

IMPACTS OF DIFFERENT TEMPERATURE LEVELS ON DEVELOPMENTAL STAGES OF SWELLOWTAIL CITRUS BUTTERFLY, PAPILIO DEMOLEUS. (LINN.)**(PAPILIONEDAE : LEPIDOPTERA)*****Yogendra Pal Singh¹ and Madhulika Singh²**

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ABSTRACT

The temperature had direct influence on the rate of development and survival of an insect. The activity and movements of insects caused by fluctuations of temperature often led to their survival. The laboratory studies made on the impact of different level of temperature (5°C, 10°C, 15°C, 20°C, 25°C, 30°C, 35°C, 40°C, 45°C and 50°C) on the incubation, larval and pupal period of *Papilio demoleus* (Linn.), a serious pest on citrus plants. It revealed that eggs were failed to hatched in to the larvae at extremely low (5°C and 10°C) and high temperature (50°C). The larval as well as pupal period were also affected by the exposure of different intensity of temperature. The larval and pupal period were increased with decreasing the temperature level and larvae goes in to hibernation at extremely low temperature with 96 to 120 days of larval period and moulted in to the pupae. Similar results were found for pupal development. Only 4th and 5th instar larvae were capable to tolerate the 5°C and 50°C temperature with maximum days as well as minimum days of larval period, respectively and moulted in to the pupae. Similar results were found for pupal developments. The optimal or moderate level of temperature was 25°C to 35°C for the development of experimental insects. Controlled experiments were conducted at the level of 30°C temperature and 60±5 percent relative humidity.

INTRODUCTION

Lemon butterfly, *Papilio demoleus* (Linn.) belongs to the genus *Papilio* which has been placed in the family *Papilionidae* of order *Lepidoptera*, firstly reported by Linnaeus (1758), Pruthi and Mani (1945), Butani (1975), Ram P., Pal R. K. & Singh J. (2000), as a voracious feeder of lemon leaf. After a major survey Bell T R (1957) was found the serious infestation of larvae of lemon butterfly from Kashmir to Cylon, East world to Burma, West world to Percia and also in China & Formosa region which cause the great loss to citrus plants. Ayer T V R (1919), Mishra C S (1919) and Desh pandey & Karanaldikar (1945) discovered some other host plants of this pest like Beal, Ber, *Glycosmis*. Saljoqi N & M.A Rafi (2006) also studied the biology and host preferences of lemon caterpillars.

The incubation period was 2.7 to 5.5 days, varies from march to November with 83.70% - 93.20% viability of eggs (Barar T S, 1959). Maheshra Babu (1988), Resham et al. (1986), Singh

and Gangwar (1989) and Radhke and Kandelkar (1989) studied the larval forms and identified the five instars and four moults with 13 – 16 days larval period but Atwal (1964), Barar & Rathaur (1973) and Ashoken (1997) stated that the larval period was vary from 8.5 – 28.8 days depending upon temperature and relative humidity. They also observed the prepupal, pupal period as well as viability was also affected by the climatic conditions, ranges from 8.2 days during June and 128 days in November to March, called as n the present investigation was under taken to study the influence of different intensity of temperature on the survival and development of various immature stages of lemon butterfly, *Papilio demoleus* (Linn.), which was not well established, therefore a detailed study was conducted.

MATERIAL AND METHODS

To study the effects of temperature on the immature stages of *Papilio demoleus*, various possible levels of temperature i.e. 5°C, 10°C, 15°C, 20°C, 25°C, 30°C, 35°C, 40°C, 45°C and 50°C, were maintained in the incubators. The 65±5 percent relative humidity was provided at all the levels of temperature in incubators. All the stages of lemon butterfly namely, eggs, I, II, III, IV, and Vth instar larvae, prepupae and pupae were selected from laboratory reared insects and kept in to various rearing jars having fresh citrus leaves which was utilized by caterpillars as food. At least 50 eggs, 30 larvae, 30 pre-pupae and 30 pupae were taken in replicate. The experiments were repeated three times. The controlled experiment was also running in another incubator at the optimum temperature (32°C) and 62±5 percent of humidity.

The observations were recorded after six hours of intervals. When eggs were hatched and caterpillar moulted in to next instar as well as pre pupae and changed into pupae and adult emerged from pupae. The data was collected hibernation period, named by Barar and Rathaur (1973). The workers like Tripathi et al. (1998) Rafi M A (2000) and Tiatula Jameer et al. (2012) also worked on the biology of citrus butterfly, *P. demoleus*. It was Sharifi & Zarea (1970) who worked on the effect of Abiotic factors in field conditions from June to September. Hays (1957) observed the effect of temperature on hatchability of *Papilio* eggs. The effects of constant temperature on the rate of development on the fourth instar, prepupae and pupae of lemon caterpillars were observed by Atwal (1971), Patil, et al. (1984), Gautam R D (1986), Mustafa T S (1988) and Garg, J R et al. (1992) found that the fluctuation of abiotic factors directly affect the development, percentage of hatching and the mortality rate of developmental stages of insects. It has been established in case of many species of insects that the rate of development is profoundly influenced by the temperature and the relative humidity. I carefully during the experiments conducted. The same data also collected from controlled experiments.

RESULT AND DISCUSSION

Temperature effect on the incubation period:

The data related to temperature impact on eggs development are placed in the table: 01.

Table–01_Effect of temperature on Incubation period of Papillio demoleous at 60+5% R.H.

Temperature °C	No. of Eggs treated	Av. Incubation Period (Days)	No. of eggs hatched	Hatching %	Observed Mortality %	Corrected Mortality %
05	50	-	NIL	00.00	100.00	100.00
10	50	12.19±2.88	09	18.00	82.00	81.63
15	50	9.08±2.26	20	40.00	60.00	59.18
20	50	5.77 ±1.84	29	58.00	42.00	40.12
25	50	4.08±1.33	37	74.00	26.00	24.49
30	50	3.62±1.19	48	96.00	04.00	02.04
35	50	3.20±1.01	49	98.00	02.00	00.00
40	50	2.59±0.72	41	58.00	42.00	40.00
45	50	2.27±0.26	20	40.00	60.00	59.18
50	50	-	NIL	00.00	100.00	100.00
Control	50	3.37 ±1.03	49	98.00	02.00	00.00

The tabulated data indicated that the incubation period of eggs depend on the fluctuation of temperature. The eggs which kept at 5°C level of temperature, failed to hatch, while some results were obtained at 10°C. It was the minimum temperature at which 18 percent of eggs were hatched in to the larvae with maximum incubation period of 12.19 days. It was further observed that at 15°C the eggs were hatched after a long time i.e. 09.08 days of incubation period with 40% of hatching and the mortality rate was noticed as 59.18%. At 20°C and 25 °C the recorded incubation period were 5.77 and 4.08 days with 58% & 74% hatching. Furthermore at 30°C and 35°C, the incubation period further reduced to 3.62 and 3.20 days which were minimum and moderate with highest percentage of hatching i. e 94% and 98%, respectively. The result reveals that at 40°C the incubation period was 2.59days and 82% eggs were hatched. But at the high level of i.e. 45°C, the recorded incubation period was shortest, 2.27 days with 40% hatching. No hatching was recorded at the extremely high temperature of 45°C because all the eggs were dried. These findings were in agreements with the study of Timberlin et al. (2002) and Mahesh Pathak (2003). In the controlled experiment, 98% eggs were hatched in moderate incubation period (3.37 days).

Temperature effect on different immature stages:

The influences of different level of temperature on the different larval stages were observed and data were placed in the table 02. as below.

Table- 03. Temperature effect on the different larval stages:

No of Larvae treated	I INSTAR			II INSTAR			III INSTAR			IV INSTAR			V INSTAR		
	Av. Larval Period (days)	Survival %	Corrected Mortality %	Av. Larval Period (days)	Survival %	Corrected Mortality %	Av. Larval Period (days)	Survival %	Corrected Mortality %	Av. Larval Period (days)	Survival %	Corrected Mortality %	Av. Larval Period (days)	Survival %	Corrected Mortality %
30	-	00.00	100.00	-	00.00	100.00	-	00.00	100.00	11.84±3.33	16.66	83.33	16.22±6.18	33.33	64.29
30	4.86±1.44	23.33	75.00	5.13±2.11	3.00	68.96	5.06±2.51	36.66	62.07	9.94±2.99	40.00	60.00	12.85±5.11	50.00	46.43
30	3.99±1.12	33.33	64.29	4.43±1.44	46.66	51.73	4.76±2.01	50.00	48.27	8.43±2.45	60.00	40.00	9.52±3.05	63.33	36.67
30	3.15±0.88	50.00	46.43	4.01±1.22	56.66	41.38	4.15±1.77	66.66	31.04	6.58±1.99	73.33	26.67	7.07±2.33	80.00	20.00
30	2.57±0.82	83.33	10.71	3.53±0.99	73.33	24.48	3.72±1.44	76.66	20.69	3.41±1.03	100.00	00.00	3.75±0.68	100.00	00.00
30	2.29±0.63	93.33	3.81	2.89±0.77	93.33	03.79	3.38±1.21	96.66	00.00	3.41±1.03	100.00	00.00	3.75±0.68	100.00	00.00
30	1.99±0.19	90.00	6.89	2.41±0.52	83.33	13.44	2.61±0.42	90.00	6.89	2.77±0.69	90.00	6.89	3.28±0.49	100.00	00.00
30	1.81±0.23	63.33	32.14	2.01±0.33	63.33	34.14	2.01±0.22	76.66	20.69	2.09±0.54	80.00	20.00	2.61±0.44	86.66	13.34
30	-	-	100.00	1.72±0.12	30.00	68.96	1.74±0.17	53.33	44.83	1.78±0.25	66.66	33.34	2.21±0.62	80.00	20.00
30	-	-	100.00	-	-	100.00	-	-	100.00	-	-	100.00	-	-	100.00
30	2.25±0.83	93.33	6.67	2.61±0.99	96.66	03.34	3.08±1.11	96.66	3.34	2.89±0.88	100.00	00.00	3.52±1.09	100.00	00.00

On first instar larvae:

The aforesaid table revealed that just hatched larvae kept at 5°C temperature failed to keep alive like the eggs. But at 10°C temperature 23.33% larvae were survive to moult with highest level of mortality (75%) and larval period (94.86 days). At 15°C survival rate increased as 33.33 percent with larval period of 3.99 days. The larval reduced to 3.15 days with 50% and 46.43% of mortality. While 10.71% mortality, 83.33% survival and 2.57 days larval period were reported at level of 25°C temperature. The moderate larval period, 2.29 and 1.99 days and 93.33% & 90.00% survival rate was recorded at 30°C and 35°C temperature, respectively. The findings indicating that 45°C temperature the larval period gradually decreased up to 1.99 days by increasing the mortality rate, 32.33% and survival rate of 63.33%. No moult was observed at 50°C because all the larvae become die and the provided food were dried at this temperature within few hours. The observations were in conformity with Balo, J. S. and S A L Haywords

(2010). In control experiment the first instar larvae were moulted into second instar in 2.25 days with 93.33% survival at 32°C temperature and 60±R. H.

On second instar larvae:

The tabulated data indicate that no survival of second instar caterpillars at 5°C temperature but at 10°C and 15°C temperature its larval period were 5.13 & 4.43 days having 30.00% and 46.66% rate of survival, respectively. The mortality rate decreased up to 41.38% & 24.48% with decreasing the larval period of 4.01 and 3.53 days at 20°C and 25°C temperature, respectively. The maximum survival were 93.33% & 83.33% and minimum mortality rate 03.79% and 13.44% as well as 2.89 & 2.41 days larval period were reported at the temperature level of 30°C and 35°C. At 40°C and 45°C temperature the larval period were further reduced a 2.01 & 1.72 days with 34.14% & 68.14% rate of mortality. Similar observations were recorded by Regniere, J. et al. (2012). The larvae could not survive at high level of temperature of 50°C due to complete dehydration. The controlled experiments results showed the 2.62 days larval period and 3.34% mortality.

On third instar larvae:

The calculated data were indicated that all third instar larvae were died at least level of temperature i.e. 5°C. Results were obtained at 10°C and 15°C the larval periods were 5.06 & 4.76 days with mortality rate 62.07% & 48.27%, respectively. At 20°C and 25°C temperature, the survival rate of caterpillars profoundly increased with 66.66% and 76.66%, respectively, moulted into fourth instar, having 4.15 and 3.72 days of larval periods. The highest level of survivality reported as 96.66% & 90.00 with 3.38 & 2.61 days larval periods was observed with treatment of 30°C & 35°C temperature intensity in conformity with the observation of Tomaz Jawoski & Jack Hilszezanski (2013). The larval period was gradually decreased as 2.01 & 1.76 days with increasing the mortality as 20.69% & 44.83% at 40°C and 45°C temperature, respectively. The extremely high temperature (50°C) dehydrated the larvae and caused 100% mortality within few hours.

On fourth instar larvae:

The findings revealed that longer larval period i.e. 11.84 and 09.94 days, higher mortality of 83.34% & 60.00% at 5°C and 10°C temperature. The required time to moult the fourth larvae into fifth instar stage at 15°C, 20°C, 25°C & 30°C were 8.43, 6.58, 4.62 and 3.41 days with 60.00%, 73.33%, 80.00% and 100% of survival rate, respectively. Mahesh pathak and co-workers (2003) also made a laboratory experiment on age specific survival and fertility table of *P. demoleus* at different sets of temperature and found similar results. The larval period again reduced as 2.77, 2.09 and 1.78 days having 90.00%, 80.00% and 66.66% mortality, when larvae were treated with 35°C, 40°C and 45°C temperature. Zero percent survivality was observed at 50°C as previous experiments. The controlled experiment conducted at 32°C temperature and 60±5 relative humidity have shown 2.89 days larval period without mortality.

On fifth instar larvae:

The calculated observations were indicated that as the temperature range increased as 5°C, 10°C, 15°C, and 20°C, the larval period were gradually decreased as 16.22, 12.85, 9.52 and 7.07 days with increasing the rate of survival as 33.33%, 50.00%, 63.33% and 80.00%, respectively. The highest rate of survival was observed at 25°C, 30°C and 35°C which were 90.00%, 100% and 100% with 5.13, 3.75 and 3.28 days of larval periods. As the temperature were increased to the higher level of 40°C and 45°C, the larval period were further decreased as 2.61 and 2.21 days with decreasing the survival rate, 86.00% and 80.00%, respectively. Timberlin, Alder and Myers (2009) observed similar trends of results in the development of Black soldier fly in relation to temperature influences. The fifth instar larvae were also unable to tolerate the extreme intensity of 50°C temperature. Zero percent mortality was observed with 3.52 days of larval period in controlled experiments.

Temperature effect on pupal stage:

The study on the impact of temperature fluctuation on the survival and development of pupal stages was carried out and findings were placed in the table-03.

TABLE – 03. Effect of temperature on Pupal period of Papillio demoleous at 60+5% R.H.

Temperature °C	No. of Pupae treated	Av. Pupation Period (Days)	No. of Pupae Emerged	Percentage of Emergence	Observed Mortailty (%)	Corrected Mortality (%)
05	30	-	00	00.00	100.00	100.00
10	30	95.26±20.19	09	30.00	70.00	76.66
15	30	59.03±11.29	16	53.33	46.67	46.67
20	30	24.85±6.66	23	76.66	23.34	23.34
25	30	11.01±3.81	28	93.33	06.67	6.67
30	30	9.38±2.27	30	100.00	00.00	00.00
35	30	8.17±1.99	30	100.00	00.00	00.00
40	30	7.62±1.66	27	80.00	20.00	20.00
45	30	6.69±1.01	20	50.00	50.00	50.00
50	30	-	00	00.00	100.00	100.00
Control	30	8.64±1.83	30	100.00	00.00	00.00

The tabulated observations were clearly indicated and found the similar results of influences of different levels of temperature on the development of pupal stage like incubation as well as larval developmental stages of experimented insect, Papilio demoleus (L). It was found that no pupae were emerged into adults at extreme low temperature of 5°C. The low level of temperature prolonging the pupal period and it was significant below 15°C. The pupae reached into the hibernation stage and required 95.26±20.19 days with 76.66% mortality at 10°C temperature. These findings were in agreements with the results of Balo and Haywords (2010). The pupal period was gradually decreased with increasing the temperature. The exposure of 15°C and 20°C temperature required 59.03 & 24.85 days and 53.33% & 76.66 pupae were emerged.

In the present investigations the 93.33%, 100% and 100% pupae were emerged in to the adults in 11.01, 9.38 and 8.17 days at the level of 25°C, 30°C and 35°C temperature. As the intensity of temperature were increased the rate of emergence were decreased with decreasing the pupal period. Therefore at 40°C and 45°C temperature the 80.00% & 50% pupae emerged into the adults and 7.62 & 6.69 days pupal period was recorded. The 100% mortality was recorded at 50°C temperature. The findings were almost similar with the results of Atwal (1955) and Thomas G. et al. (1993). No mortality was found at controlled experiments. The average pupal period was recorded as 8.64 day at 32°C temperature in controlled experiments.

Concludingly, at extremely low and high temperature no eggs were hatched into larvae as well as larvae of different instars and pupae were failed to moult into pupae and emerged into adults, respectively. During the course of laboratory works, it was proven that as the temperature increased from 10°C to 45°C the developmental period as well as mortality rate was gradually decreased. If the intensity of temperature decreased the reverse results were obtained. The optimal temperature was proven as 25°C, 30°C, and 35°C at 65+5 humidity resulting moderate developmental period and maximum rate of survival. Hence it had been established that temperature is a main important abiotic factor which directly influenced the developmental activities of an insect life.

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