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Effect of Temperature on the Estivation of
Elcysma westwoodii
 (Lepidoptera, Zygaenidae)¹

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The zygaenid moth, *Elcysma westwoodii* has a univoltine life cycle in the Kobe area where larvae start feeding on cherry flower buds in late March–early April and continue feeding on the plant until late May when they leave the feeding site for cocoon-spinning. Larvae enter estival diapause and pass the summer in a cocoon. Adults emerge from September to October and lay eggs. Newly hatched larvae spin hibernaculum in crevices of tree trunks and overwinter in the hibernaculum.

The timing of emergence was delicately adjusted by multiple modes of photoperiodic responses. Prepupae respond to both absolute day length and to shifts in daylength in unique ways (GOMI and TAKEDA, 1992). However, although photoperiodic response regulates adult emergence very precisely, changes in temperature comprise another major environmental variable and possibly cause the diapause. Yet, this effect has not been thoroughly investigated. The present paper therefore focuses on the effects of temperature changes on the regulation of diapause.

MATERIALS AND METHODS

Last instar larvae were collected on the Rokkodai

campus of Kobe University between 24 and 26 May, 1984. Larvae were reared on fresh foliage of the cherry tree until cocoon spinning under LD 18 : 6 at 20, 25 and 30°C. Corrugated filter paper was provided for the site of cocoon formation. Upon cocoon formation, some larvae were transferred to a short day condition, LD 13 : 11, and subsequent adult emergence was observed. The numbers of cocoons that were exposed to photoperiodic and temperature treatments were as follows: 71 for LD 18 : 6 at 30°C, 53 for LD 18 : 6 at 25°C, 175 for LD 13 : 11 at 25°C and 93 for LD 18 : 6 at 20°C. Out of 71 for 30°C treatment, 53 were supplementary cocoons that formed at 20°C, since the mortality at 30°C was high and only 18 cocoons were obtained.

Observations were also made in the field to clarify the life cycle and behaviors of this moth.

RESULTS AND DISCUSSION

Field observations

The feeding of overwintering larvae was synchronized with the time of flower bud bursting in spring. After feeding, mature larvae descended the host plant, or occasionally silk-hanging larvae dispersed by wind. By early June they completed cocoon formation and entered summer diapause as prepupa. Pupation started in mid-September and moth flight began in late-September and lasted until early-October. Moth flight occurred at night and mating in the morning. Moths in tandem were frequently observed before noon and many ovipositing females were observed on the trunk of host trees in the evening in early-October. Moths fed on honey-water in the laboratory. The eggs were soft and whitish yellow ellipsoid, 3 mm × 2 mm. Eggs hatched from late-October through early-November. They formed hibernaculum in crevices of the host tree.

Tachinid flies emerged from last instar larvae upon cocoon formation. The rate of parasitism was 2.7%. The wall of the cocoon was double-layered on

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Table 1. Survival rates and pre-emergence periods under different photoperiods and temperatures

	30°C	25°C		20°C
	LD 18:6	LD 18:6	LD 13:11	LD 18:6
<i>N</i>	71	53	175	93
% adults emerged	0.0	9.4	26.3	50.5
Average period till emergence (days)	—	89.8	70.5	111.1
S.D.	—	8.8	4.5	12.5

the upper side but had a single layer on the leaf side. The cocoons help pupal eclosion because when pupae were removed from the cocoon, they often failed to eclose.

The effects of temperature and photoperiod on estivation physiology

The rates of survival during the estivation and pre-emergence periods with the standard deviations are summarized in Table 1. The pre-emergence period was short under LD 13:11 at 25°C. Under LD 18:6 at 20°C, however, it was prolonged, showing a greater variance. The survival rate in this condition was the highest of all the treatments tested. No emergence was observed at 30°C.

Figure 1 illustrates the cumulative percentage of emergence after the treatments and indicates that both photoperiod and temperature effects are significant. Photoperiod, however, contributes more to the acceleration of diapause termination while temperature has a greater effect on the rate of survival.

Fig. 1. Cumulative percentage of emergence after treatment with different conditions. Unit, days; triangle, LD 13:11 at 25°C; squares, LD 18:6 at 25°C; closed circles, LD 18:6 at 20°C.

Fig. 2. Schematic representations of possible interactions between the tachytelic (broken line) and horotelic (thick line) processes in termination of summer diapause in *Pyrrhalta humeralis* (left) and *Elcysma westwoodii* (right). In *P. humeralis*, $t_1 = 30 + a$, $t_2 = 45 + a$ and $t_3 = 79 + a$, where a is the period between the initiation of diapause and the treatment. In *E. westwoodii*, $t_1 = 70.5$, $t_2 = 89.8$ and $t_3 = 111.1$.

HODEK (1983) has attempted to make general explanations for the process of diapause termination by postulating two independently proceeding processes, tachytelic and horotelic processes. The former was postulated to proceed at a standard rate while the latter proceeds at a more accelerated rate. Since we have recently published data on summer diapause regulation in a similarly estivating chrysomelid beetle, *Pyrrhalta humeralis* (NAKAI and TAKEDA, 1995), comparisons of these species in such a scheme may be appropriate for characterization of this species. We assume that the horotelic process in *P. humeralis* depends on temperature and that the tachytelic process depends on photoperiods in such a way that the former proceeds at a higher rate at lower temperatures and the latter under short days (Fig. 2, left).

Similar analysis favors the notion that the horotelic process in *E. westwoodii* is accelerated at higher temperatures (Fig. 2, right). The tachytelic process is accelerated by short photoperiods, since the emergence was greatly accelerated by a photoperiodic shift from LD 18 : 6 to LD 13 : 11.

In conclusion, the rate of termination of diapause in *E. westwoodii* depends more on photoperiodic conditions than temperature but the latter strongly affects the rate of survival. High temperature accelerated the termination of diapause but induced high mortality. The mode of dependence on temperature and photoperiod is just the opposite of what was observed in *P. humeralis*, where low temperature accelerated the termination of diapause but photoperiod affected the survival rate significantly.

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Cuticular Proteins Specific to the Last Larval Stadium in the Sweet Potato Hornworm, *Agrius convolvuli* (Lepidoptera: Sphingidae)¹

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In holometabolous insects, the larval, pupal and adult cuticles are markedly different, and each has stage specific proteins (CHIHARA et al., 1982; SRIDHARA, 1983; COX and WILLIS, 1985). Genes coding for

larval cuticular proteins of *Manduca sexta* are expressed during the feeding and growth phases and are suppressed at the initiation of moulting. Moreover, these genes are permanently suppressed by the small rise of ecdysteroid titre in the absence of juvenile hormone at the end of the larval stage (RIDDIFORD et al., 1986; REBERS and RIDDIFORD, 1988). In the cuticle of *Drosophila*, which forms a puparium from the last larval cuticle, cuticular proteins of the last instar are different from those of previous stadia (CHIHARA et al., 1982). In this study, we describe the presence of proteins specific to the last or immature larval cuticle in the sweet potato hornworm, *Agrius convolvuli*.

MATERIALS AND METHODS

Animals. *A. convolvuli* used in this study was obtained from the National Institute of Sericultural and Entomological Science. The larvae were kept under long photoperiod (16L : 8D) at 25 ± 1°C according to the methods described by KIGUCHI and SHIMODA (1994). They were reared on an SPLP-25 diet

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