Entomological Review, Vol. 83, No. 7, 2003, pp. 779–782. Translated from Entomologicheskoe Obozrenie, Vol. 82, No. 2, 2003, pp. 264–269.

Variation of the Photoperiodic Reaction in *Trichogramma embryophagum* Htg. (Hymenoptera, Trichogrammatidae)

N. D. Voinovich, T. Ya. Umarova, T. S. Kats, and S. Ya. Reznik

Zoological Institute, Russian Academy of Sciences, St. Petersburg, 199034 Russia

Received January 14, 2001

Abstract—A laboratory study of photoperiodic control of diapause in the egg parasitoid *Trichogramma embryophagum* Htg was carried out. Experiments conducted at 20°C and eight photoperiods (day-lengths of 3, 6, 9, 12, 15, 18, 21 and 24 h) revealed the long-day type response based on the maternal influence on prepupal diapause in progeny at 15°C. The maximum rate of diapause in progeny was recorded at day-lengths of 12–15 h, the estimated threshold photophases were between 6–9 h (left threshold) and 16–17 h (right threshold). A comparative analysis of photoperiodic responses in seven laboratory generations reared under constant conditions revealed endogenous variability of the tendency toward diapause. The left threshold day-length was highly variable, whereas the right threshold day-length remained relatively constant. The possible reason is that the right threshold is subject to stabilizing selection in the natural conditions, whereas the left threshold zone is a selectively neutral character revealed only in laboratory experiments.

Experimental study of the factors that control the seasonal development is one of the most important lines of investigations in modern entomology (Danilevski, 1961; Tyshchenko, 1977; Tauber *et al.*, 1986; Zaslavski, 1984, 1996; Saulich, 1999). Studying the mechanisms that underlie the diapause control is of considerable importance in mass rearing of insects, as it allows one to accumulate a biological material.

Egg parasitoids of the genus *Trichogramma* are employed for biological control of lepidopteran pests in many countries. Moreover, these parasitoids are easily reared and have short life cycle, and, therefore, they can be conveniently used as model insects (Smith, 1996).

Even the first studies concerning the control of seasonal cycles in *Trichogramma* (Maslennikova, 1959; Bonnemaison, 1972) demonstrated the prepupal diapause in these insects. Low temperature is necessary for diapause induction, i.e. a temperature response is manifested. The photoperiod under which larvae of diapausing progeny develop has no effect on the diapause induction or exerts rather week modifying influence on this temperature reaction. For instance, in *T. evanescence* Westw., one of the most abundant and well-studied species, almost all prepupae diapause at a temperature of 10°C independently of photoperiodic conditions, whereas with 15°C, a considerable fraction of diapausing individuals is observed only under a short day-length (Maslennikova, 1959; Bonnemaison, 1972). The same reactions were also revealed in other species of *Trichogramma* (Sorokina and Maslennikova, 1986, 1987; Boivin, 1994; Laing and Corrigan, 1995).

Further studies clearly demonstrated that many of *Trichogramma* species manifest the so-called "maternal influence" (Vinogradova, 1973; Zaslavski, 1984; Moousseau and Dingle, 1991). It was shown that the photoperiodic conditions during the development of the maternal generation can exert a pronounced effect on the response of larvae of the progeny, especially when a daughter generation develops under conditions close to the threshold temperature and photoperiod (Zaslavski and Umarova, 1981, 1990; Mai Phu Qui and Zaslavski, 1983; Sorokina and Maslennikova, 1986, 1987). The maternal influence on the progeny results in a considerable variability of the tendency toward diapause between generations, which was experimentally observed when studying the factors controlling the diapause in various *Trichogramma* species (Zaslavski and Umarova, 1981, 1990). The variability of the tendency toward diapause, revealed in successive generations of insects, has attracted increasing attention in recent years (Geyspits and Simonenko, 1970; Geyspits *et al.*, 1974; Simonenko, 1978; Vinogradova and Bogdanova, 1980; Vinogradova and Reznik, 2000). In most of these studies, the dynamics of the tendency toward diapause was analyzed under one or two photoperiods.

As for the partenogenetic species *Trichogramma embryophagum* Htg., examined in this study, the diapause is also induced only in larvae developing at temperatures of 15°C and lower. In this case, the response to the photoperiodic conditions of development of the parental generation is strongly pronounced, whereas a photoperiod under which larvae of the daughter generation develop has virtually no effect on their tendency toward diapause (Maslennikova and Sorokina, 1986; Sorokina and Maslennikova, 1986, 1987). It should be noted, however, that these conclusions were also based on the comparison of only two photoperiods: "long-day" (16 h) and "short-day" (10 h).

The present study had two objectives: to analyze the influence of various photoperiods on the proportion of diapausing progeny along a wide range of photoperiods (construction of a photoperiodic curve) and to estimate the variability of the photoperiodic reaction among successive generations.

MATERIALS AND METHODS

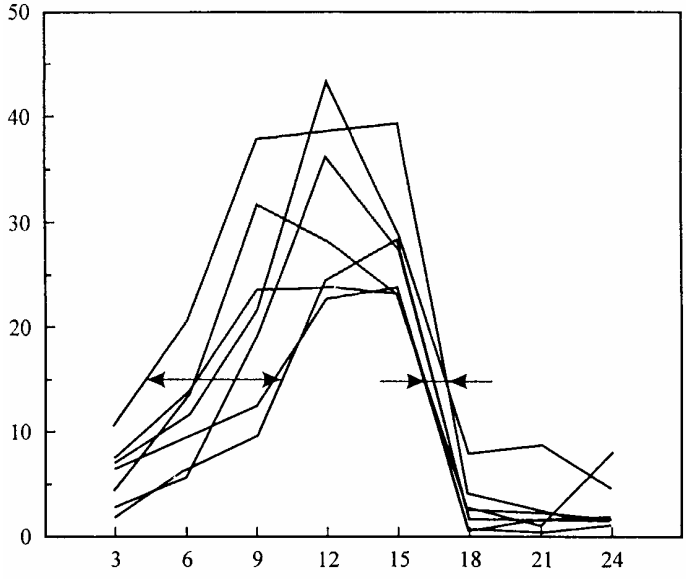
The study was conducted with a laboratory stock of *T. embryophagum* reared before the experiment for many generations on eggs of the grain moth under a 18-h photoperiod and temperature of 20°C. For each replicate of the experiment, 32 portions, each consisting of 100–200 eggs, were taken from one batch of grain moth eggs. Each portion was glued on a single paper card and the cards were subjected together for 24 h to parasitization by about 500–1000 *Trichogramma* females eclosing within 1–2 days and belonging to one generation of the main strain. Then 32 cards (maternal generation) were put in glass tubes and distributed randomly among eight groups of four tubes each. After that each group of individuals belonging to the maternal generation developed under a certain photoperiodic regimen. Experiments were conducted using 24-h light-dark cycles with photophase of 3, 6, 9, 12, 15, 18, 21 h and also permanent light, at a temperature of 20°C in all the variants.

In about one day after the beginning of *Trichogramma* eclosion (i.e., 19–20 days after parasitization and development at 20°C) a card with 200–300 grain moth eggs was placed for 24 h in each of these tubes. In 24 h, all the cards with the newly parasitized host eggs (progeny generation of *Trichogramma*) were placed in a low-temperature chamber (15°C in the dark). At this temperature, adult eclosion is completed in two months after parasitization. In two months, all the parasitized host eggs were dissected, and the number of eclosed adults and living diapausing prepupae was counted (insects that died during various developmental stages were excluded from consideration). Data for each card were regarded as a separate replicate. Thus, during this experiment seven generations of *T. embryophagum* were tested at eight photoperiods, four replicates were performed for each variant of photoperiod in each generation (100–150 parasitized eggs per replicate, a total of 29413 eggs).

For normalization of data, the fractions of diapausing individuals were arcsine-transformed by the formula $Y = 2*\arcsin(\sqrt{X})$ before the ANOVA test and comparison of means (Tukey's HSD test). Transformation and statistical processing of the results were performed using SYSTAT software. The figure shows the original (nontransformed) data.

RESULTS AND DISCUSSION

The ANOVA analysis of all the experimental results ($n = 224$) has shown that the percentage of diapausing progeny depends significantly both on the maternal photoperiod ($F = 171.3$, $p < 0.001$) and on the position of a generation under study in the sequence of generations of the *Trichogramma* laboratory strain ($F = 18.9$, $p < 0.001$). It is evident from the figure presenting the data on each of seven successive generations, that *T. embryophagum* shows a long-day type response. Two threshold photophases ranging from 3 to 9 h (left threshold) and from 15 to 18 h (right threshold) were revealed. The development of the maternal generation at day lengths of 9–15 h is the most favourable for induction of diapause in progeny, whereas a day length of more than 18 h (in some generations also less than 6 h), results in a non-diapause development of the progeny. This conclusion is in part supported by the previously published data for this species. It has been demonstrated (Sorokina and Maslennikova, 1986, 1987) that the development of the maternal generation at a day length of 10 h induced diapause in 10% of progeny developing at 15°C, whereas with a 16-h day, diapausing individuals were virtually absent (other photoperiods were not considered in this study). Most of similar experiments with other *Trichogramma* species (*T. evanescens* Westw., *T. cacoeciae* March., *T. pintoi* Voeg., *T. silvestre* Sor.) yield about the same results (Sorokina and Maslennikova, 1986, 1987).



The effect of the maternal photoperiod on the percentage of diapausing progeny in seven laboratory generations of *Trichogramma embryophagum*. Abscissa: maternal photoperiod (at 20°C); ordinate: percentage of the progeny, diapausing at 15°C. Lines correspond to the data for different generations, arrows indicate the range of variability of the left and the right thresholds of the photoperiodic curve.

The only known study concerned with the maternal influence in *Trichogramma* and conducted along a wide photoperiodic scale (Mai Phu Qui and Zaslavski, 1983) revealed a long-day photoperiodic curve in *T. euproctidis* Gir. similar to that obtained in our study.

It is also evident from the figure that photoperiodic curves constructed using data for different generations differ sharply. This refers both to the position and height of the peak in the photoperiodic curve and to the threshold photoperiod. The latter parameter is defined as the photoperiod inducing a diapause in half of individuals diapausing at the "peak" photoperiod, i.e., at the maximum average fraction of diapausing individuals equal to 30%, the threshold photophase corresponds to about 15% of diapause.

It can be well seen that the scatter of the right threshold (which has ecological importance, i.e., occurs in nature and can actually affect the seasonal dynamics) is considerably narrower than that of the left threshold (which corresponds to day lengths that can be experienced by *Trichogramma* only under the experimental conditions).

The lability of the left threshold and the relative stability of the right one (which is of ecological importance) has been repeatedly recorded in previous studies, but mainly in relation to the effect of temperature on the shape of the photoperiodic curve (Tyshchenko, 1977; Zaslavski, 1984; Saulich, 1999). In our studies, the stability of the right threshold is manifested at a constant temperature as a lower susceptibility to spontaneous endogenous variability. Thus, the differences in lability between the left and the right thresholds may be considered as not only a specific feature of the temperature dependence of the response, but also a more general phenomenon, which is evidently of adaptive value. The right threshold, which is of ecological importance, is subject to stabilizing selection, whereas the left one is a selectivity-neutral parameter manifested only in experiments (Danilevski, 1961; Zaslavski, 1984). Most probably, the differences in steepness between the left and the right portions of the photoperiodic curve are due to the same reasons.

CONCLUSIONS

(1) The investigations conducted with *T. embryophagum* in a wide range of photoperiods revealed a long-day type photoperiodic reaction based on the maternal influence on the prepupal diapause in progeny. The thresholds of the photoperiodic curve for the rearing temperatures of 20°C for maternal generation and 15°C for daughter generation range from 6 to 9 h and from 16 to 17 h.

(2) A comparative analysis of photoperiodic responses in seven laboratory generations of *T. embryophagum* revealed considerable endogenous variability of the height and position of the peak in the photoperiodic curve and of the threshold photophase. The right

threshold, which is of ecological importance, is less variable than the left threshold manifested only in the experiments. The possible reason is that the right threshold is subject to stabilizing selection in natural conditions.

ACKNOWLEDGMENTS

This study was supported by the Russian Foundation for Basic Research, grant no. 98-04-49685.

REFERENCES

1. Boivin, G., Overwintering Strategies of Egg Parasitoids, *Biological Control with Egg Parasitoids,* Wallingford: CAB International, 1994, pp. 219–244.
2. Bonnemaison, L., Diapause et Superparasitisme chez *Trichogramma evanescens* Westw. (Hymenoptera, Trichogrammatidae), *Bull. Soc. Entomol. France*, 1972, vol. 77, nos. 5–6, pp. 122–132.
3. Danilevski, A.S., *Fotoperiodizm i sezonnoye razvitie nasekomykh* (Photoperiodism and Seasonal Development of Insects), Leningrad: Lenin. Univ., 1961.
4. Geyspits, K.F. and Simonenko, N.P., Experimental Analysis of the Seasonal Changes of Photoperiodic Reaction in *Drosophila phalerata* Meig. (Diptera, Drosophilidae), *Entomol. Obozr.*, 1970, vol. 49, no. 1, pp. 83–96.
5. Geyspits, K.F., Glinyanaya, E.I., Sapozhnikova, F.D., and Simonenko, N.P., Ratio of Endogenous and Exogenous Factors in the Regulation of Seasonal Changes in the Photoperiodic Reaction of Arthropods, *Entomol. Obozr.*, 1974, vol. 53, no. 3, pp. 523–534.
6. Laing, J.E. and Corrigan, J.E., Diapause Induction and Post-diapause Emergence in *Trichogramma minutum* Riley (Hymenoptera: Trichogrammatidae): the Role of Host Species, Temperature and Photoperiod, *Canad. Entomol.,* 1995, vol. 127, no. 1, pp. 103–110.
7. Mai Phu Qui and Zaslavski, V.A., Photoperiodic and Temperature Reactions in *Trichogramma euproctidis* (Hymenoptera, Trichogrammatidae), *Zool. Zh.*, 1983, vol. 62, no. 11, pp. 1676–1680.
8. Maslennikova, V.A., On the Wintering and Diapause in *Trichogramma evanescens* Westw., *Vestnik Lenin. Univ.,* 1959, vol. 3, pp. 91–96.
9. Maslennikova, V.A. and Sorokina, A.P., Influence of Host Physiology on the Diapause and Intensity of Parasitization of *Trichogramma cacoeciae* March., *T. evanescens* Westw., *T. embryophagum* Hartig. (Hymenoptera, Trichogrammatidae), *Inf. Bull. VPS MOBB*, 1986, vol. 14, pp. 33–38.
10. Mousseau, T.A. and Dingle, H., Maternal Effects in Insect Life Histories, *Ann. Rev. Entomol.,* 1991, vol. 36, pp. 511–534.
11. Saulich, A.K., *Sezonnoe razvitie nasekomykh i vozmozhnosti ikh rasseleniya* (Insect Seasonal Development and Dispersal Potentialities), St. Petersburg: St. Peter. Gos. Univ., 1999.
12. Simonenko, N.P., The Peculiarities of Seasonal Changes in Development of the Cabbage Moth, *Barathra brassicae* L., Rearing in the Laboratory All the Year Round, *Entomol. Obozr.,* 1978, vol. 57, no. 3, pp. 473–480.
13. Smith, S.M., Biological Control with *Trichogramma*: Advances, Successes, and Potential of Their Use, *Ann. Rev. Entomol.,* 1996, vol. 41, pp. 375–406.
14. Sorokina, A.P. and Maslennikova, V.A., The Peculiarities of Phototermic Reactions in Certain Species of the Genus *Trichogramma* (Hymenoptera, Trichogrammatidae), *Vestnik Lenin. Gos. Univ., Series 3,* 1986, vol. 1, pp. 9–14.
15. Sorokina, A.P. and Maslennikova, V.A., Temperature Optimum for Diapause Induction in Species of the Genus *Trichogramma* Westw. (Hymenoptera, Trichogrammatidae), *Entomol. Obozr.,* 1987, vol. 66, no. 4, pp. 689–699.
16. Tauber, M.J., Tauber, C.A. and Masaki, S., *Seasonal Adaptations of Insects*, New York, Oxford: Oxford Univ., 1986.
17. Tyshchenko, V.P., Physiology of Insect Photoperiodism, *Trudy Vses. Entomol. O–va,* 1977, vol. 59, pp. 1–155.
18. Vinogradova, E.B., The Maternal Influence on the Progeny Diapause in Insects, *Doklady na 23 ezhegodnom chtenii pamyati N.A. Kholodkovskogo*, St. Petersburg, 1973, pp. 39–66.
19. Vinogradova, E.B. and Bogdanova, T.P., The Endogenous Cyclic Changes of the Tendency to Diapause in the Strains of Flesh Flies and Blow Flies, Developing Continuously at the Constant Conditions, *Entomol. Obozr.*, 1980, vol. 59, pp. 26–38.
20. Zaslavski, V.A., *Fotoperiodicheskiy i temperaturnyi kontrol' rasvitiya nasekomykh* (Photoperiodic and Temperature Control of Insect Development), Leningrad: Nauka, 1984.
21. Zaslavski, V.A. and Umarova, T.Ya., Photoperiodic and Temperature Control of Diapause in *Trichogramma evanescens* Westw. (Hymenoptera, Trichogrammatidae), *Entomol. Obozr.,* 1981, vol. 60, no. 4, pp. 721–731.
22. Zaslavski, V.A. and Umarova, T.Ya., Environmental and Endogenous Control of Diapause in *Trichogramma* Species, *Entomophaga*, 1990, vol. 35, no. 1, pp. 23–29.
23. Zaslavski, V.A., Diversity of Environmental Factors Controlling Seasonal Development in Insects and Possible Unity of an Underlying Physiological Mechanism, *Entomol. Obozr.*, 1996, vol. 75, no. 27, pp. 233–243.