

**EFFECTS OF TEMPERATURE ON THE DEVELOPMENT AND SURVIVAL
OF THE CREEPING WATER BUG *ILYOCORIS CIMICOIDES*
EXCLAMATIONIS (SCOTT) (HEMIPTERA: NAUCORIDAE)**

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Abstract.—The effects of temperature on the development and survival of *Ilyocoris cimicoides exclamationis* (Scott) were investigated under two photoperiods (15, 18, 20, 22.5, 25, and 28°C under 15L-9D; and 18 and 25°C under 10L-14D). Eggs did not hatch at 15°C and its development was delayed at 28°C. The survival rate of nymphs was significantly lower at 18 and 20°C than those at 22.5, 25, and 28°C. No nymphs reached adults at 18°C under either 15L-9D or 10L-14D. The developmental thresholds for the egg and 1st–5th instars were 11.8°C and 13.3°C, respectively. The thermal constants for the egg and 1st–5th instars were 270.3 and 626.5 degree-days, respectively. These results suggest that *I. c. exclamationis* is univoltine and the development of the immature stages is restricted to May to October in Kyoto, Japan.

Key Words: aquatic insect, Naucoridae, *Ilyocoris cimicoides exclamationis*, immature stage, life cycle

The family Naucoridae occurs worldwide and contains 413 described species in 39 genera (Sites 2000). Naucorids are oval, flattened aquatic bugs with raptorial front legs that are used for predation, and hairy middle and hind legs that are used for swimming (Polhemus 1979, Polhemus and Polhemus 1988).

The creeping water bug *Ilyocoris cimicoides exclamationis* (Scott 1874) is the only naucorid species in Japan. The adult is about 12 mm long and mostly brown, except for a greenish head, pronotum and scutellum; and reddish compound eyes. It occurs in China, Korea and Japan, and lives in submerged vegetation near the banks of fresh-water ponds and lakes (Ichikawa 1996, Hayaishi and Miyamoto 2005). This insect is listed as one of the near-threatened

species on the Red List of Japan (Ministry of the Environment 2000) because of recent decreases in local population numbers.

Rawat (1939) reported that the nymphal developmental period of the nominotypical subspecies *I. c. cimicoides* (L., 1758) is about 95 d at an ambient temperatures that varied from 14°C to 23°C. Japanese *I. c. exclamationis* require about 61 d from egg laying to adult emergence in an aquarium in greenhouse under natural photoperiod conditions during summer (Ichikawa 1996). However, thermal effects on developmental traits of *I. cimicoides* have not been investigated.

We study the effects of temperature and photoperiod on the development and survival of *I. c. exclamationis* to



Fig. 1. Adult female of *Ilyocoris cimicoides exclamationis* ovipositing into the root of *M. trifoliata* in aquarium.

obtain a better understanding of the life cycle of this endangered subspecies. This knowledge may be important for the conservation of this subspecies.

MATERIALS AND METHODS

Laboratory rearing.—The laboratory culture of *I. c. exclamationis* was established using three male and three female adults collected at Midorogaike Pond, Kyoto (135°46'E, 35°3'N) in summer 2003. The adults were paired in three aquaria (21 cm long, 13 cm wide, 13 cm deep) at 25°C under a 15 h, light and 9 h, dark (15L-9D) photoperiod. Each aquarium was filled with dechlorinated water (about 10 cm deep) and covered on the bottom with aquarium gravel. A section of the plant *Ceratophyllum demersum* L. (Ceratophyllaceae) was provided as a perching substrate. Because the females insert their eggs into the tissue of aquatic plants (Fig. 1), the root or stem of *Menyanthes trifoliata* L. (Menyanthaceae) was provided as an ovipositional site. Larvae of *Chironomus* spp. (Diptera: Chironomidae) were supplied as prey. Water in each aquarium was changed every 2 to 3 d. Plant sections were

replaced daily and examined for eggs under a binocular microscope.

Plant sections with eggs were transferred within 24 h to a 250 ml plastic cup and kept at temperatures of 15, 18, 20, 22.5, 25, or 28°C under a 15L-9D photoperiod. Each cup was filled with dechlorinated water that was changed daily.

Newly emerged nymphs were placed individually in plastic cups (1st–3rd instar, 90 ml; 4th–5th, 250 ml) and kept at temperatures of 18, 20, 22.5, 25, or 28°C under a photoperiod of 15L-9D or at 18 or 25°C under 10L-14D photoperiod. Each cup was filled with dechlorinated water and a plant section provided as a perching substrate. Larvae of *Chironomus* spp. were supplied as prey and water was changed daily. The developmental period and survival of each individual was recorded daily. Developmental thresholds (T_0) for the egg and nymphal instars were estimated from the regression between developmental rate ($1/D$) and temperatures, and the thermal constant (K) was calculated from T_0 and the regression equation [$K = D(T - T_0)$].

Statistical analyses.—The Kruskal-Wallis test was used to test for differences in the developmental periods of the egg and each instar among temperature treatments. When a significant difference was detected, a comparison between two temperatures in each combination was performed using the Mann-Whitney's U -test corrected by the Ryan method. A Tukey-type multiple comparison test was conducted for comparing the survival rate of instars between the five temperature treatments. A chi-square test was used to analyze differences in the survival of instars between two photoperiods at 25°C. The Mann-Whitney's U -test was used to compare differences between males and females in developmental periods of nymphs at each temperature and photoperiod, and between the photoperiods at 18 or 25°C.

Voucher specimens.—Selected samples of adults and nymphs have been deposited in the Laboratory of Applied Entomology, Graduate School of Agriculture, Kyoto Prefectural University.

RESULTS

All eggs kept at 15°C failed to hatch (n=45). Viability could not be determined because some ovipositional substrates rotted before eclosion in each treatment, especially at higher temperatures. The developmental period of eggs decreased significantly as temperature increased from 18°C (39.7 d) to 25°C (20.3 d), but was equal between 28 (25.2 d) and 22.5°C (23.9 d). Mean stadia periods for nymphs under each rearing condition were not significantly different between males and females (15L-9D: 20°C, $P=0.31$; 22.5°C, $P=0.51$; 25°C, $P=0.63$; 28°C, $P=0.27$. 10L-14D: 25°C, $P=0.48$). The 1st–5th mean stadium was significantly shorter at 25°C and 10L-14D than at 25°C and 15L-9D (Table 1). It developed significantly in 15L-9D as temperature increased from 20°C to 28°C (20°C, 92.4 d; 22.5°C, 73.3 d; 25°C, 50.8 d; 28°C, 43.6 d) (Table 1).

The survival rate of nymphs under 15L-9D was significantly lower at 18°C and 20°C than at 22.5, 25, and 28°C (Table 2). At 18°C under 15L-9D and 10L-14D, only 6.3% and 24.2% completed the 1st stadium, respectively, and no nymphs reached to the adults. About half of the nymphs died during first instar at 20°C (15L-9D). Survival of nymphs at 25°C under 15L-9D was significantly higher than under 10L-14D ($\chi^2=4.31$, $df=1$, $P=0.0379$).

The relationship between temperature and developmental rate for the egg and nymph is linear, but with an outline at each temperature extremum (Fig. 2). Data pertaining to 15°C and 28°C for the egg and 18°C for the nymph were not included in the calculation of the re-

Table 1. Developmental periods (days) of *I. c. exclamatoris* in relation to temperature and photoperiod (mean \pm S.E.).

| Temperature (°C) | Photoperiod | Egg | Nymphal stage (instar) | | | | | Total† (First–Fifth) |
|------------------|-------------|-----------------------|------------------------|---------------------|---------------------|---------------------|---------------------|-----------------------|
| | | | First | Second | Third | Fourth | Fifth | |
| 18 | 15L-9D | 39.7 \pm 0.6 (31) d | 25, 35 (2) | 21.0 (1) | 31.0 (1) | — | — | — |
| 20 | | 36.4 \pm 0.5 (29) c | 16.2 \pm 0.4 (25) | 13.7 \pm 0.2 (24) | 14.5 \pm 0.2 (23) | 17.4 \pm 0.2 (22) | 31.0 \pm 0.7 (16) | 92.4 \pm 1.0 (16) d |
| 22.5 | | 23.9 \pm 0.2 (34) b | 11.3 \pm 0.2 (39) | 10.1 \pm 0.1 (39) | 11.2 \pm 0.1 (39) | 15.0 \pm 0.2 (39) | 25.7 \pm 0.2 (35) | 73.3 \pm 0.4 (35) c |
| 25 | | 20.3 \pm 0.3 (34) a | 8.2 \pm 0.1 (35) | 7.1 \pm 0.1 (35) | 7.8 \pm 0.1 (34) | 9.9 \pm 0.1 (34) | 17.9 \pm 0.1 (34) | 50.8 \pm 0.3 (34) b |
| 28 | | 25.2 \pm 0.9 (11) b | 6.7 \pm 0.1 (31) | 5.7 \pm 0.1 (30) | 6.7 \pm 0.1 (30) | 8.4 \pm 0.1 (30) | 16.1 \pm 0.3 (28) | 43.6 \pm 0.4 (28) a |
| 18 | 10L-14D | not examined | 18.4 \pm 0.4 (8) | 15.0 \pm 0.3 (8) | 14, 15 (2) | — | — | — |
| 25 | | not examined | 8.2 \pm 0.2 (27) | 6.3 \pm 0.1 (27) | 7.0 \pm 0.1 (27) | 9.0 \pm 0.1 (26) | 16.6 \pm 0.3 (23) | 47.2 \pm 0.5 (23)‡ |

Values in parentheses represent the number of individuals examined.

Values in the same column followed by the same letter are not significantly different using the Mann-Whitney's *U*-test corrected by the Ryan method ($P>0.05$).

† Means are not significantly different between males and females in each rearing condition using the Mann-Whitney's *U*-test ($P>0.05$).

‡ Mean is significantly different in comparison to value at 25°C, 15L-9D using the Mann-Whitney's *U*-test ($P<0.05$).

Table 2. Survival rate (%) of *I. c. exclamationis* in relation to temperature and photoperiod.

| Temperature (°C) | Photoperiod | Number of individuals examined | Nymphal stage (instar) | | | | |
|------------------|-------------|--------------------------------|------------------------|-----------|-----------|-----------|-------------|
| | | | First | Second | Third | Fourth | Fifth |
| 18 | 15L-9D | 32 | 6.3 (2) | 3.1 (1) | 3.1 (1) | 3.1 (1) | 0 a |
| 20 | | 51 | 49.0 (25) | 47.1 (24) | 45.1 (23) | 43.1 (22) | 31.4 (16) b |
| 22.5 | | 40 | 97.5 (39) | 97.5 (39) | 97.5 (39) | 97.5 (39) | 87.5 (35) c |
| 25 | | 44 | 79.5 (35) | 79.5 (35) | 77.3 (34) | 77.3 (34) | 77.3 (34) c |
| 28 | | 35 | 88.6 (31) | 85.7 (30) | 85.7 (30) | 85.7 (30) | 80.0 (28) c |
| 18 | 10L-14D | 33 | 24.2 (8) | 24.2 (8) | 6.1 (2) | 0 | 0 |
| 25 | | 41 | 65.9 (27) | 65.9 (27) | 65.9 (27) | 63.4 (26) | 56.1 (23)† |

Values in the parentheses represent the number of surviving individuals.

Values in the same column followed by the same letter are not significantly different using the Tukey-type multiple comparison test ($P > 0.05$).

† Survival rate is significantly different in comparison to that at 25°C, 15L-9D.

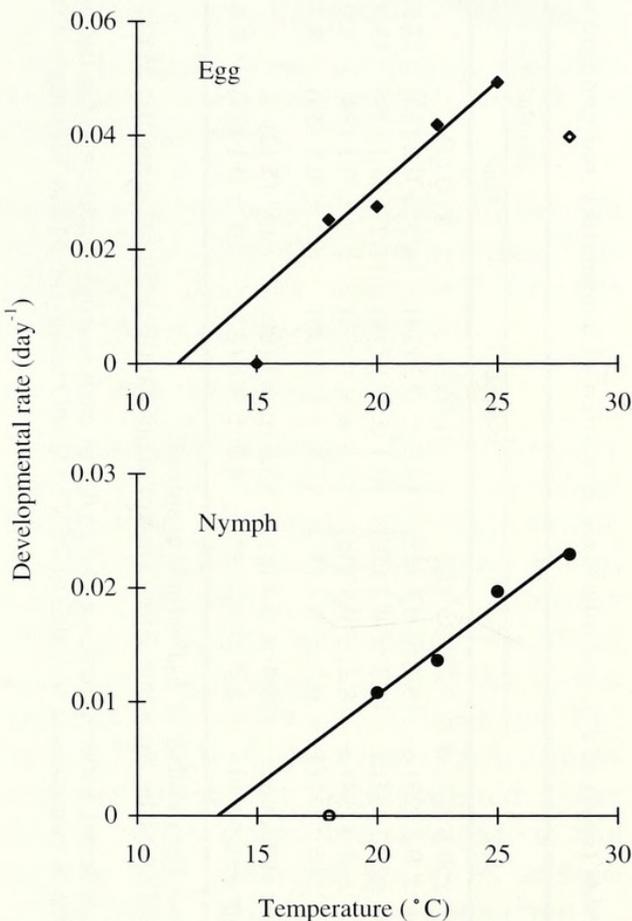


Fig. 2. Relationship between temperature and developmental rate of *Ilyocoris cimicoides exclamationis*. Open symbols represent data not included in the calculation of the regression equation.

gression equation because of developmental delay at these temperatures. The developmental thresholds (T_0) were 11.8°C for the egg and 13.3°C for the

nymph (Table 3). The thermal constants K were 270.3 degree-days (DD) for the egg and 626.5 DD for the 1st–5th instars (Table 3).

DISCUSSION

We found marked effects of temperature on development and survival of *Ilyocoris cimicoides exclamationis*. There also is earlier information on the developmental periods of other naucorid species under laboratory conditions. For example, the developmental period from egg to adult in the naucorid *Ambrysus lunatus lunatus* Usinger, a lotic species occurring in the United States of America, at 30°C was 114.3 d (egg, 20.7 d; first instar nymph, 11.0 d; second, 11.7 d; third, 19.0 d; fourth, 20.6 d; fifth, 31.3 d) (Sites and Nichols 1990). The lentic naucorid *Pelocoris femoratus* (Palliot de Beauvois) of the United States, under 16L-8D and at about 26.7°C, required 81.0 d (egg, 17.7 d; first instar nymph, 10.5 d; second, 9.7 d; third, 11.0 d; fourth, 12.6 d; fifth, 19.5 d) (McPherson et al. 1987). *Ilyocoris c. cimicoides*, at a room temperature that varied between 14°C and 23°C, required about 95 d (first instar, 12–13 d; second, 6–8 d; third, 7–9 d; fourth, 14–16 d; fifth, 21–23 d) (Rawat 1939). These reports and the result of our study suggest that the stadium at the fifth

Table 3. Developmental threshold (T_0) and thermal constant (K) of immature stages of *I. c. exclamationis*.

| Developmental stage | Regression equation | r^2 | T_0 (°C) | K (degree-days) |
|---------------------|---------------------|-------|------------|-------------------|
| Egg | $y=0.0037x-0.0435$ | 0.956 | 11.8 | 270.3 |
| First instar | $y=0.0111x-0.1606$ | 0.995 | 14.5 | 96.8 |
| Second | $y=0.0132x-0.1922$ | 0.993 | 14.6 | 75.1 |
| Third | $y=0.0105x-0.1420$ | 0.977 | 13.5 | 95.6 |
| Fourth | $y=0.0083x-0.1109$ | 0.956 | 13.4 | 119.4 |
| Fifth | $y=0.0040x-0.0481$ | 0.953 | 12.0 | 250.4 |
| First-Fifth | $y=0.0016x-0.0213$ | 0.974 | 13.3 | 626.5 |

instar is about 1.5 times that of the fourth instar. In the other species of Nepomorpha, it was reported that the stadia at later instars, especially fifth instar, were longer than at earlier instars in *Aphelocheirus vittatus* Matsumura (Ichiyanagi 2000; Ishida and Yoshiyasu 2004), *A. nawae* Nawa (Ishida and Yoshiyasu 2004), *Lethocerus colossicus* Stål (Macías-Ordóñez 2003), and *Ranatra fusca* Palisot de Beauvois (Packauskas and McPherson 1986)

Eggs of *I. c. exclamationis* did not hatch at 15°C and developed more slowly at 28°C compared to 25°C (Table 1). In contrast, nymphs developed successfully at 28°C but failed to reach adults at 18°C. These results suggest that low temperature tolerance is higher in eggs than nymphs, and that nymphs are more tolerant of high temperature than are eggs.

Although the developmental threshold of total nymphal stages was 13.3°C, no nymph developed to adult at 18°C, and 93.7% and 51.0% of nymphs died during the first instar at 18 and 20°C, respectively (Table 2). In addition, developmental thresholds of early instars (first, 14.5°C; second, 14.6°C) were higher than those of later instars, especially the 5th (12.0°C) (Table 3). Therefore, early instars apparently are more sensitive to low temperature than are later instars.

Ichiyanagi (2000) reported the effects of temperature on the development of a Japanese aphelocheirid species *A.*

vittatus, and Ishida and Yoshiyasu (2004) also noted thermal effects for development of *A. nawae*, together with the former species. The developmental threshold of nymphs of *I. c. exclamationis* (13.3°C) was higher than those of these two aphelocheirids (7.5–11.1°C) (Table 4). Both aphelocheirid species are lotic and their nymphs are found all year round according to the field surveys (Ichiyanagi 2000; Ishida and Yoshiyasu 2004). In contrast, the nymphs of *I. c. exclamationis* are discovered during late June to mid September (Ban 1985). These results indicate the breeding season of this naucorid is shorter than those of the aphelocheirids. Consequently, the higher developmental threshold of *I. c. exclamationis* will also explain their shorter and restricted breeding season.

On the basis of fragmented information derived from laboratory rearing and field surveys conducted in central Hon-

Table 4. Developmental threshold (T_0) and thermal constant (K) of immature stages of *Aphelocheirus vittatus* and *A. nawae* (Ichiyanagi 2000; Ishida and Yoshiyasu 2004).

| Species | Population | T_0 (°C) | | K (degree-days) | |
|--------------------|---------------|------------|-------|-------------------|-------|
| | | Egg | Nymph | Egg | Nymph |
| <i>A. vittatus</i> | Yoro River | 11.5 | 9.4 | 374 | 1326 |
| | Niizaki River | 11.4 | 9.0 | 388 | 1436 |
| | Kamo River | 8.9 | 11.1 | 476 | 1433 |
| <i>A. nawae</i> | Muko River | 10.6 | 7.5 | 401 | 1666 |

Table 5. Water temperature (°C) at a depth of 0.5 m at Midorogaike Pond in 2004 (Takemon, unpublished data).

| | April | May | June | July | Aug. | Sept. | Oct. | Nov. |
|-------|-------|------|------|------|------|-------|------|------|
| Min. | 11.2 | 15.8 | 19.7 | 25.7 | 24.7 | 21.8 | 13.2 | 10.4 |
| Max. | 23.1 | 26.6 | 30.7 | 35.1 | 37.4 | 34.7 | 30.7 | 20.1 |
| Mean† | 15.9 | 20.5 | 24.9 | 29.7 | 28.9 | 26.2 | 19.6 | 14.3 |

† Calculation based on temperatures recorded at intervals of 30 min.

shu, Japan, Ban (1985) and Ichikawa (1996) suggested this bug is univoltine, reproduces from late spring to early summer, and overwinters as adults. The mean water temperatures at a depth of 0.5 m at Midorogaike Pond in April and November were 15.9 and 14.3°C, respectively (Table 5), so the development of eggs and nymphs probably occurred from May (20.5°C) to October (19.6°C). In our study, total duration of the eggs and nymphs at 20, 25, and 28°C under a 15L-9D photoperiod was about 129, 71, and 69 d, respectively. Eggs of *I. c. exclamationis* that begin developing in May should reach adults in August to early September. Temperatures during September and October decrease progressively and are not high enough for immatures of a second generation to reach adults based on the results of our study under laboratory condition. The results of our study, therefore, suggest that *I. c. exclamationis* also is univoltine in Kyoto, Japan. However, the life cycle of this species should be confirmed in the future by field observations at regular intervals throughout a year.

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