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Development and reproduction of the cotton mealybug, *Phenacoccus* solenopsis (Hemiptera: Pseudococcidae) in laboratory Maha I. Abd El- Razzik

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Abstract:

The cotton mealybug, Phenacoccus solenopsis Tinsley (Hemiptera: Pseudococcidae) is an invasive pest species that has appeared in different parts of the world. The aim of this study is to investigate the influence of four seasons temperature and relative humidity on the development and reproduction of the cotton mealy bug. The results revealed that, incubation period significantly decreased from 7.43 ±0.51 hours in winter at 15.43°C with 52% RH to 1.90±0.18 hours in summer at 29.6°C with 51.24% RH. The developmental period of nymphal stage tented to be shortened with the corresponding temperature. Life period of first, second and third nymphal instars of female was decreased from 17.59, 16.02 and 17.01 days in winter at 15.43°C with 52% RH to 4.26, 4.50 and 5.004 days in summer at 29.6°C with 51.24% RH, while, the two nymphal instars (1st and 2nd instars) of male decreased from 17.89, 17.11 days to 4.50, 6.033 days, respectively, in winter and summer at the same temperature and humidity. Pupae duration decreased from 19.00 days in winter at 15.43°C with 52% RH to 6.00 days in summer at 29.6°C with 51.24% RH. The total female life cycle decreased from 102.52 days in winter at 15.43°C with 52% RH to 38.43 days in summer at 29.6°C with 51.24% RH. The total male life cycle decreased from 56.20 to days in winter at 15.43°C with 52% RH to 17.50 days in summer at 29.6°C with 51.24% RH. There was significant effect of temperature on fecundity of adult female, the mean of female fecundity was 495±0.46 eggs under perfect conditions in summer at 29.6°C with 51.24% RH. Total life cycle (both male and female) was prolonged at lower temperature and shortened at higher temperature. Optimum temperature for development of the pest was in summer at 29.6°C with 51.24% RH. It is concluded that the study P. solenopsis biology, give the understanding of mode and degree of its population growth. Hence, this information will be helpful during the development of successful integrated pest management program (IPM) for *P. solenopsis*.

Introduction

The cotton mealybug, Phenacoccus solenopsis (Tinslev) (Hemiptera: Pseudococcidae) is a major threat to agriculture and horticulture in many tropical and subtropical countries which was found to attack large number of plant species including crops, vegetables, ornamental plants and weeds (Wang et al., 2010 and Abbas et al., 2010). However, a decade ago, the evidence of mealybug was reported from Pradesh, Madhya Pradesh Uttar and Karnataka (Bambawale, 2008 a and b). Further detail studies support the strong evidences for its presence in India (Hodgson et al., 2008). The newly world species of mealybug, P. solenopsis has emerged as a serious pest of cotton in Pakistan and India and is now being as a serious threat to cotton in China. It has been reported from 173 species in 45 families and from 26 countries in different ecological zones (Abbas et al., 2010). P. solenopsis cause crinkling, twist and condense flower, bud, bolls growth and finally it cause yield loss (Sahito et al., 2009). The mealybug insects has many traits make it a serious pest like the body covered with mealy wax section reduced the insecticide effects and save it from natural enemies attack, as well as, the highly spreading because diversity of reproduction manners and various host plant (Al-Rubeae and Al-Obaidi, 2014). In Egypt, this pest was recorded for the first-time infesting Hibiscus sp. in September, 2009 by Abd-Rabou et al. (2010). This pest spread rapidly on different host plants to the extent that recorded it on 29 host plant species belonging to 16 plant families including field crops (3), vegetables (3), ornamentals (7), weeds (13) and fruits (3) (Abdel-Razzik et al., 2015).

The present work was conducted to determine the effect of temperature and relative humidity of seasons on the biology of the cotton mealybug at different sets of climatic conditions.

Materials and Methods 1.Collection and rearing of insects:

To have a culture of the cotton mealybug, Р. solenopsis. ovi-positing females were collected from infested ornamental plant, Hibiscus sp. and reared on sprouting potato tubers under laboratory conditions, Plant Protection Research Institute. Giza, Egypt. The collected mealybug was identified at Scale Insects and Mealybugs Research Department, Plant Protection Research Institute, Agriculture Research Center.

To establish initial culture of P. solenopsis, steams of the host plants infested with adult females were brought to the laboratory individuals were separated and inoculated on sprouting potato in carton cylindrical boxes (8 cm long and 12 cm diameter) and reared in the laboratory. After about three days, the female mealybugs settled on sprouting potato and started egg laying. The crawlers emerged out and started feeding on the sprouting potato tubers. For that individual sprouting potato of same size and did not exposed to any previous pesticide applications and free from any infestations, were washed with tap water, shade dried and used as food. Each sprouting was infested with an adult female mealybug individual and was individually transferred to sprouting in carton cylindrical boxes (8 cm long and 12 cm diameter).

2. Data collection:

newly When the emerged crawlers settled for feeding on sprouting potato, the crawlers were marked by drawing a circle around them and were observed daily in the morning till they attained adult stage for further aspects of biology. The eggs laid by females of P. solenopsis were examined under binocular microscope for color, shape and size. The time of egg laying was noted, laid eggs were counted freshly and transferred to fresh sprouting potato. Time taken for egg hatching was recorded to obtain the incubation period. The freshly emerged nymphs were marked individual characteristics shape, size and color on the sprouting potato and observed daily under

microscope to note moulting process. The moulting was confirmed by the presence of exuivum on the sprouting potato or on the posterior end of nymphs. Data on morphological characters and duration for relative development at various sets of temperature and humidity were recorded. The pupal stage duration, shape, color and size was observed and recorded twice a day. Adult stage characteristics, size, shape, color, fecundity, duration. longevity, preoviposition period, oviposition period and post- oviposition period were observed and recorded twice day.

The study was conducted between January 2015 to December 2015 in the laboratory when mean temperature and mean relative humidity of the study seasons ranged from 15.43, 22.50, 23.34 and 29.60°C and 52.00, 47.10, 51.24 and 57.60 % RH, respectively. Thermo-hygrograph was used to maintain temperature and humidity of experimental units. Data were statistically analyzed for analysis of variance and mean by Duncan test (P=0.05) using Costat (2005) software.

1.The eggs:

The cotton mealybug eggs were very small, oval in shape, yellowish green color with 0.4x0.1mm size. The eggs were present in the pouch made up of silken thread in the last abdominal segments of female insect. The egg duration differed at different seasons. It was 7.43 ± 0.51 hours in winter at $15.43^{\circ}C$ with 52%RH; $5.43\pm$ hours in spring at $23.34^{\circ}C$ and 47.07%RH; 1.90 ± 0.18 hours in summer at $29.6^{\circ}C$ with 51.24%RH and 4.93 ± 0.33 hours in autumn at $22.5^{\circ}C$ and 57.5%RH (Table,1). The results are similar to those of Amjad et al. (2012) who reported that eggs of the cotton mealybug were small, having yellowish green color with 0.3x0.1mm size and were present in the ovisac. The data showed the decreasing incubation period from 7.43 hours to 1.90 hours from 29.6°C, 15.43to respectively. The findings confirm with this of Chong et al. (2008) who found prolonged duration at lower temperature and shorted at higher temperature.

Table (1): Effect of various seasons temperature and relative humidity on different stages
of <i>Phenacoccus solenopsis</i> female under laboratory conditions.

Results and Discussion

	Duration(days) in various seasons							
Stags	Winter		Spring		Summer		Autumn	
	Temp. °C	%RH	Temp. °C	%RH	Temp. °C	%RH	Temp. °C	%RH
	15.43	52	23.34	47.07	29.6	51.24	22.5	57.6
Egg (LSD=0.991)	7.43±0.51 ^a		5.43±0.17 ^b		1.90±0. 18 ^c		4.93±0.33 ^b	
1 st instar (LSD=0.921)	17.89±0.21ª		7.74 ± 0.18^{b}		4.26±0.27 ^c		8.17±0.47 ^b	
2 nd instar (LSD=0.793)	16.02±0.12 ^a		7.65±0.31 ^b		4.50±0. ^{30c}		8.16±0.20 ^b	
3 rd instar (LSD=0.737)	17.01±0.18 ^a		8.24 ± 0.28^{b}		5.04±0.25 ^c		9.04 ± 0.25^{d}	
Nymphal stage (LSD=1.203)	51.00±0.16 ^a		23.62±0.38 ^b		13.78±0.63°		25.38 ± 0.41^{d}	
Pre-ovipositing (LSD=0.848)	20.16±0.35 ^a		8.47±0.25 ^b		7.00±0.25 ^c		8.00±0.251 ^b	
Ovipositing (LSD=0.903	25.00±0.31 ^a		13.16±0.30 ^b		12.26±0.39b ^c		13.03±0.37°	
Post-ovipositing (LSD=0.898)	6.40±0.18 ^a		4.26±0.37 ^a		5.42±0.34 ^b		6.48±0.15 ^c	
Adult longevity (LSD=1.2)	51.52±0.61 ^a		25.65±1.00 °		24.66±1.00 °		27.20±0.77 ^ь	
Fecundity (LSD=98.196)	107.20±20.14 ^a		318.80±49.2 ^b		495.46±32.1°		306.10±22.8 ^b	
Total life cycle (days) (LSD=2.478)	102.52±2.49 ^a		49.51±1.71 ^b		38.43±1.79°		52.58±1.69 ^d	

Mean in row sharing similar letter are not significantly different by Duncan test at P = 0.5

2. The first nymphal instar:

The 1st nymphal instar of the cotton mealybug was small and yellowish green in color, their size varied from 6

mm to 2 mm. There was no wax coating on the 1^{st} instar. The 1^{st} instar was very fast crawlers and they quickly searched the food and settled on the host. The duration of 1st instar of the cotton mealybug differed at different seasons, temperature and relative humidity. However, it declined with increase in temperature and decrease in relative humidity. The duration of 1st instar was 17.89±0.21 days in winter at 15.43°C with 52 %RH; 7.74±0.18 days in spring at 23.34°C with 47.07%RH; 4.25±0.26 days in summer at 29.6°C with 51.24 %RH and 8.17±0.47days in autumn at 22.5 °C with 57.6 %RH, respectively (Table,1). The present study is in conformity with those of Aheer et al. (2009) who reported that the 1st instar of the cotton mealybug was yellowish green in color and fast crawler. The results indicated that survival of 1st decreased at higher instar was temperature and are agree with those of Ahree et al. (2009) and Amjad et al. (2012) they reported that duration of 1^{st} instar 7-9 days at 25°C. Also, the present results can be compared with those of Child (2007) who reported that increase in damage to agriculture crop was observed due to increased environmental temperature.

3. The second nymphal instar:

The 2^{nd} nymphal instar of *P*. solenopsis was light green in color and its size varied from 0.8-.41 mm. It has two black dots on the thorax and abdomen. They found on the twigs of host plants and waxy layer developed on the body surface through a few hours after moulting. The duration of 2^{nd} instar of the cotton mealybug varied at different seasons. The duration of 2^{nd} instar female 15.02 ± 0.12 days in winter at 15.43° C with 52%RH; 7.65 ± 0.31 days in spring at 23.34° C with 47.07%RH; 4.50 ± 0.29 days in summer at 29.6° C with 51.24%RH and 8.16 ± 0.201 days in autumn at 22.5° C with 57.6%RH, respectively (Table, 1).

The duration of the 2nd instar male was 17.11±0.183 days in winter at 15.43°C with 52%RH; 9.1±0.3 days in spring at 23.34°C with 47.07%RH; 6.033±0.34 days in summer at 29.6°C with 51.24% RH and 10±0.35days in autumn at 22.5 °C with 57.6 %RH, respectively (Table,2). These results occurred that life duration of the 2nd instar both female and male decreased as the temperature increased. Thus, increase in temperature lead to rapid growth of the cotton mealybug of 2nd instar of both sexes. These findings are in conformity with those of Aheer et al. (2009) who reported light green color of 2nd instar and variation in duration of both sexes i.e. 4 to 9 days in male and 3 to 4 days in female. Similar results were also obtained by Amjad et al. (2012) who revealed that the duration of 2nd instar (female) was 4.6 days at 20 °C with 75%RH which gradually reduced to 3days at 40 °C with 40%RH.

		-10 0							
		Duration(days) in various seasons							
Stags	Wir	Winter		Spring		Summer		Autumn	
	Temp °C	%RH	Temp. °C	%RH	Temp. °C	%RH	Temp. °C	%RH	
	15.43	52.00	23.34	47.07	29.60	51.24	22.50	57.60	
Egg (LSD=0.991)	7.43±	7.43±0.51ª		5.43±0.17 ^b		1.90±0.18 ^c		4.93±0.33 ^b	
1 st instar (LSD=0.921)	17.89±	17.89±0.209ª		8.34±0.42 ^b		4.26 ± 0.268^{d}		8.17±0.47°	
2 nd instar (LSD=0.859)	17.11:	17.11±0.18 ^a		9.05±0.29 ^b		6.033±0.338°		10.00±0.354 ^b	
Pupae (LSD=0.932)	19.00:	19.00±0.32ª		8.00±0.36 ^b		6.00±0.351c		8.13±0.13 ^b	
Adult longevity (LSD=0.356)	2.20±	2.20±0.11 ^a		2.14±0.26 ^a		1.60±0.16 ^a		2.74 ± 0.16^{b}	
Total life cycle (LSD=1.472)	56.20	56.20±3.88ª		27.50±1.33°		17.51 ± 1.12^{d}		28.72±1.11 ^b	

 Table (2): Effect of various seasons temperature and relative humidity on different stages of *Phenacoccus* solenopsis male under laboratory conditions.

Mean in row sharing similar letter are not significantly different by Duncan test at P=0.5

4. The third nymphal instar: The 3rd nymphal instar of the cotton mealybug was light green in color with present two dots on the thorax and

abdomen. White waxy layer appeared on 2nd day. The third instar appeared only in the female specimens while in male specimens pupae were formed. The duration of 3rd instar was 17.01±0.18 days in winter at 15.43°C with 52%RH; 8.24±0.29 days in spring at 23.34 °C with 47.07%RH; 5±0.25 days in summer at 29.6°C with 51.24%RH and 9±0.25 days in autumn at 22.5 °C with 57.6%RH, (Table,1). respectively The results showed that life period of the 3rd instar female ranged from 5.00 to 17.01 days at season temperature range of 15.4 °C to ^oC, respectively. Thus, lower 29.6 temperature prolonged life span of the insect on opposite the higher temperature. Aheer et al. (2009) they recorded that the third instar female was light green in color and completed its life in 6.5 to 8 days at 25 °C and 70% RH. Amjad et al. (2012) reported that low temperature resulted in longer development period.

5. The pupae:

Pupae were white in color enclosed in silken sac made up of white threads. Pupae was light green; its size was 1.70 and 0.40mm. The duration of pupae was 19.00±0.3 days in winter at 15.43°C with 52% RH; 8.00±0.36 days in spring at 23.34 °C with 47.07% RH; 6.00±0.35 days in summer at 29.6°C with 51.24% RH and 8.13±0.13 days in autumn at 22.5°C with 57.6% RH, respectively (Table, 2). The present findings are similar to Aheer et al. (2009) who reported that male pupae were formed after 2nd instar and pupae duration was 7 to 8 days at 25°C. Amjed et al. (2012) pupae who reported the duration decreased from 9.57 days at 20°C to 3.78 days at 35°C.

6. The adult female:

The adult female was wingless and oblong in shape with light green in color and converted into dark brown when died, there were two pairs of dark black spots on dorsal surface of female body, it size varied from 1.60 mm to 2.25 mm. As shown in Table (1), the adult female longevity was divided into pre-oviposition, oviposition and post–oviposition periods . In general, the adult longevity for the cotton mealybug female differed at reached different temperature and relative humidity. The findings are in conformity with those of Shaymaa *et al.* (2017) who reported the female was wingless, oblong in shape and having two pairs of black spots on dorsal side of body region.

7. The pre-ovipostion period:

Results showed that the female passed through the pre-ovipositing period were varied from one season to another. Duration of pre-ovipositing was 14.36 ± 0.16 days in winter at 15.43° C with $52 \ \%$ RH; 8.47 ± 0.253 days in spring at $23.34 \ ^{\circ}$ C with 47.07% RH; 6.99 ± 0.25 days in summer at 29.6° C with $51.24 \ \%$ RH and 7.68 ± 0.25 days in autumn at $22.5 \ ^{\circ}$ C with $57.6 \ \%$ RH, respectively (Table, 1).

8. The oviposition period:

This period was found to be the longest of adult female's life throughout all seasons. The duration of this period was varied from one season to another. Duration of oviposition period was 25.00 ± 0.31 days in winter at 15.43° C with 52% RH; 13.16 ± 0.30 days in spring at 23.34 °C with 47.07% RH; 12.26 ± 0.39 days in summer at 29.6°C with 51.24% RH and 13.03 ± 0.37 days in autumn at 22.5 °C with 57.6 %RH, respectively (Table, 1).

9. The post-oviposition period:

This period was elapsed after female had stopped oviposition till the death. The post-oviposition period was found to be the shortest one. Mean durations of this period were 6.40 ± 0.18 days in winter at 15.43° C with 52% R.H.; 4.26 ± 0.73 days in spring at 23.34° C with 47.07 R.H.%; 5.45 ± 0.34 days in summer at 29.6°C with 51.24 R.H.% and 6.48 ± 0.15 days in autumn at 22.5 °C with 57.6 R.H.%, respectively (Table, 1).

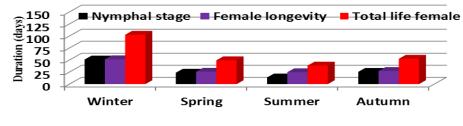
Abd El- Razzik, 2018

10. The adult female longevity:

The duration of the female longevity differed at reached different temperature and relative humidity. Mean durations of this period were 51.52±0.61 days in winter at 15.43°C with 52% RH; 25.65±0.1 days in spring at 23.34 °C with 47.07 % RH; 24.66±1 days in summer at 29.6°C with 51.24 % RH and 27.20±0.77 days in autumn at 22.5 °C with 57.6 % RH, respectively (Table,1 and Figure,1). The adult female longevity period decreased significantly with increase of temperature and vice versa. Nikam et al. (2010) who found that longevity of female was 33.67±1.19 days

at temperature and relative humidity range from 25 to 30°C and 75 to 80% RH, respectively. Similarly, studies on the biology of mealybug P. solenopsis under laboratory condition on potato sprout at Hayana Agriculture University, Hisar found that the total life span was 39.12±2.85 and 18.60±1.5 days (Kedar et al., 2011). Nikam et al. (2010) reported that longevity of female was 33.67±1.19 days at temperature and relative humidity ranged from 20 to 30°C and 75 to 80%RH, respectively. Shaymaa et al. (2017) and Shehata (2017) who reported that the adult female longevity of P. solenopsis was shorter 30°C.than 20°C.

Figure (1): Effect of various seasons temperature and relative humidity on different stages of *Phenacoccus solenopsis* female under laboratory conditions.



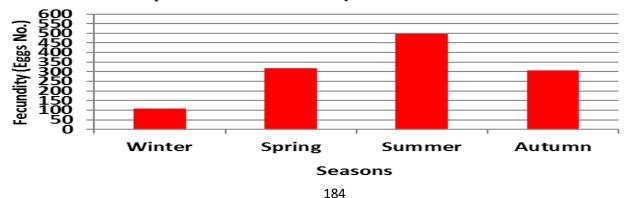


11.Fecundity of adult female:

The results showed that the total number of deposited eggs/females differed at different seasons, temperature and relative humidity during its life cycle. Data given in Table (1) and illustrated in Figure (2) showed the total number of deposited eggs/female during its oviposition period was 107.20 ± 20.14 days in winter at 15.43°C with 52% RH; 318.80 ± 49.2 days in spring at 23.34 °C

with 47.07 %RH; 495.46±32.1 days in summer at 29.6°C with 51.24% RH and 306.10±22.8 days in autumn at 22.5 °C with 57.6 %RH, respectively. The results clearly indicated that the highest number of egg in summer at 29.6°C. It could be concluded that the temperature may be considered as the thermal optimum for egg laying activity.

Figure (2): Effect of various seasons temperature and relative humidity on fecundity of *Phenacoccus solenopsis* female under laboratory conditions.

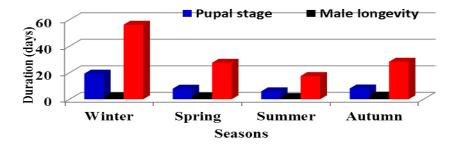


12.The longevity of adult male:

The adult male of the cotton mealybug was blackish brown in color and had four waxy filaments at the end male body and winged, wings were transparent. Size of adult male was 1.3 mmx0.4 mm. The duration of the adult male was varied from one season to another. Duration of the adult male was 2.20 ± 0.11 days in winter at 15.43° C with 52% RH; 2.14 ± 0.26 days in spring at 23.34°C with 47.07% RH; 1.60 ± 0.16 days

in summer at 29.6°C with 51.24% RH and 2.74±0.16 days in autumn at 22.5 °C (Table, 2 and Figure, 3). Prishanthini and Vinobaba (2013) found that longevity of P. solenopsis male ranged from 1 to 2 days with an average of 1.50 ± 0.5 days. Also, Aheer et al. (2009) reported that adult male is blackish brown in color, with transparent wings and four abdominal segments. They further reported that adult survived 1-2 days at 25°C with 75% RH.

Figure (3): Effect of various seasons temperature and relative humidity on different stages of *Phenacoccus solenopsis* male under laboratory conditions.



13.Total nymphal stage duration:

The results in (Table, 1 and Figure,1) indicate that duration of total nymphal stage tented to be shortened with the corresponding raise of temperature. Under any of the temperature of season, the total nymphal stage of P. solenopsiss as a whole varied greatly. The duration of total nymphal stage was 51.00±0.16 days in winter at 15.43°C with 52%RH; 23.62±0.62 days in spring at 23.34 °C with 47.07% RH; 13.00±0.63 days in summer at 29.6°C with 51.24%RH and 25.38±0.41 days in autumn at 22.5 °C. The obtained results are in agreement with (Aheer et al., 2009 and Amjad, 2012) who reported that decreased in temperature increased the duration of nymphal stage and increase in temperature decreased the duration of nymphal stage.

14.Total life cycle of female:

The results in (Table, 1 and Figure, 1) showed that duration of total life cycle of female varied significantly

with increases in temperature and relative humidity. The duration of total female life was 102.52±2.49 days in winter at 15.43°C with 52% RH; 49.51±1.71 days in spring at 23.34°C with 47.07% RH; 38.43±1.79 days in summer at 29.6°C with 51.24%RH and 52.58±1.69 days in autumn at 22.5°C. These results are similar to those of (Aheer *et al.*, 2009 and Amjad, 2012) who reported that duration of female life cycle reduced on increasing temperature and vice versa.

15. Total life cycle of male:

The total life cycle of male of the cotton mealybug duration varied significantly with increase in temperature and relative humidity. The duration of the life cycle male was varied from one season to another. Duration of the total life cycle male was 56.20±2.88 days in winter at 15.43°C with 52%RH; 27.50±1.33 days in spring at 23.34 °C with 47.07%RH; 17.51±1.12 days in summer at 29.6°C with 51.24%RH and 28.72±1.11 days in autumn at 22.5°C, respectively in (Table, 2 and Figure, 2). Also, Aheer *et al.* (2009) reported that decrease in temperature increased the duration while increase in temperature resulted in decreased duration of insect.

It is concluded that the study *P*. solenopsis biology. give the understanding of mode and degree of its growth. Hence, population this information will be helpful during the development of successful integrated pest management program (IPM) for *P*. solenopsis which is considered as major polyphagous pest in the world.

Conflict of Interest

The present study was performed in absence of any conflict of interest.

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