Population Effect on the Daily Periodic Emergence of Drosophila

Mori, Syuiti

Memoirs of the College of Science, University of Kyoto, Series B,
Vol. XXI, No. 1, Article 8, 1954

Population Effect on the Daily Periodic Emergence of *Drosophila*[1]

By

Syuiti MORI

Zoological Institute, University of Kyoto

(Received July 19, 1954)

Introduction

Various works have hitherto been published, especially during recent 30 years, on the daily periodic phenomena of animals. Improtant results among them are summarized in some reviews by Mori (1948), Kleitman (1950) and Aschoff (1954). Most of works, however, were performed methodologically with the view to clarify the phenomena at the individual level; namely, most investigators failed to recognize such phenomena as population effect, and most knowledges obtained, though fruitfull, seem to be confined to the field of physiology or physiological ecology at the individual level.

A few literatures concerning the population effect on the daily periodic behavior within the same species will be cited here. Park and Sejba (1935) studied the nocturnal activity of a beetle *Megarodacne heros*. It is conspicuous that this insect shows three activity peaks at night when reared in single (20–21, 23–24 and 3–4 o'clock respectively), but these phases are shifted 1 to 2 hours earlier when reared in a group (7 to 11 individuals). Reports on the effect of sexual partners can be found in Rodentia and *Drosophila*. Calhoun (1945) studied on *Sigmodon hispidus hispidus* and found that males as well as females, when isolated, moved most actively at night, but when sexual partners were placed in one cage they moved actively in the daytime, at least during first 7 days. A similar phenomenon was found by Ohsawa and others (1952) on *Drosophila melanogaster*, i. e., although the monosexual group, irrespective whether it is composed of males or females, shows two activity peaks in a day—morning and evening, the bisexual group shows one activity peak at midnight. Several investigators such as Hemmingsen (1932), Durrant (1935) and Slonaker (1935) have reported the characteristic influence of the female oestrus cycle

1) This research was aided by the Scientific Research Expenditure of the Department of Education.

of rat on the activity of the male (refer to Aschoff, 1954). Recently, Fujimoto (19-53) found a conspicuous phenomenon about the modification of the individual mode of daily periodic activity of mice by the social dominance order established in a group. The dominant individual among them exhibits normal periodic activity pattern, whereas the subdominant seems to move about only when the dominant rests, and thus as the social rank falls off so the modification seems to become greater and at an extreme case the lowest one seems to show no daily periodic pattern of activity.

Thanks to several works that have been performed on the daily periodic emergence of *Drosophila* (as for literatures refer to Mori, 1949), we know that this insect emerges most actively in the morning, and the light is the most important environmental factor concerned with. The purpose of the present research is to show whether there occurs, as an effect of population, any modification in the mode of daily periodic emergence of *Drosophila melanogaster* or not, and if any, to detect the possible factors related to it.

I take this opportunity of thanking the Collegues of the Research Group for Ecosystem of Kyôto University for valuable aids in many ways, and Professor Denzaburo Miyadi for his kindness in reading the original manuscript.

Materials and Methods

Drosophila melanogaster, which had been reared in our laboratory for about 100 generations with the Pearl's medium as food, was used as the material. The experiments consisted of two series.

The procedure of the *first series* was as follows: Twenty to thirty flies (1 to 4 day after emergence) were put into a glass tube (length 15 cm, diameter 3 cm) and a slide glass, on which the Pearl's medium was placed, was inserted into the tube. Twenty glass tubes were prepared at the same time. Female flies deposited the eggs on the Pearl's medium most actively in the evening and the first instar larvae used to appear in the next afternoon (Mori, 1949). The slide glasses were renewed once a day.

The first instar larvae, hatched during 15 to 18 o'clock of a day, were carefully transferred one by one during 18 to 20 o'clock with a needle to small glass tube (length 6 cm, diameter 1 cm), each containing 1 cc of the Pearl's medium and a drop of the beer yeast solution (0.5 mg/cc water) on it. These test tubes were placed in the thermostat maintained at 25°C and subjected to the normal daynight light change. About 9 days later the flies began to emerge and continued 4 to 6 days. The number of emerging flies were counted at every 3 hours till the last fly appeared. This experiment was carried out during November to December of 1953. Number of the small test tubes and number of larvae put in them are shown in Table 1.

The *second series* of experiment was designed so as to be able to compare the progress of emergence throughout a day in the normal Pearl's medium with that in the Pearl's medium containing $CuSO_4$ at the rate of 20×10^{-6} Mol. In

Table 1 Summarized data of the first series of experiment.

Number of larvae in a test tube	Number of test tubes employed	Total number of larvae used	Total number of flies emerged	Emergence rates (%)
1	285	285	112	39.3
2	179	358	136	38.0
8	47	376	133	35.4
16	18	288	93	32.3
32	21	672	90	13.4
64	11	704	57	8.1

both cases each test tube contained single larva. The experiment was performed in March of 1954. Table 2 shows the experimental data.

Table 2 Various data of the second series of experiment.

Medium	Total number of test tubes=Total number of larvae	Total number of flies emerged	Emergence rates (%)
Normal Pearl's medium	242	91	37.6
Pearl's medium containing $CuSO_4$ at the rate of 20×10^{-6} Mol.	317	80	25.2

Results and Discussions

The results of the first series of experiment are summarized in Table 3.

It may be concluded that the time of emergence in the course of a day is delayed as the population increases. That is, when the larval populations are 1, 2 or 8, the maximum emergence is observed early in the morning before sunrise; on the other hand, when the larval populations are 16, 32 or 64, the time of maximum emergence is shifted later, occuring either before or after the noon. This tendency is shown more clearly in Table 4. This table shows that the daily periodic mode of emergence is remarkably changed at about 16 population. It is interesting to find in Table 1 that the emergence rate is considerably altered between 16 and 32 populations, which coincides with the phenomenon mentioned above. The existence of the population effect is clear. Then, what cause is responsible for this phenomenon? Two major factors, related to aggregated larval life, can be considered. One is rather psychological or simple biotic factors (such as induced by collision), and the other is rather physiological factors (such as induced by lowered metabolism due to harmful excrements).

Table 3 Number of flies emerged within every 3 hours in the first series of experiment, showing the difference in emergence periodicity in different population densities.

Population in a test tube	Time								Total
	0–3	3–6	6–9	9–12	12–15	15–18	18–21	21–0	
1	**12**	**23**	**16**	**16**	**11**	**10**	**13**	**11**	**112**
	10.7	*20.4*	*14.3*	*14.3*	*9.8*	*9.0*	*11.7*	*9.8*	*100*
2	**27**	**26**	**22**	**11**	**22**	**9**	**11**	**8**	**136**
	19.8	*19.1*	*16.2*	*8.1*	*16.2*	*6.6*	*8.1*	*5.9*	*100*
8	**23**	**30**	**19**	**14**	**16**	**12**	**11**	**8**	**133**
	17.3	*22.6*	*14.3*	*10.5*	*12.0*	*9.0*	*8.3*	*6.0*	*100*
16	**9**	**18**	**6**	**10**	**12**	**20**	**12**	**6**	**93**
	9.7	*19.4*	*6.5*	*10.7*	*12.9*	*21.5*	*12.9*	*6.4*	*100*
32	**5**	**11**	**25**	**14**	**16**	**9**	**5**	**5**	**90**
	5.6	*12.1*	*27.8*	*15.5*	*17.8*	*10.0*	*5.6*	*5.6*	*100*
64	**5**	**5**	**12**	**12**	**10**	**8**	**1**	**4**	**57**
	8.8	*8.8*	*21.1*	*21.1*	*17.5*	*14.0*	*1.8*	*7.0*	*100*

Gothic figures indicate the actual number of flies emerged, and italic figures show the percentages.

Table 4 Coefficients of emergence (A and B), showing the relative abundance of emerging flies within different periods, which vary with the population density.

	Population in a tube
	1 2 8 16 32 64
A. $\dfrac{\text{No. of flies emerging during 18–6 o'clock}}{\text{No. of flies emerging during 6–18 o'clock}}$	$1.11 = 1.12 = 1.18 = 0.94 \gg 0.47 = 0.36$
B. $\dfrac{\text{No. of flies emerging during 0–9 o'clock}}{\text{No. of flies emerging during 9–18 o'clock}}$	$1.38 = 1.79 = 1.71 > 0.75 = 1.05 = 0.73$

Significancy of the difference: = non-significant, > significant at the 1% level, ≫ significant at the 0.1% level.

The second series of experiment was attempted to find some clues to the analysis of these circumstances. The toxic action of $CuSO_4$ to animals, when the concentrations of this drug are increased beyond their capacities of the excretory organs, is widely known (Poulson, 1950; McElroy and Glass, 1950) and it may be considered that this drug lowers the metabolism of *Drosophila*. If lowering of metabolism was the cause of the population effect, the similar phenomenon may be evoked by providing food contained $CuSO_4$. The result of this experiment is shown in Table 5.[2)] This table seems to show that the time of maximum emergence is delayed in the Cu-medium. This tendency is made clear in Table 6.

The similarity which is seen between the feature of the population effect and that of the Cu-effect seems to suggest that the cause of the population effect can

Table 5 Number of flies emerged at every 3 hours from both normal Pearl's medium and Cu-containing Pearl's medium.

Medium	Time								Total
	0–3	3–6	6–9	9–12	12–15	15–18	18–21	21–0	
Normal Pearl's medium	**6**	**6**	**13**	**24**	**21**	**12**	**7**	**2**	**91**
	6.5	*6.5*	*14.3*	*26.4*	*23.2*	*13.2*	*7.6*	*2.1*	*100*
Cu-containing medium	**6**	**9**	**14**	**14**	**7**	**17**	**9**	**4**	**80**
	7.5	*11.2*	*17.5*	*17.5*	*8.8*	*21.3*	*11.2*	*5.0*	*100*

For explanation of figures see Table 3.

Table 6 Coefficients of emergence (C and D) indicating the difference in the mode of daily march of emergence between the flies reared in the normal Pearl's medium and in the Cu-containing medium.

	Medium		
	Normal Pearl's medium		Cu-containing medium
C. $\frac{\text{No. of flies emerging during 6–12 o'clock}}{\text{No. of flies emerging during 12–18 o'clock}}$	2.36	$\gg$	0.85
D. $\frac{\text{No. of flies emerging during 0–12 o'clock}}{\text{No. of flies emerging during 12–0 o'clock}}$	2.37	$>$	1.22

Significancy of the difference: $>$ significant at the 1% level, $\gg$ significant at the 0.1% level.

be found in the physiological process — lowered metabolism of the larvae and not in any psychological process and so on.

To my opinion, the major internal process controlling the mode of daily periodic phenomena of animals may be physiological, notwithstanding the existence of such work as Fujimoto's (1953). The essential meaning of the sexual partnership in modifying the daily periodic activities, such as seen in the reports of Ohsawa and others' (1952) or Calhoun's (1945), may also be found rather in the physiological processes than in the psychological. In these circumstances, the present result seems to be conformed to the general principle mentioned above.

Summary

1. The experiments were attempted to see whether there is any population effect on the daily periodic emergence of *Drosophila melanogaster* or not.

2. Various number — 1, 2, 8, 16, 32 and 64 individuals of the first instar

2) The temporal mode of emergence seen about the flies reared in the normal Pearl's medium in the second series of experiment (Table 5) does not strictly coincide with that seen about flies reared singularly in a test tube in the first series of experiment (Table 3). The cause of this discrepancy is not clear at present. Perhaps the difference of the season (the difference in the time of sunrise and sunset), when the experiments were executed, may be participated in.

larvae were put into small test tubes, each containing 1 cc of the Pearl's medium. The number of tubes and larvae employed are shown in Table 1. The number of emerging flies was counted at every 3 hours. The results obtained are shown in Tables 3 and 4. It can safely be said that as the population in a tube gets larger, so the time of maximum emergence is delayed in the course of a day, i. e., the population effect exists.

3. If the cause of this population effect is found in the lowered metabolism of larvae induced by the accumulation of excrements or deficiency of food, the similar phenomenon will be induced by providing food containing toxic amounts of $CuSO_4$. The second experiment was executed to inquire on this point. The first instar larvae were placed in small test tubes (an individual a tube) each containing 1 cc of the normal Pearl's medium or 1 cc of the Pearl's medium containing $CuSO_4$ at the rate of 20×10^{-6} Mol. The number of tubes and the corresponding number of larvae employed are shown in Table 2, and the resnlts obtained are indicated in Tables 5 and 6. It is clear that the time of emergence is delayed in the Cu-medium and the cause of this phenomenon may be the lowered metabolism due to the toxic action of $CuSO_4$. Thus, it can be said that the physiological cause is participated in this modification of the mode of daily periodic activity, and such factors as psychological or simple biotic (collision, etc.) seem to be scarcely influencing.

Literatures

Aschoff, J., 1954: Zeitgeber der tierischen Tagesperiodik. Naturwiss., **14**: 49-56.

Calhoun, J. B., 1945: Diel activity rhythms of the rodents, *Microtus ochrogaster* and *Sigmodon hispidus hispidus*. Ecol., **26**, 251-273.

Fujimoto, K., 1953: Diurnal activity of mice in relation to social order. Physiol. and Ecol., **5**; 97-103.

Kleitman, N., 1950: Biological rhythms and cycles. Physiol. Rev., **29**; 1-30.

McElroy, W. D., Glass, B., 1950: Copper metabolism. A symposium on animal, plant and soil relationships.

Mori. S., 1948: Rhythmic activities of animals.

——— 1949: Daily rhythmic phenomena in the life history of *Drosophila*. Seibutu, **4**; 121-128.

Ohsawa, W., Matutani, K., Tukuda, H., Mori, S., Miyadi, D., Yanagisima, S., Sato, Y., 1942: Sexual properties of the daily rhythmic activity in *Drosophila melanogaster*. Physiol. and Ecol., **5**; 26-30.

Park, O., Sejba, O., 1735: Studies in nocturnal ecology, IV. *Megarodacne heros*. Ecol., **16**; 164-172.

Poulson, D. F., 1950: Physiological genetic studies on copper metabolism in the genus *Drosophila*. Genetics, **35**; 684-685.