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Impact of Ecological Conditions on Biology of Cotton Mealy Bug, *Phenacoccus solenopsis* (Hemiptera: Pseudococcidae) in Laboratory

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Abstract.- Effect of temperature and relative humidity on the life history of the cotton mealy bug *Phenacoccus solenopsis* Tinsley (Hemiptera: Pseudococcidae) was investigated in the laboratory. *P. solenopsis* was able to complete its life cycle at 20, 25, 30 and 35±1°C and 70, 65, 60 and 40±5% RH, respectively. Egg hatching period decreased from 32±0.22 to 0.57±0.15 hours from 20 to 40°C. But at 40°C, the male specimens were unable to pupate and the female specimens despite comparatively very short life span were unable to produce eggs. The highest fecundity was observed at 20°C with each female producing an average of 232.65±2.19 eggs. Development time from egg to adult was the longest for male at 20°C. Increasing temperature and decreasing relative humidity had profound effect on the longevity of the females whereas longevity of males was less affected. Ability of the *P. solenopsis* to develop and reproduce successfully at 20 to 35±1°C and 70 to 40±5% RH suggests that the pest can develop and build up its populations in different ecological zones within this temperature range.

Key words: Development, relative humidity, *Phenacoccus solenopsis*, cotton mealy bug.

INTRODUCTION

Cotton, *Gossipium hirsutum* (Genn), is an important cash, fiber and fodder crop in Sub-continent. Its production is facing a steady decline from 2.194 million tons in 2005 to 2.09 million tons in 2009 in Pakistan due to incidence of sucking pest complex, high cost of fertilizer and pesticides (Dutt, 2007). In 2005, a new menace, cotton mealybug, *Phennacoccus solonepsis* (Tinsley) appeared in Sindh (Sanghar, Mir pur Khas and Tando Allahyar district) and Punjab (Multan, Vehari, and other cotton growing areas of southern Punjab) provinces of Pakistan (Khushk and Bhagromal, 2005). In 2006, it appeared in Gujrat in India (Saini *et al*., 2009; Hanchinal *et al*., 2009; Nagarare, 2009). The pest feeds on more than 154 plants belonging to six different families (Arif *et al*., 2009). Damage to host crop is due to sucking of twigs and leaves. Sooty mould appears on the twisted twigs and leaves due to which young plants die and elder plants are weakened. Its damage in Punjab started from the hot regions like Vehari and Multan which might be due to the fact that hot weather and lack of rainfall encourages its development. In India Dhawan *et al*. (2009) reported positive correlation between increase in temperature whilst negative correlation between rainfall and population of pest. The present study is planned to observe the effect of temperature and relative humidity in the laboratory on biology of cotton mealy bug so that the farmer can take proper management actions against this pest keeping in view the ecological conditions.

MATERIALS AND METHODS

Experimental conditions

The cotton mealy bug specimens were collected from different host plants from Ayub Agricultural Research Institute, Faisalabad, Pakistan during 2007-2008. Plant twigs (15 cm length) containing cotton mealy bugs, were cut with plant cutter and placed in cool box to brought into the laboratory. The adult female cotton mealy bugs were separated for further rearing. The experiments were conducted in the controlled growth chamber maintained at five constant temperatures (20°C, 25°C, 30°C, 35°C and 40°C) with decrease in relative humidity (70, 65, 60, 40) and a light and dark period of 14:10 (L:D) h.

Thirty newly hatched nymphs were reared in rearing cells separately at each temperature treatment. But the sample size became different with

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death of some insects at different stages and with sex differentiation.

Laboratory studies

China rose (*Hibiscus rosasinensis*) was used as a lab host plant in this study. Fresh cuttings were obtained from the unsprayed lawns of Ayub Agricultural Research Institute, Faisalabad. Rearing of 1st instar crawlers was performed in rearing cells. The rearing cells comprised of a plastic vial of 1.5 x 0.5cm size, containing sieve of 90-mesh size, a China rose twig soaked in moist sand and covered with a plastic cover. First instar crawlers after appearing from the ovisacs of females were transferred to these rearing cells on the twig each per cell (Aheer *et al* ., 2009). The female specimens were kept in these cells for the first three instars while the males were kept up to pupal stage. Rearing of adult insects was done in glass petri dishes (9 cm diameter) containing fresh China rose twigs. The twigs were changed with new ones after every 48 hours.

Mealy bug development

Mated adult *P. solenopsis* females were kept singly in the glass petri-dishes 9cm diameter and randomly kept in each treatment. The duration from freshly laid eggs to emergence of nymphs was considered as incubation period. The nymphs on hatching were transferred in rearing cells. Duration of different nymphal instars was recorded by examining the exuvia and mean duration for development was calculated. Survival rate of each instar was calculated by counting the number of individuals that moulted successfully to the next instar. The overall survival rate from egg to adult in each treatment was determined by dividing the number of adults (male or female) emerged by the total number of eggs.

Statistical analyses

A general linear model procedure (SPSS) was performed to determine the effect of temperature on development and reproduction of *P. solenopsis*. The significant differences between means were compared by using Fisher protected significant difference test with the significance level at p = 0.05.

RESULTS

The duration of egg incubation decreased from 32 to 0.57 hours with the increase in temperature from 20°C up to 40°C. First instar cotton mealy bug was small and yellowish green in colour and their size varied from 0.5 x 0.1 mm. There was no wax coating on their bodies and were very fast crawlers. They quickly searched their food and settled on the host. From 20 to 30°C all the 1st instar nymphs were able to molt into 2nd instar. But at 35 and 40°C, 13% of total bugs died in both treatments. Higher temperature treatments were significantly shortened the developmental period in all treatments in 1st instar. The duration of 1st instar male specimens reduced three times from 20°C up to 35°C but at 40°C, slight increase in the duration was observed as shown in Table I.

The colour of 2nd instar cotton mealy bug was green. Their size varies from 0.75 x 0.33 mm. Two black streaks were present on the thorax and on the abdomen. They usually sticked to the twigs of host plants and waxy layer developed on the body surface 1h after molting. Up to 35°C, the developmental duration of 2nd instar females gradually became shorter, but at 40°C it again prolonged. There was decreasing trend in all treatments during 2nd instar of male and reduced to more than thrice at 40°C as compared to that at 20°C as appeared in Table I.

Third instar cotton mealy bugs were light green in colour. Two black streaks were present on the thorax and the abdomen. White waxy layer appeared on 2nd day. The duration of development of 3rd instar was observed to be shortened till 30°C and then it again started to increase on both of 35 and 40°C. Pupae were white in colour enclosed in silken sacs made up of white threads. Internal colour of pupae was seen light green when dissected; their size was 1.80 x 0.50 mm. Pupae were mostly found on the lid of petri dishes; duration of 3rd instar (pupae) also became shorter with the increase in temperature up to 35°C; all pupae died at 40°C as seen in Table I.

Total nymphal duration of female reduced to ½ times (from 24.12 to 12.39 days) at 20°C to 40°C, respectively in females and more than ½ times from 25.69 to 12.33 days at 20°C to 35°C, respectively in

Table I.- **Duration of development (Means± SEM) of different stages of cotton mealy bug at different sets of temperature (°C) and relative humidity (%).**

Treatments*	Eggs (Minutes)	1st instar (days)		2nd instar (days)		3rd instar (days)	
		Female	Male	Female	Male	Female	Male
T1	32.00±0.22a	8.76±0.106a	8.77±0.12a	8.35±0.12a	8.38±0.14a	7.00±0.00a	8.54±0.13a
T2	30.07±0.22b	7.78±0.879b	8.00±0.31b	7.17±0.08b	6.86±0.14b	6.00±0.00b	7.71±0.18b
T3	24.00±0.22c	4.13±0.187c	4.71±0.18c	3.65±0.10d	3.86±0.14c	4.61±0.10c	7.71±0.36b
T4	19.07± 0.19d	3.32±0.11d	2.86±0.14d	3.42±0.19d	2.86±0.34d	6.00±0.10b	5.71±0.18c
T5	0.57 ±0.151e	3.27±0.07d	3.09±0.61d	6.70±0.12c	2.45±0.16d	5.93±0.21b	-

T1, 20±1°C, 70±5% RH; T2, 25±1°C, 65±5% RH; T3, 30±1°C, 60±5% RH, T4, 35±1°C, 40±5% RH; T5, 40±1°C, 40±5% RH.

Table II.- **Total duration (days) of development (means± SEM) of different stages and fecundity of cotton mealy bug at different of temperature (°C) and relative humidity (%).**

Treatments	Total nymphal duration		Longevity		Life Span		Fecundity
	Female	Male	Female	Male	Female	Male	
T1	24.12±0.08a	25.69±0.26a	64.88±0.93a	1.92±0.05a	89.00±0.92 a	27.63±0.23a	232.65±2.19a
T2	20.96±0.13b	22.57±0.37b	57.57±0.65b	1.86±0.09a	78.52±0.72b	24.43±0.38b	195.35±1.88b
T3	15.88±0.17c	16.43±0.43c	23.43±0.76c	1.57±0.20ab	35.83±0.80c	17.86±0.63c	136.16±15.9d
T4	12.71±0.20d	11.33±0.33d	11.32±0.37d	1.43±0.20b	24.05±0.47d	12.86±0.59d	113.09±1.72c
T5	12.39±0.25d	-	4.33± 0.21e	-	20.26±0.38e	-	-

For details of treatments, see Table I.

males. Adult female size varied from 2.0 x 1.75mm and its colour is light green, which is converted into dark brown when died. In the adult life, the effect of the temperature was the most remarkable. The females completed adult life in 64.88 days at 20°C, but it completed it only in 4.33 days at 40°C. Total female life was 89 days at 20°C, which was more than twice as long as that at 30°C and more than 4 times to that at 40°C. The females were unable to lay eggs at 40°C; adult male of cotton mealy bug was winged with blackish brown colour had four abdominal filaments. The adult longevity was 1.92 days at 20°C, which was reduced to 1.43 days at 35°C, indicating no noticeable decrease as shown in Table II.

At temperature 20°C, female life span was three months which decreased significantly to 35.83 days at 30°C and decreased more than four times and was completed in 20.26 days at 40°C. Male life also exhibited decreasing trend with increase in temperature, more than two times at 35°C. Total adult life could not be calculated at 40, because all the pupae died at 40°C. Fecundity of the females reduced to ½ from 20 to 30°C, but surprisingly more eggs were recorded at 35°C than 30°C. Female cotton mealy bug did not produce eggs at 40°C due to desiccation (Table II).

DISCUSSION

Temperature is one of the most important ecological factors affecting the physiology and behavior of insects. Being ectothermic organisms, the insects are more subjected to change in rate of growth, development, life time, survival, fecundity and different biological aspects with changing temperature. Similarly relative humidity in the air in combination with temperature also influences the survival and reproduction of insects, which may due to the fact that at high temperature vapour pressure deficit (VPD) also increases. Vapour pressure deficit is the difference in the moisture of air and that of saturated air at a particular temperature (Pregner and Ling, 2001). The insects or plants loose more moisture in the air at higher VPD and their survival rates are reduced. Chong *et al.* (2003) failed to establish colonies of *P. medinrensis* at 30-40°C due to inability of eggs to hatch and mortality of crawlers before molting. They attributed this failure to the increased physiological stress exerted by high temperature and increased VPD (due to low relative humidity). In the current studies, mortality of 3rd instar males, and loss of fecundity of females at 40°C and 40% R.H., can be explained on the basis of same phenomenon. These results are also supported by the findings of Shipp and Gillespie (1993), who reported that percentage survival of flower thrips was significantly lower in the treatments of high VPDs and temperature.

Cotton mealy bug can complete 15 generations per year in field as reported by Tanwar *et al.* (2007). The population consists of overlapping generations and individuals of various developmental stages can be observed on the same time. Over- wintering of *P. solenopsis* is not well understood. Abbas *et al.* (2010) made an assumption that under unfavourable conditions the adult females near death produce ovisacs and eggs, and the crawlers live under the moribund body of female in the state of arrested growth and as soon as the conditions become favorable the crawlers start actively growing. On the other hand Aheer *et al*. (2009) observed populations of cotton mealy bug throughout the year (December, 2006-November, 2007) which indicates lack of over wintering habit in the pest, however further studies are needed to confirm to this phenomenon. In the same paper Aheer *et al*. (2009) reported fluctuation of cotton mealy bug populations with the change in the seasons, so it is of worth importance to observe the effect of different levels of temperature and relative humidity on the biology of this pest. The incubation period of *P. solenopsis* was very short as mentioned by Yong *et al.* (2011). The incubation period decreased from 32 to 0.57 minutes at 20 to 40°C, respectively. These findings are comparable with those of Aheer *et al*. (2009) and Nikam *et al.* (2010). In contrast, Akinotola and Ande (2008) was unable to observe eggs; and Hodgson *et al*. (2008) also reported the birth of young ones through the insect.

From 20 to 30°C the trends in the developmental aspects were similar, *i.e.*, the developmental periods became shorter for every instar with increase in temperature. Temperature increasing may increase the metabolic processes of the body which resulted in increase in the growth and development. But this trend changed at 40°C for 2nd instar female whose duration became longer than was observed in the first four treatments. The duration of 3rd instar female also became longer at 35 and 40°C than that observed at 20 to 30°C and all the pupae died at 40°C. This may be due to the fact that the rate of development in insects increases with increase in temperature up to optimum temperature and above that temperature the development rate again declines. So for, the 2nd instar female and pupal stage, it seems that its optimum temperature was reached before 40°C (Taylor, 1981). This hypothesis is further strengthened by the fact that the females were unable to produce eggs at 40°C. The pupal stage of males took comparatively more period to become adults as compared to 3rd instar female specimens. Due to this reason the male specimens took more time to become adult than females (Aheer *et al*., 2009).

The adult longevity of males was very short (from 1.92 to 1.43 days at 20 to 35°C, respectively) as compared to females (Kumar *et al*., 2009). Female duration was more effected by the increase in temperature (from 64.88 to 4.33 days) than the males. The fecundity of female *P. solenopsis* was recorded 232/female at 20°C, which reduced to 136 eggs at 40°C. Whereas Venneila *et al.* (2010) reported 344 eggs and Yong *et al.* (2011) described 458 eggs per female. This may be due to difference in food source because we used *Hibiscus rosasinesis* rather than cotton. In the present studies the insects were paired for mating before oviposition period because Aheer *et al*. (2009) and Hodgson *et al.* (2008) reported bisexual type of reproduction in cotton mealy bug. But Venneila *et al.* (2010) observed parthenogenesis in the cotton mealy bug.

For the successful integrated management of the *P. solenopsis*, thorough understanding of its life history is very crucial. Information from present study may be helpful in managing the susceptible stages of cotton mealy bug at different temperatures. It is evident from the present study that the development of *P. solenopsis* is dependent on temperature and relative humidity. Extended development time of immature stages and maximum fecundity from 20 to 25°C suggests the suitability of these conditions for the population build up. So in these conditions, careful inspection of the crop should be started and early stages of the cotton mealy bug should be targeted because the later stages are difficult to control due to rapid reproduction and well developed layer of waxy secretions on their body. The information regarding the ability of cotton mealy bug to develop and reproduce at 20 to 35°C and 75±5 to 60±5 % R.H. may prove useful in predicting distribution and abundance of this pest in different ecological regions of Pakistan. Similarly lack of egg laying by females and death of all pupae at 40°C and 40±5 % R.H. suggests that it is above the optimum level of temperature for this insect and there will be no more generation.

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REFERENCES

ABBAS, G., ARIF, M.J., ASHFAQ, M., ASLAM, M. AND SAEED, S., 2010. Host plants distribution and overwintering of cotton mealy bug (*Phenacoccus solenopsis*; Hemiptera: Pseudococcidae). *Int. J. Agric. Biol.,* **12**L 421–425.

AHEER, G.M., SHAH, Z. AND SAEED, M., 2009. Seasonal history and biology of cotton mealy bug, *Phenacoccus solenopsis* Tinsely. *J. agric. Res.,* **47**: 423-431.

AKINOTOLA, A.J. AND ANDE, A.T., 2008. Frist record of *Phenacoccus solenopsis* Tinsley on *Hibiscus rosasinensis* in Nigeria. *Agric. J.,* **3**: 1-3.

ARIF, M.I., RAFIQ, M. AND GHAFFAR, A., 2009. Host plants of cotton mealybug (*Phennacoccus solenopsis*): A new menace to cotton agro-ecosystem of Punjab. *Int. J. Agric. Biol.,* **11**: 163-167.

CHONG, J.H., RONALD, D.O. AND IERSEL, M.W.V., 2003. Temperature effect on the development, survival and reproduction of the Madeira Mealy Bug, *Phenaccocus madeirensis* Green (Hemiptera: Pseudococcidae), on chrysanthemum. *Ann. entomol. Soc. Am.,* **96**: 539-543.

DHAWAN, A.K., KAMALDEEP, S.A. AND SARIKA, S., 2009. Distribution of mealybug, *Phenacoccus solenopsis* Tinsley in cotton with relation to weather factors in south-Western districts of Punjab. *J. entomol. Res.,* **33**: 59-63.

DUTT, U., 2007. *Mealy bug infestation in Punjab: Bt. cotton falls flat*. Online article at http:/www.countercurrents. org/dutt210807. htm. 7th July, 2007.

HANCHINAL, S.G., PATIL, B.V., BHEEMANNA, M., HOSAMANI, A.C. AND SHARANABASAPPA, 2009. Incidence of mealy bug on cotton in Tungbhadra project area In: *Proc. Dr. Leslie C. Coleman Memorial Nation. Symp. Plant Protection* Dec.,4 - 6, 2008, University of Agricultural Sciences. GKVK, Bangalore.

HODGSON, C., ABBAS, G., ARIF, M.J., SAEED, S. AND KARAR, H., 2008. *Phenacoccus solenopsis* Tinsley (Sternorrhyncha: Caccoidea: Pseudococcidae), an invasive mealy bug damaging cotton in Pakistan and India, with a discussion on seasonal morphological variation. *Zootaxa,* 1913: 1-135. Online article at www.mapress.com/zootaxa/list/2008/zt01913

KHUSK, A. M. AND BHAGROMAL, 2005. A new bug threatens cotton crop. http://www.dawn.com/2006/06/19/ebr7.htm.

KUMAR, R., JAT, S.L. AND CHAUHAN, R., 2009. Biology of the mealy bug, (*Phenacoccus solenopsis*; Hemiptera: Pseudococcidae) on cotton in India. *Entomon*, **34**: 189-192.

NAGARARE, V.S., KRANTHI, S., BIRADAR, V.K., ZADE, N.N., SANGODE, V., KAKDE, G., SHUKLA, R.M., SHIVARE, D., KHADIA1, B.M. AND KRANTHI, K.R., 2009. Wide spread infestation of the exotic mealybug species *Phenacoccus solenopsis* (Tinsley) (Hemiptera: Pseudococcoidea) on cotton in India. *Bull. entomol. Res.,* **99:** 537-541.

NIKAM, N.D., PATER, B.H. AND KORAT, D.M., 2010. Biology of invasive mealy bug, *Phenacoccus solenopsis* Tinsley (Hemiptera: Pseudoccidae) on cotton. *Karnataka J. agric. Sci.,* **23**: 649-651.

PREGNER, J.J. AND LING, P.P., 2001. *Green house condensation control: understanding and using vapor pressure deficit (VPD)*. Fact sheet (Series) AEX-804-01. Ohio State University Extension. Columbus. OH.

SAINI, R.K., PALARAM SHARMA, S.S. AND ROHILLA, H.R., 2009. Mealybug, *Phenacoccus solenopsis* Tinsley and its survival in cotton ecosystem in Haryana. In: *Proc. Nation. Symp. On Bt-cotton:Opportunities and Prospectus*, Central Institute of Cotton Research, Nagpur, November 17-19, pp. 150.

SHIPP, J.L. AND GILLESPIE, T.J., 1993. Influence of temperature and water vapor pressure deficit on survival of Frankliniella occidentalis (Thysanoptera: Thripidae). *Environ. Ent.,* **22**: 726-732.

TANWAR, R.K. JEYAKUMAR, P. AND MONGA, D., 2007. *Mealybugs and their management*, 12p. Technical Bulletin 19, National Centre for Integrated Pest Management, New Delhi, India. Available online: http://www.ncipm.org.in/MealybugPDFs/Bulletin-Mealybugs(%20.

VENNILA, S., DESHMUKH, A.J., PINJARKAR, D., AGARWAL, M., RAMAMURTHY, V.V., JOSHI, S., KRANTHI, K.R. AND BAMABAWALE, O.M., 2010. Biology of the mealybug, *Phenacoccus solenopsis* on cotton in the laboratory. *J. Insect Sci.,* **10**: 115. Available online: insectscience.org/10.115.

TAYLOR, F., 1981. Ecology and evolution of physiological time in insects. *Am. Nat.,* **117**:1-12.

YONG, Z.Y., FANG, H. AND BIN, L.Y., 2011. Bionomics of mealybug *Phenacoccus solenopsis* Tinsley (Hemiptera: Pseudococcidae) on cotton. *Acta entomol. Sin.,* **54**: 246-252.

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