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**Balaji Nayak M**  
M.Sc. Department of  
Agricultural Entomology,  
GKVK, University of  
Agricultural Sciences, Bangalore,  
Karnataka, India

**Dr. C Chinnamade Gowda**  
Professor, Department of  
Agricultural Entomology,  
GKVK, University of  
Agricultural Sciences, Bangalore,  
Karnataka, India

**Chidananda Hirematha**  
M.Sc. Department of  
Agricultural Entomology,  
GKVK, University of  
Agricultural Sciences, Bangalore,  
Karnataka, India

**Dr. Aravinda Hubballi**  
M.Sc. (Agri.), Ph.D.,  
Department of Agricultural  
Entomology, GKVK, University  
of Agricultural Sciences,  
Bangalore, Karnataka, India

**Corresponding Author:**  
**Balaji Nayak M**  
M.Sc. Department of  
Agricultural Entomology,  
GKVK, University of  
Agricultural Sciences, Bangalore,  
Karnataka, India

## Description of different developmental stages of *Raoiella macfarlanei* Pritchard & Baker false spider mites

**Balaji Nayak M, Dr. C Chinnamade Gowda, Chidananda Hirematha and  
Dr. Aravinda Hubballi**

### Abstract

The genus *Raoiella* represents one of the important genera of the family Tenuipalpidae, which comprises half a dozen species. Members of this group have been given the status of an economic species. Present experiment was carried out on different developmental stages of *R. macfarlanei* viz., egg, larva, protonymph, deutonymph and adults were described under laboratory conditions at UAS GKVK Bengaluru. Current study revealed that, eggs are ovoid in shape, reddish in colour, soft, smooth measured 96.0 long and 62.0 wide, larva with three pairs of legs measured 174.0 long and 119.0 wide, the protonymph possessing four pairs of legs 185.0 long and 127.6 wide, the deutonymph larger in size than protonymph, red in colour, 267.0 long and 184.0 µm wide and the freshly emerged adult almost similar in size of deutonymph except for development of genital structures. Posterior end of male is conical in shape while that of the female is broader.

**Keywords:** Tenuipalpidae, *Raoiella macfarlanei* egg, larva, protonymph, deutonymph

### Introduction

The Tenuipalpidae (Acari) constitute an economically and agriculturally important group of mites ranging in size from 190 to 330 micrometer. They look like spider mite but, do not spin the webbings and hence are popularly known as false spider mites. These mites are dorsoventrally flattened, slow moving, differently coloured and have worldwide distribution.

The Tenuipalpid mites are exclusively phytophagous causing considerable damage and yield losses to many agricultural and other economic plants. Some of the false spider mites are host specific but, majority of them are polyphagous and have a very wide host range (Ghai and Shenhmar, 1984 and Sadana, 1985) [4, 8]. Many of them are serious pests of fruit trees, vegetable crops, ornamental and medicinal plants, fibre and oilseed crops. They feed on almost all parts of the plants but, usually on the underside of the leaves near the midrib, veins, leaf stalk or petiole, some may confine to twigs, floral heads and leaf sheath of monocotyledonous plants or form galls on host plants within which they confine.

The first record of the family Tenuipalpidae Berlese, dates back to 1834 when Duges described *Trombidium caudatus* on *Laurestinus* from France. This species was subsequently included in *Tenuipalpus* which was established by Donnadieu (Duges, 1834; Ghai & Shenhmar, 1984) [3, 4]. *Tenuipalpus* Donnadieu, on which the family name is based was placed under the family Tetranychidae until 1913 when Berlese separated a new group Tenuipalpini from Tetranychidae in an obscure, privately published paper in 1913 (Berlese, 1913; Donnadieu, 1875) [1, 2]. This name, Tenuipalpini, remained unknown till Sayed (1950) [10] changed the family name Phytoptipalpidae Ewing and Trichadenidae Oudemans to Tenuipalpidae.

Studies on the exploration of Tenuipalpidae are meagre across India as also in the state of Karnataka. Hence, it was felt necessary to study the fauna of tenuipalpid mites. The present study aimed at study of different developmental stages of *R. macfarlanei* viz., egg, larva, protonymph, deutonymph and adults were described.

### Material and Methods

The material used and the methodologies followed for collection, preservation, extraction, processing, description, illustration, measurement of mite specimens and for investigation on the different developmental stages.

**Collection of mites**

The plant samples harbouring mites were collected from premises of University of Agricultural Sciences, GKVK, Bangalore, and the plant samples were collected for extraction of mites. Each sample consisted of 2-3 branches of plants with leaves.

Two to three branches of plant with few leaves were sampled and placed in polyethylene bag and fastened with an elastic band, duly labelled with details of habitat, locality, date of collection *etc.*, and brought to the laboratory carefully for further processing like extraction and examination of mite specimens. Precaution was taken not to expose the sample to direct sunlight to prevent the accumulation of water droplets inside.

Each sample was assigned an accession number and the details were recorded in the field note book *viz.*, place of collection, date of collection, plant host and the collector *etc.* The plant samples in most cases were processed immediately giving priority to succulent samples, otherwise stored at 5-15 °C in a refrigerator and further processed within next 2-3 days.

**Extraction of mites from the plant material**

The plant sample was examined in the laboratory under a stereo zoom microscope and the mites present were picked with a fine needle/camel hair brush dipped in alcohol/Hoyer’s medium. Soft bodied mites were mounted directly using Hoyer’s medium on a glass slide immediately after picking from the plant material. Hard bodied mites were cleared with lacto-phenol and then mounted on Hoyer’s medium on a glass slide. The plant samples were preserved in a herbarium, photographed and identified with the help of a botanist from Mahatma Gandhi Botanical Garden at UAS, GKVK, Bangalore (Figure 1).

**Processing of mite specimens**

**Clearing**

Initially hard bodied mites were cleared in lacto-phenol in a cavity slide and kept at 40°C in a hot air oven for 2 to 7 days to enable clearing of the specimens.

**Mounting medium**

Mounting medium used in the present study was Hoyer’s medium with the following ingredients mixed in a sequence.

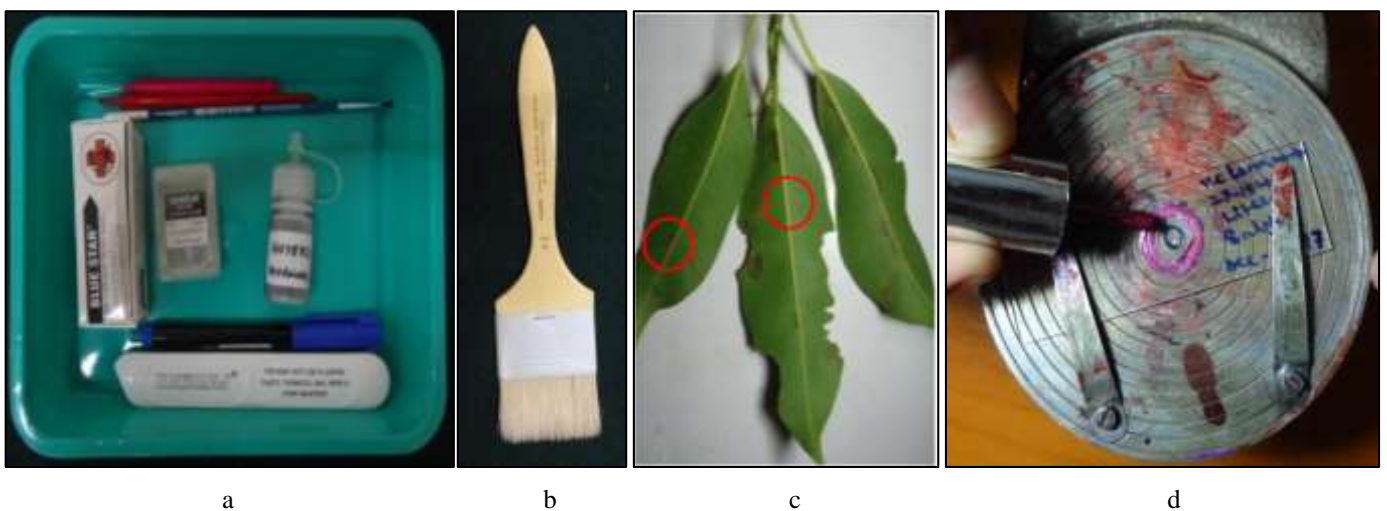
- Distilled water .....50 ml
- Gum Arabica .....30 g
- Chloral hydrate.....200 g
- Glycerol .....20 g

**Preparation of permanent slides**

Mite specimens cleared in lacto-phenol were transferred using a spatula to a cavity block with clean water for washing and rinsing with water. Specimens were then transferred on to a drop of Hoyer’s medium on a clean glass slide after removing excess water from the specimens using blotting paper. Soft bodied mites were directly placed on the Hoyer’s medium from the plant sample. After placing the specimen on the glass slide the mite specimen was pushed down the medium to rest close to the surface of the glassslide, oriented vertically with legs and gnathosoma stretched properly. Glass cover slip (1 No.) of diameter 12 mm was placed gently on the medium in such a way to avoid air bubble between the glass slide and the cover slip. Always a single specimen was mounted on each slide and up to 10 slides were prepared from each plant sample depending on the availability of mite specimens on the plant sample. Such slides were labelled on the right side with details of place, date, habitat, and collector, accession number and with the identity label on the left side of the slide. The slides were kept in hot air oven at 40-45 °C for at least two weeks or till the specimen get cleared sufficiently. After proper drying of the slides, to avoid shrinkage of the mounting medium, edges of the cover slips were sealed using nail polish with a fine brush on a ringing disc (Figure 1d). The slides were stored in slide storage box in a cool place (Figure 1).

**Materials and Methods**

A cross-bred buff reducible hernias are suitable for conservative therapies. The hernial ring can be closed using conservative treatments such as belly bandages/abdominal.



**Fig 1:** Material used for collection, extraction and preservation of mites: (a) microneedles, camel hair brush, glass slides, cover slips, Hoyer’s medium and lab cutter; (b) *Raoiella macfarlanei* infected leaves; (c) brush used to clean the leaves; (d) ringing disc/table

**Descriptions**

The morphological characters of mites were studied under Zeiss Scope A1 phase contrast microscope at 200, 400 and

1000 x magnification.

**Measurements**

The following measurements were made for individual specimen: dimensions of body, body shields, leg segments; lengths of idiosomal setae, leg setae, rostrum, chelicerae, palpal segments; length and width of female genitalia; distance between setae *etc.* Number of setae/solenidia on the body, legs, chelicerae, pedipalps and on the genital region was recorded.

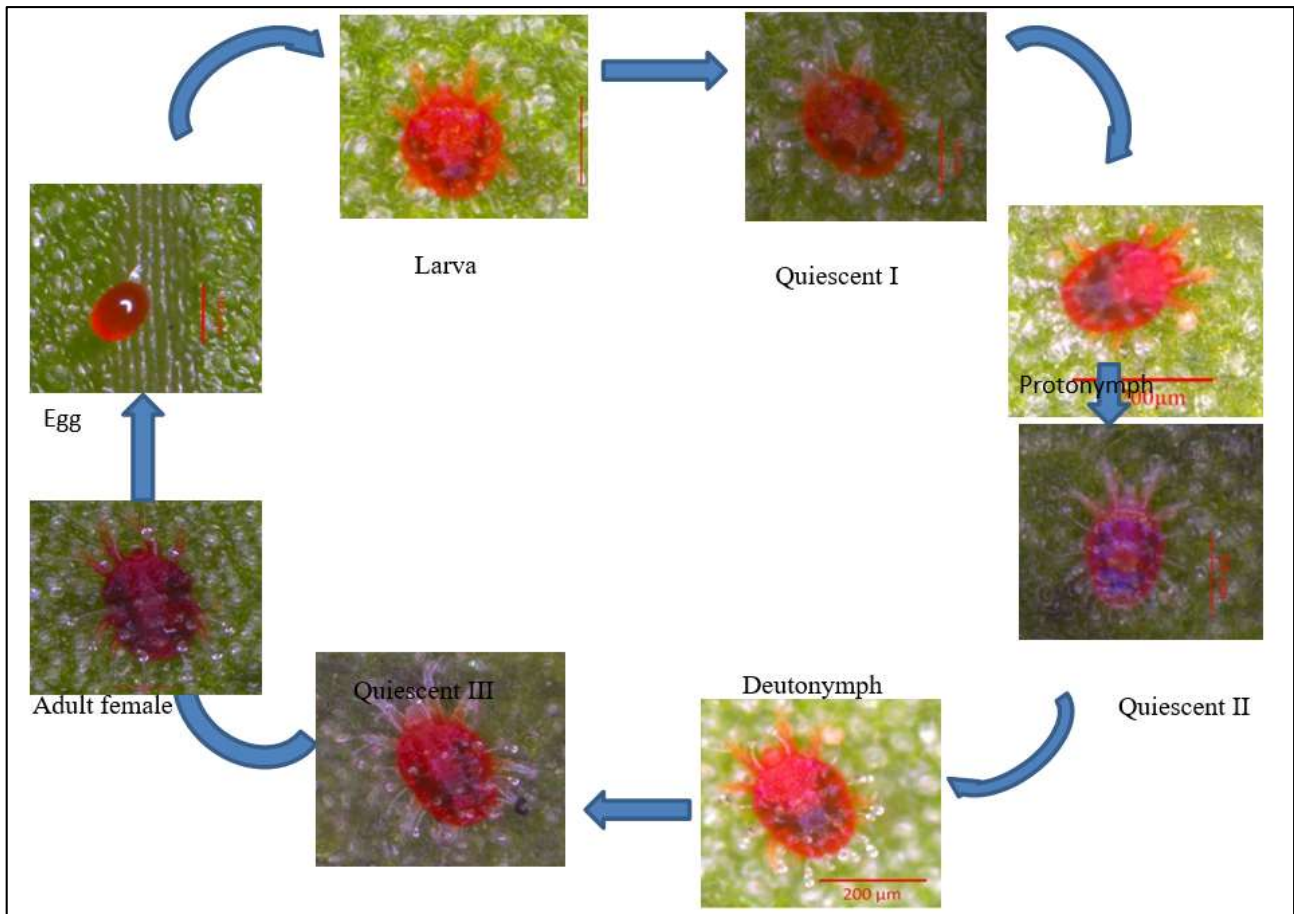
All measurements are in micrometer ( $\mu\text{m}$ ). The measurements are mean (range mentioned in parenthesis) from two to three specimens.

**Illustration**

The images of mite specimens depicting morphological characters of taxonomic importance were captured using Jenoptic digital camera attached to Zeiss Scope A1 phase contrast microscope at 400 X or 1000 X magnification.

**Description of different developmental stages of *Raoiella macfarlanei***

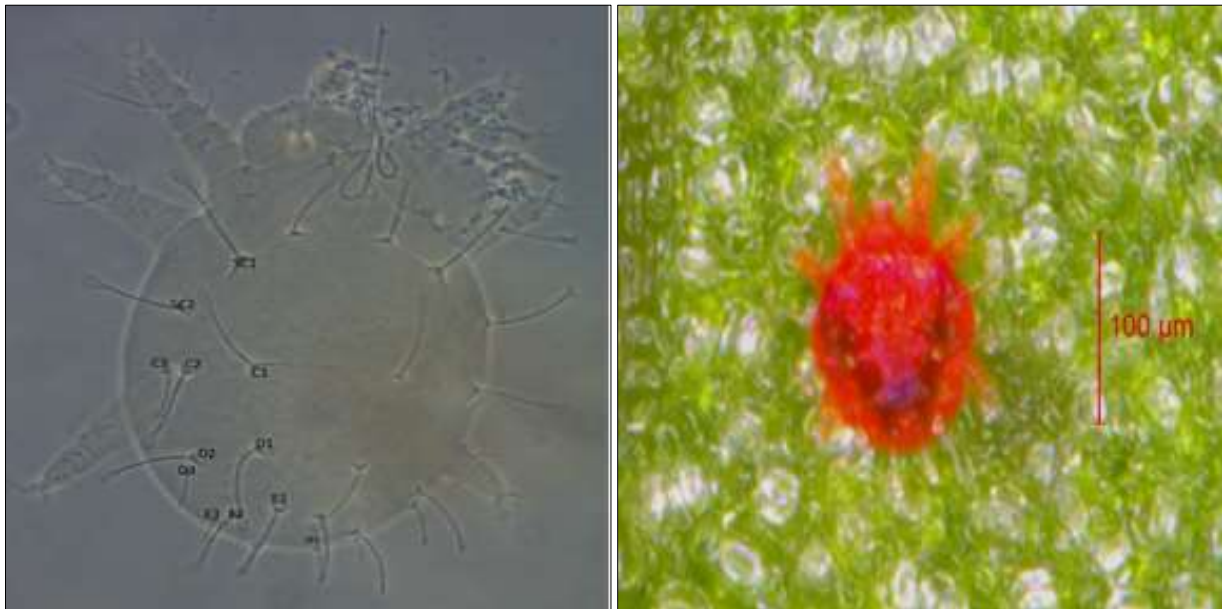
Five permanent slides for each developmental stage (larva, protonymph, deutonymph and adult male and female) of *Raoiella macfarlanei* were prepared and used to study morphological features of taxonomic importance, to measure the setae and other such characters.



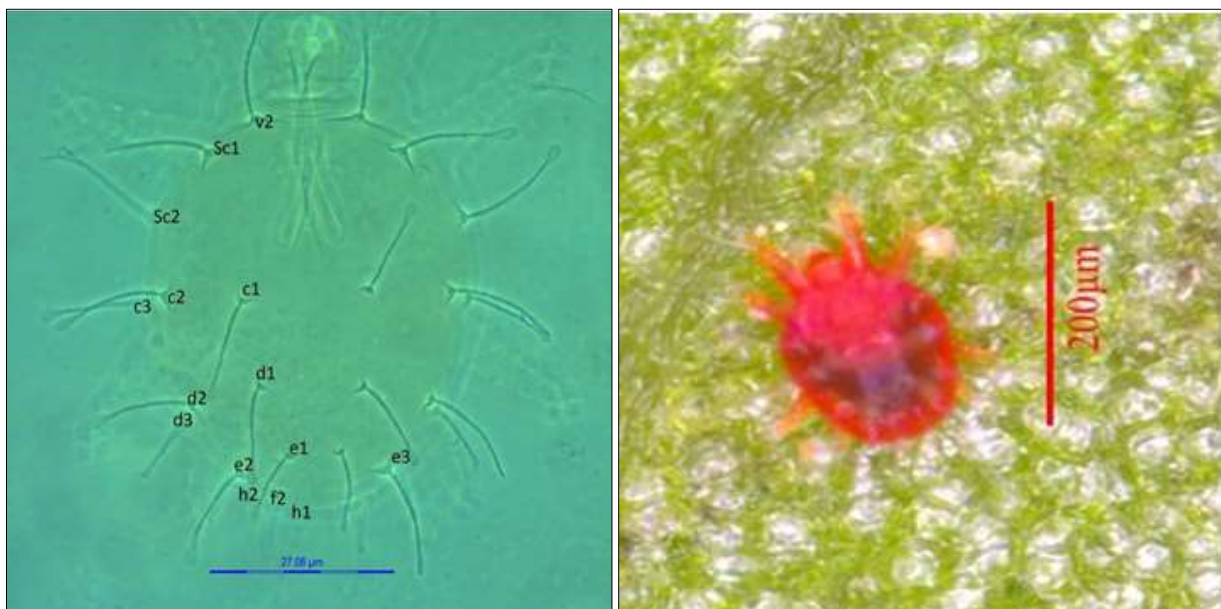
**Fig 2:** Life cycle of *Raoiella macfarlanei*



**Fig 3:** Egg of *Raoiella macfarlanei*



**Fig 4:** Larvae of *Raiiella macfarlanei*



**Fig 5:** Protonymph of *Raiiella macfarlanei*

## Experimental Results

### Description of different developmental stages of *Raiiella macfarlanei*

The different life stages of *Raiiella macfarlanei* recorded were egg, larva, Quiescent I, protonymph, Quiescent II, deutonymph, Quiescent III and adult (Figure 2).

#### Egg

Eggs were ovoid in shape with one end being slightly broad, reddish in colour, soft, smooth and sticky when laid. The eggs measured about 96 (90-104) long and 62 (55-78) wide. The freshly laid eggs were adhered to the surface of leaf and attempt to remove the eggs usually resulted in their damage. After a few hours chorion became firm. When the egg was ready for hatching, its broad end becomes swollen due to the larval pushing against the inner wall of the chorion. After a few hours, the chorion split transversely near the swollen end of the egg. The larva then crawled out with its anterior end first. The chorion appeared transparent and colourless

immediately after emergence of the larva (Figure 3).

#### Immature mobile stages

The immature mobile stages included a larval and two nymphal instars with intermediate inactive quiescent stages. Feeding, growth and limited dispersion took place during active immature phases and transformation into the subsequent stages during the inactive quiescent stages. During the inactive phase each stage is terminated by moulting.

#### Larva

The newly emerged hexapod larva was bright orange-red in colour, two days later a blackish tinge developed on the posterior end of dorsum. The newly emerged larva wandered around the egg shell for a few minutes and then settled down. Larva measuring about 174 long and 119 wide, oval outline with faint transverse striations on the dorsal hysterosoma. All setae strongly tuberculate and serrate, three pairs of dorsal propodosomal setae (v2, Sc1 and Sc2), spatulate, measuring

v2-30.9 (29.8-32.5), Sc1-42.4 (41.5-43.1) and Sc2-33.4 (32.6-34.3), the middle pair (Sc1) slightly longer than the first and third pair (v2 and Sc2) (Figure 4).

**Hysterosoma:** Hysterosoma with one pair of humeral setae (c3), measuring about 19.5(18.7-20.5). Three pairs of dorsocentral setae (c1, d1 and e1), measuring c1-34.7 (33.4-36.1), d1-32.9 (31-33.9) and e1- 29.2 (27.6-30.7). Four pairs of dorsosublaterals setae (c2, d2, e2 and f2), spatulate, measuring about c2-29.5 (28.5-31.3), d2-28.9 (27-31.3), e2-26.8 (25.9-27.4) and f2 27.7 (25.3-28.6). Five pairs of dorsolateral setae(c3, d3, e3, h1 and h2) gradually reducing in length from first to fifth measuring c3-21.5 (18.8-23.5), d3-17.7 (17-18.6), e3-17 (16-17.9) , h1- 5.7 (4.3-6.6) and h2-5.8 (5.1-6.8). Anal setae two pairs and minute.

**Length of legs:** Leg I 62, leg II 51, leg III 46. Tarsi I and II each with a single sensory rod, tibiae I with dorsal setae very long and filiform.

#### Protonymph

The protonymph differed from larva essentially being larger in size and in possessing four pairs of legs. Red in colour, the body 185 long and 127.6 wide. Propodosoma with three pairs of spatulate setae (v2, sc1 and Sc2), measuring v2-38.9 (36.7-39.5), Sc1-43.4 (42.5-44.1) and Sc2-42.8 (40.6-43.3), the middle pair (Sc1) slightly longer than the first and third pair (v2 and Sc2) (Figure 5).

**Hysterosoma:** Hysterosoma with one pair of humeral setae (c3), measuring about 24.5 (23.7-25.5). Three pairs of dorsocentral setae (c1, d1 and e1), measuring c1-36.7 (35.4-37.1), d1-34.9 (33-35.9) and e1-30.2 (29.6-30.7). Four pairs of dorsosublaterals setae (c2, d2, e2 and f2), spatulate, measuring about c2-31.5 (29.5-32.3), d2-29.4 (28-30.3), e2-29.8 (27.9-30.4) and f2 -30.7 (29.3-31.6). Six pairs of dorsolateral setae(c3, d3, e3, f3, h1 and h2) gradually reducing in length from first to fifth pair and sixth pair was minute, measuring c3-32.5 (30.8-33.5), d3- 26.7 (24.6-28.6), e3-21 (19-22.9), f3-16.5 (15.2-17.1), h1- 7.7 (6.3-7.9) and h2-5.1 (4.1-5.4). Anal setae two pairs and were minute.

**Length of legs:** Leg I 91, leg II 73, leg III 59 and leg IV 48. Tarsi I and II each with a single sensory rod, tibiae I with dorsal setae very long filiform.

#### Deutonymph

The deutonymph differed from protonymph essentially being larger in size, red in color, the body 267 long and 184 wide. Almost oval in shape, widest across the propodosoma and broadly rounded posteriorly. All dorsal setae strongly tuberculate and serrate. Propodosoma with three pairs of spatulate setae (v2, sc1 and Sc2), measuring v2-54.9 (52.7-56.5), Sc1-51.4 (50.5-52.1) and Sc2-53.8 (50.6-54.3) (Figure 6).

**Hysterosoma:** Hysterosoma with one pair of humeral setae (c3), measuring 50(45-56); three pairs of dorsocentral setae (c1, d1 and e1), measuring c1-40(38-43), d1-33 (32-35) and e1-29 (28-30); dorsosublaterals setae four pairs (c2, d2, e2 and f2), measuring c2-38 (36-40), d2-34 (31-35), e2-33 (31-35), f2- 54(50-59); dorsolateral setae five pairs (d3, e3, f3, h1 and

h2), measuring d3-48(47-50), e3-45(43-49), f3-24(20-27), h1-18(16-22) and h2-24(22-28). First pair of dorsocentral hysterosomal setae (c1) very long and spatulate. Fourth pair of dorsosublaterals hysterosomal setae (f2) very long and longer than first three dorsosublaterals pairs (c2, d2 and e2). Second and third pair of dorsosublaterals setae (d2 and e2) was equal in length. Anal and genital Figure each with two pair of minute setae.

**Length of legs:** Leg I 115, leg II 92, leg III 79 and leg IV 80. Tarsi I and II each with a single sensory rod, tibiae I with dorsal setae very long filiform.

Sex differentiation could be observed in deutonymphal stage. The female deutonymph had broad hysterosoma and the body was oval in shape, whereas, the male deutonymph had a narrow pointed or wedge shaped hysterosoma.

#### Adult female

Body length 269 (263-275), width 175 Dorsum of idiosoma smooth. Dorsal shield with 16 pairs of setae, three on the propodosoma and remaining on hysterosoma, setae long and spatulate (Figure 7).

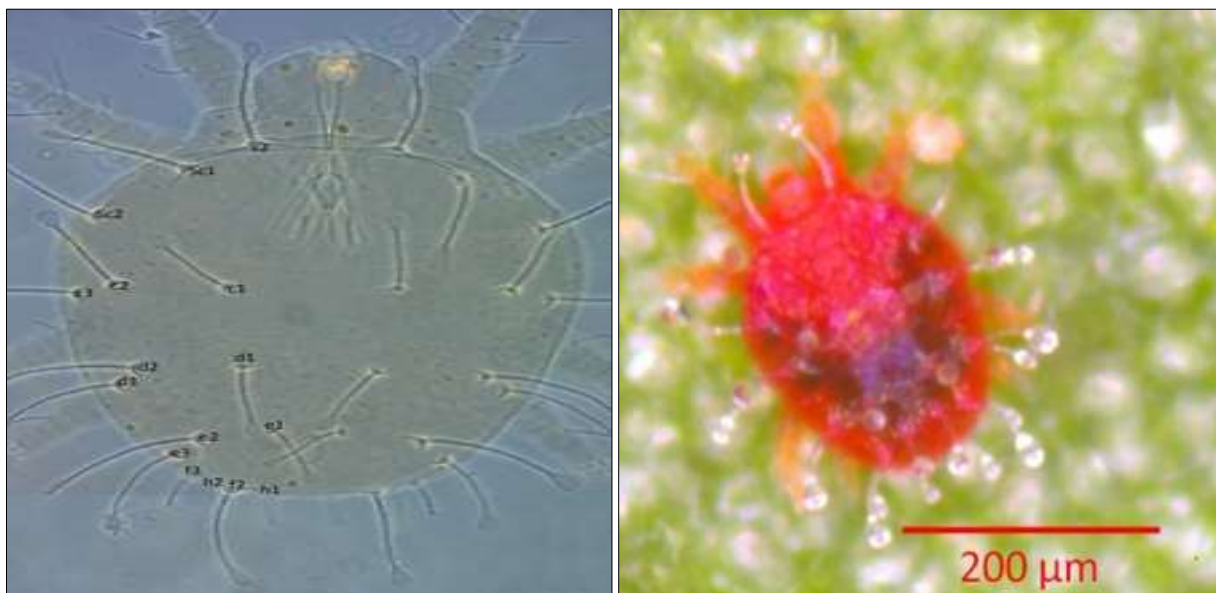
**Propodosoma:** Measurements of three pairs of propodosomal setae (v2, Sc1 and Sc2), measuring v2-59 (58-60), Sc1-51(50-51), Sc2-62 (61-64). Eyes two pairs, one pair on each side.

**Hysterosoma:** Hysterosoma with one pair of Humeral setae (c3), measuring 56(55-56); three pairs of dorsocentral setae (c1, d1 and e1), measuring c1-42(41-43), d1-35 (34-37) and e1-31 (30-32); dorsosublaterals setae four pairs (c2, d2, e2 and f2), measuring c2-42(41-43), d2-35 (34-37), e2-33 (31-35), f2- 61(59-62). Dorsolateral setae five pairs (d3, e3, f3, h1 and h2), measuring d3-52(51-53), e3-48(45-50), f3-26(25-27), h1-21(20-22) and h2-27(25-28). First pair of dorsocentral hysterosomal setae (c1) very long and spatulate. Fourth pair of dorsosublaterals hysterosomal setae (f2) very long and longer than first three dorsosublaterals pairs (c2, d2 and e2). First and second pair of dorsosublaterals setae (c2 and d2) was equal in length to the first and second pairs of dorsocentral setae (c1 and d1).

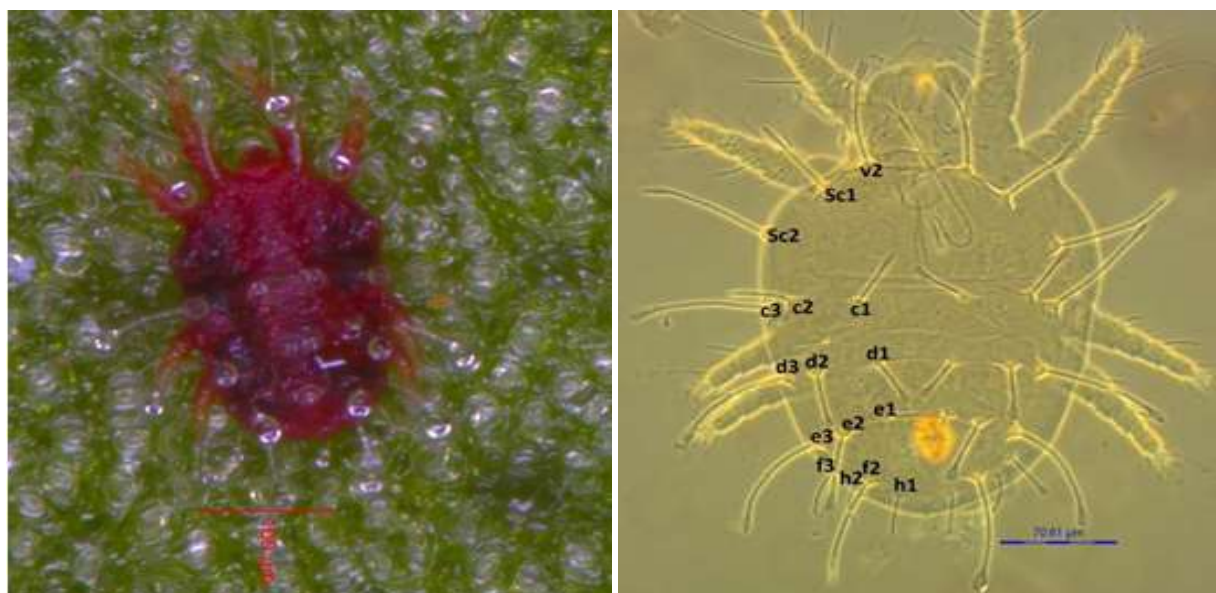
**Venter:** Venter of the body striated, striations transverse medially and semi-longitudinal laterally. Ventral propodosomal setae one pair (1a), simple and measuring about 48.6(46-50); anterior medioventral metapodosomal setae one pair (3a), simple and measuring about 11.8 (10-12); posterior medioventral metapodosomal setae one pair (4a), simple and measuring about 14.5 (13-16); ventral Figure with one pair of setae (Ag), measuring Ag-12(11-14); genital Figure setae two pairs, simple, measuring g1-13 (12-15) and g2-14 (13-15), anal Figure with wavy striations, anal setae two pairs (Ps1 and Ps2), measuring Ps1-12.5 (11-14) and Ps2-13 (12-14).

Legs four pairs, setae on legs I-IV; coxae 1-1-0-0, trochanters 1-1-1-1, femora 4-4-2-2, genua 3-3-1-0, tibiae 4-4-3-3, tarsi 6(1)-6(1)-4-4. Tarsi I and II each with a single solenidion, tibiae I with dorsal setae very long filiform.

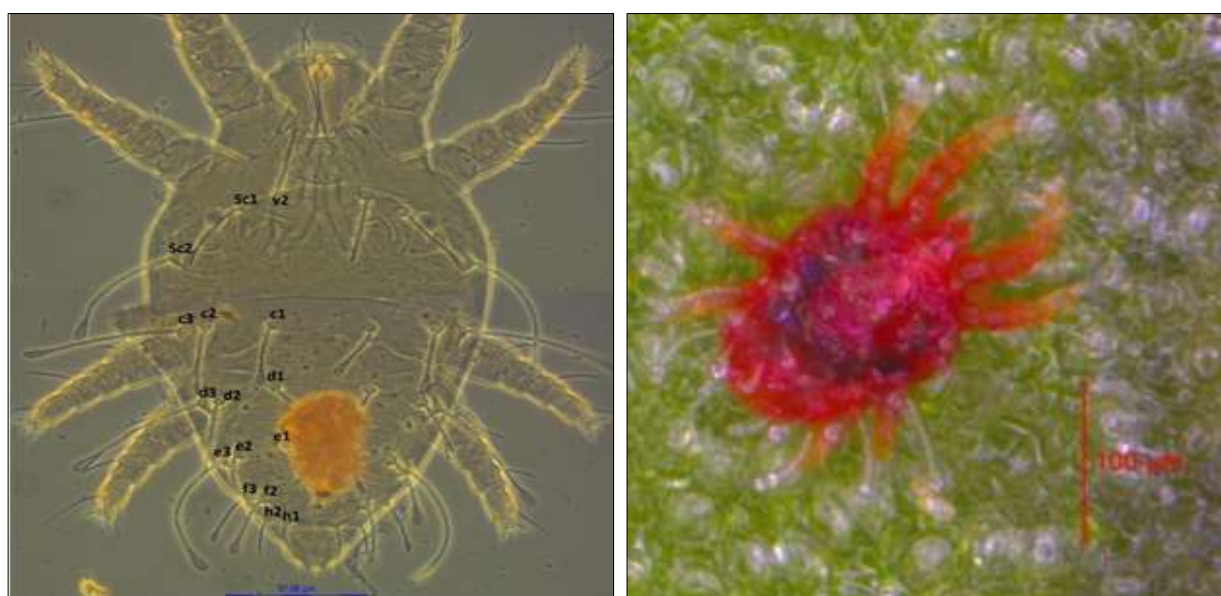
**Male:** Body smaller than female, chaetotaxy of male was same as female (Figure 8).



**Fig 6:** Deutonymph of *Raoiella macfarlanei*



**Fig 7:** Adult female of *Raoiella macfarlanei*



**Fig 8:** Adult male of *Raoiella macfarlanei*

## Discussion

### Developmental stages of *Raoiella macfarlanei*

In the present study, the descriptions were made on different developmental stages *R. macfarlanei* (egg, larva, protonymph, deutonymph and adults) and three quiescent stages (QI, QII and QIII). Eggs were ovoid in shape with one end being slightly broad, reddish in colour, soft, smooth and sticky when laid. The eggs measured about 96.0 (90.0-104.0) long and 62.0 (55.0-78.0) wide. Larvae were about 174.0 long and 119.0 wide, oval outlines with faint transverse striations on the dorsal hysterosoma. The protonymph differed from larva being larger in size and in possessing four pairs of legs which measured 185.0 long and 127.6 wide. The deutonymph was larger in size than protonymph, red in colour, body 267.0 long and 184.0 wide and the setae were similar to protonymph but little longer than on protonymph. There was no much difference between deutonymph and freshly emerged adult in their size and setal distribution except for development of genital structures in the latter. Adult male was smaller than female with similar chaetotaxy. Posterior end of male was conical in shape and that of female was more broader.

No literature is available on the description of immature stages of *R. macfarlanei* except for its faunal records. The results are discussed in the light of the information available on related species of mites. Pritchard and Baker (1958) [7] collected this species on olive from Rasal Hila, Cyrenacica and described it based on the female characteristics. *R. macfarlanei* was earlier recorded on *Jambosa vulgaris* in Karnataka, Kerala and Gujarat states (Nageshachandra, 1971; Sadana, 1997) [5, 9]. Nageshachandra (1980) [6] described the different stages of a near species *R. indica* and recorded the difference in the setal development in different stages. He observed that larvae lacking one pair of dorsolateral setae (f3), these setae developed in protonymphal stage. The similar morphometry was followed in the present study.

## Conclusion

Different developmental stages of *R. macfarlanei* viz., egg, larva, protonymph, deutonymph and adults were described. Eggs are ovoid in shape, reddish in color, soft, smooth measured 96.0 long and 62.0 wide, larva with three pairs of legs measured 174.0 long and 119.0 wide, the protonymph possessing four pairs of legs 185.0 long and 127.6 wide, the deutonymph larger in size than protonymph, red in color, 267.0 long and 184.0  $\mu\text{m}$  wide and the freshly emerged adult almost similar in size of deutonymph except for development of genital structures. Posterior end of male is conical in shape while that of the female is broader.

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