Physiological Distinction between Aestivation and Hibernation in the Lady Beetle, *Coccinella septempunctata bruckii*(Coleoptera: Coccinellidae)¹

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The lady beetle, Coccinella septempunctata bruckii is basically bivoltine in Japan, in which the 1st-generation adults aestivate while the 2nd-ones hibernate. In the aestivating adults the respiration and oögenesis were inhibited completely and the topical application of juvenile hormone analogue (JHA) to them induced the termination of aestivation. Whereas in the hibernating adults the respiration rate remained at a relatively high level and transferring them to 25°C from the outdoor induced immediately the ovarian development, but JHA stimulated slightly the oögenesis and respiration. Observation indicates that the aestivation is the true diapause controlled by corpus allatum, whereas the hibernation is not. Voltinism type of C. septempunctata in Japan is quite different from that in Europe.

INTRODUCTION

The lady beetle, Coccinella septempunctata bruckii Mulsant is the most common aphidphagous coccinellid specifically found in Japan. Viewing their utility for biological control of aphids, a full elucidation of their life cycle is required. C. septempunctata bruckii is basically bivoltine, in which the 1st-generation adults emerge in spring and aestivate during summer, while the 2nd-generation adults appear in autumn and hibernate until the next spring (Sakurai et al., 1983). Although the hibernating adults are more vital than the aestivating ones (Sakurai, 1969), it has not been clarified yet whether the aestivation and hibernation are the true diapause or not. In univoltine population of C. septempuctata Linné found in the northern Europe, the hibernation is considered to be the true diapause controlled by corpus allatum (CA) (Hodek et al., 1973). For elucidation of dormancy in C. septempunctata, comparing the physiological nature of diapause among the populations in different region in the world is important. In the present work, physiological distinction between aestivation and hibernation of C. septempunctata bruckii is described with respect to the respiration, oögenesis and juvenile hormone action.

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MATERIALS AND METHODS

Insects. C. septempunctata adults were collected in the field in Kakamigahara City, Gifu Prefecture. Active adults were collected from the weeds infested by aphids. The aestivating adults were collected from the bush of Miscanthus sinensis Anderss. where they aggregated, whereas the hibernating adults were obtained under the weeds and withered grasses. When both generation adults were collected together in the field, each generation was distinguished in accordance with the grade of elytron colour (Sakurai et al., 1983)

Measurements of respiration and oögenesis. Respiration was measured with each sex adult by Warburg respirometer for 1 hr at 25°C and expressed as μl O₂/mg wet weight/hr (Sakurai, 1969). The females were dissected in 0.9% saline under the microscope to observe the ovarian development. The stage of oögenesis was determined according to the criterion given in Fig. 2B. Measurements were done on 10 to 20 insects for each study.

Treatment of juvenile hormone analogue (JHA). The 5.0 μ g of Methoprene dissolved in 0.5 μ l peanut oil was topically applied to the sterna under the elytra of adults by microsyringe, while control was treated with peanut oil only.

RESULTS

Life cycle and respiration during dormancy

Basically there is no alteration in the life cycle of *C. septempunctata bruckii* every year if the climate conditions remain similar. In central Japan the 1st-generation adult emerges in May and aestivates after mid June. The aestivating adults aggregate around the basal part of the weeds, especially *Miscanathus sinensis* Anderss. The adults move

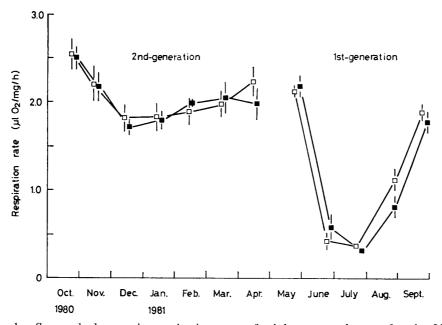


Fig. 1. Seasonal changes in respiration rate of adults. \Box , male; \blacksquare , female. Vertical line in each value shows standard deviation.

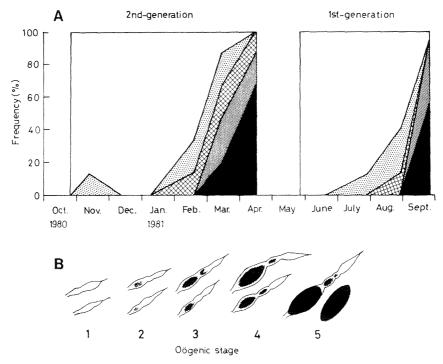


Fig. 2. Seasonal changes of oögenesis (A) and its stage criterion (B).

, Stage 1 (follicle undeveloping); , Stage 2 (follicle growing); , Stage 3 (previtellogenic); , Stage 4 (midvitellogenic); , Stage 5 (postvitellogenic and mature egg).

from the aestivating sites and become active after late August. The 2nd-generation adult emerges in October and hibernates after December. The hibernating adults exhibit preying and mating behaviors on sunny day even in midwinter.

The respiration rate of the 1st-generation adults declined sharply after entering the aestivation (Fig. 1). In late July it dropped to the minimum level, being 1/7 for the level in May. However the respiration rate of aestivating adults was much higher than that of diapausing lepidopteran pupae (cf. Mansingh and Smallman, 1967; Waku, 1965). After mid August it increased markedly and the adults awaked from the aestivation. Respiration rate of 2nd-generation adults declined slightly in early winter, then it remained at an almost constant level during the hibernation. The respiration rate was considerably higher in the hibernating adults than in the aestivating ones. But there was no difference in respiration rate between both sexes throughout year.

Oögenesis during dormancy

Figure 2 shows seasonal changes of oögenesis. Throughout the aestivation period from mid June to mid August, the oögenesis was completely suppressed to Stage 1–2. In these ovaries the germarium and 1st-follicle were undeveloped. Whereas, the oögenesis advanced gradually during the hibernation period from December to February. Enlargement of the germarium and growth of the 1st-follicle became visible by February. The oögenesis advanced noticeably after March and maturation of the 1st-follicle and

Insects ^a	Treatments ^e	Days after treatment	Respiration rate $(\mu l \ { m O_2/mg/hr}) \ (\pm \ { m S.D.})$	Oögenic stage
Aestivating ^b	Control	7	0.70 ± 0.17	1–2
	Control	14	0.48 ± 0.16	1-2
	JHA	7	1.46 ± 0.42	2-3
	JHA	14	1.24 ± 0.25	3–5
Hibernating ^c	Control	14	1.47 ± 0.39	1-2
	Control	21	1.56 ± 0.35	1–2
	$25^{\circ}\mathrm{C}$	14	2.25 ± 0.45	2-4
	$25^{\circ}\mathrm{C}$	21	1.76 ± 0.35	3-5
Hibernating ^d	Control	10	1.92 ± 0.13	1-3
	Control, JHA	10	2.04 ± 0.16	2-3
	$25^{\circ}\mathrm{C}$	10	2.16 ± 0.09	4-5
	$25^{\circ}\mathrm{C}$	10	$2.27\!\pm\!0.33$	5

Table 1. Effects of juvenile hormone analogue (JHA) and temperature on the respiration and oögenesis in the dormancy females

- ^a Insects were reared on aphids after collection under natural photoperiod.
- b Collected in early July.
- ^c Collected in early January.
- d Collected in early February.
- ^e Controls were reared at natural temperature in outdoor. For JHA-treatment, 5.0 μ g Methoprene was topically applied.

growth of the 2nd-follicle occurred. Thus the oögenesis can be initiated in the hibernating females, but not in the aestivating one.

Effects of juvenile hormone analogue and temperature on dormancy

By the treatment of JHA (Methoprene) to the aestivating females, the respiration rate increased 2 times more than that in controls, and the ovarian maturation was induced (Table 1). In the hibernating females, the respiration and oögenesis were stimulated conspicuously by rearing at 25°C and induced ovarian maturation, but affected slightly by the JHA-treatment. Results indicate that juvenile hormone is concerned in regulation of the aestivation, but not in the hibernation.

DISCUSSION

In the aestivating C. septempunctata the respiration and oögenesis were inhibited completely, and the JHA-treatment to them induced termination of aestivation. Also histological study has demonstrated that CA activity is inhibited conspicuously in the aestivating ones (Sakurai et al., 1981). These indicate that the aestivation is the true diapause caused by depression of CA activity. This view coincides with general principle that the adult diapause is controlled by CA and characterized by the reproductive diapause (Wigglesworth, 1964; de Wilde and de Loof, 1973). In the diapausing adults of Colorado beetle, Leptinotarsa decemlineata the declined respiration rate is related to the flight muscle degeneration (Stegwee, 1964; de Kort, 1969). Also in C. septempunctata the flight muscle degeneration occurs during the aestivation (Sakurai et al., 1982).

Contrary to the aestivation, the respiration rate remained in high level during the hibernation and transferring them to 25°C from the outdoor at cold brought about immediately the ovarian maturation. However the JHA-treatment to them stimulated slightly both the oögenesis and respiration. Also histological study has revealed that CA activity is inhibited slightly in the hibernating ones (Sakurai et al., 1981). These would point out that the hibernation is not under the true diapause state but only quiescent state caused by low temperature in winter. The elytron colour in the hibernating adults is more dense than in the aestivating one (Sakurai et al., 1983), indicating that CA is active in the former one.

The voltininsm of *C. septempunctata* is almost similar in central and western regions of Japan (Sakurai, 1969; Sakurai et al., 1983; Sakurai and Kubo, 1985; Kawauchi, 1985). However it varies in different region in the world (Hagen, 1962; Hodek, 1967, 1973). The univoltine or bivoltine population in Europe hibernates after early autumn, and their hibernation is regarded as the true diapause controlled by CA (Hodek et al., 1973). The bivoltine population in the region near Palestine aestivates as like the Japanese one, whereas the univoltine population in Turkey aestivo-hibernates after May to the next spring (Hagen, 1962). These differences in voltinism seem to be brought by their adaptation to different environmental conditions influenced with the factors i.e. photoperiod, temperature and prey. The present study would elucidates the physiological feature of dormancy in *C. septempunctata* adapted to the environment in Japan.

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