# Thermal energy and artificial diet for tobacco caterpillar *Spodoptera litura* (Fabricius) (Lepidoptera: Noctuidae) development

## SUBHASH CHANDER, MAZHAR HUSAIN and GUNDAPPA BARADEVANAL<sup>1\*</sup>

Division of Entomology, ICAR-IARI, New Delhi, India <sup>1</sup>Division of Crop Protection, ICAR-CISH, Lucknow, India \*Corresponding author: Gundappa@icar.gov.in

### ABSTRACT

Development and survival of tobacco caterpillar *Spodoptera litura* larvae and pupae were studied at each of seven constant temperatures,  $15\pm1$ ,  $18\pm1$ ,  $21\pm1$ ,  $24\pm1$ ,  $27\pm1$ ,  $30\pm1$  and  $33\pm1^{\circ}$ C, in incubators. Oviposition, incubation, larval and pupal periods were recorded at each of the temperatures. The survival from 1<sup>st</sup> instar to adult emergence ranged between a maximum of 60% at 24°C and 30% at each of 18°C and 33°C. Developmental duration of different larval instars and pupae declined with an increase in temperature from 15 to 33°C. The optimum temperature for larval and pupal survival in *S .litura* on artificial diet was observed to be 24°C. On the other hand, optimum temperature range for oviposition was observed to be 27-33°C. The relationship between temperature (T) and development rate (r) was established using linear regression to estimate the thermal constant (K) and development threshold (T<sub>0</sub>). Thermal constant for 1<sup>st</sup>to6<sup>th</sup>larval instars were computed as 76.9, 90.9, 125, 83.3, 62.5, 90.9 and 200 degree days (DD), respectively, with corresponding development thresholds as 12.8, 11.7, 9.8, 10.3, 10, 10.9 and 13.2°C. Single developmental threshold for larval stage was computed as 13°C. These thermal constants and development thresholds can be used in developing mechanistic population simulation model, which in turn will facilitate the assessment of climate change impact on the pest.

Key words: Thermal energy, artificial diet, development rate, Spodoptera litura, survival

Tobacco caterpillar, Spodoptera litura (Fabricius) (Lepidoptera: Noctuidae) is a perilous insect pest affecting economically important crops such as tobacco (Nicotiana tobacco L.), castor (Ricinuscommunis L.), cotton (Gossypium sp. L.), soybean (Glycinemax L.) and groundnut (Arachis hypogea L.) throughout tropical and temperate Asia, Australasia and the Pacific Islands (EPPO, 2020). Out of 112 globally recorded host plants of S. litura, 60 are known only from India (CABI, 2020; Jha et al., 2016). Due to nocturnal habit, high mobility of adult moths and ability to oviposit on a wide range of host plants, S. litura has huge potential to invade new areas and to adapt to wide range of ecological situations (Duraimurugan, 2018). In India, S. litura is widespread in almost all the states and inflicts significant losses to economically important crops like soybean, cotton and groundnut. S. litura is likely to further increase under the imminent climate change, as it has been found to consume 30 per cent more cotton leaves at elevated CO<sub>2</sub> levels than ambient (Kranthi et al., 2009). Outbreaks of S. litura were also noticed in major sunflower growing areas of Central and Southern India. During 2005, the outbreak of S. litura led to more than 90 per cent defoliation of sunflower (Sujatha and Lakshminarayana, 2007).

Insects being poikilothermic organisms, the developmental rate is highly reliant on external temperature conditions. Hence, temperature is generally considered the single most significant environmental factor influencing behavior, distribution, development, survival and reproduction in insects (Bale et al., 2002). Knowledge on the temperature-dependent population growth potential of insect pests is highly imperative for understanding their population dynamics and implementing agro-eco region specific pest management strategies, especially in the context of predicted global climate warming (Kroschel et al., 2013). Considering this, the temperature rise of 2.7–4.7°C predicted due to potential climate change (IPCC, 2013) may have drastic consequences for future incidence of S. litura. The vast majority of studies that infer the effects of temperature on developmental biology of S. litura have been undertaken under only one constant temperature in laboratory (Roa et al., 1989; Prasad 2004; Bharathi et al., 2007; Patil et al., 2014). A few studies that addressed the development of S. litura at a range of constant temperatures, were concerned with predicting developmental rates and threshold temperatures using linear degree day or heat summation models (Rao et al., 1989). The objective of this study was to develop thermal constants and developmental thresholds for *S. litura*, which can be used in developing mechanistic population simulation model, which in turn will facilitate the assessment of climate change impact on the pest.

### MATERIAL AND METHODS

The study was conducted in the laboratory of the Division of Entomology, ICAR-Indian Agricultural Research Institute (IARI), New Delhi. The laboratory experiments were conducted using Biological Oxygen Demand (BOD) incubators that were set at constant relative humidity ( $65 \pm 5\%$ ) and photoperiod (14:10 L:D) and different temperatures as required for determining thermal constants and development thresholds studyof *S. litura*.

### Establishment of a stock culture of S. litura

The *S. litura* larvae were collected from IARI fields, sorted out and reared in glass containers on fresh artificial diet that was changed daily to maintain hygienic conditions. The containers' tops were covered with perforated aluminum foil lids. The pupae were kept in separate 20x10 cm rearing jars for adult emergence. On emergence, adults were placed in 20x10 cm rearing jars with their tops covered with muslin cloth and were provided with 15% sugar solution through pieces of cotton in petri dishes. The adult food was daily changed. Fresh tomato branches dipped in 100 ml water containers were placed in adult rearing jars for oviposition. The adult rearing jars were maintained at  $25\pm1^{\circ}$ C and 65% relative humidity. The faecal matter and unused diet was removed from containers as and when required.

# Development and survival of S. litura at different constant temperatures

Development and survival of *S. litura* life stages *viz.*, eggs, larvae and pupae were studied at each of seven constant temperatures *viz.*,  $15\pm1$ ,  $18\pm1$ ,  $21\pm1$ ,  $24\pm1$ ,  $27\pm1$ ,  $30\pm1$  and  $33\pm1^{\circ}$ C in BOD incubators. Oviposition periods of females were recorded in five replicates at each of the temperatures. Likewise, freshly laid eggs were incubated at different constant temperatures in five replicates and observed for hatching between 11.00 AM-12.30 PM daily. Similarly, 10 neonate larvae were reared at different temperatures and observed for moulting, mortality and pupation every day. Similarly, pupae were observed for adult emergence. Incubation period of eggs, and development periods of different larval instars and pupae were recorded at each of the temperatures.

# Computation of thermal constants and developmental thresholds

According to rule of the constant sum of effective temperatures

 $(T-T_0) \ge D = K$  (Davidson 1944, Wigglesworth 1972).

Where T = temperature at which insect species is reared;  $T_0^{=}$  lower development threshold; D= duration of development, and K = thermal constant.

Development rates (r) of different developmental stages of *S. litura* were computed as the reciprocal of developmental periods in days (1/D) at each of the temperatures. The relationship betweenT and r was determined using linear regression where r = a + bT. The K was estimated as reciprocal of regression coefficient (b) between development rate and temperature.

K = 1/b (Kipyatkov and Lopatina 2010).

The  $T_0$  was determined as ratio of regression intercept (a) and b.

 $T_0 = -a/b$ 

Thermal constant and lower developmental thresholds were determined for eggs, larvae and pupae of *S. litura.* 

#### **RESULTS AND DISCUSSION**

Oviposition by S. litura was observed only at 27°C and 33°C and at other constant temperatures either adults did not emerge or they died before oviposition. Neonate larvae completed their development from 1<sup>st</sup>instar to pupal stage at 18-33 °C (Table 1). However, larval development was not completed at 15°C as larvae died in 5th instar at 15°C. This might be due to the fact that at constant temperatures of  $\circ^{\circ}C$ , larvae required considerably more time to develop and favorable physiological conditions could not be maintained for such a long time. Developmental periods of different S. litura instars declined with an increase in temperature from 15 to 33°C. However, at 15°C larvae did not complete their life cycle. Development period of pupae also declined with temperature increase from 18°C to 33°C. Combined period for the larval and pupal development was 101.67±0.67 days and 30±1 days at 18 and 33°C, respectively (Table 3). The larval and pupal development periods of S. litura were thus influenced by temperature, which has also been observed earlier (Prasad 2004; Bharathi et al., 2007; Patil et al., 2014). Each species has an ecological maxima and minima within which its development rate increases with rise in temperature

Table 1: Developmental period in days (mean±SE) of S. litura at different constant temperatures

| Temp.     | n  | I instar   | II instar       | III instar | IV instar  | V instar  | VI instar | First            | Pupal      | Total                         |
|-----------|----|------------|-----------------|------------|------------|-----------|-----------|------------------|------------|-------------------------------|
|           |    |            |                 |            |            |           |           | instar to        |            | Develop-mental                |
|           |    |            |                 |            |            |           |           | final instar     |            | period                        |
|           |    |            |                 |            |            |           |           |                  |            | (1 <sup>st</sup> instar-pupa) |
| 15°C      | 10 | 22.50±0.65 | 21.14±1.70      | 18.33±3.15 | 19.33±3.38 | -         | -         | -                | -          | -                             |
| 18°C      | 10 | 20.33±1.20 | 19.56±0.75      | 11.38±1.03 | 10.83±0.98 | 6.00±0.12 | 8.67±0.33 | 69.67±1.20       | 32.00±1.53 | 101.67±0.67                   |
| 21°C      | 10 | 3.22±0.15  | $7.00{\pm}0.50$ | 10.40±0.93 | 6.60±0.93  | 6.40±0.60 | 8.25±1.03 | 41.25±1.18       | 20.25±0.95 | 61.50±0.65                    |
| 24°C      | 10 | 4.89±0.31  | 6.89±0.45       | 8.50±0.76  | 4.50±0.71  | 4.50±0.19 | 4.88±0.35 | 33.67±0.92       | 12.14±0.51 | 45.83±0.79                    |
| 27°C      | 10 | 4.63±0.32  | 5.25±0.52       | 6.80±1.36  | 4.60±0.81  | 3.00±0.55 | 5.75±0.75 | $30.00 \pm 2.52$ | 13.67±0.33 | 43.67±2.85                    |
| 30°C      | 10 | 5.00±0.31  | 5.43±0.65       | 7.71±0.29  | 5.83±0.31  | 3.83±0.40 | 5.17±0.31 | 33.17±0.79       | 11.00±1.15 | 45.33±2.03                    |
| 33°C      | 10 | 3.30±0.15  | 4.20±0.97       | 4.00±0.58  | 3.00±0.00  | 2.33±0.33 | 4.33±0.67 | 22.00±1.00       | 8.00±1.00  | 30.00±1.00                    |
| CD (0.05) |    | 1.512      | 3.344           | 3.251      | 2.139      | 0.931     | 1.088     | 2.288            | 1.623      | 2.438                         |

Table 2: Larval and pupal survival (%) in Spodoptera litura at different constant temperatures

| Temp. | n  | I instar | II instar | III<br>instar | IV<br>instar | V instar | VI instar | Total larval<br>survivorship  | Pupal<br>survivorship | Total<br>survivorship    |
|-------|----|----------|-----------|---------------|--------------|----------|-----------|-------------------------------|-----------------------|--------------------------|
|       |    |          |           |               |              |          |           | (1 <sup>st</sup> instar-final | (final instar-        | (1 <sup>st</sup> instar- |
|       |    |          |           |               |              |          |           | instar)                       | pupa)                 | adult)                   |
| 15°C  | 10 | 80.00    | 87.50     | 85.72         | 50.00        | 0.0      | 0.0       | 0.0                           | 0.0                   | 0.0                      |
| 18°C  | 10 | 90.00    | 100.00    | 88.89         | 75.00        | 83.33    | 60.00     | 30.00                         | 30.00                 | 30.00                    |
| 21°C  | 10 | 90.00    | 100.00    | 55.56         | 100.00       | 100.00   | 80.00     | 40.00                         | 40.00                 | 40.00                    |
| 24°C  | 10 | 90.00    | 100.00    | 88.89         | 100.00       | 100.00   | 100.00    | 80.00                         | 70.00                 | 60.00                    |
| 27°C  | 10 | 80.00    | 100.00    | 62.50         | 100.00       | 100.00   | 80.00     | 40.00                         | 30.00                 | 30.00                    |
| 30°C  | 10 | 70.00    | 100.00    | 100.00        | 85.72        | 100.00   | 100.00    | 60.00                         | 30.00                 | 30.00                    |
| 33°C  | 10 | 100.00   | 50.00     | 60.00         | 100.00       | 100.00   | 100.00    | 30.00                         | 30.00                 | 30.00                    |

but decreases beyond it. Generally, lower temperatures result in a decrease in the rate of development and an increase in duration of a developmental stage (Manimanjari *et al.*, 2014). Present study revealed that *S. litura* could develop under constant temperatures ranging between 20°C and 30°C. We also found that the females of *S. litura* were able to lay eggs only at constant temperatures of 27°C and 33°C. Present results are in agreement with those reported by Rao *et al.* (1989), however, with a deviation that they reported egg laying at low temperature of 15°C, which we did not observe. Rao et al. (2014) reported decrease in development time of different stages with an increase in temperature from 20-35°C that was similar to the decrease in developmental time from 15-33°C in the present study too. Likewise, thermal requirement from first to sixth instar in present study occurred to be 527DD in comparison to 538.5 DD from egg to adult development in case of Rao *et al.* (2014).

Incubation, larval and pupal developmental periods of *S. litura* as recorded in present study were proximal to those reported earlier under similar temperature conditions (Fand *et al.*, 2015; Nandita *et al.*, 2017). In current study, *S. litura* completed its larval development in 6 instars that has also been reported earlier (Rao *et al.* 1989). The developmental durations of *S. litura* immature stages observed at different constant temperatures were largely in conformity with earlier reports (Rao *et al.*, 1989; Patil *et al.*, 2014).Deviations in the developmental periods in the present study vis-à-vis earlier work may be attributed to the different

| Life stages | b     | a      | DD    | То    | R <sup>2</sup> |
|-------------|-------|--------|-------|-------|----------------|
| I instar    | 0.013 | -0.167 | 76.92 | 12.85 | 0.900          |
| II instar   | 0.011 | -0.129 | 90.90 | 11.73 | 0.958          |
| III instar  | 0.008 | -0.079 | 125   | 9.88  | 0.794          |
| IV instar   | 0.012 | -0.124 | 83.33 | 10.33 | 0.783          |
| V instar    | 0.016 | -0.160 | 62.5  | 10.00 | 0.785          |
| VI instar   | 0.011 | -0.120 | 90.91 | 10.91 | 0.869          |
| I-VI instar | 0.001 | -0.013 | 1000  | 13.00 | 0.848          |
| Pupa        | 0.005 | -0.066 | 200   | 13.20 | 0.904          |

 Table 3: Linear regression equation, lower developmental thresholds and thermal constants of larval and pupal developmental stage of *Spodoptera litura*.

host plants used as food for rearing the insect species (Xue *et al.*, 2010).

In present study, the larval survival was observed to be maximum (80%) at 27°C and minimum (30%) at both 18 and 33 °C as indicated by pupal formation. Overall, survival from 1<sup>st</sup> instar to adult emergence ranged between a maximum of 60% at 24°C and <sup>7</sup>/<sup>°</sup> at each of 18°C and 33°C(Table 2). The 24°C was thus observed to be the optimum temperature for the survival and development of S. litura . The survival rates of S. litura immature stages as well adult stage varied significantly at various constant temperatures. The constant temperatures below 20°C and above 35°C were highly unfavourable for survival of all the life stages, at which increased mortality rates were observed. Earlier, Fand et al. (2015) observed highest percentage of survival (65.0%) of S. litura larva when reared at constant temperature of 30°C in laboratory conditions. Manimanjari et al. (2014) reported the similar trend in survivorship of S. litura where in they found lowest and highest survival at 20°C and 30°C temperatures, respectively, when reared on sunflower (Helianthus annus L.). Further, Xue et al. (2010) reported that survival rates of immature stages of S. litura are highly dependent on host plants used for larval feeding.

Based on regression of developmental rates on corresponding temperatures, thermal constant for  $1^{st}$  to  $6^{th}$  larval instars and pupal stage of *S. litura* were computed as 76.9, 90.9, 125, 83.3, 62.5, 90.9 and 200 degree days (DD), respectively, with corresponding development thresholds as 12.8, 11.7, 9.8, 10.3, 10, 10.9 and 13.2°C (Table 3; Fig 1). In present study, developmental threshold for different larval instars reared on artificial diet ranged from 9.8-13.2°C and single developmental threshold for larval stage was determined as 13°C. The lower developmental threshold temperatures for

*S. litura* larval stages estimated in the present study were in closer agreement with the earlier reports (Rao, 1989). The slight deviations that exist between current thresholds and those reportedearliermay be ascribed to different rearing conditions and host plants used as larval food. In present study, thermal constant for pupal stage was computed as 200 DD with developmental threshold as  $13.2^{\circ}$ C. Earlier Fand *et al.* (2015) reported lower developmental threshold for pupal stage of *S. litura* as  $9.8^{\circ}$ C.

It has also been opined earlier that variable values obtained for thermal constants in different environments probably are indicative of different ecotypes of the pest. Fecundity and adult longevity in *S. litura*are influenced by temperature, humidity, and larvaland adult nutrition (Zhu *et al.*, 2000; Xue *et al.*, 2010). In present study, although larvae did not complete their development at 15 and 18°C, yet developmental thresholds for different development stages ranged from 9.8– 13.2°C. This was possible due to extrapolation of developmental rates obtained for temperatures from 18-33°C.

In this study, thermal constants for different larval instars ranged from 62.5-90.9 DD, while their developmental thresholds ranged between 9.8-12.8°C. On the other hand, pupal development required more amount of degree days for its completion with higher developmental threshold. Pupa being a transformational stage in insect development thus required higher amount of heat energy and its development was also triggered at higher developmental threshold.

### CONCLUSION

The growth and development of *S. litura* were significantly influenced by temperature. Results from this study showed that both low and high temperatures limited

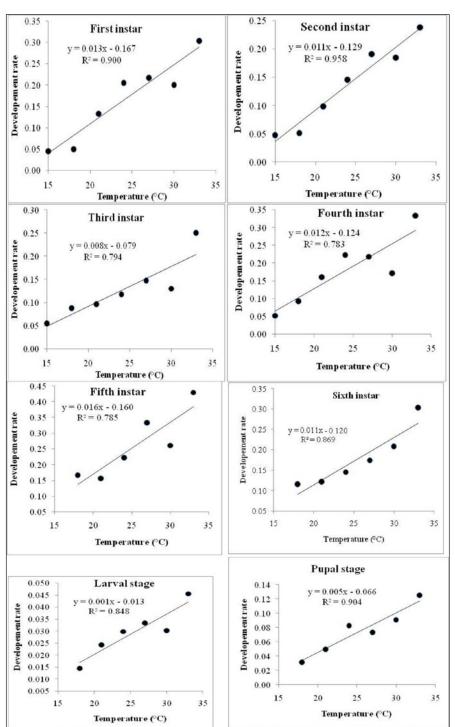


Fig. 1: Linear relationship between the temperature and developmental rate of different developmental stages of S. litura.

the survival and development of this insect pest and based on development, survival and fecundity, optimum temperature for the larval and pupal survival was observed to be around 24°C, though the development rates increased with temperature up to 30°C. On the other hand, optimum temperature ranges for oviposition seems to be 27-33°C. Data on thermal constant and developmental thresholds of different developmental stages of *S. litura* can be utilized to formulate a population simulation model of the pest. Different abiotic and biotic mortality factors can also be incorporated in to the simulation model. Such a model will be useful to investigate the impact of global warming on biological processes that are responsible for changes in population levels of the pest. The model will thus prove more useful than empirical insect-weather regression models that do not explain the mechanisms behind insect population change.

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