Kontyû, Tokyo, 53 (3): 516–519. September 25, 1985

Sperm Displacement in *Henosepilachna pustulosa* (Coleoptera, Coccinellidae)

Susumu NAKANO

Zoological Institute, Faculty of Science, Hokkaido University,
Sapporo, Hokkaido, 060 Japan

Abstract Sperm displacement in the phytophagous ladybird, *Henosepilachna pustulosa*, was studied using two morphologically distinct local forms. The percentage of offspring fertilized by the second male was 71.7%.

Multiple mating by females is common in various groups of insects (PARKER, 1970; WALKER, 1980). Since the females of these species can store a large amount of sperm for a long time without losing fertilization capacity, competition for fertilization between sperm from different males often occurs and usually results in the last sperm precedence in many insect species (PARKER, 1970). In the coccinellid subfamily Epilachninae, last sperm precedence was suggested in *Epilachna varivestis* by using chemosterilized males (WEBB & SMITH, 1968). In this paper, I show the sperm displacement in another epilachnine species, *Henosepilachna pustulosa* (KÔNO) (henceforth abbreviated as Hp), based on crossing experiments between two different phenotypes.

Materials and Methods

Hp consists of four morphologically distinct local forms (KATAKURA, 1981a). Both males and females of Hp are known to mate repeatedly, though a single mating is sufficient to fertilize eggs for more than one month (NAKANO, 1985). Two forms of Hp, named the nominate form (abbreviated as P-II) and the Sapporo form (P-III) (KATAKURA, 1974, 1981a), were chosen for the present study. P-II was collected at Kamuikotan, central Hokkaido, and P-III at Sankakuyama in Sapporo, both from thistle as eggs or larvae in late June, 1981. Adults obtained were sexed within two days after emergence. Then, males and females were reared in isolation more than two weeks for sexual maturation (*cf.* KATAKURA & NAKANO, 1979). The experimental procedure was as follows: A virgin female was put together with a male in a plastic case ($6.5 \times 5.5 \times 2.0$ cm). When the mating lasted more than 40 min., the female was isolated after the completion of mating, reared individually with sufficient food plant, and allowed to lay several egg masses. Percentage hatching of each egg mass was recorded. After having confirmed the fertility of eggs, the female was again mated with a male of the type different from the first

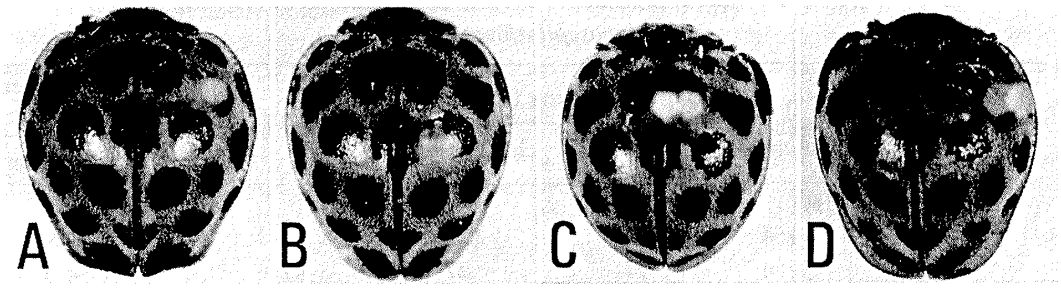

Fig. 1. Two local forms of *H. pustulosa* and their F_1 hybrids. A, P-II; B, F_1 hybrid between P-II♀ and P-III♂; C, P-III; D, F_1 hybrid between P-III♀ and P-II♂.

male in the same manner as before, and the percentage hatching of subsequent eggs was recorded untill death. All the larvae from the egg masses laid after the second mating were reared. The following four combinations of mating were tested: (A) P-II♀(P-II♂+P-III♂), (B) P-II♀(P-III♂+P-II♂), (C) P-III♀(P-III♂+P-II♂), (D) P-III♀(P-II♂+P-III♂), where the first and second male forms involved are shown in brackets. P-II, P-III and their F_1 hybrids can be distinguished on emergence by their elytral shapes (Fig. 1), though not during the immature stages. Thus, the degree of sperm precedence (the percentage of the eggs fertilized by the second male) was estimated by the numbers of hybrid and normal individuals in the offspring obtained after the second mating. Rearing and crossing were made under uncontrolled room conditions. The food plant used for rearing was *Solanum megacarpum*, one of the host plants of Hp in the field (KATAKURA *et al.*, 1977).

Results and Discussion

As shown in Table 1, there was no reduction of percentage hatching caused by the mating between different forms. In 12 out of 14 females, sperm displacement occurred. Although the degree of the second sperm precedence [b/(a+b)] was variable even in the same combination, the averages for the four combinations were similar. In females Nos. 7 and 10, sperm displacement did not occur at all. Ejaculation by the second male could not be confirmed in the present study. Further, females of Hp sometimes fail to accept sperm despite a prolonged mating (KATAKURA, unpubl.). Therefore, these females might not succeed in the second mating. When they are excluded and the four combinations combined, the overall average of the second sperm precedence is 71.1%. A similar result was obtained in *Epilachna varivestis* by using chemosterilized males (WEBB & SMITH, 1968).

Reviewing the sperm utilization in 30 insect and one mite species, WALKER (1980) pointed out the apparent correlation between the gross morphology of spermathecae or other sperm storage organs (e.g. seminal receptacle in *Drosophila*) and the degree of sperm precedence. According to him, species with spheroid

Table 1. Sperm precedence in four combinations of crossing in *Henosepilachna pustulosa*.

Type of combination and female no.	No. of eggs hatched	Offspring from		b/(a+b)	Average percent. hatching
		1st male (a)	2nd male (b)		before M_2*/after M_2*
P-II♀(P-II♂+P-III♂)					
1	164	15	122	89.1	80.5/85.2
2	64	17	27	61.4	61.5/76.3
3	75	21	17	44.7	64.4/62.6
average				65.1	68.8/74.7
P-II♀(P-III♂+P-II♂)					
4	83	15	55	78.6	43.8/52.0
5	77	11	31	73.8	72.4/79.8
6	47	19	18	48.6	38.6/43.6
7	89	69	0	0.0	60.1/51.4
average				50.3 (67.0)**	53.7/56.7
P-III♀(P-III♂+P-II♂)					
8	46	3	21	87.5	37.8/53.8
9	135	26	91	77.8	52.4/74.8
10	115	99	0	0.0	64.7/84.4
average				55.1 (82.7)**	51.6/71.0
P-III♀(P-II♂+P-III♂)					
11	83	1	30	96.8	79.0/78.6
12	67	11	43	79.6	89.9/73.3
13	185	51	80	61.1	75.8/87.1
14	34	9	6	40.0	75.7/87.8
average				69.4	80.1/81.7

* M_2=second mating. ** Average degree of sperm precedence when excluding females showing no displacement.

spermathecae show the first sperm precedence, while those with elongate or tubular ones show the last sperm precedence (WALKER, 1980). This trend is understandable if it is assumed that in elongate spermathecae it is easier to displace prior sperm into a region of the spermathecae where they cannot easily reach the spermathecal duct (WALKER, 1980). In the case of *E. varivestis* quoted by him or Hp in the present study, his inference is not supported since they show the last sperm precedence in spite of having spheroid spermathecae. According to KATAKURA (1981b), however, the so-called spermatheca of the Epilachninae is generally spheroid but vestigial and seems to have lost its primary function. As a substitute sperm storage organ, they have a pair of swellings located in the common oviduct (KATAKURA, 1981b). The inner side of this organ is roughly divided into two larger lateral parts (corresponding to the swellings) and a narrower part connecting them (KATAKURA, 1981b). This unique sperm storage organ may be similar in function to the elongate type.

Acknowledgments I am grateful to Prof. M. YAMADA, Zoological Institute, Hokkaido University and Prof. Sh. F. SAKAGAMI, Institute of Low Temperature Science, Hokkaido University, for their critical reading of the manuscript. Thanks are extended to Dr. H. KATAKURA and an anonymous reviewer whose valuable comments improved the manuscript.

References

KATAKURA, H., 1974. Morphology and distribution of the forms of *Henosepilachna vigintioctomaculata* complex in Hokkaido. *J. Fac. Sci. Hokkaido Univ. Ser. VI. Zool.*, **19**: 657–680.

———— 1981 a. Classification and evolution of the phytophagous ladybirds belonging to *Henosepilachna vigintioctomaculata* complex (Coleoptera: Coccinellidae). *Ibid.* **22**: 301–378.

———— 1981 b. Sperm storage place in adult females of the phytophagous ladybirds belonging to *Henosepilachna vigintioctomaculata* complex (Coleoptera: Coccinellidae). *Kontyû, Tokyo*, **49**: 477–481.

————, H. HINOMIZU, K. HOSHIKAWA & T. KIMURA, 1977. Problems and methods on the food habits of *Henosepilachna vigintioctomaculata* complex. *Seibutsu Kyôzai*, **12**: 61–92. (In Japanese.)

———— & S. NAKANO, 1979. Preliminary experiments on the crossing between two puzzling phytophagous ladybirds, *Henosepilachna vigintioctomaculata* and *H. pustulosa* (Coleoptera: Coccinellidae). *Kontyû, Tokyo*, **47**: 176–184.

NAKANO, S., 1985. Effect of interspecific mating on female fitness in two closely related ladybirds (*Henosepilachna*). *Kontyû, Tokyo*, **53**: 112–119.

PARKER, G. A., 1970. Sperm competition and its evolutionary consequences in insects. *Biol. Rev.*, **45**: 525–567.

WALKER, W. F., 1980. Sperm utilization strategies in nonsocial insects. *Amer. Natur.*, **115**: 780–799.

WEBB, R. E. & F. F. SMITH, 1968. Fertility of eggs of Mexican bean beetles from females mated alternately with normal and apholate-treated males. *J. Econ. Ent.*, **61**: 521–523.