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Observations on the termination of the facultative diapause in the Codling Moth (*Cydia pomonella* L., Lepidoptera: Tortricidae).

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INTRODUCTION

Effective Temperature Sums (ETS) are often calculated to predict biological events in insects; e.g. the duration of development, that of different developmental stages, the emergence of the first and of the 50% adults, etc. In spite of this generally accepted method, it has already been shown that different populations of the same species can have different ETS requirements (Saunders, 1977), and that the ETS of development can be influenced by food quality (Dittrick & Chiang, 1981) or changed by the length of the photoperiod (Deseö, 1973b; Hayes & Cawley, 1978). We are going to demonstrate that, in addition to the cumulative effect of a certain ETS necessary in spring until adult emergence, higher temperatures can trigger the resumption of active development (in this case, in the Codling Moth (*Cydia pomonella* L., Lep.: Tortricidae), referred to as « CM » in the following text). It is known that the inhibiting role of the short photoperiod in the diapausing CM larvae is nearly abolished in the first three months of diapause and so resumption of active development in spring in the orchard is influenced mostly, if not only, by temperature (Russ, 1966; Jermy, 1967; Wildbolz & Riggenbach, 1969). Therefore, to predict the beginning or the 50% of spring emergence of the adults, the effective temperatures are usually summed up. The sum (expressed in DD, effective Degree-Days required for the beginning of flight) vary from 34 to 450 DD respectively (Labanowski, 1981), varying also in the same area, as a consequence of microclimatical differences (Jermy, 1964; Sàringer, 1971, 1977).

These spring ETS for CM are always calculated until eclosion: i.e. this period includes diapause termination, resumption of active development, pupation and the temperature-dependent period of pupal development, and finally the emergence of the adults.

MATERIALS AND METHODS

CM larvae were collected in an apple orchard near Ravenna in the autumns (Sept.-Nov.) of 1979 and 1980 by winding corrugated paper bands around the trunks. The larvae were held until the following spring at 4-7°C in darkness. For the observations on the effect of temperature on diapause termination/eclosion, about 300 CM larvae were exposed to different temperatures the following springs: on March 22, 1980 and March 3, 1981. The temperatures used were: 9.5°C, 11.1°C, 14.2°C, 17.4°C, 20.5°C, 23.6°C, 27.0°C, 30.2°C, (±1°C); with a long photoperiod of 17/7:L/D and 60% relative humidity. The numbers of adults emerged were recorded every 48 hours. After 100 days, the experiment was finished by checking the number of non-developed individuals at the different temperatures. Larvae found were exposed to 26°C. Emergence within 40 days, mortality and causes of death of these larvae and pupae were recorded.

A « laboratory » population was established with larvae collected in 1980. A slightly modified artificial diet of Huber et al. (1972), and Sender (1970), (Deseö et al., 1981), was used at 26°C and 17/:L/D. Experiments were carried out with sixth generation larvae to check the effect of different diapause inducing conditions on adult emergence in spring: i.e. CM were reared from egg stage to cocoon spinning at 20°C and 26°C, under different photoperiods (14/10, 15/9, and 16/8: L/D). For each regime, 250-300 specimens were used. Diapausing larvae were held for 18 weeks at 4-7°C in darkness, then transferred to 17/7:L/D at 26°C, with 60-80% relative humidity. Emergence was reported every 24 hours. In all experiments, the reproductive behaviour of the adults was considered the parameter of a physiologically complete diapause.

Towards this aim, individual pairs were held in small plastic jars and fed honey-water soaked cotton. Afterwards, the females were dissected for the presence of spermatophores and the eggs laid were counted (Deseö, 1971).

SOME CHARACTERISTICS OF THE POPULATION

The CM has two entire and a partial third generation yearly in this area (Briolini et al., 1972).

The diapause-inducing critical photoperiod at 26°C is 15 hours 40 minutes long. These data correspond with the earlier findings of Ivancich-Gambaro in Italy (1958). This means that in the Ravenna-Bologna area, the first diapausing larvae can be found in the first week of August and diapause occurs in the whole population after the second half of the month.

Twenty percent of the first generation diapauses and thus has only one generation per year; this agrees with the observation of southern CM populations described by Shel'deshova (1965). This genetic characteristic was found even in the 7th laboratory generation after diapause. However, in outdoor conditions, only 90% of these diapausing larvae had a one-year-long diapause; the other 10% emerged sporadically in the second half of the same summer.

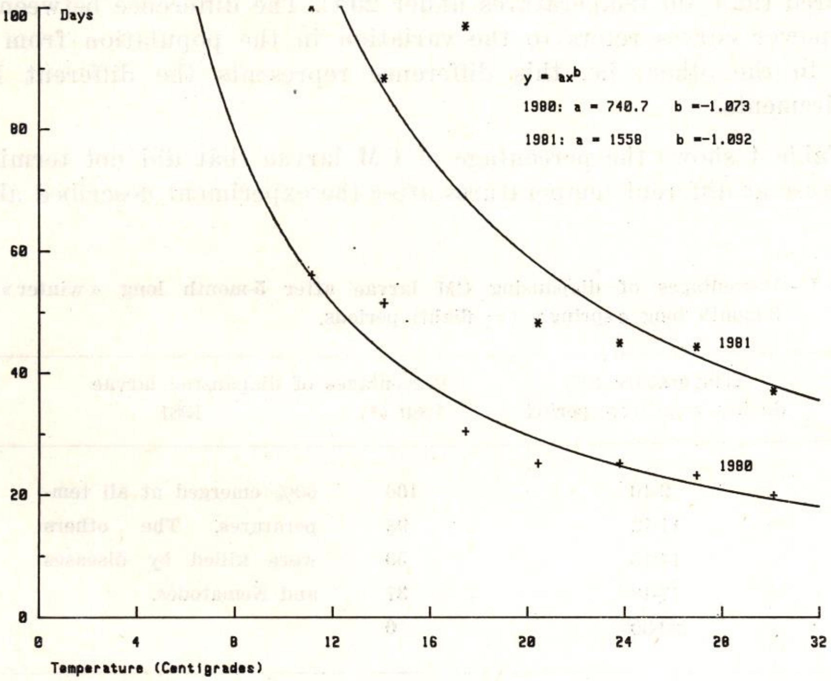

FIG. I

Length (Days) of the diapause termination period until 50% emergence in *C. pomonella* orchard populations (at different temperatures) under 17/7:L/D photoperiod. (Curves: $y = ax^b$. 1980: a = 740.7; b = −1.073; 1981: a = 1557.8; b = −1.092).

The mean fecundity of the orchard and laboratory reared moths is about 90 eggs/female.

Diseases found in 10% of the orchard population in 1980 were Granulosis Virus and Microsporidia. In 1981, the diseased larvae were 40%. Besides the above mentioned pathogens, the fungus *Beauveria bassiana* Balsam. (Vuill.) and an entomophagous Nematode (*Steinernema feltiae* Filip.) were found. In the laboratory-reared population, the diseased larvae were about 2%.

RESULTS

1. Effect of different spring temperatures on the adult emergence in orchard population.

A) Figure 1 shows that the number of days required for adult emergence is influenced by temperature. The correlation can best be expressed by power curves. These curves show that an increase in temperature above 20°C caused smaller differences in the number of days of development required than did temperatures under 20°C. The difference between the two power curves refers to the variation in the population from one year to the other; i.e. this difference represents the different ETS requirements.

B) Table 1 shows the percentage of CM larvae that did not terminate diapause at different temperatures after the experiment described above.

TABLE I - Percentages of diapausing CM larvae after 5-month long « winter » and 3-month long « spring » (= flight) periods.

Temperature (°C) during « spring » period	Percentages of diapausing larvae 1980 (*)	1981
9-10	100	60% emerged at all temperatures. The others were killed by diseases and Nematodes.
11-12	93	
14-15	50	
17-18	37	
20-30	0	

(*) Nearly all these larvae developed into adults in 17-38 days when exposed to 26°C.

Data in Table 1 show that part of a healthy CM population resumed active development under 20°C; the lower the temperature, the larger the number of inactive larvae. These diapausing larvae, however, could be « triggered » by a higher temperature (26°C) to resume activity.

2. Adult emergence after inducing diapause with various photoperiods.

Figures 2 and 3 show that the date and pattern of emergence of adults after diapause under constant temperature (26°C) and photoperiod (17/7:L/D) is influenced by the photoperiod and perhaps also by the temperature during prediapause development. The first adult eclosion took 13-15 days. 50% of tre group exposed to longer photophases during prediapause (15/19:L/D) emerged 4 days earlier than the group held at 14/10:L/D. That is, the diapause lasted longer when the prediapause photoperiod was shorter. Short photoperiods lengthened the larval life before diapause. At 26°C, prediapause larval life was 1 day longer with 15/9:L/D than with 16/8:L/D and it was lengthened

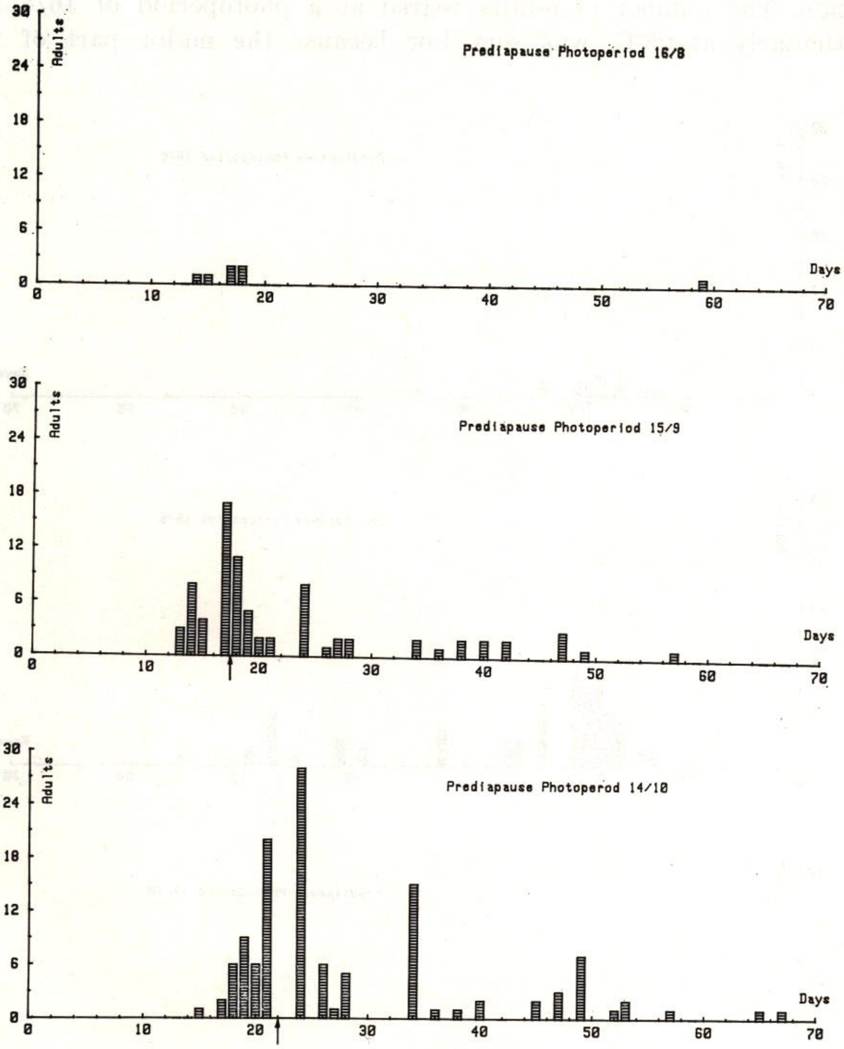

Fig. II

Emergence of *C. pomonella* « laboratory population » after an 18 weeks long diapause. (Emergence conditions: 26°C and 17/7:L/D photoperiod). The diapause was induced at 26°C but by different long photoperiods: 16/8:L/D = 15/9:L/D = 14/10:L/D. Arrows show the 50% emergence of the groups with different prediapause photoperiods. No arrow given for 16/8:L/D because a very small percentage went into diapause at this photoperiod and 26°C.

by 3 days with 14/10 :L/D. At 20°C, the differences were 3 days and 5 days respectively.

Figures 2 and 3 also demonstrate the long-lasting effect of the prediapause temperature. Groups entering diapause at 20°C showed, after diapause, smaller differences in 50% emergence (4 days) than those reared at 26°C (6 days). However, this observation needs confirmation. The number of adults reared at a photoperiod of 16/8 :L/D, particularly at 26°C, was very low because the major part of these

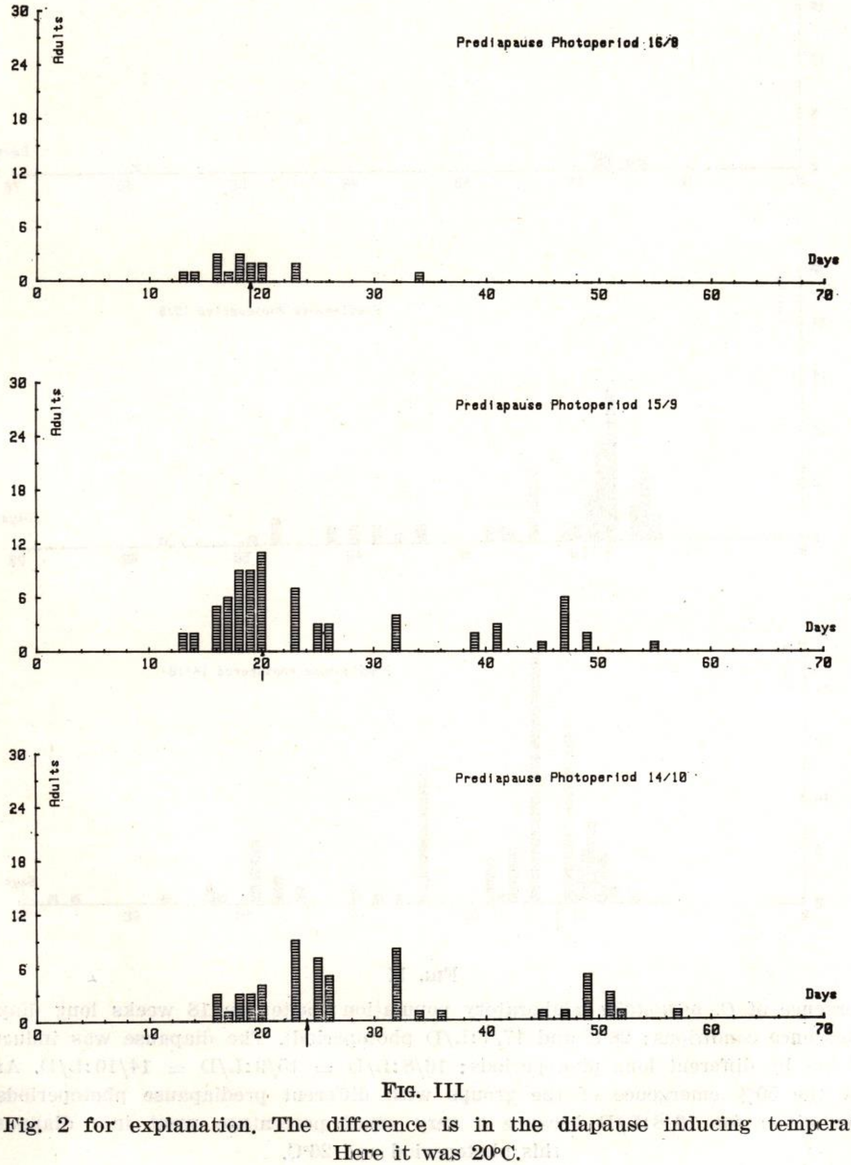

FIG. III

See Fig. 2 for explanation. The difference is in the diapause inducing temperature. Here it was 20°C.

groups did not enter diapause. Interestingly, all emergence curves had 2 peaks, except the case of a longer diapause observed in the laboratory (longer than 9 months), when they had only a single peak.

3. Effects of prediapause photoperiods and temperatures on the reproductive activity of the adults.

The fecudity of the spring (orchard) population and that of the continuous laboratory rearings were about 90 eggs/female. Fig. 4 shows

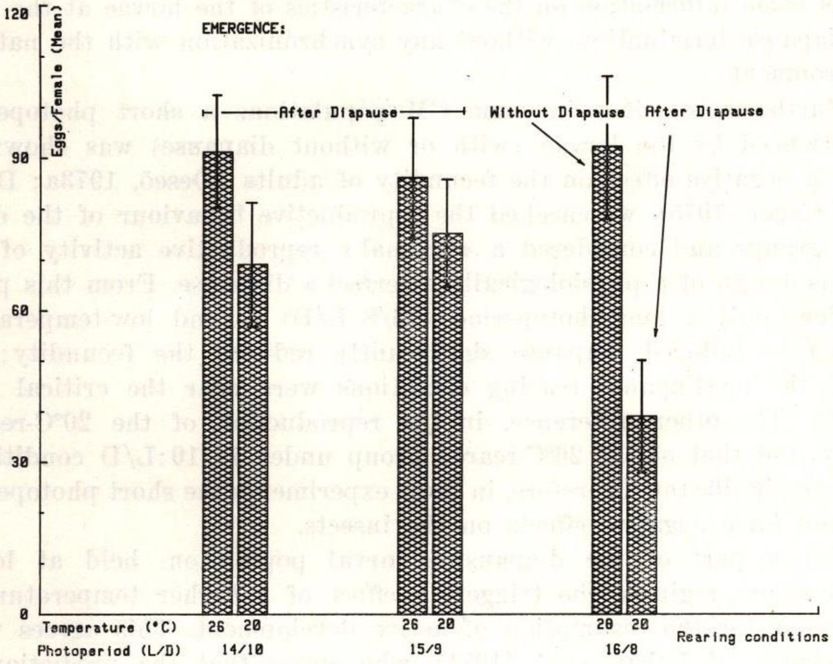

FIG. IV

Mean number and standard error of eggs per female of *C. pomonella* reared under different temperature and photoperiod conditions before diapause. In case of 16/8:L/D nearly all larvae pupated without diapause.

that this was also the mean fecundity of most of the above mentioned experimental groups. Although there was an evident difference between the populations which had been reared at different temperatures (20°C and 26°C) during prediapause at 14/10:L/D, this difference is not significant. A significant difference in the fecudities was found only between moths which emerged without and those which emerged after diapause from groups reared under 16/8:L/D photoperiodic conditions at 20°C. Low mean fecundities were caused by females that did not mate or by mated females which did not lay eggs.

DISCUSSION

Many authors have studied the factors that influence the period from diapause termination to adult emergence in the CM. Those experiments were carried out, in large part, with orchard populations diapausing in outdoor conditions. Our observations were made with orchard populations as well, but diapause development occurred at a constant temperature, in darkness, in two successive years.

Thus, the effect of the natural winter conditions, (i.e. the varying temperatures and photoperiods) was eliminated. In this way we hoped to get some information on the characteristics of the larvae at the time of diapause termination, without any synchronization with the natural environment.

Furthermore, since in some CM populations a short photoperiod experienced by the larvae (with or without diapause) was shown to have a negative effect on the fecundity of adults (Deseö, 1973a; Deseö & Sàringer, 1975), we checked the reproductive behaviour of the different groups and considered a « normal » reproductive activity of the CM as a sign of a physiologically « perfect » diapause. From this point of view, only a long-photoperiod (16/8 :L/D) — and low-temperature (20°C) — induced diapause significantly reduced the fecundity; i.e. when the prediapause rearing conditions were near the critical daylength. The other difference, in the reproduction of the 20°C-reared group and that of the 26°C-reared group under 14/10 :L/D conditions, was not significant. Therefore, in these experiments the short photoperiod did not have negative effects on the insects.

For a part of the diapausing larval population, held at lower temperature regimes, the triggering effect of a higher temperature is necessary for the resumption of active development. This agrees with the results of Labanowski (1981), who states that the prediction of 50% emergence is possible only if calculated after some warm spring days. The influence of temperature on development is more evident under about 20°C than it is above it. At higher temperatures regimes, development seems to be somewhat « independent » of the temperature. It is known that pupal development is a temperature-dependent process (Glenn, 1922; Geier, 1963; Russ, 1966). So, evidently diapause termination must, to a certain extent, have « self-running » characteristics. perhaps up to pupation.

The most important characteristic of this latter period is that it can be manipulated by prediapause abiotic factors. The prediapause rearing temperature was found to influence the length of the diapause termination, including adult emergence (Sieber & Ben, 1980). On the other and in our experiments the prediapause photoperiod was shown to be decisive in the postdiapause processes. A shorter photoperiod during diapause induction prolonged the time including diapause development and the interval from diapause termination to eclosion. In other words, the depth of diapause differs between these groups: « The lower the metabolic rate, the greater the duration of the diapause, the greater its intensity » (Danilewski, 1965).

Since the continuation of important developmental processes during diapause was proven (Chippendale & Yin, 1973; Beck, 1980; de Wilde & Lefèvre, 1982), it cannot be excluded that even during diapause these low metabolic processes happen in the photophase, bound perhaps to a free-running circadian rhythm (i.e., the shorter the photophase the longer the diapause). When certain physiological processes are complete or when the circadian time measurement expires, larvae begin to resume postdiapause activity, helped or triggered in nature by temperature.

In any case, the effective temperature sums, necessary for 50% emergence of the adults after diapause, are different in the groups of larvae with different prediapause photoperiod and temperature experiences.

The interpretation of these data becomes even more complex when the heterogeneity (Russ, 1966; Jermy, 1967) of the population (which leads to differences of up to 60 days in the pupation dates and usually to two peaks in flight) is taken into consideration. However, these differences are also the results of the differences in the physiological condition at the time of diapause termination. In case of a long-lasting diapause, the adults emerge nearly instantly and in a considerably shorter period (Wildbolz & Riggenbach, 1969; Sàringer, 1977). Thus it is highly probable that in spring the CM larvae, being in various physiological levels of the reactivation processes, have different sensitivities to temperature. This diversity may be abolished with time, during a long diapause, resulting in one peak during swarming.

To conclude, it can be stated that temperature has a dual effect on the resumption of active development of the postdiapause CM larvae:

1. it works by accumulating the effective degree-days, but different temperatures have different promoting effects on the speed of development;
2. high temperature can act as triggering factor for the activity resumption.

In any case, the activity resumption in spring calls for further studies, as has already been argued by Tauber and Tauber (1976).

SUMMARY

A Codling Moth (CM) population from an Italian orchard near Ravenna was held during diapause at a constant temperature in darkness. Diapause termination and adult eclosion were observed at 8 different temperatures under long-day conditions in two successive years. Results showed that temperature has a dual effect on the resumption of activity after diapause: 1. It works, as is known, by accumulating the effective degree-days (however, the temperatures below and above approximately 20°C differ on promoting development). 2. Higher temperatures can trigger the activity resumption.

In the same, but continuously reared CM population, diapause was induced by different long photoperiods and temperatures. The shorter the photoperiod was, the longer the prediapause development and diapause termination periods were. So, the depth of the diapause could be manipulated in this way. For these processes a free-running circadian system is supposed. It is also evident that the Effective Temperature Sums required for the 50% emergence of adults after diapause, differ according to the length of the photoperiods experienced during the prediapause development.

Furthermore, the reproductive activity of the differently reared CM lots was always checked as a parameter for a physiologically healthy diapause. Significant difference in the mean fecundity was found only in those CM which developed around the critical day-length at 20°C and emerged without or after diapause. The fecundity of the latter group was very low.

The characteristics of this Italian population are also described.

Osservazioni sull'uscita dalla diapausa facoltativa in *Cydia pomonella* L. (Lepidoptera: Tortricidae).

RIASSUNTO

Una popolazione di *Cydia pomonella* proveniente da un frutteto nei pressi di Ravenna è stata mantenuta, durante la diapausa, a temperatura costante e all'oscurità. Per due anni si sono osservati la fine della diapausa e lo sfarfallamento degli adulti, ad 8 diverse temperature e con fotoperiodo a giorno lungo.

I risultati mostrano che la temperatura ha un duplice effetto sulla ripresa dell'attività dopo la diapausa: 1. Essa agisce, come noto, per accumulo dei gradi-giorno efficaci (ma le temperature al di sopra e quelle al di sotto di circa 20°C hanno effetti diversi sull'avanzamento dello sviluppo). 2. Temperature più alte possono dare il via alla ripresa dell'attività.

Nella stessa popolazione di *C. pomonella*, in allevamento continuo, si è indotta la diapausa in diverse condizioni di fotoperiodo e di temperatura. Quanto più breve era il fotoperiodo, tanto più lunghi erano il periodo di sviluppo pre-diapausa e quello di completamento della diapausa. In tal modo, è possibile influire sull'intensità della diapausa. Si ipotizza che tali processi siano regolati da un sistema autonomo di orologio fisiologico circadiurno. Si conclude inoltre che la somma termica efficace, richiesta per lo sfarfallamento del 50% degli adulti dopo la diapausa, differisce secondo la lunghezza del fotoperiodo a cui sono state sottoposte le larve durante lo sviluppo pre-diapausa.

Come indice di una diapausa fisiologicamente normale, per i gruppi di larve allevati in differenti condizioni, si è sempre considerata la loro attività riproduttiva. Si sono notate differenze significative nelle fecondità medie, solo nei gruppi sottoposti a temperature di 20 gradi e a fotoperiodi vicini a quello critico, sia che essi fossero sfarfallati senza diapausa, sia che avessero avuta una diapausa. In particolare, la fecondità di questi ultimi è stata molto bassa.

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