Effects of Food Plants on Voltinism in *Neptis sappho*
PALLAS (Lepidoptera: Nymphalidae)

**Hideo Banno**

*Institute of Biological Sciences, University of Tsukuba,*
*Sakura-mura, Ibaraki 305, Japan*

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The durations of egg, larval and pupal stages of *N. sappho* were studied at 16, 19, 22 and 25°C. Larvae were reared on three kinds of food, *Wisteria floribunda*, *Pueraria lobata* and *Robinia pseudo-acacia* at each temperature. The thermal constants and thresholds of development for the three stages, the critical day-length for larval diapause, the longevity of adults and the time required for ovarian eggs to mature were determined. The seasonal life cycle of *N. sappho* was estimated from the results of these experiments and climatic data in Tokyo. It was inferred that food plants may influence the voltinism of *N. sappho*.

**INTRODUCTION**

In many lepidopterans, the life cycles are controlled by temperature and day-length (Danilevsky, 1961). Temperature mainly influences the developmental velocity (Ito et al., 1968; Bailey, 1976 a; Mishima, 1976; Ohkaki, 1982). On the other hand, the day-length synchronizes the life cycle with the seasonal changes (Danilevsky, 1961; Hong and Platt, 1975; Beck, 1981).

The range of food plants varies among different species of butterflies both monophagous and polyphagous. Most butterflies are oligophagous and feed on several species of plants of a particular family (Ehrlich and Raven, 1964). Several authors reported that the food plants influenced the developmental velocity of larvae in several lepidopteran species (Beckwith, 1970; Sharifi and Zaera, 1970; Bailey, 1976 b). It is expected that variation in duration of the larval stage due to food plant differences influences the life cycle.

*Neptis sappho* PALLAS utilizes many plant species belonging to Leguminocae as larval food (Shirōzu, 1965; Fukuda et al., 1972). The duration of its larval stage varies with food plant species (Banno, 1984). This study was carried out in order to examine the influence of different food plants on the life cycle of *N. sappho*.

**MATERIALS AND METHODS**

*N. sappho* is one of the most common and widely distributed butterflies in Japan. The adults occur from early May until late September. It has probably 3 generations a year in the lowland Kanto area. By late autumn, larvae develop fully and hibernate, and the next spring they pupate without feeding and then emerge as adults.

The developmental duration of each immature stage was examined from June to
August in 1976 and 1979. Females were collected from Fujino, Kanagawa in 1976 and from Tsukuba, Ibaraki in 1979. They were released in small cages in which leaves of the larval food plant (*Wisteria floribunda* D. C.) were provided as oviposition sites. Eggs from each of these females were distributed into 12 equal groups. The eggs, larvae and pupae were reared individually in glass vials (3 cm in diameter and 10 cm in length) immersed in thermo-baths controlled at 16, 19, 22 and 25°C under natural light conditions. Three species of plants, *W. floribunda* D. C., *Pueraria lobata* Orw. and *Robinia pseudo-acacia* L., were used as larval food; these plants are utilized as larval food in the field (Shirōzu, 1965; Fukuda et al., 1972). The food plants were renewed at one or two day intervals. Cotton plugs of the vials were moistened to prevent the food leaves from withering. The equation \( K = D (T - t) \) was used to determine the thermal threshold and constant for each immature stage, where \( K \) = thermal constant (day-degrees), \( D \) = developmental duration (days), \( T \) = temperature and \( t \) = threshold of development.

The larvae used in the photoperiodic experiment were offspring of females collected in June, 1982, from Tsukuba, Ibaraki. Several larvae were reared together in a plastic vessel (12 cm in diameter and 4 cm in depth) and exposed to 12.5, 13, 13.5, 14, 14.5, 15 or 16 hr of light per day. They were supplied with fresh leaves (*P. lobata*) at an interval of one or two days. The temperature ranged from 21°C to 26°C during this experiment. The number of pupating larvae was recorded every day.

The longevity of adult butterflies and the time required for maturation of ovarian eggs were also studied, using butterflies that emerged from the pupae in the photoperiodic experiment. Adults were numbered individually with a marker on the underside of the hind wing and were kept in a nylon cage in an incubator at 25±1°C under natural day-length. They were fed on thin sucrose solution every day and often on juice of watermelon. Dead individuals were recorded every day for 30 days. Dead females were dissected in order to examine the degree of egg maturation. Surviving females at the end of the 30 day period were killed and dissected to count the mature eggs.

**RESULTS**

There was a difference in developmental time between the male and female larvae, the females pupating later than the males in all experiments. The duration of larval stage in each group was represented by the means of the two sexes. The duration of larval stage was slightly longer in 1979 than in 1976 at almost all temperatures tested. The relationships between the duration of larval stage and temperature are shown in Fig. 1, and those between the durations of egg and pupal stages and temperature are shown in Fig. 2. Since the duration of pupal stage did not differ among the three food groups, Fig. 2 shows the means of the three groups. The velocities of development (the reciprocals of the durations) are also plotted in both figures. The threshold of development \( t \) and the thermal constant \( K \) for each developmental stage were calculated as shown in Table 1. These values were similar in the *Pueraria*-fed group and the *Robinia*-fed group. However, the values of the *Wisteria*-fed group were different from those of the other groups, except for the threshold of development in 1979.

The results of photoperiodic experiments are shown in Table 2. Non-diapausing larvae pupated in 17 to 23 days. Diapausing larvae grew more slowly and were still in the 4th instar even when non-diapausing larvae pupated. It was therefore easy to determine the occurrence of larval diapause. The critical day-length was estimated at
Estimation of Voltinism in *Neptis sappho*

Fig. 1. Relation of temperature to the developmental duration (hollow circles) and velocity (solid circles) in larvae of *N. sappho* reared on different foods in 1976 and 1979.

Fig. 2. Relation of temperature to the developmental duration (hollow circles) and velocity (solid circles) in eggs and pupae of *N. sappho*.

about 13 hr 40 min.

The result of adult rearing is shown in Fig. 3. Most individuals survived for more than 10 days. At the end of 30 days of rearing, about half of them survived. The relationship between the surviving period of females and the number of mature ovarian eggs is shown in Fig. 4. Mature eggs were greenish and almost equal in size to those deposited. They were counted under a dissecting microscope. Females had no eggs at emergence and required at least 5 days to mature eggs. Some females had no eggs even 10 days after emergence.
Table 1. The threshold temperatures ($t$) and thermal constants ($K$) for development of immature stages of *N. sappho* reared on different food plants

<table>
<thead>
<tr>
<th>Food plant</th>
<th>Larva (1976) $t$</th>
<th>Larva (1979) $t$</th>
<th>Egg (1976) $t$</th>
<th>Pupa (1976) $t$</th>
</tr>
</thead>
<tbody>
<tr>
<td><em>Wisteria</em></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td><em>floribunda</em></td>
<td>7.4</td>
<td>464.9</td>
<td>9.2</td>
<td>412.7</td>
</tr>
<tr>
<td><em>Pueraria</em></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td><em>lobata</em></td>
<td>9.8</td>
<td>304.1</td>
<td>9.3</td>
<td>350.3</td>
</tr>
<tr>
<td><em>Robinia</em></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td><em>pseudo-acacia</em></td>
<td>9.7</td>
<td>306.6</td>
<td>9.3</td>
<td>354.6</td>
</tr>
</tbody>
</table>

$n$ The values for egg and pupa are calculated from pooled data for three food groups.

Table 2. Incidence of larval diapause in different photoperiods and duration of non-diapausing larval stage at 21–26°C

<table>
<thead>
<tr>
<th>Photoperiod</th>
<th>No.</th>
<th>No. of pupation</th>
<th>Rate of diapause (%)</th>
<th>Duration of larval stages (days)</th>
</tr>
</thead>
<tbody>
<tr>
<td>12.5L–11.5D</td>
<td>12</td>
<td>0</td>
<td>100</td>
<td>17.4±0.5</td>
</tr>
<tr>
<td>13 L–11 D</td>
<td>10</td>
<td>0</td>
<td>100</td>
<td>17.3±2.1</td>
</tr>
<tr>
<td>13.5L–10.5D</td>
<td>12</td>
<td>5</td>
<td>38.3</td>
<td>17.4±0.5</td>
</tr>
<tr>
<td>14 L–10 D</td>
<td>12</td>
<td>11</td>
<td>8.3</td>
<td>17.3±1.8</td>
</tr>
<tr>
<td>14.5L–9.5D</td>
<td>12</td>
<td>12</td>
<td>0</td>
<td>17.3±1.8</td>
</tr>
<tr>
<td>15 L–9 D</td>
<td>11</td>
<td>11</td>
<td>0</td>
<td>17.8±1.7</td>
</tr>
<tr>
<td>16 L–8 D</td>
<td>9</td>
<td>9</td>
<td>0</td>
<td>18.9±2.9</td>
</tr>
</tbody>
</table>

Fig. 3 Survivorship curve of adult butterflies from emergence to day 30.

Fig. 4 Number of ovarian eggs as a function of adult age in *N. sappho*.
DISCUSSION

The seasonal life cycles are predicted using the thermal threshold and constant in the three food groups and the meteorological data (temperature and day-length) in Tokyo (Fig. 5). The time from sunrise to sunset plus 40 min, the duration of twilight, was used as day-length. In this prediction, it is assumed that the females of the first generation begin to oviposit on May 10th and continue to do so till June 10th, because adult butterflies emerge in early May in the lowland Kanto area and they do not have mature eggs at emergence. The emergence time probably varies among individuals in the first generation. Adult longevity is assumed to be about 2–3 weeks. Oviposition by the second generation females is assumed to begin 5 days after emergence, since at least 5 days were necessary for the maturation of eggs as already described. In the Kanto area, the critical day-length for this species occurs in early September. The photoperiod-sensitive stage of the larva is not precisely determined in N. sappho; it varies from species to species (Danilevsky, 1961; Beck, 1981). In this experiment, the diapausing larvae developed at the same rate as the non-diapausing larvae up to the 3rd instar, after which their growth decelerated. Thus, the stage sensitive to photoperiod is assumed to be before the 4th instar in predicting the life cycles of this species.

Although the thermal threshold and constant were somewhat different between 1976 and 1979, the predicted seasonal life cycles of different food groups were similar. The number of generations per year was different between the Wisteria-fed and Pueraria-fed or Robinia-fed groups. When the larvae feed on W. floribunda, 3 generations can be realized in a year. By the time the larvae of the 4th generation reach the mid-instar, the day-length becomes shorter than the critical level, and they enter diapause. On the other hand, when the larvae feed on P. lobata or R. pseudo-acacia, most of them would grow beyond the mid-instar before early September. Such larvae would complete development and emerge as 4th generation adults.

It is possible, therefore, that food plants influence the voltinism of this species through their effect on larval development.
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REFERENCES


