Comparative Biology of the Two Scelionid Egg
Parasites of *Nezara viridula* L.
(Hemiptera: Pentatomidae)

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The general bionomics of the principal egg parasites of the southern green stink bug, *Nezara viridula* L., namely *Asolcus mitsukurii* ASHMEAD and *Telenomus nakagawai* WATANABE, were studied. *A. mitsukurii* differs considerably from *T. nakagawai* in the following points. 1) *A. mitsukurii* is polyphagous and prefers the pentatomids which deposit eggs in a small egg mass, while *T. nakagawai* is oligophagous and prefers large egg mass of pentatomids such as *N. viridula* or *N. antennata*. 2) *A. mitsukurii* is bisexual and the first egg deposited by a mated female was always a male, but *T. nakagawai* is unisexual producing female progeny by parthenogenesis. 3) Both sexes of *A. mitsukurii* have aggressive behavior, but *T. nakagawai* is not aggressive. 4) The developmental period of the female *A. mitsukurii* at 25° C was shorter by about 3 days than that of *T. nakagawai*, but the female of the latter was more prolific and lived longer than the former. These specific properties were discussed in comparison with those of other egg parasites of *Nezara*.

INTRODUCTION

KIRITANI and HOKYO (1962) stressed the importance of the two scelionid egg parasites *Asolcus mitsukurii* ASHMEAD and *Telenomus nakagawai* WATANABE as one of the possible controlling factors of the population of *Nezara viridula* L. This is particularly true in the first generation of the host where these parasites cause a high mortality of *Nezara* eggs. HOKYO and KIRITANI (1963), however, suggested that a severe interspecific interference between adult females might result in a decrease of parasitic efficiency of *T. nakagawai* in the field. Furthermore, there exist marked differences between the two concerning their ovipositional behavior (HOKYO and KIRITANI, 1966). In the present paper, the general bionomics of these parasites will be reported mainly based on the results obtained from laboratory study.

RESULTS

1. *Egg parasites and their host range*

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Three minor egg parasites other than *A. mitsukurii* and *T. nakagawai* have been recorded from *Nezara* eggs (Table 1). The first, *Telenomus gifuensis* is known

Table 1. A LIST OF HOSTS OF THE EGG PARASITES OF THE SOUTHERN GREEN STINK BUG, *Nezara viridula* L.

Parasite species	Body length(mm)		Sex ratio	Hosts
	male	female	(♀/♂+♀)	
<i>Asolcus mitsukurii</i> ASHMEAD	1.03	1.12	0.75 ^b	<i>Nezara viridula</i> L. ^b , <i>N. antennata</i> SCOTT ^{b,d} , <i>Dolycoris baccarum</i> L. ^d , <i>Gonopsis affinis</i> UHLER ^d , <i>Lagynotmus elongatus</i> DALLAS ^d , <i>Piezodorus hybneri</i> GMELIN ^d , <i>Scotinophara</i> <i>lurida</i> BURMEISTER ^b , <i>Eusarcoris</i> sp. ^b
<i>Telenomus nakagawai</i> WATANABE		1.16	0.98 ^b	<i>N. viridula</i> L. ^b , <i>N. antennata</i> SCOTT ^{b,d} , <i>S. lurida</i> BURMEISTER ^b
<i>Telenomus gifuensis</i> ASHMEAD	1.11	1.25 ^a	0.80 ^a	<i>S. lurida</i> BURMEISTER ^d , <i>D. baccarum</i> L. ^d , <i>Eusarcoris parvus</i> UHLER ^a , <i>Eusarcoris</i> <i>guttiger</i> THUNBERG ^a , <i>N. viridula</i> L. ^b , <i>N.</i> <i>antennata</i> SCOTT ^a , <i>Anacanthocoris</i> <i>concoloratus</i> UHLER ^{*a} , <i>Riptortus clavatus</i> THUNBERG ^{*a}
<i>Ooencyrtus nezarae</i> ISHII	0.83 or less ^c		see text	<i>N. viridula</i> L. ^c , <i>N. antennata</i> SCOTT ^{c,d} , <i>A. concoloratus</i> UHLER ^{*d}
<i>Anastatus japonicus</i> ASHMEAD	1.37 ^{***c}	2.81 ^{***c}	see text	<i>Lymantria dispar</i> L. ^e , <i>Dendrolimus spectabilis</i> BUTLER ^e , <i>Malacosoma neustria testacea</i> MOTSCHULSKY ^e , <i>N. viridula</i> L. ^e , <i>N.</i> <i>antennata</i> SCOTT ^e

a : HIDAOKA (1958) ; b : HOKYO and KIRITANI (1963) ; c : HOKYO (1965) ;

d : YASUMATSU and WATANABE (1964) ; e : HIROSE (Personal communication).

*These two hosts belong to the family Coreidae, but the others belong to Pentatomidae excluding lepidopterous hosts: *L. dispar*, *D. spectabilis* and *M. neustria testacea*.

**Only males emerged from the parasitized eggs of *N. viridula*.

***Females collected from the field.

as an effective egg parasite of *Scotinophara lurida* (HIDAOKA, 1958). The second, *Ooencyrtus nezarae* is the smallest species among the three but its detailed biology will be investigated in future. The third, *Anastatus japonicus*, which is closely resemble *Anastatus disparis* in Europe, is known as an egg parasite of the gypsy moth, *Lymantria dispar*. This species also parasitizes the eggs of the pine moth, *Dendrolimus spectabilis* and has been wrongly identified as *Anastatus bifasciatus* (HIROSE, personal communication). The above two lepidopterous hosts lay eggs much larger in volume than those of *Nezara*. *A. mitsukurii* and *T. nakagawai* both parasitize pentatomid eggs, but *A. mitsukurii* is far wider in host range than *T. nakagawai*. Excluding *Nezara* which lays eggs of 70–100 as a mass in *N. viridula* and 50–80 in *N. antennata*, the other hosts lay eggs in a small egg mass, e. g. 14 eggs per mass in *S. lurida* (KIRITANI and HOKYO, 1965 and unpublished data). *T. gifuensis* as *A. mitsukurii* has a wide range of hosts which lay eggs in small masses.

2. Morphology and other specific differences

1) Variation in the coloration of parasitized eggs, meconium and cocoon membrane, and the shape of emergence hole

VOEGELÉ (1964) pointed out that the pigmentation which occurred inside the host egg with development of the parasite is specific and characteristic of each species of *Asolcus* which parasitizes the eggs of the pentatomids: *Eurygaster integriceps* and *Graphosoma lineata*. The creamy white egg of *N. viridula* normally turns gradually deep red with the development of the embryo. When parasitized, the color of the egg turns dark grey within a few days. It was, however, difficult to distinguish between *A. mitsukurii* and *T. nakagawai* by the external coloration of parasitized eggs, although the eggs parasitized by *A. mitsukurii* are somewhat brownish in color in contrast with blackish color of the eggs parasitized by *T. nakagawai*.

The most clear cut difference between the two is the color of the meconium and cocoon membrane and the shape of emergence hole of the parasite. Meconium and cocoon membranes are respectively brown and transparent in *A. mitsukurii*, while creamy white and brown in *T. nakagawai*. On emergence, the egg cap is broken along the circular arrangement of micropyle. The shape of emergence hole of *A. mitsukurii* is circular but it is semi-circular in *T. nakagawai* (Fig. 1).

Fig. 1. Shapes of operculum broken on hatching of the host and on emergence of the parasite. 1. host, 2. *A. mitsukurii*, 3. *T. nakagawai*.

The mean lengths of the shortest diameter of emergence hole are 0.56 mm in *A. mitsukurii* and 0.40 mm in *T. nakagawai*. These coincided well with the mean widths of the prothorax of the respective species (Table 2).

Table 2. THE MEAN WIDTH OF PROTHORAX AND THE MEAN LENGTH OF THE SHORTEST DIAMETER OF EMERGENCE HOLE OF THE PARASITE

Species	No. examined	Width of prothorax (mm) mean $\pm$ 99% C.L.	Length of the shortest diameter (mm) mean $\pm$ 99% C.L.
<i>A. mitsukurii</i>	10	0.55 $\pm$ 0.02	0.56 $\pm$ 0.02
<i>T. nakagawai</i>	10	0.41 $\pm$ 0.01	0.40 $\pm$ 0.02

2) Ovaries and ovarian eggs

A diagrammatic presentation of the ovaries and ovarian eggs is given in Fig. 2. The ovaries of both species are similar in appearance. Egg tubes or ovarioles were not detectable, and the matured eggs filled the ovaries at the time of emergence.

Fig. 2. Shapes of ovary and ovarian egg of the parasite.

The type of ovarian eggs of both species belonged to the stalked type which is common in Scelionidae (CLAUSEN, 1962). The ovarian eggs of *A. mitsukurii* were significantly greater both in length and width than those of *T. nakagawai* (Table 6).

3. Duration of developmental period, sex ratio and sex determination

The mean durations of the two species required for the development from egg deposition to emergence are shown in Table 3. The mean duration of males of

Table 3. MEAN DURATIONS REQUIRED FOR THE DEVELOPMENT FROM EGG DEPOSITION TO THE FIRST DAY OF THE PARASITE EMERGENCE ^a

Species	Year	Sex	No. of replications	Duration (days) mean $\pm$ 99% C.L.
<i>A. mitsukurii</i> ^b	1962	male	6	11.17 $\pm$ 0.67
		female	7	12.29 $\pm$ 0.70
	1965	male	13	10.31 $\pm$ 0.72
		female	15	12.20 $\pm$ 0.60
<i>T. nakagawai</i> ^c	1962	female	7	15.29 $\pm$ 1.06

^a Experiment was conducted at 25°C and 15 hr illumination in both years of 1962 and 1965. Relative humidity was kept at 75 per cent in 1965 but it per cent fluctuated between 70-90 per cent in 1962.

^b Bisexual species.

^c Unisexual species.

A. mitsukurii was 1-2 days shorter than that of females. The development of females of *T. nakagawai* required a significantly longer time than those of *A. mitsukurii*.

A. mitsukurii is an arrhenotokous species; fertilized eggs giving rise to females, but males are produced by parthenogenesis. On the other hand, *T. nakagawai* is a deuterotokous species; usually females are produced by parthenogenesis. The sex ratios ($\frac{\text{♀}}{\text{♂} + \text{♀}}$) of the two species are shown in Table 1. As regards the other parasites, *T. gifuensis* is arrhenotokous. And *Ooencyrtus nezarae* is bisexual (HOKYO, 1965). *Nezara* eggs parasitized by *Anastatus japonicus* gave

rise to only males (see foot-note of Table 1). When a field born female of *A. japonicus* was allowed experimentally to oviposit on *Nezara* eggs, all the ensuing progeny were males. But, when the same female was allowed to parasitize the eggs of *Halyomorpha picus*, whose eggs are three times as great as those of *Nezara*, 7 females and 5 males emerged from 12 parasitized eggs. Usually, this species gives rise to both sexes when laid on the eggs of *L. dispar*, and of *D. spectabilis*, but only males have been obtained from the eggs of the tent caterpillar, *Malacosoma neustria testacea* that are smaller than the eggs of the former two hosts (HIROSE, personal communication).

Fresh egg masses of *N. viridula* were divided into three groups of smaller size: one, two and twelve eggs per group. In each experiment, one mated female of *A. mitsukurii* of one or two days old, was allowed to oviposit on the egg(s) for 24 hr at 25°C with a 15 hr illumination period (Table 4). The percentage

Table 4. SEX DETERMINATION OF *A. mitsukurii* IN RELATION TO THE NUMBER OF HOST EGGS ALLOWED TO PARASITIZE

No. of host eggs per mass	No. of replications	Total No. of eggs	No. of eggs parasitized (%)	No. of adults emerged		Sex ratio (♀/♂+♀)
				male	female	
1	32	32	5(15.6)	5	0	0.00
2	6	12	4(33.3)	3	1	0.25
12	14	168	158(94.0)	19	128	0.87

parasitism of eggs and the sex ratio (♀/♂+♀) of the progeny increased with an increase in size of egg mass. The low percentage parasitism of isolated and paired eggs was due to the difficulty for the parasite to oviposit on such an extraordinarily small egg mass. The fact that only males emerged from the isolated egg group indicated that the first egg deposited by the parasite was a male. This was also supported by the cases in which egg pairs were used; When only one of the two eggs was parasitized, only one male emerged. But, when both eggs were parasitized one male and one female emerged. The fact that males arise from first egg deposited during an ovipositional cycle is again illustrated in the experiment in which egg masses containing 12 eggs were used. In this experiment, a total of 14 egg masses were used, and in 9 of these masses, all the eggs were parasitized. When adults emerged and the sexes were checked, 1 male and 11 females had emerged from each of these 9 egg masses.

4. Longevity and fecundity

The mean longevity and fecundity of the parasites are shown in Table 5. When only water was supplied, the mean adult longevity of the parasites was 2-3 days irrespective of the species and the sex. A marked prolongation in the mean longevity was observed when honey was given. The increase in mean longevity was as much as 2-3 times in *A. mitsukurii* and about 5 times in *T. nakagawai*. The mean longevity of the females is significantly longer in *T. nakagawai* than in *A. mitsukurii*. The mean longevity of the females was shortened by about 5 days in *T. nakagawai* when a fresh egg mass of *N. viridula* was supplied daily for oviposition. This was not the case with *A. mitsukurii* which showed no ill

Table 5. THE MEAN LONGEVITY IN DAYS AND FECUNDITY OF THE PARASITE AT 25°C AND 75% R.H. UNDER 15 HR ILLUMINATION

Treatment	Item	<i>A. mitsukurii</i>		<i>T. nakagawai</i>
		male	female	female
Water without hosts	longevity	2.14±1.26 ^a (7) ^b	2.50±0.76(26)	3.36±0.75(14)
Honey without hosts	"	5.25±2.86 (8)	8.94±1.70(31)	15.45±1.15(20)
Honey with hosts	"		8.40±1.13(5)	10.60±1.84(5)
—ditto—	fecundity		65.40±17.45(5)	102.60±52.46(5)

^a Mean±99% C.L.

^b Number of replications.

effect due to the oviposition.

The mean fecundity of females fed on honey was estimated from the total number of host eggs parasitized during the life span of individual females (Table 5). *T. nakagawai* lays 1.6 times as many eggs as *A. mitsukurii*, but the difference between the mean is not significant probably due to the small number of replications. The survival curve and the daily oviposition trends of both species are shown in Fig. 3. The latter is expressed by plotting the mean numbers of eggs/

Fig. 3. Daily oviposition trend and survival curve of the parasite.

female/day with their standard errors. The mean numbers of eggs laid on the first day were 43 in *T. nakagawai* and 40 eggs in case of *A. mitsukurii*. These figures coincide well with those for the ovarian eggs of each species (Table 6). The oviposition rate of both species abruptly decreased after the second day. Thereafter a small number of eggs were laid continually until the sixth day for *A. mitsukurii* or in the case of *T. nakagawai*: the eighth day. All the females used lived a short period after the end of oviposition. These facts suggested that the subsequent formation of some ovarian eggs after the first oviposition does take

Table 6. NUMBER AND SIZE OF OVARIAN EGGS ON EMERGENCE OF THE PARASITE

Species	No. dissected	No. of ovarian eggs ^a	No. of eggs measured	Length of stalk ^a (mm)	Length of egg body ^a (mm)	Width of egg body ^a (mm)
<i>A. mitsukurii</i>	16	42.96±4.21	10	0.087±0.007	0.137±0.006	0.063±0.010
<i>T. nakagawai</i>	12	47.25±5.98	5	0.073±0.013	0.103±0.011	0.033±0.003

^a Mean±99% C.L.

place if the parasites were allowed to feed on honey. The survival curves of both species belonged to the so-called "*Drosophila* type" (PEARL and PARKER, 1921).

DISCUSSION

A. mitsukurii and *T. nakagawai* are the two major egg parasites of *N. viridula* as it occurs in the southern parts of Japan. The present study has shown that they are different from each other in several points: sex ratio, developmental period, fecundity, longevity etc.

It seems that the biological and behavioral characteristics of *A. mitsukurii* resemble those of *T. gifuensis* than *T. nakagawai* except for the host preference (Table 1). 1) The former two species are arrhenotokous but the latter is deuterotokous. 2) Both sexes of the former two have aggressive behavior but this is apparently absent in the latter (HIDAKA, 1958; HOKYO and KIRITANI, 1966). 3) The former two species are polyphagous and prefer to parasitize small egg masses of several pentatomids but the latter is oligophagous and prefers to parasitize large egg masses of *Nezara*. In this respect, HOKYO and KIRITANI (1966) have shown that ovipositional behavior of *T. nakagawai* is better adapted to large size of host egg masses, but that of *A. mitsukurii* to small size of host egg masses.

CUMBER (1964) noted that in New Zealand, a native *Asolcus* sp. is unable to attack egg masses of *N. viridula* but will attack small egg masses of five other pentatomids. An introduced parasite of *N. viridula*, *Asolcus basalis*, on the other hand, can also utilize all these species as hosts. Further, females of the native *Asolcus* are more aggressive than those of *A. basalis*. All these lines of evidence suggest that the development of aggressive behavior in females is closely related to the size of host egg mass.

On the sex determination of arrhenotokous hymenopterous parasites, FLANDERS (1939) stated that an increase in the rate of oviposition probably increases the number of eggs escaping fertilization. However, the reverse is the case for *Caraphractus cinctus*, an egg parasite of Dytiscidae (JACKSON, 1958). In this case an increase in the rate of oviposition causes an increase in the proportion of fertilized eggs. The present study suggests that the sex of *Anastatus japonicus* is probably determined by the volume of the host eggs. OKADA and MAKI (1934, cited from CLAUSEN, 1962) noted that in *Telenomus dignus* (*Phanurus beneficiens*), an egg parasite of the rice stem borer, *Chilo suppressalis* in Japan, males develop abundantly from the first and the last eggs laid by the female parasite. CUMBER (1964) mentioned that male producing eggs of *Asolcus basalis* might be laid in the

earlier stages of oviposition. In *A. mitsukurii*, the first egg laid by a mated female was consistently male. This is very interesting taking into account the fact that the first male of *Asolcus* to emerge from a parasitized egg mass drives the other males away in order to mate with females which emerge later (SAFAVI, 1964).

The percentage parasitism of *Nezara* eggs by these two species is high in the first generation of the host, but it drops abruptly in the following two generations. This is considered to be due to the differential ability of dispersal between the host and parasites (KIRITANI and HOKYO, 1962). In the laboratory, the shortest duration from egg to the egg deposition of the female of *N. viridula* is 56 days at 25°C (KIRITANI, 1963). While, the duration from egg to the death of the female parasite is at most 21 and 26 days for *A. mitsukurii* and *T. nakagawai*, respectively at the same temperature (see Table 5). Therefore, *A. mitsukurii* may repeat at least 11 generations and *T. nakagawai* 9 generations a year.

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