Studies on the Diapause in the Planthoppers

I. Effect of Photoperiod on the Induction and the Completion of Diapause in the Fourth Larval Stage of the Small Brown Planthopper, *Delphacodes striatella* FALLÉN

By Ryôiti KISIMOTO

Entomological Laboratory, Kyoto University

INTRODUCTION

The small brown planthopper hibernates as the 4th instar larvae among weeds grown near the paddy fields, and the adults appear in early spring. A short day length, e. g. 8 hours, during the whole developmental period induces a retardation of development in the 3rd, 4th and 5th larval stage, especially presenting a clear elongation of the 4th larval period (MIYAKE, 1932, 1951: KISIMOTO, 1956). But, the arrest of development in the 4th larval stage which is considered to be a true diapause (MIYAKE, 1932 etc.) is never sustained for such a long period as usual in other insects entering diapause in the egg or pupal stage. In fact, even in a short day length of 8 hours the 4th moulting occurs gradually among the insects considered to be in diapause and after a certain duration of the 5th larval stage adults emerge, showing a large variation of developmental period. It was also revealed that the growht was accelerated whenever the larvae kept under a short photoperiod were transferred to a long photoperiod (KISIMOTO, 1956).

Dendrolimus pini which hibernates as larva displays the similar diapause (DANILYEVSKY, 1948; GAYSPITZ, 1949; LEES, 1955). The arrest of growth remains under the imminent control of day length throughout the period of dormancy, and growth is again resumed if the diapausing larvae are kept in appropriate photoperiods.

The diapause in the larval stage seems to differ from the ordinary cases, not confining itself to certain Lepidoptera. But details of the phenomenon are remained to be solved.

The author acknowledges the advice of Prof. S. Utida of this laboratory.

METHODS

Larvae were put into test-tubes (2 cm in diameter and 17.5 cm in length) within 24 hours of hatching with a leaf blade or a seedling of rice plant in each for food. A few drops of water were poured into each tube. Food and container were renewed every few days to avoid the wilting of the food. In most cases the number of larvae per tube was 5 individuals and no harmful effect of overcrowding was found. Long photoperiods over the natural day length were supplemented by artificial illumination. As ordinary in many cases, high temperature, such as 30°C, prevented the onset of diapause. So the rearing was carried out in a glass house of a constant temperature of 20~22°C. The intensity of light employed in supplementing the natural day length seems to be appropriate for the photoperiod response. Other technique in detail will be given in each case.

RESULTS

The effect of photoperiod on the onset of diapause. Seven series of different day length were made, namely 0, 4, 8, 10, 12, 14 and 24 hours of illumination per day. The

Contributions from the Entomological Laboratory, Kyoto University, No. 295.

Received for publication, December 17, 1957.

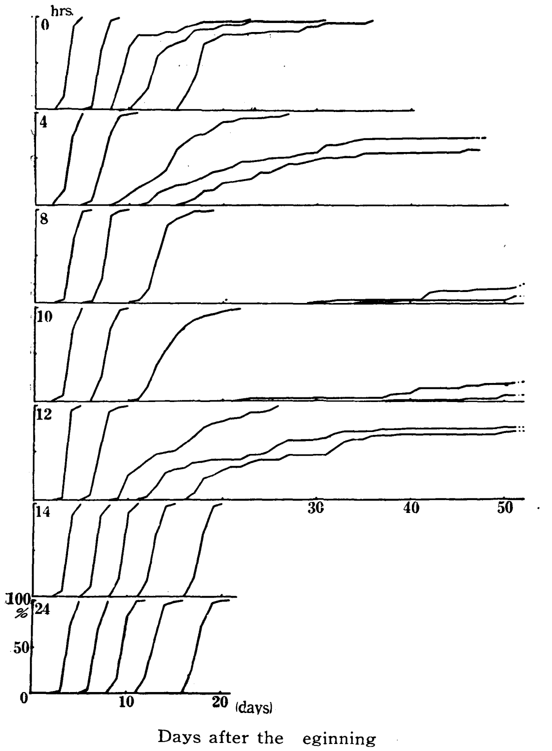

Fig. 1. Accumulated percentage moulting curves of each instar and emergency curves in various photoperiods at 20～22°C. The rearing was stopped at the 50～60th day of beginning.

rearing was started on June 26th to 28th 1956. The accumulated curves of percentage moulting of every instar obtained in each series are shown in Fig. 1. The rearing was stopped at the 50th to 60th day from the beginning, then the trends of onset and completion of the diapause in each photoperiod having been well shown. The mortality was almost negligible.

The relationship between the photoperiod and the incidence of diapause is rather similar to the ordinary cases, namely short day lengths ranging from about 8 to 10 hours induce 100 per cent diapause, over and below this range the incidence of diapause falls. The decrease of the incidence of diapause is abrupt from 10 to 14 hours day length and no diapause is found in long day lengths over 14 hours. A clear-cut distinction can not be made between diapausing

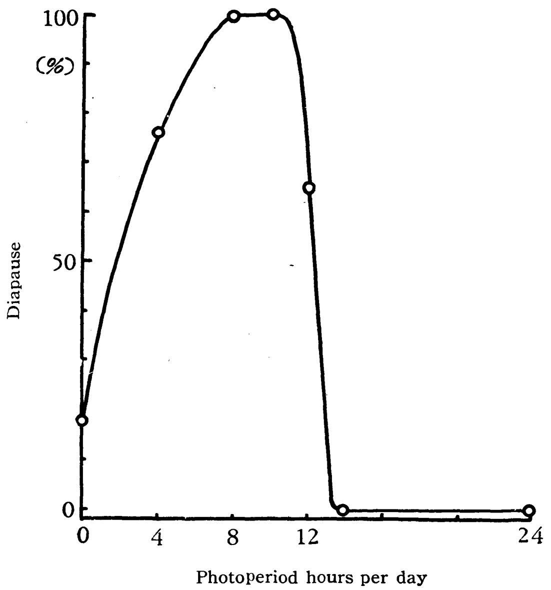

Fig. 2. The effect of photoperiod on the induction of diapause at 20～22°C.

and non-diapausing larvae in the day lengths of 0, 4 and 12 hours, the 4th moulting and the emergency curves overlapping from a non-diapausing range to diapausing one. But if a tentative discrimination is made at 20～21 days after the beginning of rearing when all adults finished to emerge in long days of 14 and 24 hours, the relationship between photoperiod and the percentage incidence of diapause can be shown in Fig. 2.

The general pattern is similar to the examples studied by several workers (DANILYEVSKY, 1948; DICKSON, 1949; TANAKA, 1950; WAY, & HOPKINS, 1950; LEES, 1953; OTUKA, & SANTA, 1955; etc.), but differs in several points. The incidence of diapause falls promptly at a photoperiod of 12 hours, which is shorter than the examples obtained by the former workers. An elongation of the developmental period is found to occur from the 3rd instar larvae and the elongation is clearer in photoperiods of 4 and 12 hours which induce a partial diapause than in photoperiods inducing a 100 per cent diapause. Of course, the larvae having longer period in the 3rd instar enter diapuse in the 4th larval stage and the non-diapausing larvae have shorter period in the 3rd instar. Therefore, the 3rd moulting curve of the shortest duration is found, paradoxically at first sight, in a photoperiod of 8 hours, those in 14 and 24 hours photoperiods being left out of consideration. In other words, the variation of the 3rd larval duration is least in a photoperiod inducing 100 per cent diapause. A little difference is shown even between photoperiods of 8 and 10 hours.

The incidence of diapause of the individuals emerged after 20~22 days from the beginning in 0, 4 and 12 hours illumination is shown also by the fact that some brachypterous males appeared. The appearance of the brachypterous male was considered to be a characteristic of the individuals experienced the diapause (KISIMOTO, 1956), and in fact, no brachypterous male appeared in photoperiods of 14 and 24 hours.

The 4th instar larvae entered diapause do not become completely inactive but their motion becomes a little slower and show a trend of resting on dried parts of food. The feeding and excretion decrease which is clear from the fact that the food and container are kept fresh and clean for longer time than those with the non-diapausing larvae. The non-diapausing larvae can not endure for a long time on dried food. A dark pigmentation can be found in most of the diapausing larvae.

The larvae in diapause begin to moult gradually after a certain period of arrest of development even under the constant conditions of temperature and photoperiod which are appropriate in inducing the diapause. After a certain period of the 5th larval stage adults emerge. In this case no special agent, such as a low temperature, is necessary for the completion of the diapause, at least concerning the intensity of the diapause in the present case.

The sensitivity to photoperiod during the 4th larval stage and the effect of high temperature for the completion of the diapause. The larvae which entered diapause under the conditions of temperature of 20~22°C and photoperiod of 8 hours were put under 0, 4, 8, 12, 14, 20 and 24 hours photoperiods at 20~22°C and 30°C within 24 hours of the 3rd moulting. The larvae were offered seedlings of rice plant. The rearing was started at 20th to 22nd, July 1956. The accumulated curves of the percentage 4 th moulting and emergence are shown in Fig. 3.

The 4th instar larvae considered to be in diapause retain their sensitivity to photoperiod, as shown in Fig. 3. As already stated, the larvae in diapause resume the development sooner or later under the present condition, the sensitivity to photoperiod is to be shown by the duration necessary for the completion of the diapause. The general trend is similar to that of the former experiment, that is, the photoperiod shows a similar effect on the completion of diapause to that on the induction of diapause. The average duration of the 4th and 5th instar larvae were longest in a photoperiod of 8 hours per day and decreased as daily photoperiod is extended or shortened. Under continuous illumination the variation of the 4th and 5th larval periods became least of all. In complete darkness also the acceleration of development was clearly observed. The duration of the 5th larval stage was also under the control of the photoperiodism, the 5th larval period being longest in a photoperiod of 8 hours among all photoperiods and decreases as the photoperiod is extended or shortened. The average durations of the 4th and the 5th larval stage in each photoperiod are shown in Table 1. The durations in photoperiods of 4, 8 and 12 hours at 20~22°C in which the 4th and the 5th larval periods extended clearly were to be underestimated as the mortalities were considerable among the individuals having longer developmental period. The larvae which died before emergence were found to be well fed and in fact much swelled.

At 30°C the 4th and the 5th larval periods are shortened in considerable degree in comparison with those at 20~22°C in each photoperiod. But the response to the different photoperiods was similar to that at 20~22°C, namely, the duration of the 4th larval stage is extended in photoperiods of 4, 8

Table 1. Average durations of the 4th and the 5th larval stage and mortalities in various photoperiods. The larvae used were reared in a photoperiod of 8 hours per day until the 3rd moulting at 20～22°C.

photoperiod	20～22°C			30°C		
per day	4 th	5 th	mortality	4 th	5 th	mortality
0 hrs.	15.2 d.	6.1 d.	7.5 %	6.9	3.2	3.3
4	20.4	8.1	32.5	9.2	3.4	6.6
8	27.8	16.4	37.5	10.9	3.4	0.0
12	25.8	7.5	20.0	10.0	3.5	6.6
14	11.1	5.8	7.5	5.6	3.9	13.1
20	14.6	6.0	2.5	6.4	3.6	10.0
24	9.9	5.3	5.0	5.5	3.1	3.1

and 12 hours and decreased as the photoperiod is extended or shortened, reaching the minimum in permanent illumination. At 30°C, however, the 5th larval stage is not under the control of the photope riod in every photoperiods, showing a uniform figure. It is supposed that the 5th larval stage is completely out of the diapause state, at least 3.1 to 3.9 days are the minimum value of the 5th larval period which had been obtained during the successive rearing.

An irregularity in the trend of response to photoperiod was found in a photoperiod of 20 hours, that is, certain larvae show a longer 4th larval period than that in the others at both rearing temperatures. The reason for this fact is not assumed.

Interrelation between the 4th larval period

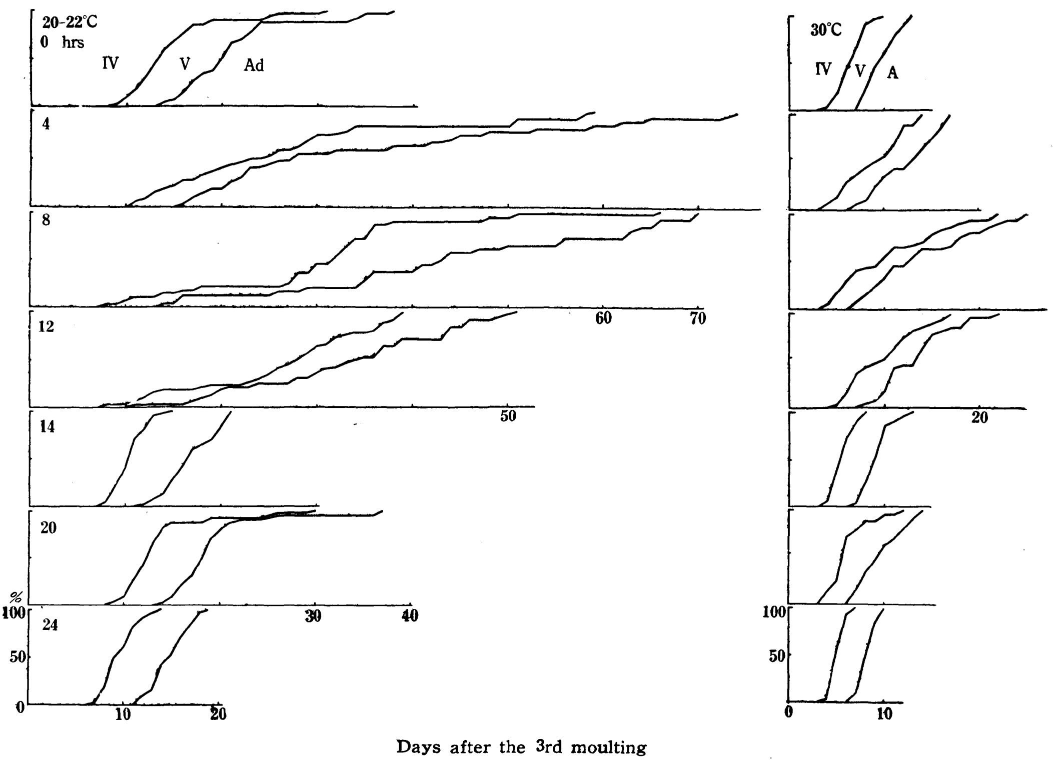

Fig. 3. The effect of photoperiod on the completion of diapause at 20～22°C and 30°C. The diapausing larvae used were reared in a photoperiod of 8 hours per day at 20～22°C.

and the 5th larval period in various photoperiods at 20～22°C. It was shown in the preceding chapter that the 5th larval period is also under the control of the photoperiodism as well as the 4th larval period at 20～22°C. The interrelation between the 4th and the 5th larval period was taken individually in each photoperiod. The result is shown in Fig. 4.

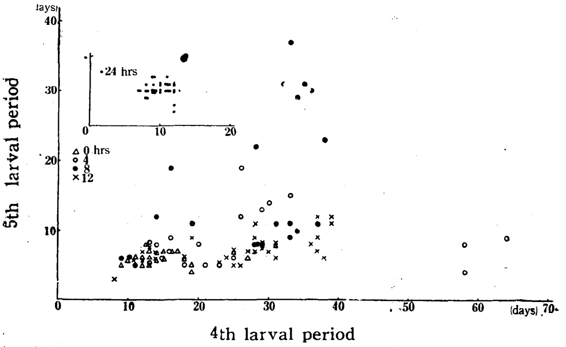

Fig. 4. Correlation between the 4th larval period and the 5th larval period of the diapausing larvae reared in various photoperiods at 20～22°C (c. f. Fig. 3).

Most larvae in photoperiods of 0, 16, 20 and 24 hours have the 4th larval periods concentrated in a range from 7 to 15 days, which is longer than those of the non-diapausing larvae at this temperature. The 5th larval period is also concentrated in a range of 4 to 8 days, a little longer than that of the non-diapausing larvae. In the non-diapausing larvae the ratio: 4th larval period/5th larval period is 0.67～0.70, namely, the 5th larval period is longer than the 4th larval period. But in the diapausing larvae the ratio is in the least 1.87 in a photoperiod of 24 hours in which the diapausing larvae complete the diapause most promptly. In other photoperiods the ratio exceeds this figure.

In photoperiods of 4, 8 and 12 hours the 4th larval period shows a large variation as mentioned above. However, the 5th larval period is not necessarily elongated with the 4th larval period, namely, most larvae in photoperiods of 4 and 12 hours and those larvae which have the extraordinarily long 4th larval period in a photoperiod of 20 hours present 5th larval period in a range of 8 to 11 days. A clear elongation of the 5th larval period is shown by most larvae in a photoperiod of 8 hours and about a half of the larvae having longer 4th larval periods in a photoperiod of 4 hours. In extreme, the 5th larval period become equivalent to the 4th larval period. No example that the 5th larval period much exceeded the 4th larval period was found.

The effect of the night break. The effect of the night break, a short interruption of the dark phase, was studied under the similar conditions of the preceding experiments, namely, a photoperiod of 8 hours at 20～22°C. Eight hours after the beginning of the dark phase 1, 5, 20, 60 and 120 minutes of illumination was given every day from the beginning of reaing. But no effect of the night break to prevent diapause was found.

DISCUSSION

The general pattern of the photoperiodism in the induction of diapause obtained in the small brown planthopper is very similar to that in several other arthropods. But in detail several differences are found. The optimal day length for the onset of diapause is narrower and a little shorter than those obtained in *Acronycta rumicis* (DANILYEVSKY, 1948), *Grapholitha molesta* (DICKSON, 1949) and *Barathra brassicae* (UCHIDA & MASAKI, 1954: OTUKA & SANTA, 1955) and others. In the present case, a photoperiod of 12 hours prevents diapause in about 33 per cent of the larvae. The fact means that the small brown planthopper can display a normal development until the autumn equinox, a little later than in the preceding examples.

The developmental process of the diapausing larvae completely overlaps to that of the non-diapausing larvae. No clear discrimination between the diapausing and the non-diapausing larvae can be made, at least through the developmental period. Nevertheless the incidence of diapause is clear especially in those larvae which have larval periods of over two months.

Moreover, the sensitivity to the photoperiod is sustained in the 4th larval stage considered to be in diapause and the larvae respond to photoperiod in the similar way to that in the induction of diapause. A high temperature of 30°C which prevents the larvae to enter diapause is also responsible for the completion of diapause though varying with photoperiod. But even the longest 4th and 5th larval period which was realized in a photoperiod of 8 hours is a same order of that in long photoperiods of over 14 hours at 20~22°C.

These facts may lead us to the conclusion that the arrest of development in the 4th larval stage is not a real diapause but merely a photoperiodically suspended development, so to speak. However, the facts that the insect hibernates as 4th instar larvae, shows a clear elongation of the 4th larval period in a short day length and spasmodic feeding during the period, and also have cold-hardiness at 0°C (unpublished result) etc, seem to be enough for concluding that the arrest of development is due to the diapause. The present result offers an example to show the difficulties in distinguishing the real diapause from the facultatively suspended development. Lees (1955) said that the definition of 'diapause' and 'quiescence' of Shelford (1929) was no longer wholly appropriate, and presented an example of *Dendrolimus pini* (GAYSPITZ, 1949, 1953) showing the difficulties of distinction between the two types of arrest. The present result is another example and may be one of many examples remained to be solved.

Whether or not a more profound diapause is induced by other environmental factors such as low temperature or food shortage, and if obtained, whether such diapause needs a low temperature for the completion are in the course of experiment.

SUMMARY

The effect of the photoperiod on the induction and the completion of the diapause in the 4th larval stage was studied at 20~22°C with the small brown planthopper, *Delphacodes striatella* FALLÉN.

1. 100 per cent diapause was induced in a short day photoperiod ranging from 8 to 10 hours. The incidence of diapause falls rather abruptly as the duration of illumination shortened or extended over this range, reaching 100 per cent non-diapause in photoperiods of over 14 hours illumination. But in complete darkness the percentage diapause was about 18 per cent.

2. The diapausing larvae never continue to be in the arrest of developement indefinitely in conditions of temperature and photoperiod inducing the diapause. The 4th moulting and emergence occurs ranging from a few days to a few months after the onset of diapause.

3. The diapausing larvae retain their sensitivity to photoperiod. The general trend of the effect of photoperiod on the completion of diapause is similar to that of the induction, the emergence being performed uniformly after 2 to 3 weeks in a long day photoperiod at 20~22°C.

4. At 30°C the completion of diapause was accelerated though varying with the photoperiod subjected.

5. In spite of these facts which are negative for considering the present type of arrest as the true diapause, some other affirmative attributes are presented. Difficulties for a clear-cut definition of the diapause was discussed.

6. No effect of the night break from 1 to 120 minutes was obtained.

LITERATURE CITED

DICKSON, R. C. (1949) Ann. Ent. Soc. Amer. **42** (4): 512~37.

KISIMOTO, R. (1956) Oyo-Kontyu **12** (4): 202~210.

LEES, A. D. (1953) Ann. Appl. Biol. **40**: 449~486, 487~497.

LEES, A.D. (1955) The Physiology of Diapause in Arthropods. Cambridge Univ. Press.

MATSUMOTO, S., H. SANTA and M. OTUKA (1953) Oyo-Kontyu **9** (2): 45~51.

MIYAKE, T. (1932) Kontyu **6** (1/2): 20~36, 47~65.

MIYAKE, T. (1951) Reports Hiroshima Agr. Exp. Stat. **1**: 1~21.

OTUKA, M. and H. SANTA (1955) Bull. Nat. Inst. Agri. Sci. Japan [C] **5**: 49~56.

SHELFORD, V.E. (1929) Laboratory and Field Ecol-

ogy. Baltimore; Williams and Wilkins.
TANAKA, Y. (1950) J. Seric. Sci. Japan **19**: 580～590.
UCHIDA, T. and S. MASAKI (1954) Mem. Fac. Agr. Hokkaido Univ. **2** (1): 85～95.
WAY, M. J. and B. A. HOPKINS (1950) Jour. Exp. Biol. **27** (3/4): 365～76.

摘　　要

ウンカ類の休眠に関する研究

I. ヒメトビウンカの4令幼虫における休眠の誘起および消去におよぼす日長効果

岸　本　良　一
京都大学農学部昆虫学研究室

1) ヒメトビウンカ幼虫を初令から 20～22°C で種々な日長の下に飼育した。

2) 日長が 8～10 時間では休眠率は 100% になるが，日長がこの範囲からはずれると休眠率は減少する。12時間では約 35% の非休眠個体を生じ，14時間ではすでに 100% 非休眠となる。全期間暗黒状態では約 18% の休眠率となる（第1図，第2図）。

3) 休眠した個体は4令期間が延びるが，同じ条件下でも徐々ながら発育し，4令期間の個体変異は非常に大きい。休眠個体と非休眠個体との区別は，少なくとも発育日数からは明白にはつけにくい。

4) 休眠に入った幼虫も日周期に対する感受性をもっており，長日や極端な短日下では急速に発育を始め，かなり斉一な羽化が見られる。最も長い休眠状態は休眠誘起に好適な日長下で維持される。このような場合には5令期間も長くなり，4令期間と匹敵するほどになる個体も多くなる（第3図）。

5) 休眠に入った幼虫を 30°C のような高温において好適な餌を与えると，日周期によって多少違いがあるが，発育は促進され，斉一な羽化が見られる（第3図）。

6) 8時間日長下で，暗黒相を 1～120 分間破ったが，休眠誘起を阻害する効果はなかった。

7) ヒメトビウンカ幼虫における休眠は他の種の卵や蛹における休眠とは様相を異にしている。このような型の休眠では従来の定義は再検討する必要があると思われる。

抄　　録

erythomycin および他の数種の抗生物質によるミツバチ腐疽病の防除

WILSON, W. T. and J. O. MOFFETT (1957) The effect of erythomycin and other antibiotics on the control of European foulbrood of honeybees. J. Econ. Ent. **50**(2): 194～196.

ミツバチの腐疽病（European foulbrood）の防除に sulfathiazole が有効であることが発見されて以来，薬品や抗生物質でこの病気を防除しようとの試みが行われてきている。

この研究では純品の erythomycin およびその製剤である glymycin を用いて腐疽病の防除を行ったところ卓効をしめすことがわかった。すなわち，MK-65 および penicel は防除効果ほとんどなく，aurefac, tetracycline, streptomycin-penicin は相当程度の防除効果を示したが，erythomycin は最もすぐれており，かつミツバチに対して無害であり，その製剤の glymycin は使用に便利な形態である。なお蜂蜜中にこれらの抗生物質が貯えられる危険性をさけるために，初期のコロニーに対して予防的に用いるのが良い方法であろう。

（農技研　湯嶋　健）