BIOLOGY AND POPULATION GROWTH PARAMETERS OF TOMATO LEAF MINER, (*Tuta absoluta*, MEYRICK) (GELECHIIDAE: LEPIDOPTERA), ON TOMATO IN THE LABORATORY

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ABSTRACT

Tuta absoluta Meyrick *recent invasion, morphological and biological variations, spreading capacity and the urgency for substantiate potent management strategy makes this pest researchable in Nepal. An experiment was carried out in the laboratory of Entomology Division, Khumaltar, Nepal at August 2016 to June 2017 to access biology and population growth parameters of this pest under different temperatures and relative humidity. Parameters such as developmental stage, adult longevity, fecundity and mortality were recorded. Considering the life cycle of T. absoluta* Meyrick, *the results revealed that the increase of temperature and relative humidity shortened the overall development period of the pest. Rise in temperature caused decreased pupal development period by three times i.e. 15.8±0.3 days at 20.9˚C to 5.6±0.2 days at 29.01˚C. The adult longevity and fecundity also decreased with increasing temperature. The mortality rate was recorded the highest at egg stage (46%), while the lowest rate was recorded at adult stage (11.3%) at mean temperature 24.4˚±0.16˚C and RH 74.4 ±1.3%.*

Keywords: Fecundity, longevity, relative humidity, temperature, *Tuta absoluta*

INTRODUCTION

Tomato, *Solanum lycopersicon* L. (syn. *Lycopersicon esculentum* Mill.) is one of the most consumed vegetables in the world, its production accounts for about 4.8 million hectares of harvested land area globally with an estimated production of 162 million tons (FAOSTAT, 2014). The total production of tomato in Nepal is about 331736 mt from 19725 hectares of land with productivity 17mt/h (MoAD, 2016). Productivity of tomato in Nepal is lower than half by the world average may be due to several biotic and environmental constraints. According to Kafle and Shrestha (2017) the major limiting factor on production of plastic house tomato in Nepal are disease and insects/pests. Global agriculture and trade introduced new pests into the country frequently (Bhandari *et al.*, 2016). Recently, the most destructive harmful one is *Tuta*

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absoluta Meyrick (Lepidoptera: Gelechiidae) (Sabbour and Nayera, 2014). The South American tomato leaf miners an invasive pest, native to South America which was detected in eastern Spain at the end of 2006. The intensity of its attack and also occurrence during all cropping period have has made *Tuta absoluta* Meyrick a very serious agricultural threat to tomato production in both greenhouse and on-field tomato crops (Oliveira *et al.*, 2008). This pest spreads galloping and its status in the world has completely changed within only a few years from a South American tomato insect to a major threat to tomato world production (Duarte *et al.,* 2015).

This pest was reported for the first time, from Kavrasthali in Kathmandu valley of Nepal in 2016 (Bajracharya *et al.,* 2016) and then distributed in many parts of the country. The survival and adaptation potential is considered a prime component that affects the level of genetic diversity in introduced populations of an invasive species (Sakai *et al.,* 2001). The widened infestation pressure of the pest due to seasonal variation causes economic losses of up to 100% (Ayalew, 2015). Moreover, in Nepal where the pest was noticed in farmer's fields recently, it seemed doubtlessly that tomato growers were not prepared for the pest thus they never had any appropriate control strategy against it. Preliminary information suggests 25-30% of tomato crops have been lost in the hardest hit areas. That amounts to approximately a loss of $7,200 a hectare (IDE, 2016). All the successful tools of pest management used in other countries may not be directly applicable and successful in Nepalese agro- ecosystem. Notable new management strategies against pests can be achieved by knowing the population ecology of the target species. Tomato Leaf Miner (TLM) pheromone lure @ 1 trap per ropani was recommended for management of *T. absoluta* in Nepal (PPD, 2017). However, Caparros *et al.,* (2012) reported the evidence of deuterotokous parthenogenesis in *T. absoluta*. This might hinder the efficacy of pheromone based management approach. An experiment carried out at plastic house condition in kathmandu valley showed that Chlorantraniliprole was the most effective pesticide in managing *Tuta absoluta* whereas Spinosad and Abamectin plus *B. thuringiensis* combination was also equally effective (Simkhada *et al.,* 2018). Life table information is crucial while focusing to manage an insect species as it gives the most comprehensive description on the growth, survival and fecundity briefly which implies for scientific management of the pest. For this reason, study on its biology and population growth parameters is crucial to develop and implement effective management tactics of this pest.

OBJECTIVE

The objective of the present work is to study the biology and population growth parameters of *Tuta absoluta* Meyrick on the tomato plants at laboratory condition.

METHODOLOGY

INSECT SOURCES

Populations of the *Tuta absoluta* Meyrick were collected from Horticulture Research Division (HRD), Khumaltar, Lalitpur. The infested leaves, along with the eggs, larvae and pupae were collected and reared to adult stage; and were subsequently established under laboratory of Entomology Division of Nepal Agriculture Research Station (NARC), Khumaltar, Lalitpur situated at 1326 masl, within 27˚38.315'N and 85˚22.883'E. Minimum/Maximum temperature and relative humidity (RH) of the laboratory was recorded daily.

INSECT REARING

Newly emerged *T. absoluta* Meyrick adults were placed in a net cage (46.5 × 46.5 × 46.5 cm). *T. absoluta* Meyrick was reared for one generation and eggs laid by females from the second generation were used in the experiments.

EGG DEVELOPMENT AND SURVIVAL

T. absoluta Meyrick adults were placed in transparent acryliccups (6.5 cm of height, 8.0 cm of diameter) covered with black muslin cloth. Two droplets of honey water solution were added through muslin cloth in the cup as food and were kept in the acrylic cup to oviposit for twenty four hour. Next, the eggs laid on the underside of muslin cloth were counted and after that, eggs were collected daily by fine brush and placed in a plastic box lined with moist tissue containing a single piece of tomato leaf. When eggs were hatched, the emerged larvae were provided with fresh tomato leaves, egg development time and egg survivals were recorded.

LARVAL DEVELOPMENT AND SURVIVAL

The larvae were reared in a plastic container (30 × 21 × 7 cm) with a piece of tissue paper at the bottom of the lid. Larvae were developed through four instars in tomato leaves and then pupated. Leaf pieces were changed every day in order to feed the emerging larvae. The old piece of leaf was cut around the larva using a scalpel blade and the new one was inserted in the container. Newly emerged larvae were counted daily. Very little water was also added to humidify the tissue paper until pupation. Mature larvae were pupated at the bottom of larval rearing containers making folds inside the lined tissue paper in the laboratory. Pupae were counted and collected daily by carefully examining the rearing containers.

PUPAL PERIOD AND ADULT LONGEVITY

Counted number of pupae was placed individually in a Petri plate containing a filter paper. This allowed adult moths emerging from individual pupa to be counted. The observations were made till the adult emergence. The period between pre-pupae till the adult emergence was noted as pupal period. Adults obtained from the culture were enclosed in screen cages, fed with a honey solution (10%). The period from the adult emergence to death was taken as adult longevity. During the development of the insects, egg-laying date, hatching date, pupation date, adult emergence date was daily recorded for each individual.

FECUNDITY

To determine the fecundity of *T. absoluta* Meyrick, a pair of newly emerged female and male moths were introduced into a plastic container (11.5 cm diameter, 9.5 cm depth), which was sealed at the top with a fine mesh net for ventilation. Each container was supplied with a 10% honey solution for adults feeding. Also the containers for oviposition were covered with thin black muslin cloth for the ease in counting the eggs produced. The number of eggs laid in each day was recorded until the death of the last adult.

STATISTICAL ANALYSIS

The collected survey data was classified according to the required information and analyzed using various statistical tools like mean and standard deviation. All data recorded from the laboratory were tabulated and drawn in the Microsoft Excel. The information regarding number of insects dying as a percent of number entering a particular stage can be obtained from the apparent mortality, which was calculated as:

Apparent mortality = (dx / lx) × 100 where

lx = Number surviving at the beginning of each interval

dx = Number dying during the age interval

RESULTS AND DISCUSSION

DEVELOPMENT RATE OF TUTA ABSOLUTA MEYRICK IN LABORATORY

Results of the comparative study to evaluate effect of temperature and relative humidity for South American tomato leaf miner, *T. absoluta* Meyrick on the developmental parameters at eggs, larvae and pupae stages are shown in Table 1. All experimental cohorts of *T. absoluta* Meyrick were able to complete their development from 20˚C to 29˚C mean temperatures conditions. With increasing temperature, developmental duration for different stages of the pest decreased (Figure 1).

Table 1: Developmental periods (days) of *T. absoluta* Meyrick (Mean±SE) at three different mean temperatures in laboratory

Mean Temperature (˚C)	Mean Relative Humidity (%)	Developmental Stages		
		Egg Dev. period	Larval Dev. Period	Pupal Dev. Period
20.9	52.87	7.4±0.24	18.8±0.9	15.8±0.3
24.3	74.4	4.6±0.3	13.2±0.4	10.4±0.2
29.01	75.54	3.2±0.2	8.4±0.4	5.6±0.2

The value before ± indicates average number (days) and after ± indicates standard error (S.E)

Egg stage

The freshly deposited eggs were translucent, smooth and shining, were oval in shape and creamy white in color then turn to yellow and finally black before hatching. The incubation period decreased from 7.4±0.24 days to 4.6±0.3 and to 3.2±0.2 days with increase of the mean temperature from 20.9˚C, 24.3˚C to 29.01˚C respectively (Table 1).

Larval stage

Just after hatching, the soft bodied larvae in the initial instars were cream colored with prominent dark brown head. The larvae seemed to be full grown as their size became remarkably large and fed more voraciously and become greenish in color and the dorsal region turns to pinkish when they are close to pupate. The mean length of full grown larvae was 7.37±0.05mm and width was 1.2±0.08mm. Data presented in Table (1) shows that the increase of temperature shortened the larval duration being 18.8±0.9, 13.2±0.4 and 8.4±0.4 days at 20.9˚C, 24.3˚C and 29.01˚C mean temperature respectively.

Pupal stage

After development of 4th larval instars the insect pupates. The pupa was the ultimate transformation of pre-pupa which was also a non feeding stage, the pupa was cylindrical shape, the body color in initial stage was green and finally it turned brown. The mean length of pupa was 3.97±0.58mm and width was 1.09±0.07mm. Rise in temperature leads to the decreased pupa development period by three times i.e. from 15.8±0.3 days at 20.9˚C to 5.6±0.2 days at

29.01˚C. The longest pupal period also occurred at 20.9°C while the shortest was in the 29.01°C. Following the pupation, the moth emerges to give male and female adults. Both pre-pupal and pupae stage took place outside the mine

Adult

Adult moths were silvery brown in color and have black spots on the forewings. They have fiiform antennae. The abdomen of male moth was slender and sharp while comparing with the wider abdomen of female. The mean length of adult was 9.98±0.72mm and width 1.41±0.58mm.

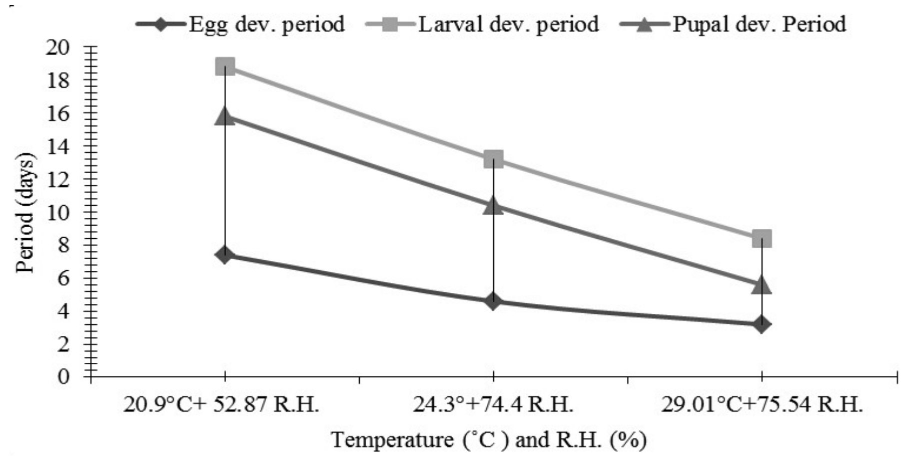

Figure 1. Developmental stages of *Tuta absoluta* Meyrick at three mean temperatures in laboratory

Temperature regime and relative humidity influenced almost all parameters we determined for the various stages of *T. absoluta* Meyrick. Our findings revealed that the mean egg development period of *T. absoluta* Meyrick varied from 3.2 days at 29.01°C to 7.4 days at temperature 20.9°C. The defensive compounds and nutritional qualities of host plants might influence the fecundity and oviposition duration of herbivore insects.

In the current study, the mean larval periods varied between 8.4 days to 18.8 days on different temperature and relative humidity regime. At 25 ± 1°C the larval periods of the tomato leaf miner were 12.14 days on tomato (Pereyra and Sanchez, 2006). It was determined that that the period of larvae instar of *T. absoluta* Meyrick was 10.97 days at 25-26°C (Erdoghan and Babaroglu, 2014) which is nearly similar to this results.

ADULT LONGEVITY AND FECUNDITY

Table 2. shows the longevity and reproductive parameters of the adults of *T. absoluta* Meyrick at three different mean temperatures. Adult longevity durations decreased with the temperature increase and lowest figures were recorded at the highest temperature (29.01°C). The variation in totals of eggs laid by female moths fluctuated with the changing temperature. Results (Table 2) reveal that females reared at mean temperature 20.9 ˚C had the highest fecundity (159.8±6.2 eggs) whereas those from the 29.01°C mean temperature had the lowest (113.8±4). The length of the oviposition period, in general, decreased from 20.9 to 29.01°C. At mean temperature/ relative humidity of 24.3˚C/ 74.4% respectively, production was 127.6±5 eggs/female. The adult longevity decreased to be 8.6±0.2 days at mean temperature 29.01˚C from18.2±0.3 days with temperature 20.9˚C.

Table 2: Adult longevity (days), and fecundity (eggs/female) of *T. absoluta* Meyrick at three mean temperature and relative humidity under laboratory conditions

Mean temperature (˚C)	Mean relative Humidity (%)	Adult longevity (days)	Fecundity (eggs/female)
20.9	52.87	18.2±0.3	159.8±6.2
24.3	74.4	11.4±0.4	127.6±5
29.01	75.54	8.6±0.2	113.8±4

The value before ± indicates average number (days) and after ± indicates standard error

Tuta absoluta Meyrick is well able to develop under temperatures that would commonly be experienced in plastic house of Nepal. The current work has determined that between 20 and 29°C is the most favorable temperature for moth development. It cannot be confirmed that a population would die out if laboratory temperatures were lowered to below 20°C. The longevity of adult decreased with the accelerated temperature. At temperature and relative humidity of 29.01°C (75.5% R.H.) in the laboratory, adult *T. absoluta* Meyrick lived for 8.6±0.2 days. In this temperature range, the longevity of *T. absoluta* Meyrick is shorter compared to the other temperatures. In higher temperature conditions, the development of egg, larva and pupa shortens, which is the characteristic phenomenon for large group of forest species (Szujecki, 1998). Fast temperature changes and particularly exposure of insects to low temperatures result in a reduction of the metabolic rate and, consequently, in slower development (Roy *et al.,* 2002).

The egg production was highly affected by temperature variation and the highest production occurred at 20.9°C producing 159.8±6.2 eggs/female, while, at higher temperatures, the mean production of eggs was decreased i.e. 113.8±4 eggs/female at 29.01°C. The result is in line with Pereyra and Sanchez (2006) who reported that the fecundity (number eggs/female) of *T. absoluta* Meyrick was 132.78 eggs on tomato plants. Similarly, Erdoghan and Babaroglu (2014) reported that the mean fecundity of this insect was 141.16 eggs on tomato. Number of eggs laid by female insects on leaf surfaces may vary due to the repellent activity of volatile compounds, causing irritability in the females at the time of oviposition (Mordue and Blackwell, 1993). Observation of repellent action was beyond the scope of this study. It was noted that oviposition at its peak during the first days of egg lying. Similar observation was made by Bogorni and Carvalho (2006) at 25°C on susceptible tomato cultivars.

MORTALITY ANALYSIS

Table 3. shows mortality rate of *T. absoluta* Meyrick with respect to its different life stages. Results clearly indicated that the mortality rate (dx) was recorded highest (46%) at egg stage, particularly in the early stages; followed by a gradual decrease in the population densities throughout its life span over the study period. The lowest mortality rate was recorded at adult stage (11.3%) at mean temperature 24.4˚±0.16˚C and RH 74.4 ±1.3%. Considerable number of mortality was recorded in the early instars larvae and pupae stages; 32.4% and 27.3% respectively. Low numbers of adult moths were developed in the laboratory experiments compared to the starting number of eggs. The values of the population parameters can vary under different field and laboratory conditions.

In the current study, *T. absoluta* Meyrick showed high natural mortality. During its life cycle the larval stage is known to be the most critical (Miranda *et al.*, 1998). Mortality percentage of eggs was recorded during life cycle: 46%, at 24.4±0.16°C (74.4±1.3% R.H.). To the contrary, Cuthbertson (2011) stated that in the development stage, the survival of the egg stage was 100%; no mortality rate was recorded. For *T. absoluta* Meyrick, the weak points appear to be the egg and the first instar larva before it tunnels into the plant.

Table 3: Mortality rate (%) of T. *absoluta* Meyrick at mean temperature 24.4±0.16 (RH 74.4±1.3%)

Mean Temperature ˚C		Relative humidity %		Age	Surviving at start (lx)	Number dying (dx)	Mortality rate (%)
24.4±0.16		74.4±1.3					
Max Temp	Min Tem	Max RH	Min RH				
30.8˚C	25.3˚C	99%	71.3%	Egg	200	92	46
				larvae	108	35	32.4
				Pupa	73	20	27.3
				Adult	53	6	11.3

The possible reasons might be due to unsuitable environment such as moisture and temperature coupled with the artificial diet. Possibly, due to their small size, the eggs and the first instar larva are more susceptible to changes in temperature than other developmental stages. Accordingly, the pupal mortality rates during the study periods were recorded as 32.4% under same laboratory conditions. This data is in agreement with the findings of Erdogan and Babaroglu (2014), who found that the survival rate of pupa was 63.10% that is 36.9% mortality. When the average environment temperature has increased, higher mortality has been observed for caterpillars of *Lymantria monacha* Linnaeus (Karolewski *et al.,* 2007). Higher mortality rate of all development stages in laboratory may be partly due to the stress caused by the experimental conditions. Furthermore, under natural conditions, this insect can complete its development in a single leaf, as it takes longer to deteriorate under these conditions than in the experimental conditions.

CONCLUSION

Large host range and innate ecology has made *Tuta absoluta* Meyrick, a crucial pest of tomato around the world. The results obtained in this study demonstrated that the incubation period of the eggs, larval and pupal duration were significantly shortened with the rise in temperature. Since this experiment explores the biology and population growth parameter of *Tuta absoluta* in the context of Nepalese climatic condition, this information can be of use while projecting the population growth and development of management strategy against this pest in field condition.

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