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Original Research Article

Effect of Elevated Temperatures on Mortality of Different Life Stages of *Lasioderma serricorne*

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Keywords

Cigarette beetle, temperature, lethal temperature and lethal time

A B S T R A C T

An experiment was conducted at the Department of Entomology, S. V. Agricultural College, Tirupati on mortality response of different life stages (egg, early, middle, late larval instar, pupal and adult) of cigarette beetle, *Lasioderma serricorne* to various temperature regimes (-5 to 35^0 C). The results indicated that all the life stages viz., egg, early instar, middle instar, late instar, pupa and adult were susceptible to low temperature at -5°C. Among all the life stages tested, eggs and early instars were susceptible to temperatures of 0°C and 5°C. Middle instars, late instars, pupa and adult were not susceptible to these temperatures. There was no effect on different life stages of cigarette beetle, at 15°C, 25°C and 35°C temperatures.

Introduction

Managing grain in storage from insect damage is just as important, or more so, than managing the crop while it is growing in the field (William and Dwight, 2016).

The cigarette beetle, *Lasioderma serricorne* (Fabricius) (Coleoptera: Anobiidae) is one of the several beetle pests that commonly infest warehouses and retail stores. It is known to successfully breed and develop on a variety of grain-based products, spices, tobacco and infest these commodities during storage, manufacturing and at the retail level (Dimetry *et al.*, 2004).

Stored grain insect pest are mainly managed traditionally with synthetic chemicals. However chemical management techniques have lead to various potential problems such as high traces of synthetic pesticides on treated stored food grain coupled with development of insecticide resistance in these insect pests owing to indiscriminate use of chemical pesticides. Resistance to malathion is widespread in Canada, USA and Australia (Subramanyam and Hagstrum, 1995). Currently, eco-friendly agriculture and demand for organic foods are gaining worldwide attention due to potential threat associated with the agricultural chemicals (Sangavi and Edward, 2017)

A number of alternative ways have been envisaged by various workers for managing

store grain insects of which modified atmosphere provides a way to eliminate insects from stored commodities without polluting the atmosphere and is considered as a safer method of managing stored insect pests.

Temperature treatment of stored grains is one of a best physical method which successfully kills several life stages of insects. Most of the stored product insects cannot tolerate extreme temperature, heating and cooling and shows high mortality. Low temperature reduces insect development and kills large number of immature stages of stored grain insects (Upadhyay and Ahmad 2011).

Materials and Methods

The investigation on effect of temperature on the mortality of various life stages of cigarette beetle under laboratory conditions was conducted at the Department of Entomology, S.V. Agricultural College, Tirupati and Institute of Frontier Technology, RARS, Tirupati.

Insect cultures and collection of life stages

Different life stages (egg, grub, pupa and adult) of *Lasioderma serricorne* used in the present investigations were taken from the base culture that was derived from a nucleus culture of *L. serricorne* maintained on turmeric under laboratory conditions (Temperature 26 ± 2^0C and 60-95 per cent R.H) at Department of Entomology, S. V. Agricultural College, Tirupati, since 2014.

Pupae were sexed on the basis of genital papillae (Halstead 1963). Known number of males and females of *L. serricorne* from nucleus culture were taken, paired and kept on coriander for oviposition. Eggs that were deposited were reared continuously on coriander till adult emergence. The required stages (egg, grub, pupa and adult) were taken from this base culture and used in experiment.

The various life stages of *L. serricorne i.e.,* egg, early instars, middle instars, late instars, pupae and adults were exposed to different temperatures and the effect of these temperatures on the mortality of each stage of *L. serricorne* was determined.

Growth Chambers

Six growth chambers (Kemi (Model No. KBOD-10S), Bros Scientifics (Model No. 77510S), REW (Model No. BOD-6S), Equitron (Model No. #7142), Labline (Model No. 05124C) and Spectrum (Model No. SDS-170) were used for exposing different life stages of *L. serricorne* to various constant temperature of 35°C, 25°C, 15°C, 5°C, 0°C and -5°C. A control was set at normal room temperature. Growth chambers were set at required temperature 24 h before the start of experiment.

Exposure of various stages to elevated temperature

Ten numbers of eggs (24 h old) were separated from the food material using fine camel hair brush under Binocular Microscope and were carefully placed in to Petri Plates (9 × 3 cm) with 1 gr of fine coriander powder and properly covered with lid Petri Plates were carefully placed in incubators. The same procedure was followed for other life stages viz., grubs (early, middle and late instars), pupae (24 h old) and adults (24 h old) of *L. serricorne*.

Three replicates of each treatment was maintained for each level of temperature with an equal number of untreated controls. The insects in Petri Plates were exposed to

the following temperatures for 24 h and temperatures in the incubators were monitored regularly.

Assessment of mortality (LTemp99)

After 24 h exposure, the observations on the mortality of the insects were recorded. Per cent mortality was calculated using the following formula.

$$\text{Per cent mortality} = \frac{\text{No. of insects dead}}{\text{Total no. of insects}} \times 100$$

Determination of LT50 and LT99 at LTemp99

Various life stages (egg, grub, pupa and adult) of *L. serricorne* at their respective LTemp99, were exposed to different time intervals, and the mortality data was recorded. The mortality data was subjected to probit analysis (Finney, 1971) and LT50 and LT99 values (in hours) were calculated.

The exposure period for egg, early instars and adult were as follows

Results and Discussion

Mortality response of eggs at various constant temperatures

When the eggs (24 h old) were exposed to the various constant temperatures viz., 35°C, 25°C and 15°C for 24 h there was no effect of these temperatures on the mortality of eggs and they hatched normally. However when the eggs were exposed to temperatures of -5°C, 0°C and 5°C for 24 h there was 100 per cent mortality of eggs and no hatching took place at these temperatures. From the results, we have taken temperature of 5°C as lethal temperature that causes 100 per cent mortality of eggs.

LT50 and LT99 (in hours)

The LT50 and LT99 values for eggs of *L. serricorne* at 5°C (LTemp99) were 12.43 and 28.54 h, respectively (Table 2).

The results are in conformity with previous works of Toshihiro and Haruyasu (2006) who concluded that eggs were most susceptible to low temperatures of lower than -5°C. At 20°C, most eggs (80 per cent) normally hatched within 4 weeks, but all eggs died within 6 weeks at temperatures less than 18°C. This fact indicates that the post embryonic development in eggs can be blocked at temperatures less than 18°C.

Mortality response of early instar grubs at various constant temperatures

Early instar grubs (5 days old) of cigarette beetle that were placed in Petri Plates were exposed to various constant temperatures viz., 35^0C, 25^0C, 15^0C, 5^0C, 0^0C and -5^0C for 24h There was no survival of grubs, when they were exposed to low temperature *i.e.*, -5°C, 0°C and 5°C and died due to cooling effect. When early instar grubs exposed to temperatures more than 5°C *i.e.*, 15°C, 25°C and 35°C there were 100 per cent survival and grubs continued to feed and developed normally.

LT50 and LT99 (in hours)

The LT50 and LT99 values for early instars of *L. serricorne* at 5°C (LTemp99) were 12.09 and 29.41 h, respectively (Table 3).

Mortality response of middle instar grubs at various constant temperatures

After 24 h of exposure at 35°C and 25°C the grubs were active and fed actively. However, at 15°C temperature, immediately after 24 h exposure the grubs were sluggish

and fed slowly with no mortality. Similarly, when grubs were exposed at 5°C and 0°C temperature the grubs stopped feeding and became moribund. However, when these moribund grubs were kept at room temperature they returned to normal stage and continue to feed on food. When middle instar grubs exposed at -5°C for 24 h, there was 26.66 per cent mortaliy.

LT50 and LT99 (in hours)

The LT50 and LT99 values for middle instars of *L. serricorne* at -5°C were 45.78 and 102.54 h, respectively (Table 4).

Mortality response of late instar grubs at various constant temperatures

Grubs when exposed to temperature 35°C and 25°C showed normal development. At 15°C exposure for 24 h, the grubs were sluggish and fed slowly compared to control, with no mortality. When the grubs were exposed to low temperatures viz., 5°C and 0°C temperature the activity of exposed grubs was stopped.

However, once removed from these temperatures to normal temperature the grubs become active and continued to feed and developed normally. When the grubs were exposed to sub-zero temperature *i.e.,* -5, there was 23.333 per cent mortality

LT50 and LT99 (in hours)

The LT50 and LT99 for late instars of *L. serricorne* at -5°C were 66.22 and 121.54 h, respectively (Table 5).

These results were in conformity with Childs *et al.,* (1970) who found that at 4.4ºC the third and fourth instars of *L. serricorne* died in three weeks; at 7.2ºC third instars died in three weeks and fourth in stars in 5 weeks. At 10ºC, 60 per cent of third instars and 20 per cent of fourth instars died in 11 weeks.

Howe (1957) reported that larvae of *L. serricorne* were very resistant to low temperatures. At a low temperature (below 17ºC), the development of *L. serricorne* was not completed. Loschiavo (1960) explained that a six day exposure at –1, 10 and 20°C to mature larvae of *T. parabile* did not kill them.

Toshihiro and Haruyasu (2006) reported LT99 values of acclimated larvae of *L. serricorne* as 7.2 h at -15°C, 23.7 h at -10°C, 376 h at -5°C, 1,140 h at 0°C and 1,880 h at 5°C. A period of 47 and 92 h were required to get 50 and 100 per cent mortality of middle larva of *L. serricorne*, respectively when exposed to -5°C.

The acclimated larvae required 11 week for eradication at 5°C, whereas other stages were eradicated within 5 week.

Runner (1919) stated that the larvae of *L. serricorne* become dormant and do not damage at low temperature but can survive long enough to pass the winter.

Mortality responses of pupa at various constant temperatures

Temperature of 35°C, 25°C, 15°C, 5°C and 0°C didn't had any effect on the survival of pupae of *L. serricorne*. Adults emerged from these exposed pupae normally. However when pupae were exposed to -5°C, there was 6.66 per cent mortality of pupa.

LT50 and LT99 (in hours)

The LT50 and LT99 for pupa of *L. serricorne* at -5°C were 85.23 and 152.60 h, respectively (Table 6).

Exposure of various stages to elevated temperature

S. No.	Treatment	Temperatures
1	T1	35°C
2	T2	25°C
3	T3	15°C
4	T4	5°C
5	T5	0°C
6	T5	-5°C
7	T7	Control

Determination of LT50 and LT99 at LTemp99

S. No.	Treatment	Exposure Periods (Hours)
1	T1	4
2	T2	8
3	T3	12
4	T4	16
5	T5	20
6	T6	24

The results were in conformity with Adler (2002) who exposed unacclimated cocoons and acclimated cocoons of *L. serricorne* to -15°C and -20°C in insulated boxes.

There was no adult emergence from unacclimated cocoons following exposure to the respective temperatures for 2 h and 1 h. With acclimated cocoons there was no adult emergence after 2 h at -15°C and 1 h at -20°C, but at -10°C, there was adult emergence after 8, 12 and 24 h exposures.

Collins and Conyers (2010) reported that there was no adult emergence from unacclimated cocoons of *L. serricorne* to -10°C, -15°C and -20°C following exposure to the respective temperatures for 4, 2 and 1 h. With acclimated cocoons there was no adult emergence after 2 h at -15°C and 1 h at -20°C, but at -10°C, there was adult emergence after 8, 12 and 24 h exposures. Even at field-scale experiments there was no adult emergence, when exposed to at least temperature of -18°C for periods ranging between 3.75 h and 39.25 h or when exposed to -25°C for a period between 2.4 h and 3.7 h.

Mortality responses of adults at various constant temperatures

When the adult cigarette beetle exposed to temperature 35°C and 25°C, they were normal and active. At other temperatures *i.e.*, 0°C, 5°C and 15°C, there were no mortality of exposed adults. However, when the adults were exposed at -5°C, there was 100 per cent mortality and hence -5°C was considered as LTemp99.

LT50 and LT99 (in hours)

The LT50 and LT99 for adult *L. serricorne* at -5°C as lethal temperature (LTemp50) were 13.56 and 32.47 h, respectively (Table 7).

The results obtained with mortality response of different life stages of cigarette beetle, *L.*

serricorne exposed to various temperature levels indicated that all life stages viz., egg, early instars, middle instars, late instars, pupa and adult were susceptible to low temperature below -5°C. However, among all stages tested, eggs and early instars were more susceptible to temperatures at0°C and 5°C. However, middle instars, late instars, pupa and adult were not susceptible to these temperature. At 15ºC, 25ºC and 35ºC temperatures there was no of temperature on different life stages of *L. serricorne*. At LTemp99 (5°C) of eggs stage of *L. Serricorne*, the LT50 and LT99 was 12.43 h and 28.54 h, respectively. At LTemp99 (5°C) of early instar grubs of *L. Serricorne*, the LT50 and LT99 was 12.09 h and 29.41 h, respectively.

At LTemp 99 (-5^{0}C) of middle instars of *L. serricorne*, the LT50 and LT99 was 45.78 and 102.54 h, respectively. At LTemp 99 (-5^{0}C) of Late instars of *L. serricorne*, the LT50 and LT99 was 66.22 and 121.54 h, respectively. At LTemp 99 (-5^{0}C) of pupae of *L. serricorne*, the LT50 and LT99 was 85.23 and 152.60 h, respectively. At LTemp 99 (-5^{0}C) of middle instars of *L. serricorne*, the LT50 and LT99 was 13.56 and 32.47 h, respectively.

The results of present investigations indicate the tolerance of various life stages of *L. serricorne* as; pupae > late instars > middle instars > adult > early instars > egg (most tolerant to least tolerant) to low temperatures.

The results are in agreement with Toshihiro and Haruyasu (2006) who reported that acclimated larva and cocoons were resistant among all other life stages of *L. serricorne*.

Howe (1957) reported that development and hatching of eggs to occur between 20 and 34ºC. The optimum temperature for rapid larval development is 32.5ºC and development slows down at 20ºC. Development of pupae takes the shortest time at 32.5–35.0ºC. Development of *L. serricorne* cannot be completed at 17.5°C or 40°C. The temperatures below and above to optimum the growth and development of insect get seized. Minimum conditions of −18°C for 24 h and -25°C for 4 h are recommended for control of *L. serricorne.*

According to Strang (1992), cooling to just above 0°C is also known to have a fatal effect on some insect species (e.g.,, Cigarette Beetle, *L. serricorne*), provided the exposure to cold lasts many days. Eliopoulos *et al.,* (2011*)* reported at temperature -16°C larvae and adults of grain beetles, *T. confusum*, *O. surinamensis* and *T. granarium* values of LT50 ranged between 0.7 and 1.0 h for *T. confusum*, 0.7–1.9 h for *O. surinamensis* and 1.2–3.4 h for *T. granarium.*

Collins *et al.,* (2010) reported that the diapausing larvae are more cold tolerant than the eggs of *E. elutella*, with days of exposure required for complete mortality, compared to hours. Complete mortality of eggs was achieved after 1 and 7 h at -15oC and -10oC, respectively, whereas complete mortality of diapausing larvae was achieved after 1, 3 and 22 days at -20oC, -15oC and -10oC, respectively.

Becket *et al.,* (2007) reported three temperature zones as significant for growth or death of stored-product insects. At optimal temperatures (25 to 32°C), insects have maximum rate of increase. At suboptimal temperatures (13 to 24°C and 33 to 35°C), development slows and at lethal temperatures (below 13°C and above 36°C), insects stop feeding, develop slower and eventually die. The more extreme the temperature, the more quickly they die. The

insect freezing point varies by species, stage and physiological state between -4.0 and -22.0°C.

According to Fields, (1992) temperatures above 35°C lethal to insects where the insects die due to maximum rate of population increase, 25 to 35°C is optimum temperature for growth and development of insects wherein maximum reproduction and maximum rate of population increase takes place, below 25 to 5°C there is slow increase in population, slowly lethal and development of insect stops, 5 to 0°C the activity of movement gets ceases at sub-zero temperatures the insects die in hours, weeks and months if acclimated. One factor that is often neglected when considering insect survival at low temperatures is the ability of insects to acclimate to cold.

At temperatures between 20 and 0°C insects increase their tolerance to low temperature. There are a number of physiological changes, such as, higher concentrations of cryoprotectants, clearing of ice nucleators, changes in the cell membrane, within the insect responsible for this increase in cold hardiness (Burks *et al.,* 2000).

At temperatures below the development threshold, insect will eventually die. The length of time this takes depends on many factor; temperature, insect species, life stages, moisture content of the grain and acclimation to cold. At temperatures slightly above those that promote the faster rate of development, the fitness of insects rapidly fall with increasing temperature (Birch, 1953).

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