rosemond, Agricultural Entomologist; Mr.FarzanHosein Plant Pathologist; Ms. Petal Ram Agricultural Officer I; Mr MukashRamdhanie, Agri. Officer I; Ms.CareleneLakhan, Agricultural Officer I; Mr Nigel Allen, Laboratory Attendant; Mr. Jain Babooram, Laboratory Attendants; Ms. Felicia Forbes and Ms. Emma Thomas OJT Trainees whose untiring efforts made this project successful.

Also deserving of special mention are the administrative staff from MFP who helped directly and indirectly with the RPM project.

Dr A. Sujatha ITEC Expert  
Principal Scientist [Entomology] and ITEC Expert,

**Final Report of the Project entitled “Development of Integrated Pest Management with special emphasis on Biological Agents for the control of RPM on coconut in Trinidad and Tobago”.**

**Duration:** One year

**Period:** 1st March 2012 to February 2013

**Location:** Central Experimental Station, Ministry of Food Production (MFP)  
Caroni North Bank Road, Centeno

**Associates:** Mr. Farzan Hosein  
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## 1. INTRODUCTION

### *Global*

Coconut is an important tropical perennial crop which is an economically important predominant crop in Asia, Pacific and African countries. Coconut palm, a crop of small holders in many countries has become an integral and inseparable part of many a culture, tradition and religion. In Sanskrit, coconut palm is known as “KALPA VRIKSHA” which means “the tree which gives all that is necessary for living” as every part of the coconut tree is useful to man in many ways. It not only provides sustainable income to millions who are directly and indirectly depending on this crop but also helps to provide food security, nutritional security, and alleviate poverty. Coconut is grown in more than 93 countries around the world in an area of 12.29 million ha producing 57,943 million nuts or 11,439 million tons of copra equivalent as of 2004. Though coconut oil is the major export product in the global market, the share of other products like copra meal, coco-chemicals and activated carbon have been on increasing trend in the export products. Virgin coconut oil (VCO), because of its wider use in functional foods, pharmaceuticals, nutritional and cosmoceutical fields has got good market potential.

### *Trinidad and Tobago*

In Trinidad and Tobago, coconut was one of the major export tree crops which occupied approximately 4,000 ha in small and large holdings and contributed its share in the country's economy. The coconut crop being an integral part of beaches in Caribbean region, found a potential source of income in the tourism industry. After the entry of Red Palm Mite (RPM), *Raoiella indica* into the Caribbean region in 2004 and Trinidad in 2006, it is considered one of the factors hampering coconut production. Further, impact of this Invasive Alien Species (IAS) has been reflecting in agriculture, cultural heritage, ecotourism, biodiversity and the industrial sectors of the country. This backdrop lead to the Memorandum of Understanding (MOU) between Indian Government and the Government of Trinidad and Tobago and eventually, Dr. A. Sujatha, Principal Scientist (Entomology) has been deputed as an Indian Technical Economic Cooperation (ITEC) expert for one year to assist in the control of Red Palm Mite in Trinidad and Tobago with the following Terms of Reference (TOR).

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## Terms of Reference (TOR)

### **Pre-Visit (preparation) - November 2011 – January 2012**

- Review the current available information on coconut cropping systems and the ecology of the Red Palm Mite (RPM) in Trinidad and Tobago
- Identify potential exotic natural enemies for the management of the RPM in Trinidad and Tobago.

### **During Visit - Visit starting early 2012 and continuing for a minimum of 1 year, possibly two years**

- Liaise with technical personnel and producers to review the agro-ecology of coconut cropping systems in Trinidad and Tobago
- Liaise with key stakeholders within the coconut industry
- Review the current status of the endemic Natural Enemies (NE) identified
- Determine the efficacy of the endemic NE (predatory-prey interactions)
- Develop rearing techniques for the host and NE
- Document rearing protocols developed
- Training technical staff in various aspects of biological control – e.g. rearing, release, efficacy, assessments

*If classical biological control is necessary then the following activities will be pursued*

- Advise on the quarantine handling of the potential exotic NE including the upgrade of the quarantine facilities, permits and the agencies from which clearance is required (in India)
- Assist with the development and initiation of Pest Risk Analysis (PRA/s) for the exotic NE/s that are to be imported
- Develop protocols for laboratory and field simulated evaluations on the efficacy of the exotic NE
- Conduct laboratory and field simulated evaluations on the efficacy of the exotic NE

Consequently, the project “Development of Integrated Pest Management with special emphasis on Biological Agents for the Control of the Red Palm Mite (*Raoiella indica* Hirst) on coconut in Trinidad and Tobago” was designed for one year reflecting the following objectives:

## 2. OBJECTIVES

- Estimation, identification of pest and natural enemies
- Mass multiplication, release and monitoring of natural enemies
- Standardization and documentation of a rearing technique of natural enemies
- Capacity building of staff
- Strengthening linkages with the coconut farmers association and other stakeholders
- Development of an Integrated Pest Management (IPM) protocol

In view of the fast approaching closing date of the project, few objectives were redefined in the third quarter to fit into the time frame.

- Development of protocol for rearing of RPM, *Raoiella indica*
- Development of protocol for rearing of *Amblyseius largoensis*, a natural enemy of Red Palm Mite
- Development of protocol for studying feeding efficiency of *A. largoensis*
- IPM demonstrations with cultural and pest control technologies in different locations can be organized by the Ministry of Food Production (MFP) (initially proposed in farmer’s field)
- Guidelines for release of natural enemy in the RPM infested fields
- Preparation of information material (factsheet, pamphlet, manual etc.) for farmers on cultural practices in context of pest management



### 3. ABOUT RED PALM MITE

#### <sup>1</sup>Scientific Classification of Red Palm Mite

- Kingdom: Animalia
- Pylum: Arthropoda
- Subclass: Acari
- Order: Trombidiformes
- Family: Tenuipalpidae
- Genus: *Raoiella*
- Species: *indica*
- Binomial Name: *Raoiella indica* Hirst, 1924

The Red Palm Mite, *Raoiella indica* Hirst., also known as the coconut mite, coconut red mite, red date palm mite, leaflet false spider mite, frond crimson mite, scarlet mite is also an important pest of coconuts, date palm, other palm species (infesting over 32 species of palms) and bananas, heliconias, gingers, beans, and durian in different parts of the world. It is considered that the pest has serious consequences for the coconut, ornamental palm and banana industries of the Caribbean islands. Damage to coconuts results in 70% yield reduction and possibly job losses leading to a major socio-economic problem for some of the islands.

#### **Distribution:**

The RPM is indigenous to Egypt, India, Iran, Israel, Mauritius, Oman, Pakistan, Philippines, Réunion, Saudi Arabia, Sri Lanka, Sudan, Thailand, and the United Arab Emirates. Within the Caribbean, the mite has been detected in most islands (Fig.1).

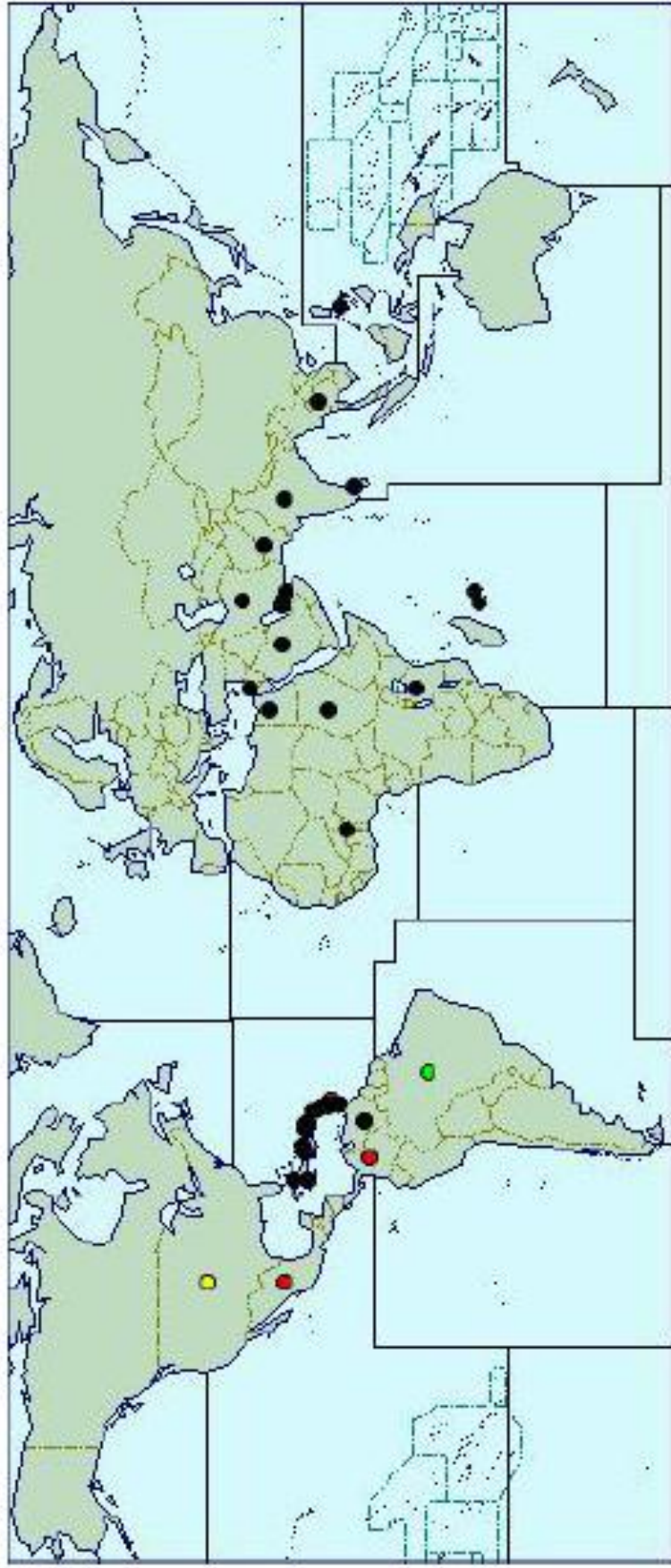
#### **Damage:**

The RPM forms colonies on the undersides of leaves, feeds on the contents of the cells causing localized yellowing. Infestation starts in the lower whorl of the leaves and as the population increases it spreads to upper whorls; this results in the yellowing of the plant and later necrosis.

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<sup>1</sup> From internet

**Figure 1: Distribution of Red Palm Mite in different countries**



Severe infestation leads to fruit dropping, loss of vitality, aborted inflorescences and eventually a reduction in yield.

**Dispersal:**

The mites disperse by wind as well as on infested plants or plant material, landscape utensils, handicrafts, Caribbean palm souvenirs fashioned from coconut leaves such as hats, baskets, rugs, bowls, purses etc. Tropical storms and hurricanes can distribute this mite over wide areas.

**Description:**

The RPM can be identified by its bright red colour, flat spatulate body, long spatulate setae, and droplets on the dorsal body setae. There is also a noticeable absence of the webbing associated with numerous other spider mites. During all stages of life, this species is red, with adult females often showing black patches on their backs after feeding.

#### **4. ACTIVITIES**

##### **A. Field Studies**

###### *(i) Survey in Trinidad and Tobago:*

A roving survey was conducted in Trinidad and Tobago to determine the status of coconut production system, the extent of damage, the population levels of RPM and its natural enemies; species of natural enemies present. The palms sampled for the survey were located in different eco-systems which include hills, valleys, slopes, swamps, sea coasts and normal cultivated lands. Locations were selected randomly on the roadside. Two palms were selected randomly from each location. From each palm, six leaflets were sampled from three different levels of palm, @ two leaflets/ top, middle and lower whorls, respectively.

Population counts of RPM and its natural enemies on each leaflet were assessed in two ways (1) Manual counting by observing the abaxial surface of entire leaflet through the stereo zoom microscope and recording different life stages of RPM and its natural enemies present (2) Digital counting by focusing the scalar digital camera with 25 times magnification at 12 random spots on the abaxial surface of the leaflet and counting on the

monitor by uploading the pictures into the computer. The average number of RPM and its natural enemies/leaflet was determined by manual counting whereas per unit area i.e.  $\text{cm}^2$  was obtained in digital pictures by counting on the monitor through the computer. Egg, larval, protonymphal, deutonymphal and adult stages of RPM put together were considered as RPM count and similarly all the stages of natural enemies of individual species were taken as count of natural enemies. The average count of RPM and natural enemies per  $\text{cm}^2$  area of leaflet was worked out from the counts of 12 spots of each leaflet, based on the leaf area exposed in digital camera. Later average count for leaflets from top, middle and lower levels of the crown was arrived individually and average count for unit area i.e.  $\text{cm}^2$  on single leaflet for each sample palm was derived. Finally population counts of RPM and its natural enemies per leaflet as well as per unit area of leaflet were assessed for each location surveyed from two palms data obtained in the respective locations.

The extent of damage caused by RPM on the foliage was expressed as percentage of yellowing or percentage of infestation of leaflet and it was assessed by comparing the damaged area to the healthy area (green colour) of the leaflet. The infestation levels were graded as zero, low, medium and high as per the scale given hereunder:

S. No.	% of Yellowing or Infestation of Leaflet	Scale
1	0.0	Zero
2	0.1 to 15.0	Low
3	>15.0 to 50.0	Medium
4	> 50.0	High

As per the scale of infestation, the particular location was categorized under zero, low, medium or high scale of infestation.



A location was considered as hot spot area where percentage of yellowing or infestation of leaflet was >20 per cent or Red Palm Mite population count exceeds 400/leaflet.



Plate 1: Survey in Constantine estate in Cedros shores



Plate 2: Data collection in the field

*a. Estimation of RPM damage levels and collection of natural enemies in Trinidad:*

During the survey, leaf samples were collected for estimation of extent of Red Palm Mite damage, population levels of RPM and natural enemies, and collection of natural enemies from thirty-one locations in seven counties. The RPM infestation in different locations varied from 1.65 to 46.8%, population count ranged from 7.0 to 11,452 per leaf let whereas natural enemy population recorded was 1 to 19 no/leaf let. The occurrence of RPM population/unit area was recorded from 0.008 to 24.26/cm<sup>2</sup> leaf area (Table: 1) in Trinidad.





**Plate 3: Yellowing due to Red Palm Mite**



**Plate 4: RPM colony**



**Plate 5: Yellowing symptoms of coconut trees on leaves due to RPM damage**



**Plate 6: Yellowing and necrotic symptoms**



Information drawn from the survey provided the status of RPM as well as natural enemies of RPM in different locations. It was found that 7 locations *i.e.* Siparia, 19<sup>3</sup>/<sub>4</sub> Blanchisseuse, Freeport, Golconda, Paria Main Rd., Las Cuevas and La Fillette are free from RPM infestation, and 13 locations namely Valencia, Madamas, Maracas Bay, Wallerfield, Maloney, Valsayn, Maraval, Chaguaramas, Kelly Village, Charlieville, Macaulay, Waterloo and Golconda Ring Rd. with low infestation and 11 locations (Cumana, L'Anse Noire, Guiaco, Manzanilla, Guayaguayare, Mayaro, Blanchisseuse, Bonasse, Columbus Bay Rd., Icos and Diego Martin) with medium infestation of RPM out of 31 locations surveyed in Trinidad (Table: 2)

**Table 2: List of locations indicating status of Red Palm Mite in Trinidad**

Scale of infestation	Nil	Low	Medium	High
% infestation of leaflet	0.0	(0.1% to 15.0%)	(>15.0% to 50.0%)	(> 50.0 %)
Name of the location	1. Siparia 2. 19 <sup>3</sup> / <sub>4</sub> Blanchisseuse 3. Freeport 4. Golconda 5. Paria Main Rd. 6. Las Cuevas 7. La Fillette	1. Valencia 2. Madamas 3. Maracas Bay 4. Wallerfield 5. Maloney 6. Valsayn 7. Maraval 8. Chaguaramas 9. Kelly Village 10. Charlieville 11. Macaulay 12. Waterloo 13. Golconda Ring Rd.	1. Cumana 2. L'Anse Noire 3. Guiaco 4. Manzanilla 5. Guayaguayare 6. Mayaro 7. Blanchisseuse 8. Bonasse 9. Columbus Bay Rd. 10. Icos 11. Diego Martin	Nil

*b. Hotspot areas of RPM in Trinidad:*

Based on the RPM population and the percentage yellowing of leaf, the following 13 locations from 6 counties (North, South and Central parts of Trinidad) are found to be the hot spot areas of the pest high levels of RPM populations i.e. more than 400 per leaflet or high infestation per cent of leaflet (>20%) were recorded in 13 locations; Bamboo, Blanchisseuse, Valencia, Paria Main Road, Las Cuevas, Diego Martin, Cumana, L'Anse Noire, Bonasse, Columbus bay, Manzanilla, Mayaro and Waterloo which are spread over in all types of geographical area in North, South, East and Central parts of Trinidad. It should be highlighted that surveys area snap shot in time and as such further roving as well as fixed plots surveys at periodical intervals throughout the year for longer periods are required to get better understanding of the pest scenario, seasonal abundance/ occurrence and nature of pest (Fig.2) (Table: 3).

Figure - 2 : Hot spot areas of Red Palm Mite in Trinidad



**Table 3: Hotspot areas of red palm mite pest in Trinidad**

S. No	County	Name of location	Intensity of RPM	
			pop./leaflet (No.)	Infestation of leaflet (%)
1.	<b>NORTH</b> St. George	i. Bamboo	4258	10.0
		ii. Blanchisseuse	0	27.1
		iii. Valencia	2628	6.6
		iv. Paria Main Road	11452	0
		v. Las Cuevas	1011	0
		vi. Diego Martin	438	35.6
2.	St. David	i. Cumana	977	24.3
		ii. L'Anse Noire	980	46.8
3.	<b>SOUTH</b> St. Patrick	i. Bonasse	1163	20.0
		ii. Columbus bay	2110	39.3
4.	<b>EAST</b> St. Andrew	i. Manzanilla	154	20.9
5.	Mayaro	i. Mayaro	8284	16.7
6.	<b>CENTRAL</b> Caroni	i. Waterloo	397	1.65



*Cataloguing, identification and status of the indigenous natural enemies:*

The natural enemies collected from the field surveys are catalogued in the laboratory. *A. largoensis* was identified a predatory mite (order: Acari, family: Phytoseiidae) as the most predominant natural enemy among the field collected natural enemies. It's occurrence in the field was found in the ratio of 1:3 to 1: >100 of host population and noticed in 19 locations out of 31 locations surveyed. The other predatory mite, the *Bdella* sp. a member from Bdellidae family collected from RPM colonies in 8 locations out of 31 locations surveyed and is present in the ratio of 1:6 to 1: 23. The *A. largoensis* is associated with higher populations of red palm mite whereas the *Bdella* sp. is associated with lower populations of red palm mite. In few locations eggs and immature stages of chrysopid which is an insect predator were found in RPM colonies (Table: 4).



Plate 7: Natural Enemy of the RPM  
*Amblyseius largoensis*



Plate 8: Natural Enemy of the RPM  
*Bdella* sp.

*d. Estimation of RPM damage levels and collection of natural enemies in Tobago:*

A total of 22 locations were surveyed in Tobago covering six counties; St. Andrew, St. David, St. John, St. Mary, St. Patrick and St. Paul. The presence of RPM was recorded in all the 22 locations surveyed (Fig.3). From the data, it was found that the percentage yellowing ranged from 4 to 90 per cent of the palm and 2.5 to 81.0 per cent of leaflet. Population counts of RPM ranged from 2.0 to 87.0/cm<sup>2</sup> and that of natural enemies from 0.03 to 3.0/cm<sup>2</sup>. Whereas, that of RPM number per leaflet ranged from 892 to 8366 and natural enemies from 0.33 to 11.0 (Table: 5).

**Table 4: Status of Natural Enemies of RPM in Trinidad and Tobago**

Name of Natural Enemy	Trinidad (31 locations)				Tobago (22 locations)			
	Occurrence (% of Locations)	No/ Leaflet	No/cm <sup>2</sup>	Ratio	Occurrence (% of Locations)	No/ Leaflet	No/cm <sup>2</sup>	Ratio
<i>Amblyseius largoensis</i> (Acari: Phytoseiidae)	61.2	1-25	0.01-0.08	1:3	77.3	0.33-11.0	0.01-3.0	1:17 to 1:800
<i>Bdella</i> sp. (Acari: Bdellidae)	9.7	1-16	0.002-0.08	1:6 to 1:23	9.0	0.16-0.3	-	-
Chrysopid	6.5	-	-	-	9.0	0.3-4.16	0.3	-
Coccinellid	-	2	0.02	-	4.5	0.16	0.04	-

TABLE - 5: SURVEY IN TOBAGO: POPULATION COUNTS OF RPM AND NATURAL ENEMIES AND % YELLOWING

NO.	LOCATION	POPULATION /15sam <sup>2</sup> (No)				POPULATION /sam <sup>2</sup> (No)				MANUAL COUNT			YELLOWING %		REMARKS	
		PEST		NATURAL ENEMIES		PEST		NATURAL ENEMIES		Ambly	RPM	Ambly	RPM	Leaflet		Palm
		RPM	Ambly	Ambly	Others	RPM	Others	Ambly	Others							
1	Fig Farm Rd, Goldborough	0.00	0.00	0.00	-	0.00	0.00	0.00	0.00	0.33	0.70-1.5k	3.50	-	RPM present		
2	Fig Farm Rd, Goldborough	0.00	0.00	0.00	-	0.00	0.00	0.00	0.00	0.00	-	3.30	4.00	Seen Chrysopid, RPM present. Many Chrysopids & Ambly (10-15 Ambly per leaflet), RPM present		
3	Kendal, Roxborough	2.30	0.00	0.00	-	2.00	0.00	-	-	0.00	-	2.50	-	RPM present		
4	Indian Well, Moriah	19.81	0.08	0.08	-	87.00	1.00	-	-	1.00	-	80%	90.00	More Ambly & mites (8 per leaflet)		
5	#143 North Side Rd, Moriah	22.25	0.17	0.17	-	19.00	0.10	-	-	5.16	-	60%	60.00	RPM present		
*6	Moriah Rd	0.00	-	-	-	-	-	-	-	-	-	-	>50.00	RPM Euvivae present		
7	Costers Recreation Facility	9.67	0.01	0.01	-	8.00	0.01	-	-	0.30	-	16.67	30.00	RPM present		
8	English Man's Bay, North Side Rd	0.00	-	-	0.30-1.00	-	-	-	-	0.16	0.30-1.00	-	-	RPM absent		
9	North Side Rd, Parliament	5.13	0.11	0.11	-	4.00	0.10	-	-	5.00	-	44.17	-	RPM present		
10	North Side Rd, Parliament	24.16	0.92	0.92	-	21.00	1.00	-	-	5.83	-	80.82	-	RPM present		
*11	L'Anse Fourmi	-	-	-	-	-	-	-	-	-	-	-	-	Lots of RPM & Ambly		
*12	Hemitage	-	-	-	-	-	-	-	-	-	-	-	-	RPM present		
*13	Charlottesville	-	-	-	-	-	-	-	-	-	-	-	-	RPM present		
		70sam <sup>2</sup>														
14	Spotside Main Rd	15.40	0.00	0.00	0.03-1.00	20.00	1.00	0.30-1.00	1.20	0.03-1.00	28.3	25.00	-	-	RPM present	
*15	L'Anse D'Or	-	-	-	-	-	-	-	-	-	-	-	-	RPM present		
*16	Richmond Village	-	-	-	-	-	-	-	-	-	-	-	-	RPM present		
17	Big Buolet/Minister Bay	12.71	0.00	0.00	-	17.00	0.00	-	1.20	1.20	-	27.5	30.00	RPM present		
18	Lambert Beach, Old Midford Rd	4.78	0.00	0.00	-	6.00	0.00	-	3.00	3.00	-	7.50	17.50	Seen Chrysopid, RPM present		
19	Magdalenia Resort	38.5	0.00	0.00	-	50.00	3.00	-	5.60	5.60	-	13.33	30.00	Seen Coccinellids, RPM present		
20	Croon Reef Rest House	8.80	0.03	0.03-1.00	0.04-1.00	11.00	0.40	0.04-1.00	3.70	4.16-1.00 0.16-1.00	21.7	40.00	-	Plenty Ambly, Coccinellids, RPM & RPM Euvivae		
21	Pigeon Pt Beach Resort	11.0	0.02	0.16-1.00	0.16-1.00	14.00	0.03	-	11.00	0.16-1.00	11.7	25.00	-	Plenty Ambly, RPM present, Ambly, Chrysopid, lacewing present & RPM present		
*22	Mt. Irvine Hotel	-	-	-	-	-	-	-	-	-	-	-	-	-		

\* All Locations 6, 11, 12, 13, 15, 16 & 22 no. leaf samples could not be collected as they are on the hill  
Average area of a leaflet = 44cm<sup>2</sup>



**Plate 9: Symptoms of RPM damage**



**Plate 10: Road side location in Tobago**

From the survey in Tobago it was derived, two locations (Moriah Road and English Man's Bay) were free from RPM, 6 locations (Pig Farm Rd. Goldsborough a, Pig Farm Rd. Goldsborough b, Kendal, Roxborough, Lambeau Beach, Old Milford Rd., Magdalena Resort and Pigeon Pt. Beach Resort) were found with low infestation, 5 locations (Castara Recreation Facility, Parlatuvier, Speyside Main Rd., Big Bacolet/ Minister Bay and Cocoa Reef Rest House) medium level of infestation and 5 locations (Indian Walk, Moriah, #143 North Side Rd. Moriah and Parlatuvier) with high infestation. Most of the locations are with few numbers of palms on slopes of hills growing naturally (Table: 6).



**Table 6: List of Locations indicating status of Red Palm Mite in Tobago**

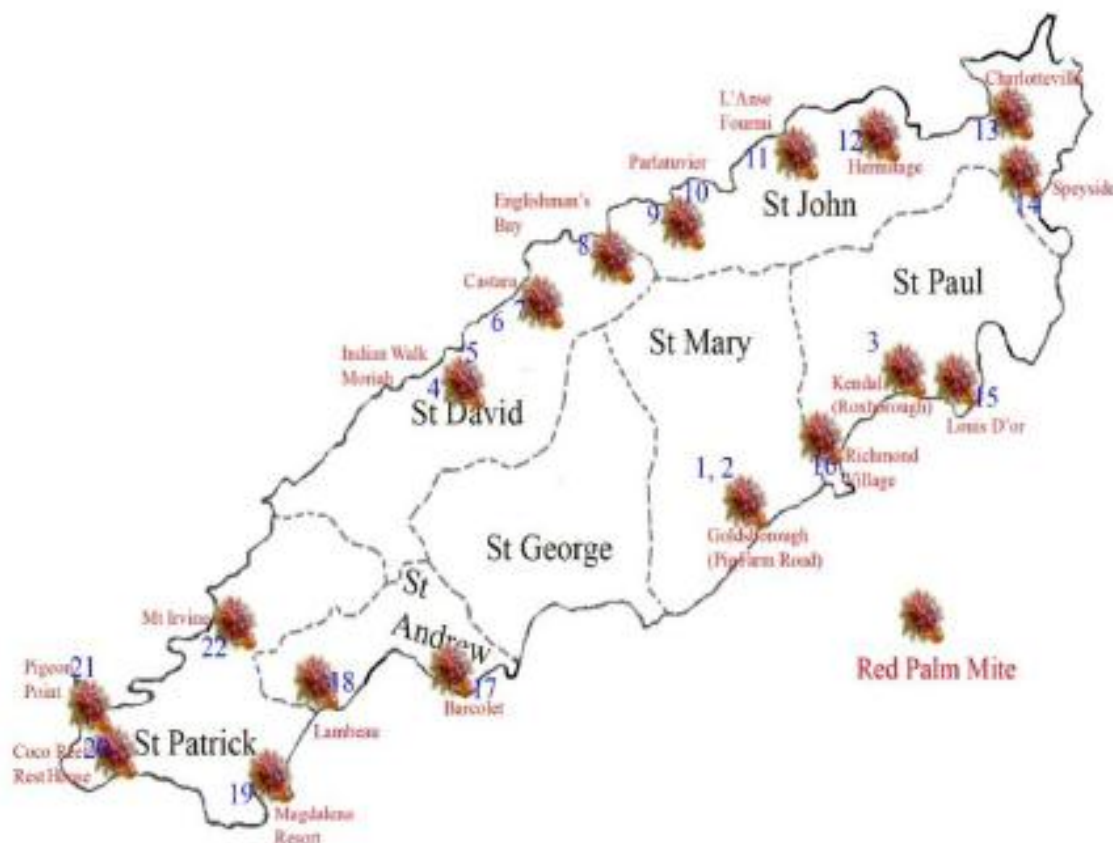
Scale of infestation	Nil	Low	Medium	High
% infestation of leaflet	0.0	(0.1% to 15.0%)	(>15.0% to 50.0%)	(> 50.0 %)
Name of the location	1. Moriah Road 2. English Man's Bay 3. Mt. Irvin Hotel 4. Louis D'or 5. Hermitage 6. Richmond Village	1. Pig Farm Rd. Goldsborough 2. Pig Farm Rd. Goldsborough 3. Kendal, Roxborough 4. Lambeau Beach, Old Milford Rd. 5. Magdalena Resort 6. Pigeon Pt. Beach Resort	1. Castara Recreation Facility 2. Parlatuvier 3. Speyside Main Rd. 4. Big Bacolet/ Minister Bay 5. Cocoa Reef Rest House	1. Indian Walk, Moriah 2. #143 North Side Rd. Moriah 3. Parlatuvier 4. L'anse Fourmi 5. North Side Road, Parlatuvier

*e. Hotspot areas of Red Palm Mite in Tobago:*

Based on the RPM population and the percentage yellowing of leaf, 20 locations from 5 counties are found to be the hot spot areas of the pest (Table: 7). The average population of RPM in counties is 6,690/leaflet with 51.1 per cent leaflet yellowing, 892/ leaflet with 2.5 per cent leaflet yellowing, 5307 /leaflet with 52.5 per cent leaflet yellowing, 5,129/ leaflet with 17.5 per cent leaflet yellowing and 9619/leaflet with 15.5 per cent leaflet yellowing in St. John, St. Paul, St. David, St. Andrew and St. Patrick, respectively (Fig.4).



**Figure 3: Distribution of Red Palm Mite in different Counties of Tobago**



### Locations

- |                                    |                                     |
|------------------------------------|-------------------------------------|
| 1 Pigfarm Rd, Goldsborough         | 12 Hermitage                        |
| 2 Pigfarm Rd, Goldsborough         | 13 Charlotteville                   |
| 3 Kendal, Roxborough               | 14 Speyside Main Rd.,               |
| 4 Indian walk, Moriah              | 15 Louis D'or                       |
| 5 143 North Side Rd, Moriah        | 16 Richmond Village                 |
| 6 Moriah Road                      | 17 Big Bacolet/ Minister Bay        |
| 7 Castara Recreation Facility      | 18 Lambeau Beach, Old Mildford Rd., |
| 8 English Man's Bay, North Side Rd | 19 Magdalena Grand Beach Resort     |
| 9 North Side Rd, Palatuvier        | 20 Coco Reef Rest House             |
| 10 North Side Rd, Palatuvier       | 21 Pigeon Point Beach Resort        |
| 11 L'Anse Fourmi                   | 22 Mt Irvine Hotel                  |

**Table 7: Hotspot areas of Red Palm Mite in Tobago**

No.	Name of the county	Name of the location	Intensity of RPM	
			Pop/Leaflet (No)	Yellowing of Leaflet (%)
1	St. Andrew	i. Minister Bay	7582	27.5
		ii. Lambeau Beach	2676	7.5
2	St. David	i. Indian Walk Moriah	3880	80.0
		ii. Moriah #143, North Side Rd	8474	60.0
		iii. Castara Recreation Facility	3568	16.7
		iv. English Man's Bay	—	—
3	St. John	i. Palatuvier, North Side Rd	1784	44.2
		ii. Palatuvier, North Side Rd	9366	80.8
		iii. L'Anse Fourmi	—	—
		iv. Hermitage	—	—
		v. Charlotteville	—	—
		vi. Speyside Main Rd	8920	28.3
		ii. Pig Farm Rd, Goldsborough	—	3.3
4	St. Patrick	i. Magdalena Resort (Tobago Plantations)	22300	13.3
		ii. Coco Reef Rest House	1650	21.7
		iii. Pigeon Point Beach Resort	4906	11.7
		iv. Mt. Irvine Hotel	—	—
5	St. Paul	i. Kendal, Roxborough	892	2.5
		ii. Louis D'or	—	—
		iii. Richmond Village	—	—

\* Average Leaf area from 5 counties: 446cm<sup>2</sup>

Figure 4: Hot Spot Areas of Red Palm Mite in Tobago



It is important to note that few times symptoms of yellowing or infestation was observed without live populations of RPM but more frequently with high numbers of exuvia. On few occasions leaves appeared quite green in colour with RPM live populations. Many times no correlation between pest population and symptoms was observed. These instances are leading to a scope for detailed studies about population dynamics, appearance of symptoms, influence of abiotic factors, seasonal variations, plant reaction *etc.*

*f. Cataloguing, identification and status of the indigenous natural enemies in Tobago:*

Survey data in Tobago highlighted the presence of natural enemies in all the locations surveyed. The natural enemies recorded are *A. largoensis*, *Bdella* sp., Chrysopids and coccinellids. Among the natural enemies collected, the predominant natural enemy is *A. largoensis*. Out of 22 locations surveyed, *A. largoensis* was noticed in 17 locations. The ratio of its occurrence to RPM ranged from 1:17 to 1: 800 (Table: 4).

Comparison of present survey data with earlier reports from Trinidad and Tobago on the damage, population levels of RPM and its natural enemy *A. largoensis* inferred that many folds increase in the damage per cent of leaflet as well as population counts from 8.52 % to 81.0% (2011 to 2012) and 5.42 to 275.3/cm<sup>2</sup> leaf area (2008 to 2012), respectively. Similarly the natural enemy count per leaflet also increased from 3.03 to 25.00 (2009 to 2012) over a period of 3 years. From this information it is evident that even though pest population increasing there is a scope for the build-up of natural enemies (Table: 8).

Review of earlier reports of Trinidad and Tobago about the status of *A. largoensis* revealed increase in a number over a period of time indicating its acclimatization to feed on RPM.

**Table 8: Trend of RPM damage on coconut and occurrence of natural enemies in Trinidad and Tobago**

Year of observation	Population of RPM		Population of natural enemy <i>A. largoensis</i>				Damage (%)	
	No./Leaflet	No./ cm <sup>2</sup>	No./Leaflet	No./ cm <sup>2</sup>	Ratio	Palm	Leaflet	
2008	-	1.71 to 5.42 (3.82)	-	3.00 to 38.50 (9.8)	-	-	-	
2009	-	0.99 to 2.22 (1.53)	2.58 to 3.03 (2.45)	-	-	-	-	
2011	1481	2.65	6.17	-	-	25%	8.52%	
Trinidad (2012)	7.0 to 11,452	0.01 to 275.3	1.00 to 25	0.01 to 0.08	1:3	-	1.65 to 46.8 %	
Tobago (2012)	892 to 9311	2.00 to 87.0	0.33 to 11.0	0.01 to 3.0	1:17 to 1:800	4.00 to 90%	2.5 to 81.0%	



It was known that natural predators may offer a good means of controlling RPM populations. The reports from different countries revealed the natural enemies that occur in respective country; in India the phytoseiid mite, *Amblyseius channabasavanni* and beetle, *Stethorus keralicus* Kapur (Coleoptera: Coccinellidae) were considered to be the most important predatory species. In Mauritius, the principal predator of *R. indica* in coconut plantations was *Typhlodromus caudatus* Chant (*Amblyseius caudatus* Berlese). In the Western hemisphere, *Neoseiulus longipinosus*, also native to the East has been found to prey on *R. indica* in the Caribbean. Also, there are several coccinellid and phytoseiid potential, endemic predators that are found preying on similar species in the Western Hemisphere. A fungus, possibly, *Hirsutella* spp. has been observed infecting the RPM. According to the available literature, each site has a different natural enemy complex with only one predator species, *A. largoensis*, present in all the geographical areas. *R. indica* was found on 36 palm species and *A. largoensis* was found on half of them. Through various surveys, *A. largoensis* was identified as the most abundant predator and often as the only phytoseiid species associated with *R. indica*.

The characteristics of *A. largoensis* namely;

- High survival
- High reproductive rate
- Presence in all areas
- Shorter life cycle
- The numerical response to pest increase
- Abundant presence
- Presence throughout the year
- Can feed, develop and reproduce on a diet consisting only of *R. indica*

and the information gathered from the surveys shows *A. largoensis* has the most potential as bio-control agent of RPM.

The low abundance of the predatory mite, *Bdella* sp. observed in the field indicates unlikely to be promising predator, further studies are necessary to know its predator status. It would be desirable to intensify search on natural enemies from other classes also that

could prey on all stages of RPM and other mortality factors to use in combination for effective suppression of RPM.

*g. Field Observations in Trinidad and Tobago during Survey:*

- The yellowing symptoms of coconut palms were observed in many locations, but the reasons for yellowing were found not only due to RPM infestation but also because of red ring disease, coconut leaf scales (yellow and brown), nutritional disorder, (confused symptoms) stress conditions in addition to the original yellow color of leaves which is a varietal character. The most prominent observation was 10-40% death of the palms caused by red ring disease.
- Lack of irrigation sources in many places subjecting the palms to stress conditions.
- Farmers are not aware of coconut cultivation technologies like agronomical, plant production and protection and not following good agricultural practices.
- Shortage of human resources is another important drawback for good agriculture in Trinidad and Tobago.
- Over-viewing of coconut crop condition in Trinidad and Tobago revealed the factors like Red Ring Disease, senility, stress conditions, salty high humid wind velocity at shores, soil erosion by tidal waves etc., were also contributing to the decline of coconut crop.
- Other minor pests that were observed include sucking insects, lepidopterans, grass hopper, mites, rat *etc.*, (Table: 9).

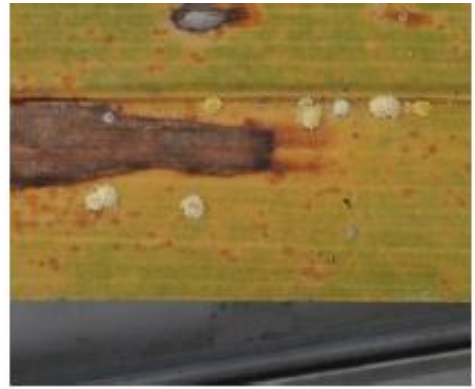
**Table 9: Pests associated with coconut palm in Trinidad and Tobago observed during survey**

Common Name	Scientific Name
<b>Major Pests</b>	
Red Ring disease – nematode coupled with Palm Weevil	<i>Rhadimaphelenchus cocophilus</i> <i>Rhynchophorus palmarum</i>
Red Palm Mite	<i>Raoiella indica</i>
<b>Minor Pests</b>	
Spider Mite	<i>Tetranychus urticae</i>
Cedros Grasshopper	<i>Tropidacris cristata</i>
Whitefly	<i>Aleurodicus trinidadensi</i> ; <i>A. cocois</i>
Coconut Scale	<i>Aspidiotus destructor</i> ; <i>Aonidiella orientalis</i> ,
Brown Scale	<i>Saissetiacoffeae</i>
Mealybug	<i>Dysmicocu sbrevipes</i> ; <i>Nipaecoccus nipae</i>
Coconut Mite	<i>Aceria guerreronis</i>
Palm Aphid	<i>Cerataphis brasiliensis</i> <i>Toxoptera aurantii</i>
Coconut Moth	<i>Batrachedra muciferae</i>
Tree Termite	<i>Naustitermes costalis</i>
Rat	<i>Rattus rattus</i>
Brown Hairy Caterpillar	<i>Brasolis sepherae</i>

- Nutritional Disorders: deficiency of Boran and potassium were observed



**Plate 11: Snow scale on coconut leaf**



**Plate 12: Mealy bugs on coconut leaf**



**Plate 13: Coconut scale on coconut leaf**



**Plate14: Yellowing due to Brown Scale**



**Plate 15: Death of palm due to red Ring disease**



*(ii) Demonstration of Coconut Cultivation Technologies as a part of Integrated Pest Management (IPM) of RPM:*

The use of practices that impact the life stages of the pest as well as assist the crop to withstand infestations are important in the management of the RPM.

Good agricultural practices like sanitation; removal and destruction of severely pest infested and dried leaves, ploughing, regular irrigation/adaption of moisture conservation measures; removal of senile palms and replantation with elite coconut germplasm, application of organic manures like neem cake (a product obtained from neem seed which has the antifungal, insecticidal properties and also serves as nutrient input); farm yard manure green manure etc.; application of fertilizers namely, urea, single super phosphate, Muriate of potash, Magnesium Sulphate, Boron etc., growing inter-crops like banana or root crops or cocoa or vegetables or pineapple or flowers or medicinal and aromatic plants or ornamental plants etc., need based use of botanical pesticides, placing yellow sticky traps in the palm crown for trapping mites, checking RPM population on alternate hosts, release of bio-agents etc., are components for integrated pest management strategy recommended for the successful management of RPM.

In addition to bio-control, a holistic approach with good agricultural practices is essential for successful suppression of Red Palm Mite as bio-control alone would not be able to control the pest.

*a. Establishment of demonstration plots:*

Two coconut blocks located in the Research Division of Centeno, were selected for demonstrating the coconut cultivation technologies. The plot - 1 is in the **northern block** with 35 coconut palms of about 10yrs age. The plot - 2 is in **eastern block** with 56 coconut palms in age group of 4 to 20 years. The following cultivation technologies were implemented in the plots:

- Brush cutting



- Crown cleaning (removal of dried leaves, dried inflorescences, fibres *etc.*)
- Ploughing of the land up to plough depth of 6 inches with rotovator
- Removal of diseased and dead palms
- Preparation of 2 metre radius basin around the trunk
- Making of drainage channels for improving drainage facility in low lying plots
- Application of manure at the rate of 50kg/palm in the basin
- Application of fertilizers namely urea 1kg, MOP (Muriate of Potash) 2.5kg, Mg so<sub>4</sub>(Magnesium Sulphate) 500g / palm /year
- Application of boron at the rate of 50g/palm/year
- Application of Neem Cake @ 10 kg/palm/year

Therefore, these cultivation technologies would improve the crop condition in the fields, minimize stress conditions and make the palms strong enough to withstand pest or disease attack. If bio-agents are released in addition to these practices, the suppression of Red Palm Mite would be an easy task. Periodical monitoring and re-releasing of bio-agents would help for sustaining the good levels of bio-agents in the field. Once this package is adopted and maintained regularly, over a period of time the coconut crop would be revived and yield potential increased.

## Demonstration of Coconut Cultivation Technologies as a part of IPM of RPM



Plate 16: Demonstration plot



Plate 17: Brush Cutting



Plate 18: Crown Cleaning



Plate 19: Ploughing



Plate 20: Removal of Dead Palms



Plate 21: Drainage channel



**Plate 22: Two metre radius basin**



**Plate 23: Application of FYM**



**Plate 24: Application of fertilizer**



**Plate 25: Covering of basin**

### **Recommendations:**

- The causes for yellowing are need to be addressed with correct diagnosis.
- All the coconut plantations along the coastal belts need to be revitalised or rejuvenated with elite germplasm by removing the senile, unproductive and dead palms
- Most of the locations are not with assured irrigation; hence an alternative source of irrigation is needed to be worked out
- Plant protection being a crucial part of the cultivation, a specific recommendation for managing the red ring disease, palm weevil, RPM and other sucking pests is essential
- As there was no standing recommended package for coconut cultivation, a research based recommendation suitable for local conditions for nursery management, spacing, planting, input application like organic manures and chemical fertilizers and maintenance of the crop is necessary.
- Several natural enemies of the Red Palm Mite were found active in most of the locations. Therefore, conservation of the natural enemies with good agricultural practices would help to manage the Red Palm Mite problem
- Training for extension staff as well as stakeholders in areas of coconut crop production and protection technologies is crucial for addressing the coconut problems.
- Creating awareness about the scientific coconut technologies through information bulletin is another important element to improve the coconut crop conditions.



## B. Laboratory Studies

Information generated through the survey paved a path to attempt the mass culturing of RPM and its natural enemy *A. largoensis* under the laboratory conditions.

To attempt mass culturing of prey and predators in the laboratory it is necessary to know the details of biology as well as life stages of respective species. The knowledge relating to feeding efficiency of predatory species is also essential to make use of the particular predator for the pest suppression under field conditions. Hence laboratory studies were conducted to determine the life cycle of RPM and its natural enemy; and feeding potential of natural enemy. Mass culturing of mites in the laboratory involved the following activities:

- Establishment of coconut seedlings
- Mass culturing of Red Palm Mite
- Mass culturing of *A. largoensis*, a natural enemy of RPM

### (i) *Establishment of a 'Greenhouse' Coconut Seedling Nursery*

Six months old coconut seedlings were collected from the field, washed with running water and dried with a cotton cloth. Seedlings were planted in a pot having pot mixture. Adequate watering was given regularly and seedlings were placed in the green house with adequate natural light.



Plate 26: Coconut seedling nursery in the Green House



**(ii) Mass Culturing of RPM, *Raoiella indica*:**

**a. Culturing of Stock Colony of RPM on Coconut Seedlings in the Greenhouse:**

Five coconut seedlings were cleaned carefully with a brush to remove all other arthropods. Yellow tagging tape covered with petroleum jelly was tied around the base of the palms to exclude any crawling arthropod.

Three hundred RPM, females collected from the field samples were placed on a rectangle piece of coconut leaf (15cm x 2.5cm). The rectangle leaf bits with 300 RPM females were stapled to the lower leaf of the potted coconut seedlings at one leaf bit/plant to facilitate RPM infestation of the coconut seedling. The potted coconut seedlings were covered with a nylon net fixed with the help of PVC pipes. Plants were maintained undisturbed for 45 days to allow the multiplication of RPM on the seedling. The procedure was repeated every week to infest a new batch of potted plants. Different stages of RPM were collected from these seedlings and used to inoculate new seedlings for laboratory multiplication of RPM, feed *A. largoensis* and other studies in the laboratory.



**Plate 27: Inoculation of RPM**



**Plate 28: Seedling with Stapled Leaf Bit**



**Plate 29: Caging of Coconut Seedlings**

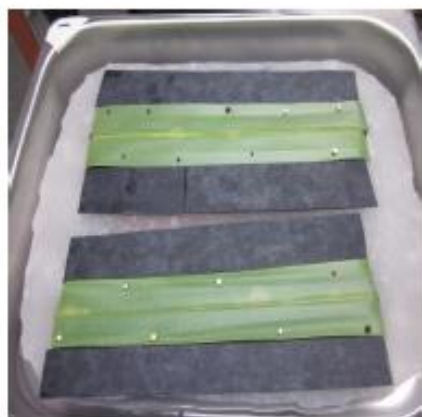
*b. Culturing of RPM, Raiella indica in the Laboratory as Prey Mites:*

Various trials were pursued for rearing of RPM in the laboratory. The materials required for making test arenas, the dimensions of the leaf bit and the container to be used, the types of sponge, black paper, as well as the tips for maintaining the life span of leaf bit *etc.*, were studied thoroughly and the following types were found suitable for culturing of mites in the laboratory:

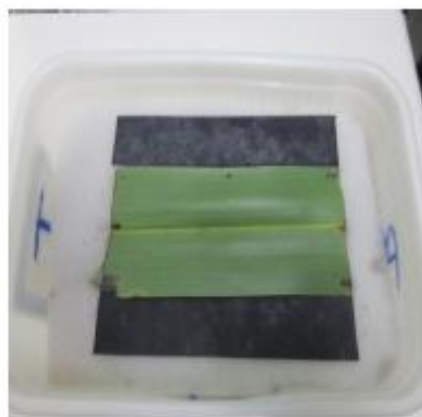
*b.(i) Types of containers and arena sizes:*

No	Details	Container size	Sponge size	Size of Leaf bit	Purpose
Type 1	Metal tray	32.5cm x 26.5cm x 4.5cm	27.0cm x 20.0cm x 3.5cm	24.0cm x 2.5cm	Mass culturing of RPM
Type 2	Plastic tray	18.0cm x 12.0cm x 7.0cm	16.0cm x 10.0cm x 3.5cm	8.0cm x 2.5cm	Mass culturing of RPM/arenas for laying females of RPM
Type 3a	Petri dish	8.0cm diameter	6.0cm x 6.0cm x 2.0cm	8.0cm x 2.5cm	Biology Studies of RPM/ natural enemy, <i>Amblyseius largoensis</i>
Type 3b	Petri dish	4.0cm diameter	3.0cm x 3.0cm x 2.0cm	4.0cm x 2.5cm	Biology Studies of RPM/ natural enemy, <i>Amblyseius largoensis</i>

## Different Types of Arena



Type 1 - Arena in Metal Tray



Type 2 - Arena in Plastic Tray



Type 3a - Arena in Petri Dish



Type 3b - Arena in Petri Dish

### *b.(ii) Arrangement of Arena:*

RPM infested coconut leaf samples were collected from the field. Another sample of fresh leaves was collected from field. Fresh leaves were washed with running tap water and then cotton dried.

Different containers i.e., the metal tray (32.5cm x 26.5cm x 4.5cm), plastic tray (18cm x 12cm x 7cm), and petri dishes (size 4cm, 8cm diameter), were sanitized and dried. The

sponge was cut into pieces of similar size to the respective container and pre-soaked in water for 20 minutes. Sponge pieces were placed according to the size of container i.e. metal tray (27cm x 20cm x 3.5cm), plastic tray (16cm x 10cm x 3.5cm) and petri dishes (6cm x 6cm x 2cm) to fit exactly into the container. Fresh leaves collected from the field were measured with a ruler and cut into required sizes (24cm x 2.5cm – Type 1, 8cm x 2.5cm – Type 2, 8cm x 2.5cm – Type 3a and 4cm x 2.5cm – Type 3b). The cut leaf bits were placed on the respective size sponges and fastened with insect pins. The required number of RPM females (50) was transferred to leaf arenas from field sample or stock colonies reared on potted coconut plants in green house with the help of a pig hair brush. Black construction paper was placed around each arena to prevent mite escape. The containers were filled with water to the level of the sponge. Arenas were prepared at periodical intervals to make continuous availability of prey mites. These arenas were maintained in laboratory at 27° C Temperature, 70% RH and 12:12 photoperiod.

*b.(iii) Study of RPM Biology:*

RPM life cycle was studied under laboratory conditions by releasing 5 laying females on arena Type 3a and 3b. Eggs of same day laying were pooled on to a separate arena of same Type and monitored till adult emergence. Observations on date of egg laying, date of eclosion, date of first moult, date of second moult and third moult and duration of each life stage, longevity of adult, average egg laying, fecundity *etc.*, were recorded.

**Biology**

Eggs are red in colour, smooth and laid in groups on the arena. The number of eggs in a group or colony varied from 11 to 52. Each female laid on an average, 6.67 eggs with a minimum of 3 to maximum 11 numbers in a life period of 17 to 21 days under laboratory conditions. Pre-oviposition period ranged from 5 to 10 days with an average period of 8.3 days and oviposition period varied from 4 to 9 days averaging 6 days. Incubation period was recorded from 4 to 9 days with an average period of 5.87 days. Larval stage is having three pairs of legs. The larval, protonymphal and deutonymphal periods ranged from 2 to 11, 2 to 6 and 2 to 9 days, respectively. The average duration of larval, protonymphal and deutonymphal stages was 6.48, 4.39 and 6.04 days respectively. The total duration of

immature stages varied from 6 to 26 days with an average of 16.91 days. The life cycle of RPM was completed in 18 to 33 days with an average of 22.78 days. The female adult laid on an average one egg/day and it varied from 1-2 eggs/day. Female adult lived up to 17 to 21 days whereas male lived up to 9 to 16 days. Mating duration was observed for about 2 to 4 days (Table: 10).

**Table 10: Developmental period of Red Palm Mite**

No.	Incubation Period (Days)	Larval Period (Days)	Protonymphal Period (Days)	Deutonymphal Period (Days)	Total Period (Days)
1	5	7	4	4	20
2	6	9	4	3	22
3	6	9	6	2	23
4	4	8	5	3	20
5	4	10	5	3	22
6	6	8	4	2	20
7	3	6	4	7	20
8	3	6	4	8	18
9	5	4	4	5	19
10	6	4	4	5	21
11	6	6	4	5	21
12	7	7	2	5	21
13	9	4	4	5	22
14	7	4	4	5	20
15	6	2	4	9	21
16	7	2	4	9	22
17	6	5	4	8	23
18	5	2	4	9	20
19	6	4	4	9	23
20	6	10	6	8	30
21	8	11	5	9	33
22	8	11	6	8	33
23	6	10	6	8	30
Total	135	149	101	139	524
Average	5.87	6.48	4.39	6.04	22.78



## LIFE CYCLE OF RED PALM MITE (RAOIELLA INDICA HIRST)

### b.(iv) Mass Culturing of RPM on Arena:

For mass culturing of RPM, arena type 1 was found to be suitable. Fifty pairs of female and male RPM were placed on each arena of Type 1 for egg laying and further multiplication. The arenas were replaced with fresh leaf bits when the leaf dried. From each arena with 50 females, 120 eggs were obtained in 10 days. The required RPM were collected from these arenas for making new arenas. All the arenas were monitored and maintained for getting further cultures.

### RPM in the Laboratory



Plate 30: Different Types of Arenas in the Lab



Plate 31: RPM Colony on the Arena



Plate 32: Microscopic observations



Plate 33: Monitoring of arenas

**(iii) Mass Culturing of *A. largoensis*, a natural enemy of RPM:**

**a. Studies on Life Cycle of *A. largoensis*:**

The eggs of *A. largoensis* were obtained by keeping the female adults in individual glass vials. As soon as the eggs hatched, larvae were individually transferred to experimental arenas. Arena Type 3b was used for studying the life cycle of *A. largoensis*. The small leaf bit containing RPM eggs was placed over the arena having *A. largoensis* to replenish the diet (RPM) daily. Arenas were replaced when leaf was dried. Observations were recorded daily from arenas to obtain details of life stages of *A. largoensis*.

**Biology**

Eggs are in cream colour and laid singly. Each adult laid 1 or 2 eggs/day. Eggs were hatched after a period of 3.52 days on an average; which varied from minimum one day to maximum seven days. Larvae were hyaline in colour with three pairs of legs and developed into protonymph with 4 pairs of legs in 3.38 days on an average. Larval period was ranged from 2 to 6 days. Protonymph moulted into deutonymphal stage which was also having 4 pairs of legs and bigger than a protonymph. The protonymphal stage was completed in 2.2 days on an average. The deutonymph took 4.9 days to become adult which was ranged from 5 to 11 days. Protonymph is hyaline in colour and red coloured intestinal contents could be seen distinctly from deutonymphal stage which indicates feeding on RPM. Duration of immature stages was occupied on an average 10.46 days. Deutonymph developed into adult with 4 pairs of legs and little bigger than deutonymph in size. Total life cycle from egg to adult was completed on an average of 14.00 days. Minimum period of life cycle was 10 days whereas that of maximum was 17 days. Adult *A. largoensis* lived for a maximum of 22 days under laboratory conditions (Table: 11).

**Table 11: Developmental period of *Amblyseius largoensis***

No.	Incubation Period (Days)	Larval Period (Days)	Proto and Deutonymphal Period (Days)	Total Period (Days)
1	2	2	6	10
2	3	3	11	17
3	2	4	5	11
4	3	-	-	-
5	2	-	-	-
6	3	2	5	10
7	6	4	-	-
8	5	2	9	16
9	7	3	7	17
10	7	3	6	16
11	1	3	9	13
12	2	-	-	-
13	1	3	9	13
14	4	4	5	14
15	5	5	7	17
16	4	6	6	16
17	5	-	-	-
18	3	-	-	-
19	4	-	-	-
20	3	-	-	-
21	2	-	-	-
<b>Total</b>	<b>74</b>	<b>44</b>	<b>85</b>	<b>170</b>
<b>Average</b>	<b>3.52</b>	<b>3.38</b>	<b>7.10</b>	<b>14.00</b>

## **Life Cycle of *Amblyseius largoensis* a Natural Enemy of the Red Palm Mite**

### *b. Studies on Feeding Potential of A. largoensis:*

*A. largoensis* adults were starved for 24 hours by isolating individually into 20 glass vials (8.5 cm x 2.3 cm). Leaf bit containing 40 RPM eggs was introduced into the vial and the number of eggs consumed in 24 hours was recorded. Eggs were provided continuously until *A. largoensis* died. Data were recorded daily and tabulated for determining the feeding potentiality of *A. largoensis*.

From the studies on feeding potentiality, it was observed that field collected *A. largoensis* when fed 24hrs after starvation lived up to 3-10days. Feeding potential of *A. largoensis* was varied from 6.14 to 24.38 RPM eggs in 24hrs. On an average 15.53 number of RPM eggs were consumed by single *A. largoensis* female within 24hrs (Table: 12).

**Table 12: Studies on Feeding Potential of *Amblyseius largoensis***

Serial #	FEEDING DAYS																	Total	Average
	1	2	3	4	5	6	7	8	9	10	11	12	13	14	15	16	17		
1	0	1	25	27	39	40	36	27	-	-	-	-	-	-	-	-	-	195	24.38
2	0	51	15	5	5	22	40	40	6	5	3	5	-	-	-	-	-	197	16.42
3	23	14	15	10	3	29	15	32	20	37	21	20	32	13	15	10	10	296	17.41
4	0	3	32	7	18	25	18	7	8	9	-	-	-	-	-	-	-	127	12.70
5	13	17	27	14	5	14	5	23	14	5	24	-	-	-	-	-	-	161	14.64
6	26	4	21	-	-	-	-	-	-	-	-	-	-	-	-	-	-	51	17.00
7	16	23	0	24	26	31	40	40	17	6	8	-	-	-	-	-	-	231	21.00
8	0	1	30	27	7	30	29	2	4	10	13	-	-	-	-	-	-	153	13.91
9	39	10	13	13	9	4	-	-	-	-	-	-	-	-	-	-	-	88	14.67
10	40	26	27	5	22	0	11	10	14	-	-	-	-	-	-	-	-	155	17.22
11	4	3	9	27	12	24	13	10	14	15	-	-	-	-	-	-	-	131	13.10
12	0	3	5	13	2	13	7	-	-	-	-	-	-	-	-	-	-	43	6.14
13	9	24	2	0	0	10	32	30	3	6	13	26	38	6	11	11	6	227	13.35
14	10	8	6	10	6	9	30	25	-	-	-	-	-	-	-	-	-	104	13.00
15	0	0	35	18	14	20	9	10	-	-	-	-	-	-	-	-	-	106	13.25
16	0	16	20	9	0	20	8	10	20	-	-	-	-	-	-	-	-	93	10.33
17	40	16	26	40	28	12	16	8	7	33	40	28	37	10	8	23	14	386	22.71
18	9	11	8	-	-	-	-	-	-	-	-	-	-	-	-	-	-	28	9.33
19	0	24	11	23	34	31	10	5	26	-	-	-	-	-	-	-	-	164	18.22
20	24	23	31	14	8	14	20	33	40	34	23	40	38	13	10	14	4	383	22.53
21	2	3	4	4	10	20	12	8	24	36	40	-	-	-	-	-	-	163	14.82
<b>Total</b>																		<b>326.13</b>	
<b>Average</b>																		<b>15.53</b>	



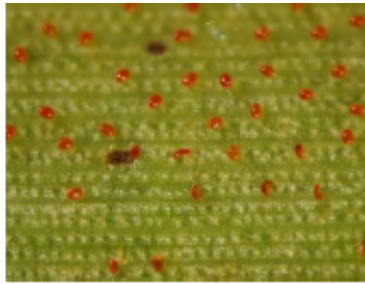


Plate 34:RPM Eggs Eaten by *A. largoensis*



Plate 35: RPM Nymph eaten by *A. largoensis*

*c. Standardisation of Rearing Protocols for Natural Enemy, A. largoensis:*

Mass culturing of the natural enemy, *A. largoensis* in the laboratory was attempted in three methods:

- (i) Culturing on potted coconut seedlings having stock colony of RPM
- (ii) Culturing on RPM infested coconut leaf bits in a plastic container
- (iii) Culturing in the arena – Type 3

*c.(i) Culturing on potted coconut seedlings:*

RPM colonized potted coconut seedlings were inoculated with 25 pairs of *A. largoensis* adults on each seedling by placing a glass vial containing *A. largoensis* adults at the collar region of seedling. Seedlings were protected with caging and maintained with adequate water and sunlight. *A. largoensis* population on the seedling was monitored at fortnightly intervals by observing 5 randomly selected areas on leaves. The same procedure was followed to inoculate the other seedlings. After 30 days *A. largoensis* can be harvested for release in the fields.

*c.(ii) Culturing on RPM infested coconut leaf bits in a plastic container*

Coconut leaf bits of 30cm length were cut from the field collected RPM infested leaf samples or potted coconut seedlings. Leaf bits were cleaned to ensure that they were free from other arthropods. Ten leaf bits were placed in each plastic container (2<sup>o</sup> x 1<sup>o</sup> x 2<sup>o</sup>) and maintained under laboratory conditions. Leaf bits were monitored at 3-day intervals and dried leaf bits were replaced with fresh leaf bits containing RPM. The increase in the number of *A. largoensis* was recorded from 60 to 100 in 7 days in a single container. The

containers can be maintained for allowing 30 days for the multiplication of *A. largoensis*. After building the population of *A. largoensis* in the container, they can be used for release in the field.

*c. (iii) Culturing in the arena – Type 3*

Culturing of *A. largoensis* on leaf arenas was also tried with arena of Type 3b. In spite of series of approaches, the culture could not be established on the arena.



Plate 36: Inoculating *A. largoensis* on Coconut Seedling



Plate 37: Culturing of *A. largoensis* in plastic container

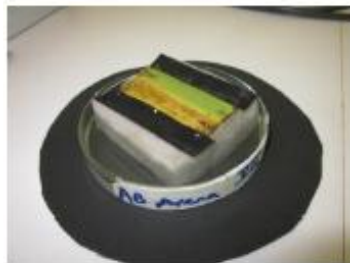


Plate 38: Culturing of *A. largoensis* on arena



Plate 39: Pair of *Amblyseius largoensis*

Even though monitoring of samples was found to be critical and difficult, culturing on the potted coconut seedlings and containers method were found to be better options for the mass culturing of *A. largoensis* over arena method. Monitoring of *A. largoensis* culture on seedling revealed suppression of RPM population but *A. largoensis* population could not be seen from sampling leaf area. Further investigations are required to determine best method of approach. The potentiality of natural enemy also needs to be evaluated under field conditions with repetitive and long term experiments field studies like rate of release, number of releases, type of release (inoculative or inundative) are necessary to evaluate a

potential bio agent. To conduct the above field studies, the continuity of the culturing of mites in the laboratory is necessary. The scale of bio agent production in a laboratory with existing facilities and economics of mass production are yet to be worked out.

### **C. Suggested Protocols for Field Release of Natural Enemy, *A. largoensis***

- Collect leaf bits (6cm) with 100 pairs of *A. largoensis* from seedling or plastic container
- Clip the leaf bits to the leaves of coconut palm in opposite directions in the RPM infested field at the rate of 2 leaf bits per whorl of leaves.
- Or carry the leaf bits in zipper locked polythene bags and place in between the fronds
- Open the zipper lock to facilitate the *A. largoensis* to move on to palm
- Evening hours would be congenial for release
- Periodical releases can be made at 15 to 20 days intervals

### **D. Capacity Building of Staff in Ministry of Food Production (MFP)**

Agricultural Officers, Field Assistants and On-the-Job Trainees were trained in:

- Skills for identification of RPM pest, diagnosis of RPM infestation symptoms in the field, natural enemy fauna available in Trinidad and Tobago, identification of *A. largoensis* etc.
- Establishment of a coconut nursery in the green house
- Greenhouse maintenance
- RPM inoculation to seedling, RPM stock colony maintenance on the seedling
- Preparation of different types of arenas for laboratory rearing of mites
- Handling and transferring of mites by observing through microscope
- Microscopic observations for identification of eclosion of eggs, moulting of life stages, identification of immature stages of mites etc. (RPM and its natural enemy, *A. largoensis*)

- Monitoring/maintenance of laboratory cultures
- Daily laboratory activities

### **E. Conducting Awareness Campaigns, Seminars and Training Programmes**

Presentations on coconut production and protection technologies for dissipation of knowledge among stakeholders and updates about the RPM project were given during the project period by taking opportunity of different occasions like

1. Business seminar organized by the High Commission of India
2. Seminar to stakeholders of coconut in Trinidad organized by Ministry of Food Production
3. Seminar on Organic Farming organized by the High Commission of India
4. Third Meeting of the Palm Pest Complex Technical Working Group held at Centeno, Trinidad on July 19<sup>th</sup>, 2012

Open Day seminar on “RPM of coconuts” was conducted on 29<sup>th</sup> and 30<sup>th</sup> January 2013 in which the Senator the Honourable Devant Maharaj, Minister of Food Production, representatives from the Ministry of Food Production, Indian High Commission, CARDI, the Coconut Growers’ Association, stakeholders and delegates from other Caribbean islands (Venezuela, Antigua, St. Kitts & Nevis, Jamaica, Dominica and St. Lucia) have participated.

The first day seminar was organized in two sessions.

- Session I:        Opening ceremony in which the Senator the Honourable Minister, Devant Maharaj delivered the feature address and released the technical information
- a. Technical Manual
  - b. Factsheet
  - c. Pamphlets for distribution among technical and extension personnel, stakeholders for the guidance in crop and pest management of coconut



## 5. GENERAL CONCLUSION:

Entry of the Invasive Alien Species (IAS), Red Palm Mite (RPM), *Raoiella indica* into the Caribbean region in 2004 and Trinidad in 2006, significantly impacted the agriculture sector in Trinidad and Tobago, lead to the Memorandum of Understanding (MOU) between Indian Government and the Government of Trinidad and Tobago and eventually, Dr. A. Sujatha, Principal Scientist (Entomology) has been deputed as an Indian Technical Economic Cooperation (ITEC) expert for one year to assist in the control of Red Palm Mite in Trinidad and Tobago. Estimation of RPM damage, identification of pest and natural enemies, development of protocol for rearing of RPM, *Amblyseius largoensis*, a natural enemy of RPM and capacity building of staff are the major objectives of the project.

The estimated RPM infestation in different locations of Trinidad through the survey was varied from 1.65 to 46.8 per cent of leaf let. RPM population count ranged from 7.0 to 11,452 per leaf let, whereas the *A. largoensis*, a natural enemy of RPM recorded was 1 to 19 per leaflet. From the survey in Tobago it was found that the percentage infestation ranged from 2.5 to 81.0 per cent of leaflet. The RPM number per leaflet ranged from 892 to 8366 and natural enemies from 0.33 to 11.0. The predatory mite *A. largoensis* (phytoseiidae-Acari) was identified as a potential indigenous natural enemy of the RPM. In addition to RPM pest, prevalence of Red Ring Disease, sucking pest, nutritional disorders, stress to the palm, soil erosion with palms at shores of the ocean, lack of awareness about coconut cultivation technologies among the farming community *etc.*, were found to be factors contributing to the decline of the coconut industry in Trinidad and Tobago.

Laboratory protocols for rearing RPM, *A. largoensis*, a natural enemy of RPM were standardized. Recommended components of Integrated Pest Management (IPM) for the RPM were implemented in the demonstration plots at Research Division, Centeno. Training of staff in the Ministry of Food Production in terms of pest diagnosis, skills for laboratory rearing of RPM and its natural enemy, *A. largoensis*, bio-control laboratory maintenance were accomplished as a part of capacity building of staff. The outcome of the project was brought in the form of publications; extension activities coupled with distribution of printed material would be helpful



for creating awareness about coconut cultivation among the technical staff, agricultural officers and extension workers.

Continuity of mass culturing of *A. largoensis* in the laboratory, field efficacy studies, confirmative studies and bio-agents were recommended. Field maintenance of demonstration plots with sequential application of inputs in the long run would result in the crop improvement and suppression of RPM. A large scale rejuvenation programme of coconut crop to replace dead, senile and unproductive palms with elite germplasm was suggested for plant health improvement and increased productivity. Establishment of farm level processing units or cottage industries for value addition to the coconut products would create opportunities for youth employment and boosts the crop economy. Sharing of information was strongly emphasized to build up the knowledge and make the coconut industry more productive to sustain the ‘Tree of Life’ for the benefit of mankind.

### **Recommendation**

The bio-control studies which are interlinked with life cycle of respective bio-agent/prey organism needs specific period for developing the cultures in the laboratory. Basic bio-control studies needs repetitive confirmations both in the lab as well as under field conditions involving long time process to identify a successful bio-agent. The bio-control through endemic indigenous bio-agents has got a limitation of having a wide range of prey material available in natural habitat which makes the bio-agent non-specific to target prey organism (pest). In view of this situation, means of classical bio-control would be a more efficient option for quick suppression of the pest within a specified period. Field efficacy studies with inoculative or inundative releases of indigenous bio-agent, *A. largoensis* are also needed, to find out the rate of release in the infested field. It should be noted that the objectives proposed under this project for one year were too many to undertake given the nature of the crop-pest-natural enemy complex that was being targeted. For a perennial crop like coconut, any input application investigations requires a minimum of three years to determine the impact on the crop condition and yield. Hence the project proposals for biological control studies should be planned for a period of three to five years to observe successful suppression of the pest and improvement in the crop

condition. An upgrade of the current bio-control laboratory and green house facilities with suitable infrastructure would hasten the attainment of the objective.

## 6. OUTPUT OF THE PROJECT

- The predatory mite *A. largoensis* (phytoseiidae-Acari) was reconfirmed as a potential indigenous natural enemy of the Red Palm Mite.
- Developed protocols for mass culturing of Red Palm Mite and its natural enemy *A. largoensis*
- Demonstrated sustainable IPM technologies for RPM suppression in the field and studies are in progress
- Capacity building of staff in the Ministry of Food Production – Agricultural Officers, On-the-Job Trainees and Field Assistants were trained in:
  - Skills for identification of RPM pest and natural enemy fauna.
  - Diagnosis of RPM pest symptoms in the field.
  - Establishment of a coconut nursery in the green house
  - Greenhouse maintenance
  - RPM inoculation to seedling and stock colony maintenance in the greenhouse
  - Preparation of different types of arenas
  - Handling and transferring of mites
  - Microscopic observations on eclosion of eggs, life stages, moulting, identification of immature stages of mites *etc* (RPM and its natural enemy, *A.largoensis*).
  - Monitoring/maintenance of laboratory cultures
  - Daily laboratory activities
- Training programme and seminars to improve crop and pest management skills amongst stakeholders were organized
- Information material was prepared as publications namely:
  - a. Technical Manual
  - b. Factsheet
  - c. Pamphlets for distribution among technical and extension personnel, stakeholders for the guidance in crop and pest management

- Basic protocols required for establishing a bio-control laboratory was developed – further confirmative studies are needed for standardization

## 7. WAY FORWARD

The Coconut Industry has to tap the full potential of coconut as a renewable resource, which could be used to generate a range of environmentally friendly, natural products, with a wide variety of end-uses and applications like oleo chemicals, virgin coconut oil, functional foods, functional drinks, convenience foods etc. Likewise the country need to exploit to the fullest, their individual comparative advantages in cultivation, processing and marketing of coconuts.

### **Micro-scale (Farm or crop level management):**

Enhancement of suitable technology adoption under different agro ecological situations is needed to overcome the present trends in coconut sector.

Suitable technologies available:

- Fertilizer requirement - methods
  - Irrigation methods – water requirement
  - Moisture conservation methods
  - Green manure crops
  - Coconut waste material utilization
  - Coconut based cropping system
  - Drought tolerant management
  - Integrated Pest Management
  - Integrated Disease Management
  - Pre and Post-harvest technologies
- Generation of technologies with due emphasis on integrated farming systems in different agro ecological situations with inter/mixed/multiple/multi storied cropping and farming system; Agriculture, Poultry, Dairy, Aqua culture etc.
  - Increasing productivity of coconut is one way to reduce the cost of production and increase in the net returns to the producer. The productivity increase of coconut plantations can be achieved through rejuvenation of existing plantations, replanting and under-planting to

eliminate old, senile and unproductive plantations with high yielding varieties and hybrids; optimum input management through organic recycling nutrient and water conservation; integrated pest management, adopting integrated farming in coconut and proper organic recycling of farm wastes to maintain soil health.

- Identification and effective utilization of available generic resources with quantitative traits is essential. Characters like dwarf varieties, early bearing, varieties suitable for tender nut water and copra, high yielding can be looked for Trinidad and Tobago conditions.
- Location specific, need based application of resources to increase the nutrient used efficiency and increase the productivity, better management of pest and diseases is needed to be intensified.
- Converting the weakness of not applying the inorganic fertilizers into strength by adopting organic farming with holistic approach integrating nutrient management and integrating pest management practices with involvement of efficient bio agents, botanical pesticides, pheromones and bio fertilizers to produce niche organic products and to get more price and income.

#### **Industry Scale:**

##### **Extension activities**

- Strengthening of extension efforts by setting up farmers' field school, farmers participatory demonstration stations to achieve faster dissemination of technologies is another important factor to get the impact from different areas of the country.
- Development of information system with latest media material and cultivation practices of coconut for the farmer and extension personnel would be another important area to be emphasised.
- Providing training facilities for the stakeholders and unemployed youth for the production of diversified and value added products from coconut crop can be formed as an integral part of policies to uplift the coconut crop situation in the country.

##### **Strengthening of research**

- Research activity and breeding programs for disease resistance would pave a path for sustainability in yields



### Coconut industry development

- Rejuvenation of sick, senile and unproductive palms would be helpful to improve the crop condition and yields
- Establishment of small scale cottage industries at farm level processing and large scale industrial units would be an important milestone for employment as well as income generation
- Linking farmers to the market and making intensive market promotional efforts
- Formulation of schemes for area expansion on the coconut, rejuvenation of coconut crop, establishment of seed gardens and nurseries, financial support for home units, small scale and large scale industries, input subsidies; for fertilizers, plant protection chemicals, suitable farm tools and implements for cultivation *etc.*, is an area for bringing up the income and sustainability of the crop.
- Promoting coconut based eco-tourism in islands/back water areas and establishment of coconut world.
- To derive maximum economic returns by exploring crop potentiality for the farmers and the industry product diversification of coconut and development of value added products become very much essential. Various products and value added products that can be obtained from coconut crop are furnished below:

#### (1) From fruit

**Coconut Water:** (i) Tender coconut water, Snow ball tender nut and Coconut water beverages

(ii) Water from matured nut – Coconut vinegar, Nata-De-Coco

**Copra:** (i) Wet - Virgin Coconut Oil (VCO) such as Body oil/Baby oil and Pharmaceutical, Desiccated coconut, Coconut Cream, Coconut milk, Spray Dried Coconut milk powder, Coconut chips, Coconut dishes, Coconut cake, Coconut biscuits, Coconut candy etc.

(ii) Dry – Copra and Coconut oil, Dietary oil and Bio diesel/Bio fuel/Bio-lubricants/Cosmoceuticals.



- Shell products:** Shell Flour, Shell Charcoal, Activated Carbon and Handicrafts.
- Fibre Products:** Brushes, Foot mats, Rubberised mats, Husk such as Coir Geotextile, Coir ropes, other Coir products and Handicrafts.
- Coir Dust:** Planting material, Coir Pith Compost, Coco peat etc.
- (2) **Coconut Inflorescence:** Toddy, coconut jaggery and Confectionery jelly.
- (3) **From Wood:** Housing Material, Furniture and handicrafts.
- (4) **Coconut Leaves:** Mats, Brooms, Hats and other handicrafts.

**Appendix I**  
**List of gardens and farmers visited during the Red Palm Mite survey in Trinidad, 2012**

No.	Name of Farmer	Contact	Location	County	Size of farms (ha/%, of trees)	Age of the palms	Variety	Remarks
1	Mr. K. Eavis	368-8906	Volcania	St. George	1.31	<5yrs	Yellow	R.R.-25%, RPM, lots of weeds
2	Mr. E. Williams/Mr. Peter Peabes	676-6007	Canouan	St. David	60.7%	6 yrs& 50-60 yrs	Yellow, green, tall	R.R. RPM
3	Mr. Ray Ramdoyal	763-3268	L'Asse Noire	St. David	207 trees	12 yrs	Yellow, green	Severe RPM, pea measure, weed control with chemicals
4	Mr. Harold Gavett	741-6638	Sanger Grande	St. Andrew	2.02	1-13 yrs	Yellow	R.R.-10%, RPM
5	Sea Coast (East)	-	Manzanilla	St. Andrew	3000 trees	2-5 yrs-60 yrs	Yellow, tall	R.R.-35%, scalle, RPM
6	Mr. Stanton	680-3521	Mayaro	Mayaro	3.04	2 to <10 yrs	Yellow, tall	R.R.-35%, RPM
7	Mr. Bumperal	354-8190	Guayaguayare	Mayaro	100 trees	3 to >13 yrs	Yellow	RPM
8	Mr. Jason Bissessar	-	4 1/2 mi Blanchisseuse Rd	St. George	15 trees	3 yrs	Yellow, tall	R.R.-30%, RPM
9	Mr. Ronald Tribune	669-3484	Didimas	St. George	15 trees	3 yrs	Yellow	RPM
10	Ms. Bunde/Haricharan	763-3795	19 1/2 mi Blanchisseuse Rd	St. George	2.92	1 to 15 yrs	Yellow, green, orange	R.R.-67%, RPM
11	Mr. Gilbert	383-3037	No. 20 Paris Rd	St. George	25 trees	15-20 yrs	Yellow, green	R.R. RPM
12	Mr. Robert Asar	-	Maracas	St. George	4.15 (on border)	>8 yrs	Yellow, green, Chinese	R.R. RPM
13	-	-	La Platte	St. George	128 trees	3 to >25 yrs	Yellow	RPM
14	-	-	Maracas Bay (Beach)	St. George	158 trees	3 to >10 yrs	Yellow	R.R. RPM
15	-	-	Bonisse (Beach)	St. Patrick	-	<5 yrs	Green	RPM
16	Mr. Chandro/Ranjitkuma	-	Lp 154 Columbus Rd, Celvos	St. Patrick	15 trees	2-5 yrs	Yellow	RPM
17	Mr. Phillip Agostini	680-2477	Condance Estate Terrace	St. Patrick	600	2 to >5yrs	Yellow, tall, orange Naturally Crossed Dwarf	R.R.-40%, RPM
18	Mr. Pato/Gaffner	-	Walkerfield	St. Patrick	4.85	5 to 7 yrs	Green	Intercropped, R.R.-30%
19	Mr. B. B. Gogoi	727-5751	Maloney South	St. George	50 trees	3-5yrs	Green, yellow	R.R.-60%, RPM
20	-	-	Vibron	St. George	100 trees	3-8yrs	Green, yellow	RPM
21	Mrs. Ines/Annam Church	-	Maraval	St. George	15 trees	4-6yrs	Green	R.R.-20%
22	-	-	Ava Brook Ave, Diego Martin	St. George	15 trees	7-8yrs	Yellow	RPM
23	-	-	Williams Bay Chaguanas	St. George	20 trees	8-8yrs	Yellow	RPM
24	Mr. Bantol/Bachan	333-9961	Kelly Village	Caroni	60 trees	2-25yrs	Yellow	R.R.-30%, RPM
25	Ms. Lallan	752-9361	Charterville	Caroni	20 trees	1-25yrs	Yellow	R.R.-30%, RPM
26	Mr. Phillip Edward	746-5839	Miscawby	Caroni	2.45	1-3yrs	Tall	RPM
27	Mr. Heera	673-2257	Uquare Rd F repeat	Caroni	2 1/2 acres	3-4yrs	Green	R.R.-30%
28	-	-	Waterloo	Caroni	189 trees	4-5yrs	Tall	RPM
29	Mr. Michael Somaroo	308-1783	Galesada San Fernando	Victoria	26 trees	2-15yrs	Chinese, yellow, green, orange	R.R.-20%, RPM
30	Mr. Rajjant/Ramnarain / Ms. Esha	357-3725	Galesada Ring Rd	Victoria	1.21	5-10yrs	Yellow, Chinese, tall	R.R.-20%, RPM
31	Mr. Tom Singh	647-1921	Siparia	St. Patrick	268 trees	3-20yrs	Yellow, Chinese	R.R.-20%

\* R.R.- Red Ring disease

\* RPM- Red Palm Mite

## Appendix II

### 1. Protocols for Establishment of a 'Greenhouse' Coconut Seedling Nursery

- Collect coconut seedlings between 4 – 6 months age (optimal age, 5 months)
- Obtain top soil, manure and promix to use as potting material in plastic planting pots (24 cm height and 26cm diameter)
- Fill the plastic planting pots with mixture of top soil, manure and promix up to the rim
- Plant the seedling in a plastic planting pot and cover the seed nut with the soil at the level of collar region
- Water the seedlings sufficiently
- Place the potted coconut seedlings in greenhouse with adequate light source

### 2. Protocols for Culturing of Stock Colony of RPM on Coconut Seedlings

- Clean five potted coconut seedlings carefully with a soft brush to remove all other arthropods
- Tie a yellow tagging tape covered with petroleum jelly around the base of the palms to exclude any crawling arthropod
- Collect RPM infested coconut leaves from the field
- Cut the field collected RPM infested leaf into 15cm sections (leaf bits)
- Examine leaf bits to remove all other arthropods with a camel hair brush
- Adjust RPM population on 15 cm leaf bit to ensure that 300 RPM females are present
- Clip or attach the leaf bit to the abaxial surface of the lower leaf of the potted coconut seedling to facilitate movement of RPM from leaf bit to lower leaf of seedling
- Enclose plants in a cage which will keep out all other insects but will allow adequate sunlight
- Leave the seedlings undisturbed for 45 days until the RPM population is established and multiplying on the seedlings
- Monitor and water the seedlings adequately once or twice/week
- After 45 days collect RPM from one seedling and use as stock to infest a new seedling
- Label the seedlings with index cards for recording the observations
- Repeat the procedure every week to infest another batch of five seedlings

## 1. Types of Arenas

No	Details	Container size	Sponge size	Size of Leaf bit	Purpose
Type 1	Metal tray	32.5cm x 26.5cm x 4.5cm	27.0cm x 20.0cm x 3.5cm	24.0cm x 2.5cm	Mass culturing of RPM
Type 2	Plastic tray	18.0cm x 12.0cm x 7.0cm <b>Maintenance of Natural Enemy <i>Amblyseius largoensis</i> in the Laboratory</b>	16.0cm x 10.0cm x 3.5cm	8.0cm x 2.5cm	Mass culturing of RPM/arenas for laying females of RPM
Type 3a	Petri dish	8.0cm diameter	6.0cm x 6.0cm x 2.0cm	8.0cm x 2.5cm	Biology Studies of RPM/ natural enemy, <i>Amblyseius largoensis</i>
Type 3b	Petri dish	4.0cm diameter	3.0cm x 3.0cm x 2.0cm	4.0cm x 2.5cm	Biology Studies of RPM/ natural enemy, <i>Amblyseius largoensis</i>

## 2. Protocols for Arrangement of Arenas

- Cut mature coconut leaves into rectangles of required sizes
- Cut sponge pieces of a similar size to the container
- Pre-soak sponge pieces for 20minutes in water and place in the respective container

- Place coconut leaf bit (rectangles) with abaxial surface facing up on sponge saturated with water in the container
- Insert cut ends of the coconut leaf bit into the sponge by making a slit in the sponge
- Secure the leaf bit on the sponge with the help of straight pins
- Place black paper strip along the edges of the leaf section to minimise mite escape
- Add water to the container to prevent predator or prey from escaping and keep leaf bit fresh
- Maintain water in the container up to the level of sponge constantly
- Prepare required number of arenas daily over a period of 7 days

### **3. Protocol for Release of RPM into Arenas to Deposit Eggs**

- Set up the laboratory conditions at 27 °C Temperature, 70% RH and 12:12 Photoperiod
- Release 50 RPM females/arena of Type 2 (plastic tray)
- Prepare 25 arenas of Type 2 in 5days@5arenas/day (required number of arenas/day can be made based on the scale of need)
- Allow the females to lay eggs for 5 days
- After 5 days, take out females and use eggs

### **4. Protocol for RPM Life Cycle Study**

- Prepare required number of arenas of Type 3a and 3b
- Release 5 RPM females on each arena using a camel hair brush
- Leave the females for 5 days to lay the eggs
- After 5 days, remove females from the arenas with the help of camel hair brush
- Pool eggs of same day laying from different arenas by cutting the leaf area
- Prepare minimum 10 to maximum 20 arenas with eggs @ 20 eggs/arena
- Monitor and maintain these arenas daily till the adult stage
- Observe the arenas daily under a dissecting microscope for: date of eclosion, 1<sup>st</sup> moult, 2<sup>nd</sup> moult and 3<sup>rd</sup> moult
- Record these observations on data sheet
- Transfer the RPM onto fresh arena when the leaf dies up by keeping the dried leaf arena on the fresh arena or by transferring the RPM stages with the help of camel hair brush



- Determine period of different immature stages and total life cycle by tabulating the data
- Place pair of RPM (one ♀ and one ♂) on each of 5 arenas
- Record details of fecundity, average egg laying/day and longevity by daily observation under the dissecting microscope until adults die

#### 5. Protocol for Mass Culturing of RPM

- Prepare required number of arenas of Type 1 (metal tray)
- Place 50 pairs of female and male RPM on each arena
- Monitor and maintain the water level in the arena daily
- Transfer the RPM onto fresh arena when the leaf dies up by keeping the dried leaf arena on the fresh arena or by transferring the RPM stages with the help of camel hair brush
- Collect RPM from one arena and
  - i. Use for making subcultures on new arena
  - ii. Use as prey for *A. largoensis* cultures
- Based on the test arena (Type 1) from 50 females, 120 eggs can be obtained in 10 days from a single arena

#### 6. Protocol for Life Cycle Study of *A. largoensis*

- Transfer 5 - 10 *A. largoensis* females from the field collected leaf samples to a one end open glass vial (size 8.5cmx2.3cm) and close the open end with cotton lid
- Starve the predators by isolating *A. largoensis* adults with no food, for 24 hours
- After 24 hours provide RPM stages (eggs, larva, protonymph $etc.$ ,) daily to feed *A. largoensis* adults in the glass vial
- Observe adults every 12 hours for egg laying and keep the newly deposited eggs in the glass vial along with female till the eclosion of egg
- Prepare arena Type 3b with RPM population to maintain emerged larvae of *A. largoensis*
- As soon as eggs hatch, transfer emerged larvae of *A. largoensis* to experimental arenas individually
- Observe at 12 hour intervals under a dissecting microscope for: 1<sup>st</sup> moult, 2<sup>nd</sup> moult and 3<sup>rd</sup> moult

- Record the survivorship and duration of the immature stages
- R
- Replenish the diet (RPM) on each arena daily by placing the leaf bit containing RPM eggs (ensure that leaf bit contains 40 RPM eggs)
- R
- Replace arena when it dries up
- Record observations - daily from arenas to obtain life stages of *A. largoensis*
- After completing development, determine sex of the predator and transfer male from the colony into each arena where an adult female has emerged
- Replace males if mortality occurs
- Observe adults daily and determine their pre-oviposition and oviposition periods, and the longevity of both sexes
- Remove eggs and transfer to new experimental arenas to determine the sex ratio of the offspring

#### 7. Protocol for Feeding Potential Study of *A. largoensis*

- Obtain RPM from stock colonies reared on potted coconut seedlings in green-house.
- Prepare arenas with laying females of RPM for 5 days @5 arenas/day by placing 50 RPM females in oviposition upon Type 2 arenas daily
- Remove females from arena after 5 days to use the eggs laid by the females
- Obtain desired number of eggs (40) by cutting the leaf bit from the arena
- Obtain *A. largoensis* from the field population
- Transfer a single *A. largoensis* female into a one end open glass vial (size 8.5cm x 2.3cm)
- Similarly, isolate 20 *A. largoensis* individually in 20 vials
- Close the open end of glass vial with a cotton lid
- Starve the *A. largoensis* for 24 hours
- Introduce leaf bit with 40 RPM eggs into the vial which contains starved *A. largoensis* female
- Record the number of eggs consumed in 24 hours
- Replenish the eggs every day to make up 40 eggs in every vial

- Continue provision of eggs till *A. largoensis* dies
  - Tabulate the daily observations recorded to get feeding efficiency of natural enemy
- 8. Protocol for Mass Culturing of *A. largoensis* on Potted Coconut Seedlings**
- Take RPM colonised potted coconut seedlings from green house to laboratory
  - Transfer *A. largoensis* from field population into a glass vial (8.5cm x 2.3cm) using pig hair brush
  - Transfer 25 pairs of *A. largoensis* into a glass vial and close the vial with cotton lid
  - Place the vial containing 25 pairs of *A. largoensis* at the collar region of the seedling and remove the cotton lid
  - Allow *A. largoensis* to walk unto the plant and settle
  - Repeat the procedure on other seedlings
  - Maintain the seedlings in the lab by securing with cages and watering regularly
  - Leave the seedlings undisturbed for 15 days for the establishment and multiplication of *A. largoensis* on the seedlings
  - Monitor the seedlings at fort-nightly intervals
  - Observe at 5 random spots on the leaves of seedling
  - Record *A. largoensis* population with the help of a 10X lens
  - Re-release *A. largoensis* if the number is low or fails to be established
  - After 30 days, harvest *A. largoensis* from the seedling to release in the field
  - Harvest *A. largoensis* from the seedling by cutting leaf bits of 8cm size with high number of *Amblyseius largoensis*

**9. Protocol for Culturing of *A. largoensis* in the Containers**

- Wash the plastic containers measuring 36.5cmx25.5cm and dry
- Select the leaves which are infested with RPM from the field sample and remove all other arthropods
- Remove all natural enemies except *A. largoensis* from the selected leaves
- Cut the leaf bits of required size (30cm length) to fit into the plastic container
- Place 10 leaf bits in a container
- Collect 10 pairs of *A. largoensis* from field samples and place in a glass vial

- Place the glass vial in the container and open the cotton lid to allow *A. largoensis* to walk onto the leaf bits
- Cover the plastic container with lid and maintain in the lab at 27°C Temperature, 70% R. H. and 12:12 photoperiod
- Monitor and maintain the containers by placing fresh RPM infested leaf bits whenever the leaves dry up or RPM populations have declined
- Monitor the cultures in the container at 3-day intervals
- Replenish with *A. largoensis* if its count goes down in the leaf sample
- Allow *A. largoensis* to multiply for a period of 30 days in the container but continue to monitor at 3-day interval
- Harvest *A. largoensis* from the container by taking out the leaf bits containing *A. largoensis* and release in the field once the *A. largoensis* number/leaf bit exceeds 10 females
- Repeat same procedure to maintain more containers

#### **10. Protocol for Culturing of *A. largoensis* on Arenas**

- Cut leaf bit 6cm length from field collected RPM infested leaves
- Secure the leaf bit onto the sponge in a petri dish
- Place black paper strips along the edges to minimize mite escape
- Place each rearing unit in a petri dish and add water daily to keep sponge wet
- Isolate and transfer 25 - 50 *A. largoensis* females into rearing arenas
- Provide the predatory mites with RPM (all stages brushed onto the arena)
- Give honey-water solution (1:1) as a food source 3 times a week
- Sub-culture the colonies by transferring a minimum of 25 - 50 adult female predators to a new arena every 3 weeks

#### **11. Protocols for Field Release of *Amblyseius largoensis***

- Maintain RPM stock colonies on the potted coconut seedlings (6 months old) under green house conditions for about 45 days
- Introduce 25 pairs of *A. largoensis* on single seedling or 10 pairs in a plastic container containing RPM infested leaf bits or arenas in petri dish

- Allow for 30 days for multiplication of *A. largoensis* on coconut seedlings
- Collect leaves (leaf bits-6cm size) from potted seedlings or plastic container or arenas with *A. largoensis*
- Clip the leaf bits with *A. largoensis* to the RPM infested coconut palm in the field @2 leaf bits/one whorl of leaves in opposite direction with 100 pairs of *A. largoensis* if the palm height is reachable

OR

- Keep the leaf bits with *A. largoensis* cultures in the vials or small zipper lock polythene bags for transporting to the field
- Select size of the zipper locked bag (small, medium or large) depending on the size of the leaf bit
- Use ice box to carry the cultures to the field
- Evening hours would be congenial for release of the natural enemy
- Place these vials or zipper locked bags, in between the fronds in opposite directions at the rate of 2 vials or zipper locked bags/whorl with 100 pairs of *A. largoensis*
- Remove the lids of vials or open the zipper locked bags to facilitate the *A. largoensis* to walk away to reach the RPM on the palm
- Periodical releases can be made at 15-20 days intervals based on the levels of RPM infestation

## 12. Critical Factors for Successful Mass Culturing of Mites

- Minute size of mites needs microscopic observation
- Microscopic observation for long hours
- Strenuous to the eyes
- Colour of the natural enemy, *A. largoensis* (hyaline)
- Swift movements of the natural enemy, *A. largoensis*
- Natural enemy, *A. largoensis*, frequently moving away from the arenas
- Keeping the leaf arena fresh for a long time
- Daily watering of arenas
- Monitoring and maintenance of cultures demands 24 x 7 hour requirement of human resources