LARVAL DIAPAUSE IN FANNIA CANICULARIS (L.)

HIDEAKIRA TSUJI

F-409, 2-1 Nishino-Rikyu-cho, Yamashina-ku, Kyoto 607, Japan

Abstract—When parent adults were kept and allowed to oviposit at 20°C in a long-day (14L:10D) regime, and the eggs and hatching larvae were reared at 20°C under the same conditions or in darkness, no or very few (less than 12%) fully-grown larvae entered diapause.

When parent adults were kept and allowed to oviposit at 20°C and in a short-day (8L:16D or 10L:14D) regime, the incidence of larval diapause under the same condition or in darkness was high (usualy 32% to 100%). This suggests that the short-day regime, especially in parent flies, is an important diapause inducing factor for fully grown larvae of *F. canicularis*.

Chilling treatment of diapausing larvae at 6°C-8°C, longer than enough to terminate the larval diapause, allowed the larvae to pupate under the chilling temperature, even giving rise to emergence of a few adults at between 127-134 days. More than 30% of non-diapause pupae could emerge into adults under the same chilling temperature.

INTRODUCTION

Tsuji *et al.* (1992) have already shown that fully-grown larvae of *Fannia canicularis* can be divided into two groups, one pupating on the 16th–25th day, another on the 60th–100th at 20°C after egg laying by their mothers (Fig. 1).

The latter group of larvae survived 78 days of chilling treatment at 5°C and promptly pupated after being returned to 20°C, indicating that they were in a diapause state (Fig. 2).

The present paper describes preliminary experiments designed to test the effect of photoperiod on the incidence of diapause in this species. The results of much longer exposures of the diapause larvae to a low temperature are also described.



Fig. 1. Two-group pupation in Fannia canicularis larvae at 20°C in darkness.

Cumulative pupation curves
Larval mortality curves

n Number of larvae used.



Fig. 2. Effect of chilling treatments on the fully-grown larvae of *Fannia canicularis* in arrested development _____ Cumulative pupation curves

- Larval mortality curves

C Length (in days) of chilling treatment at 5°C

n Number of larvae chilled - killed at 5°C.

MATERIALS AND METHODS

Insects

A stock colony of *Fannia canicularis* was started from about 60 adult flies collected at a poultry farm in Ashimori, Okayama-city on April 10th, 1990.

Rearing

Adult flies were kept in fly-cages $(33 \times 26 \times 27 \text{ cm}, \text{ made of metal frame and nylon gauze; or } 15 \times 15 \times 15 \text{ cm}, \text{ made of wooden frame and nylon gauze})$. The flies were provided with 3% water solution of sugar soaked in absorbent cotton and food. The food served for egg-laying and larval medium as well.

The medium was exchanged for a new one together with the container (5 cm diameter, 1 cm depth). When taken out of the fly-cage, the medium together with eggs laid on it, or with larvae hatching, was gently moved onto fresh medium in another container for further rearing of larvae.

The rearing containers for larvae were two types of plastic cups (one 9.5 cm upper diameter, 8 cm bottom diameter, 4.5 cm depth; and another 6 cm upper, 4 cm bottom, 4 cm depth). Each of them was covered with a sheet of cotton fabrics or nylon gauze.

The food medium was composed by mixing the following constituent. Wheat bran—1 part in weight. Powdered animal food for mice and rats (F-1, Funabashi farm, Funabachi)—1 part in weight. Tap water—2 parts in weight.

Containers and food were changed or renewed according to the number and the devlopment stage of the larvae.

Temperature and photoperiod.

Adult flies, eggs, and larvae were kept under and, if necessary, transferred to one of the following conditions.

(1) $26^{\circ}C$ —Long day: A constant-temperature room, with a temperature of $26 \pm 1^{\circ}C$, 50% RH, and the regime of 14-hours light and 10-hours darkness per day (14L:10D) or 16-hours light and 8 hours darkness per day (16L:8D).

(2) $30^{\circ}C$ —Darkness: A constant-temperature room, with a temperature of $20 \pm 1^{\circ}C$, uncontrolled RH, and no light except when the insects were observed.

(3) 20°C—Long or short day: (a) An incubator box with the regime of 14:10D, 10L:14D, or 8L:16D and kept in the above 20°C–Darkness room.

Chilling treatment

Several groups of diapausing larve and pupae from non-diapause larvae (obtained as pupae together with diapausing larvae) separated from the food medium were confined in new plastic cups with moistened pieces of folded filter paper. The cups were placed in a moistening container (a larger glass container) in which relative humidity of the air was kept at nearly 100% by adding water into the bottom area.

The other two groups of diapausing larvae (Table 4, No. 4 and 5) were not separated from the food medium and were also placed in a similar moistening container.

The moistening containers were placed in a low-temperature room where the temerature was kept at 6-8°C. After the chilling treatment, the insects were returned to the 20°C-Darkness room.

RESULTS AND DISCUSSION

Inception of diapause under a long-day regime at 20°C

New adult flies emerging at 20°C-Darkness room from the pupae obtained after the chilling treatment of the diapausing larvae at 5°C (Tsuji et al. 1992) were kept at 20°C and 14L:10D. Their eggs and hatched larvae were reared under the same conditions. Results are shown in Table 1-B-D. The results indicate that no or very few larvae entered diapause.

Similar results were obtained (Table 1-C), using new adult flies of generation B emerging at 20°C and 14L:10D. Eggs obtained were placed, and hatched larvae were reared, under the same conditions.

Inception of diapause under a short-day regime at 20°C

New adult flies, obtained through the non-diapause route at 20°C and 14L:10D, were transferred to a fly-cage kept at 20°C and 8L:16D. Their eggs and resulting larvae were reared under the same conditions (8L:16D). Results of two series of the same experiment are shown in Table 2-EF.

The incidence of diapause ranged from 46.2 to 100% in most of the test cups (9 cups among 11), though being 9.5 and 6.7%, respectively, in other two cups F-1 and F-2. The results indicate that the short-day regime given favoured the inception of larval diapause in this species. As 20 to 100 parent flies were released into each fly-cage, the low incidence of diapause in the two cups (F-1 and F-2) may reflect the possible egg-laying by only one or two adut flies with non-diapause tendency.

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Table 1. Percent inception of larval diapause in *Fannia canicularis* L. under a long-day light regime at 20°C. Both parent flies and their progency were reared under a light regime of 14L:10D per day at 20°C. Generation B parents were from insects of generation A, emerging into adults after being chilled at 5°C for 57 or 78 days (Table 3). Generation C parents were flies of B-1, emerging into adults without entering larval diapause. Generation D parents were from diapause insects, emerging into adults at 20°C.

Container (Generation lot and egg-laying order)	Number of insects when examined	% Diapause in Container	Lot
B-1	22	0	
B-2	3	0	
B- 3	22	4.5	
B-4	2	0	2.0
C-4	5	0	
C-5	20	10	
C-6	19	0	0
D-1	46	0	<u></u>
D-2	54	0	0

Table 2. Percent inception of larval diapause in *Fannia canicularis* L. under a short-day light regime at 20°C. Both parent flies and their porgeny were reared under a light regime of 8L:16D per day at 20°C. Generation E parents were from non-diapause larvae of Generation D, emerging into adults at 20°C. without being chilled. Generation F parents were from non-diapause larvae of Generation E, emerging into adults at 20°C. without being chilled.

Conta (Gene egg-la	iner ration lot and ying order)	Number of insects when examined	% Diapause in Container	Lot
E-1		2	100	
E-2		126	97.6	
E-3		175	62.9	
E-4		347	65.1	71
F-1		42	9.5	
F-2		15	6.7	
F-3		384	78.7	
F-4		106	52.8	
F-5		26	46.2	
F-6		87	77.2	
F-7		111	73.9	68.0

Table 3. Percent inception of larval diapause in *Fannia canicularis* L., egg and larval stages being reared in darkness at 20°C. Parent flies and oviposition were under a long-day (14L:10D) or a short-day (10L:14D) light regime at 20°C. Generation C parents were from non-diapause larvae, emerging into adults at 20°C without being chilled. Generation D parents were from diapause larvae, emerging into adults at 20°C after being chilled at 5°C for 88 days. Generation A parents were from non-diapause larvae, emerging at 26°C in 14L:10D regime.

Container (Generation lot and replicate)	Light regime for Parents & oviposition	Number of insects when examined	% Diapause in Container	Lot
C-1	14L:10D (Long day)	38	0	
C-2 C-3		25	0	0
D-3	14L:10D (Long day)	80	2.5	
D-4		9	0	
D-5		59	11.5	6.1
A-1	101*14D (Short day)	22	31.8	
A-2		55	47.3	

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Container No.	1	2	3	4	5	6	7
Days of exposure	134	134	134	127	127	85	85
No. larvae exposed	2	110	123	> 90	228	359	149
% individuals surviving	100	*	*	*	*	*	84.6
Adults emerging at 6-8°C	0	6	0	0	0	0	0
Adults surviving before 20°		2					
% pupation at 6-8°C	100	100	100	*	*	> 50	71.4
No. adults emerging after			•				
returning to 20°C	2	43	> 58	> 63	>66	273	104
% adults emerging at 20°C							
per larvae exposed	100	39.1	>47.2	*	> 28.7	76.0	67.8

Table 4. Results of extended exposures of diapause larvae to a chilling temperature of 6-8°C.

*Not counted.

Inception of diapause in darkness

New adult flies emerged at 20° C and 14L:10D were allowed to oviposit under the same conditions, and their eggs were transferred to the 20° C-Darkness room. Results are shown in Table 3C. Table 3C indicates that no larvae entered diapause even when they were kept in dark conditions from the egg stage through the larval one. Similar results were obtained (Table 3, D) in another series of experiments in which parent flies had emerged through the diapause course at 20° C and 14L:10D after being chilled at 5° C.

When parent flies and their oviposition were under a short-day regime (10L:14D, 20°C), the inception of larval diapause was substantial (Table 3A). About 32 and 47% of the surviving insects on the 34th day were diapausing larvae. These diapausing larvae were used in the chilling treatment in a previous paper (Tsuji *et al.*, 1992), and the resulting flies were used in the generation B experiment (Table 1B).

Extended exposure of diapause larvae to a chilling temperature

The treatments and results are shown in Table 4.

When chilled at $6-8^{\circ}$ C for 134 days, all the surviving larvae pupated, a few even emerging into adults before returning to 20°C. Most of them emerged into adults within 7 days after returning to 20°C.

No adults, however, emerged during 127 days of the same chilling treatment.

When chilled for 85 days, more than 50% of the surviving larvae pupated but no individuals reached the adult stage before returning to 20°C. They emerged into adults 7 to 8 days after returning to 20°C.

Chilling treatment pupae

Pupae obtained together with diapausing larvae at the same time were chilled. The pupae, therefore, had experienced no diapause in their larval stage. Results are shown in Table 5.

Table 5. Results of	f an extende	d exposure of	`pupae f	from non-diar	bause larvae to a	a chilling tempe	rature of 6-8°C.
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<u> </u>	Container No.	1	2	
	Days of exposure to 6-8°C	134	134	
	No. pupae used	30	3	
	Adults emerging at 6-8°C	11	1	
	Adults surviving before 20°C	0	0	
	No. adults emerging after			
	returning to 20°C	0	0	

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More than 33% of the pupae were able to emerge into adults under the chilling temperature, showing that the pupal development of this species could proceed even at $6^{\circ}-8^{\circ}C$.

General conclusions

The results described above show that the short-day photoperiod, especially in parent flies, is an important diapause-inducing factor for fully-grown larvae of F. canicularis.

The results shown also suggest that the post-diapause development of the fully grown larvae, as well as the non-diapause development of pupae, is able to proceed even under relatively low temperatures.

REFERENCE

Tsuji, H., Taneike, Y. and Tabaru, Y. (1992). Adult longevity and larval diapause in the little house fly, Fannia canicularis (L.). Jpn. j. Environ. Entomol. Zool. 4: 180-184. (In Japanses, with English abstract).

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