Journal of Insect Physiology 103 (2017) 78–85

Contents lists available at ScienceDirect

Journal of Insect Physiology

journal homepage: www.elsevier.com/locate/jinsphys

Effects of temperature, soil moisture and photoperiod on diapause termination and post-diapause development of the wheat blossom midge, *Sitodiplosis mosellana* (Géhin) (Diptera: Cecidomyiidae)

Weining Cheng[a,*], Zhiren Long[a], Yudong Zhang[a], Tingting Liang[a], Keyan Zhu-Salzman[b,*]

[a] *Key Laboratory of Integrated Pest Management on Crops in Northwestern Loess Plateau, Ministry of Agriculture, Northwest A & F University, Yangling, Shaanxi 712100, China*

[b] *Department of Entomology, Texas A & M University, College Station, TX 77843, USA*

ARTICLE INFO

Keywords:
Sitodiplosis mosellana
Diapause termination
Post-diapause development
Temperature
Soil moisture
Photoperiod

ABSTRACT

Sitodiplosis mosellana, one of the most important wheat pests, goes through larval diapause in a cocooned form. It is univoltine, but some individuals exhibit prolonged diapause. In this study, we documented diapause termination rate of cocooned larvae at different diapausing periods and time required for adult emergence when they were brought to 25 °C from the field in northern China. We found that field larvae all entered diapause by June, but none terminated diapause until late September when the daily average temperature dropped to below 20 °C. Furthermore, termination rate increased significantly as diapausing larvae underwent increasing chilling duration, reaching > 95% from early December to early March. Our results suggest that chilling was necessary for diapause to terminate and that field diapause termination ended in early December. To explore low temperature and duration required, we cold-treated field diapausing larvae of different periods at different low temperatures for various lengths of time prior to the 25 °C incubation. Chilling at 4–8 °C for 60–90 days resulted in a higher termination rate (> 90%) and shorter adult emergence time in general. Additionally, we investigated the combined effect of temperature (18–30 °C) and soil moisture (20–60%, on dry weight basis) as well as the effect of photoperiod (24:0–0:24 L:D) on post-diapause development. While photoperiod did not affect adult emergence, soil moisture ranging from 30 to 50% and temperature from 22 to 26 °C resulted in the highest adult emergence rates (> 46%) within relatively short time (< 18 days). Mortality and/or prolonged diapause rate drastically increased when incubation conditions were outside the optimal range, especially at 30 °C. These findings provide new insight into the diapause process of *S. mosellana*, and information will be useful for development of field forecasting and laboratory rearing techniques of this pest insect.

1. Introduction

Diapause is a seasonally programmed state of developmental arrest, which enables insects and related arthropods to synchronize their life cycles with the seasonality of their environment (Denlinger, 2002). Once diapause begins, it requires a certain time period for insects to reactivate development, and the length of this period depends on the conditions insects experience during diapause (Danks, 1987). Diapause typically contains several successive physiological phases, i.e. diapause initiation, maintenance, termination, and sometimes post-diapause quiescence (Košťál, 2006). This process is followed by post-diapause development. Every stage is strongly affected by one or more environmental factors. For example, chilling is considered the most common factor promoting termination of winter diapause (Denlinger and Armbruster, 2014; Dong et al., 2013). In some species, photoperiod can also affect diapause termination (Bradshaw and Lounibos, 1972; Eizaguirre et al., 2008). Temperature, photoperiod and soil moisture also directly impact initiation of post-diapause development in many species (Denlinger, 2008; McDonald et al., 2015; Tang et al., 2012; Zhuo et al., 2011). A better understanding of the impact of these factors is pivotal for facilitating laboratory insect rearing, and also pest population forecasting in the field.

The orange wheat blossom midge, *Sitodiplosis mosellana* (Géhin) (Diptera: Cecidomyiidae), is one of the most destructive wheat pests in Asia, Europe, and North America (Chavalle et al., 2015; Gaafar and Volkmar, 2010; Jacquemin et al., 2014; Miao et al., 2013). This species is univoltine and has evolved a 10-month diapause period which aligns its development with the phenology of the wheat. In most northern

* Corresponding authors.
E-mail addresses: chengwn@nwsuaf.edu.cn (W. Cheng), ksalzman@tamu.edu (K. Zhu-Salzman).

http://dx.doi.org/10.1016/j.jinsphys.2017.10.001
Received 1 July 2017; Received in revised form 26 September 2017; Accepted 1 October 2017
Available online 04 October 2017

China areas, adults emerge and oviposit on wheat ears in late April and early May (Wang et al., 2015). Hatched larvae feed on developing kernels. Around late May to early June, mature 3rd instar larvae fall onto the soil surface from wheat ears, burrow into the soil and spend summer, autumn and winter in round diapausing cocoons they form. Not until mid-March of the following year, is post-diapause development activated in response to rising temperatures, indicated by larvae exiting cocoons, pupating on the soil surface and emerging as adults. A portion of overwintering larvae, however, do not exit cocoons to pupate and emerge. Instead, they remain in prolonged diapause for more than one year, especially in dry soil (Doane and Olfert, 2008).

Studies on *S. mosellana* have implied that low temperature plays a role in diapause termination and that temperature and soil moisture influence post-diapause development (Basedow, 1977; Elliott et al., 2009; Hinks and Doane, 1988; Wise and Lamb, 2004). However, very few experiments directly demonstrate effects of these factors on diapause termination and post-diapause development, and many questions remain to be answered. For example, when does diapause termination in nature begin and end in northern China? What is the optimal low temperature and minimal exposure time for diapause to terminate? Will these parameters vary dependent on different diapausing periods of the insect? Furthermore, it would be interesting to know the optimal ranges of temperature and soil moisture that trigger post-diapause development. Finally, since *S. mosellana* pupates and emerges on soil surface, effects of photoperiod should be explored.

In the present study, we monitored the course of diapause in *S. mosellana* under field conditions in northern China from 2013 to 2014 in order to provide seasonal information on time and temperature at which diapause termination begins and ends for this insect species. Furthermore, we determined the optimal conditions for diapause termination by manipulating low temperature and exposure time using larvae at different diapausing periods. In addition, we investigated effects of different temperature and soil moisture combinations as well as the effect of photoperiod on post-diapause development. Information derived from this research will certainly contribute to development of pest population prediction and laboratory rearing techniques for *S. mosellana*.

2. Materials and methods

2.1. Source and handling of experimental insects

To obtain experimental insects, a field insectary was built in Yangling (34°16′N, 108°4′E), Shaanxi Province, China. Wheat ears containing mature *S. mosellana* larvae were harvested in late May 2013 and placed on soil in the field insectary. The soil was sprayed with water to maintain moisture, which facilitated larvae to crawl out of glumes and burrow into soil. Successful diapause initiation is indicated by formation of larval cocoons. Cocooned larvae collected from this field insectary during late June 2013 to late May 2014 were used for the study. A portion of the samples was subjected to different time periods of chilling treatment prior to further processing (Sections 2.3 and 2.4) whereas others were not, representing natural field conditions (Sections 2.2,2.5 and 2.6). The experimental design is shown in Fig. 1.

Collected cocooned larvae were placed approximately 3 cm below the soil surface in plastic pots (12 cm in diameter), 100 larvae per pot. The soil substrate was mixture of midge-free field soil from local farmland and small amounts of peaty soil (Pindstrup Mosebrug A/S, Denmark). Unless otherwise stated (Section 2.5), soil substrate was adequately moist (around 40% water content on dry weight basis). These pots were covered with clear conical plastic hoods, and transferred to controlled environmental incubators at temperatures (± 1 °C) indicated in the following sections and relative humidity 70% (± 5%). To maintain the initial soil moisture, water was periodically added. Adults that emerged from soil were counted and removed daily. Once emergence completed, i.e. no adults were detected within a week, the soil was sieved to check for larvae that were still in diapause (Wu et al., 2005).

2.2. Determination of diapause termination under field conditions

To determine the process of diapause termination of *S. mosellana* in the field with changing temperature, approximately 300 cocooned larvae were collected from the field insectary each time for 10 times from late June 2013 to early March 2014 at 20–30 day intervals. In Shaanxi Province, *S. mosellana* generally enters diapause in June, and initiates post-diapause development in mid-March of the following year. In addition, 300 cocooned larvae were also collected in late May 2014, which represented the population undergoing prolonged diapause. Collected larvae were transferred to 25 °C incubators with photoperiod of 16L:8D. Due to the small size and soil dwelling habit of this insect, adult emergence was used to serve as the indicator of diapause termination (Wise and Lamb, 2004). Additionally, viable individuals are orange whereas dead individuals turn black and quickly decay in soil. The latter groups were excluded during calculation. After adult emergence ended, the number of larvae still in diapause was counted, and mean duration required for adult emergence (from the time larvae were transferred to environmental chambers) and diapause termination percentage was calculated.

Field air temperatures (daily maximum, minimum and mean) during the trials were obtained from Yangling local meteorological station.

2.3. Effect of chilling (4 °C) duration on diapause termination

To elucidate the effect of length of cold exposure (4 °C) on diapause termination of *S. mosellana* larvae at various diapausing periods, soil samples gathered from the field insectary in July, August and September of 2013 and May of 2014, respectively, were incubated at 4 °C for 0 (control), 30, 60 or 90 days. For each diapause period × - chilling duration combination, 300 cocooned larvae were collected, and incubated at 25 °C under photoperiod 16L:8D. After adult emergence ended, the number of larvae still in diapause was counted, mean duration required for adult emergence (from the time larvae were transferred to environmental chambers after exposure to chilling) and diapause termination percentage were calculated.

2.4. Effect of different low temperature treatment on diapause termination

To compare effects of chilling at various low temperatures on diapause termination, soil samples gathered from the field in late September 2013 were incubated at 0, 4 and 8 °C respectively, with those experiencing the natural field low temperature as control. After being chilled for 30, 60 or 90 days, 300 cocooned larvae were collected from each chilling temperature × chilling duration combination and incubated at 25 °C under photoperiod 16L:8D. Mean duration required for adult emergence and diapause termination percentage were calculated as described in Section 2.3.

2.5. Effects of temperature and soil moisture on post-diapause development

To determine the optimal combination of temperature and soil moisture to trigger post-diapause development, cocooned larvae collected in January of 2014, which had terminated diapause (see Section 3), were incubated at all combinations of five different soil moisture levels (20, 30, 40, 50 and 60% water content on a dry mass basis) and four temperatures (18, 22, 26 and 30 °C). Each treatment combination was repeated three times with 100 individuals per replicate. After adult emergence ended, the number of larvae still in diapause was counted, mean time required for adult emergence, percentage of adult emergence, and percentage of prolonged diapause individuals were calculated.