JARQ 31, 101–108 (1997)

Use of Coccinella *septempunctata brucki* Mulsant as a Biological Agent for Controlling Alfalfa Aphids

Keiichi TAKAHASHI*

Department of Environment, National Grassland Research Institute
(Nishinasuno, Tochigi, 329-27 Japan)

Abstract

Ecology, mass production and release of a lady beetle, *Coccinella septempunctata brucki*, were investigated in Japan to use the beetle as a biological agent for controlling alfalfa aphids. In the alfalfa field, the number of overwintered aphids, *Acyrthosiphon kondoi*, increased on alfalfa in April and the occurrence from late April to mid-May showed a marked peak. Without *C. septempunctata brucki*, the reduction of alfalfa yield by aphids amounted to 40 to 70%. The overwintered adults of C. *septempunctata brucki* appeared in early March on the alfalfa field and began predation and oviposition. In total about 20% of the egg population developed to adults successfully. For mass production, A. pisum population was reared on Vicia faba as prey for the lady beetle. The overwintering adult lady beetle was used to obtain the eggs for mass production. The optimum development stages for release to the alfalfa fields where C. *septempunctata brucki* was absent were the 2nd, the 3rd and the 4th instar larvae in early March when the maximum temperature was around 10°C. The Japanese pampas grass, *Miscanthus sinensis*, was planted near the alfalfa field for keeping lady beetle populations in winter and summer as their overwintering and estivating sites.

Discipline: Insect pest
Additional key words: biological control, *Acyrthosiphon kondoi*

Introduction

Alfalfa, *Medicago sativa*, is a superior forage crop because of its high content of nutrients, high dry matter production and high palatability. Alfalfa was introduced to Japan in 1874 from the USA. The area of alfalfa cultivation in Japan which is about 12,000 ha is increasing[8]. In Japan, heavy rain is conducive to outbreaks of pest insects and diseases for alfalfa cultivation[1,5]. *Therioaphis trifolii* and *Acyrthosiphon pisum* have been the major pest aphids of alfalfa for a long time in the world. However, recently *A. kondoi* has replaced these aphids and has become the most harmful insect for alfalfa[3]. *A. kondoi* damages alfalfa by sucking and virus transmission, and its honeydew induces the occurrence of spring black stem and leaf spot by *Phoma medicaginis*.

Application of chemicals for controlling aphids is limited in grasslands and there is no effective method of cultural control except for harvesting the aphid-infested alfalfa. Although some resistant varieties, CUF101 for example, have been developed[2], they do not cover the resistance to all races of *A. kondoi*. Many natural enemies of aphids are known, including lady beetles, chrysophids, syrphids, parasitoids and fungi[4]. Biological control of aphids using lady beetles appears to be sound in the absence of other effective methods of control. In this paper *Coccinella septempunctata brucki* which occurs mainly in grasslands was investigated as a suitable biological agent for controlling alfalfa aphids. Most of the following studies were carried out in fields of the National Grassland Research Institute in Tochigi Prefecture, Japan.

Aphids in alfalfa fields

Fig. 1 shows the seasonal occurrence of aphids

Present address:
* Okinawa Subtropical Station, Japan International Research Center For Agricultural Sciences (Ishigaki, Okinawa, 907 Japan)

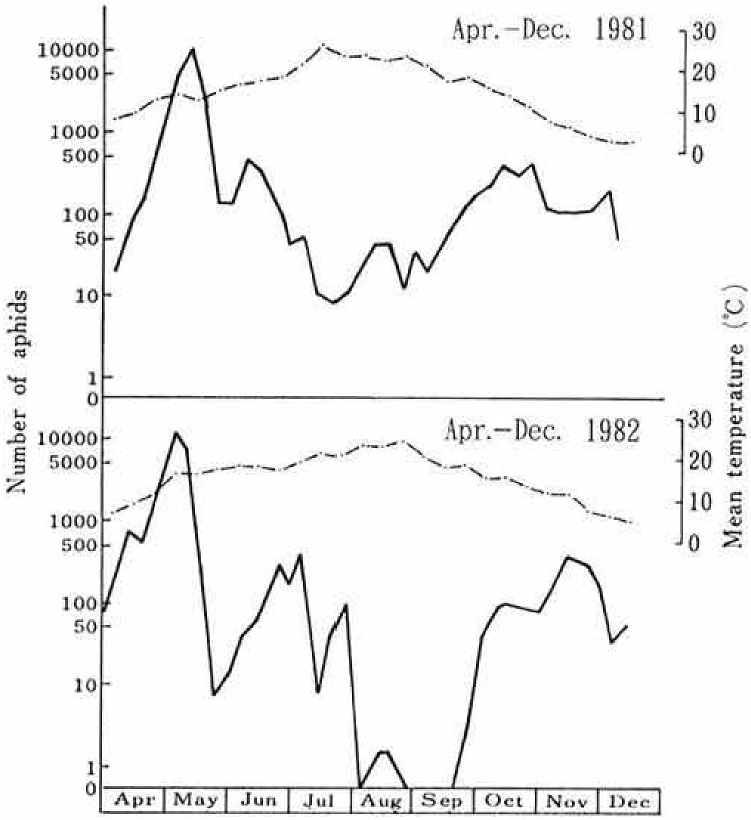

Fig. 1. Seasonal occurrence of aphids, mainly *Acyrthosiphon kondoi*, in the alfalfa field
Average number of aphids per row (0.6 × 8 m) collected by beating.

in 1981 and 1982 in the alfalfa field. Almost all of the collected aphids in the alfalfa field consisted of *A. kondoi*[9]. The number of overwintered *A. kondoi*, mainly viviparous females, increased in April. Occurrence from late April to mid-May showed a marked peak and then decreased. In summer few aphids were observed. A small peak was observed in late autumn. The optimum temperature for reproduction of this aphid was in the range of 10-15°C.

The yield loss of alfalfa caused by aphids was investigated in Tsukuba, Ibaraki Prefecture from 1992 to 1993. Without *C. septempunctata brucki*, the yield loss of alfalfa amounted to 40 to 70%. Earlier attack of the aphids enhanced the damage[7].

C. septempunctata brucki in alfalfa fields

The major species of lady beetles preying on alfalfa aphids was *C. septempunctata brucki* (Fig. 2). The overwintered adults appeared early in March in the alfalfa field and began predation and oviposition. *Harmonia axyridis* was also common and appeared in the field from May. The larvae of *H. axyridis* began to appear in late May when the population of aphids decreased drastically. *Propylea japonica* was also found but not as abundantly as the other 2 species.

Other natural enemies, chrysophids, syrphids, parasitoids and fungi were also recognized. However, the most effective natural enemy for alfalfa aphids was assumed to be *C. septempunctata brucki* because of its early emergence and high population density.

Overwintering *C. septempunctata brucki* adults were detected in mounds and bunches of Japanese pampas grass, *Miscanthus sinensis*, in February[14]. Most of the adults were found in the south- and east-facing mounds, where the air temperature was 2-6°C higher and there were more aphids than in the north-facing mound. Adult beetles started to migrate to the alfalfa field infested with aphids in early spring, when the daily maximum temperatures exceeded 12°C, but only a few adults migrated to

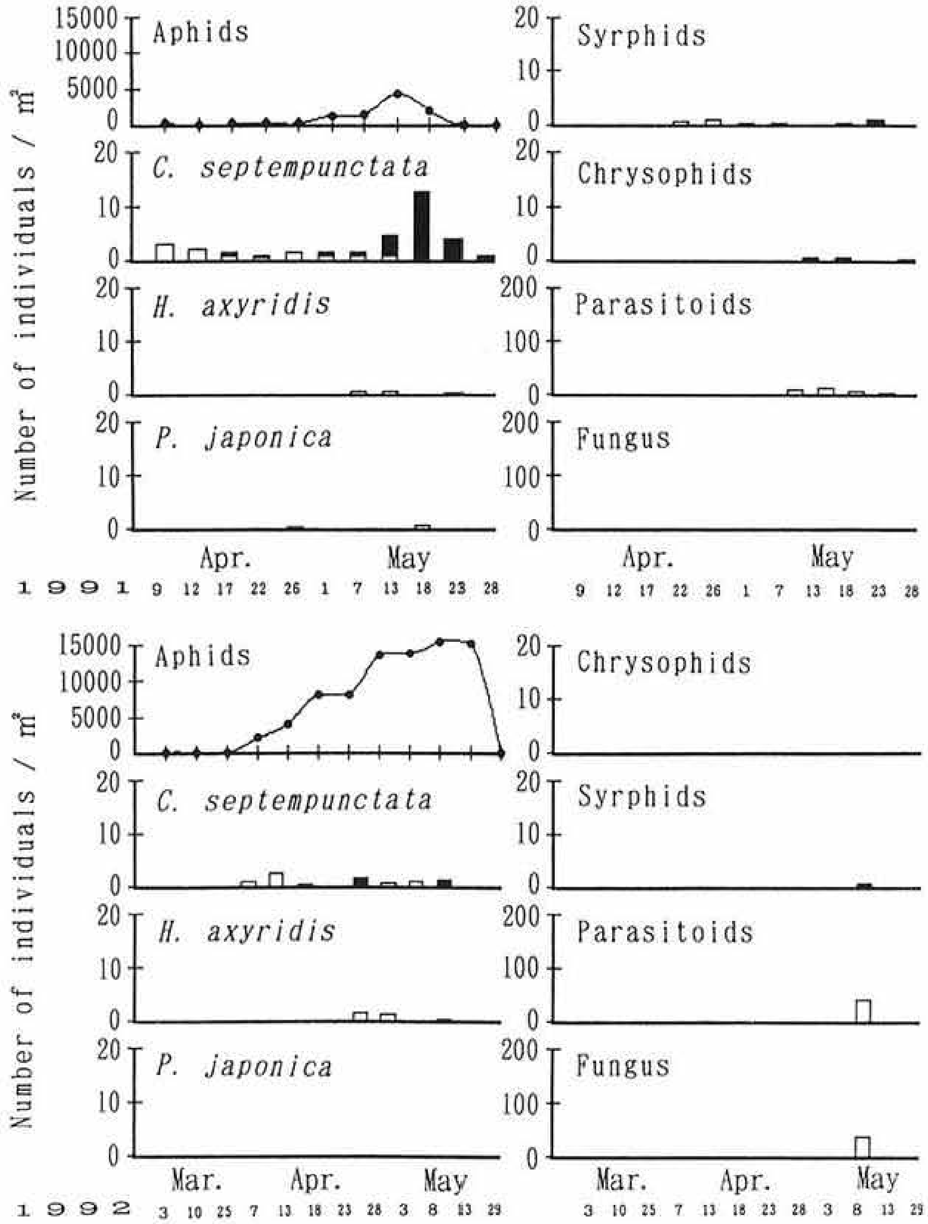

Fig. 2. Natural enemies of aphids in the spring alfalfa field
□ : Adults, ■ : Larvae.

the field where the aphids had been eliminated. Some of the adults marked in their overwintering sites were recaptured in the alfalfa fields. The recapture ratio was higher in the population marked in the hibernation sites near the alfalfa fields. In August, estivating adults were found mainly in bunches of Japanese pampas grass and some of the adults marked in August were recaptured in the alfalfa fields in the next spring.

The activity of the adults of *C. septempunctata brucki* in the spring alfalfa field was affected by several factors[15]. Above 10°C, adults were active only in the daytime when the luminosity exceeded 0.2 $MJ/m^2/0.5$ h, and the wind velocity was less than 10 m/s. Predation, oviposition and mating were observed constantly in the daytime. After sunset, the insects remained at the base of the alfalfa plants. Density of the aphid population also affected the intensity of their activity: the lower the density, the more active they were.

The overwintered adults of *C. septempunctata brucki* laid their eggs mainly on the undersides of dead leaves, mainly those of *Quercus serrata* and other plants (Fig. 3). *C. septempunctata brucki*

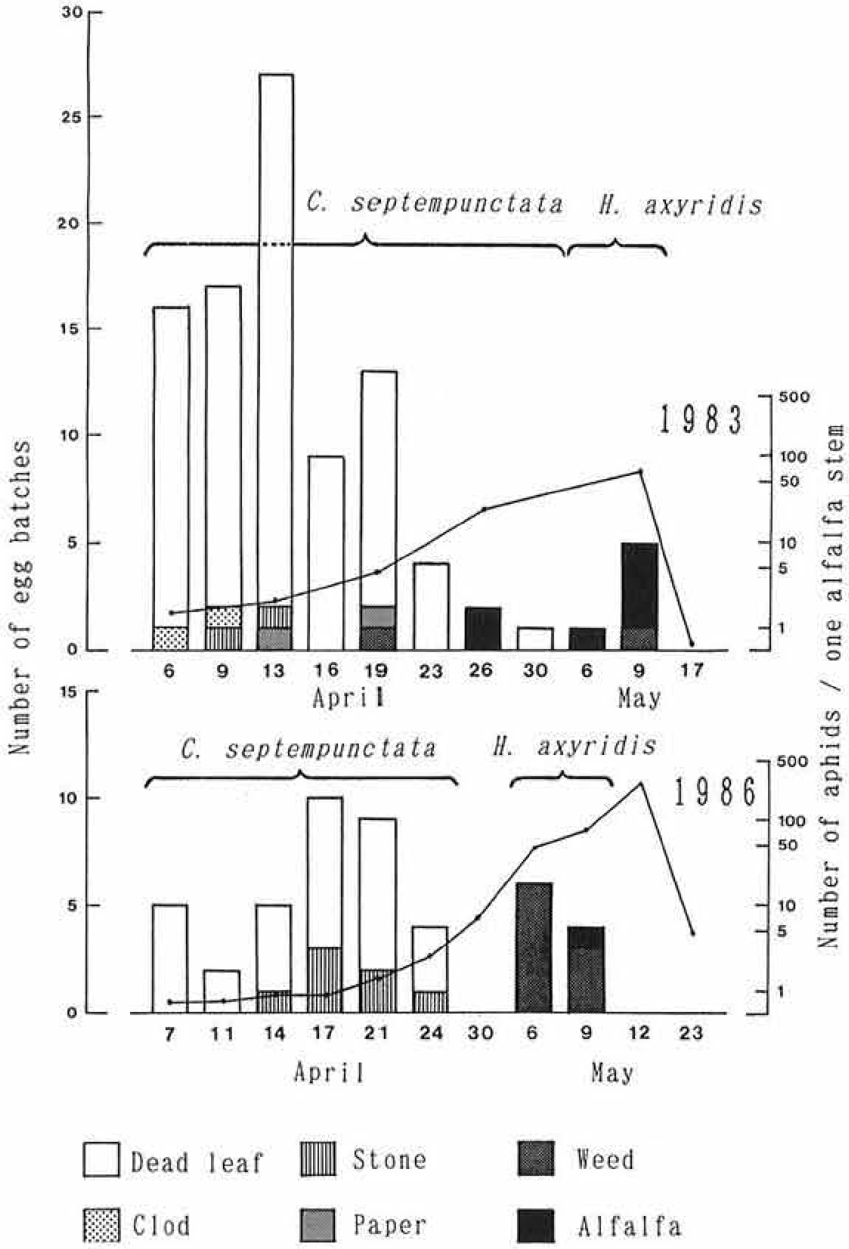

Fig. 3. Seasonal changes of oviposition sites of lady beetles in the alfalfa field

oviposited when the aphid density was still low, less than 1 aphid per stem. This oviposition behavior appeared to enable them to avoid the shortage of prey on a small plant and then the larvae could disperse rapidly to find aphids on different plants[12]. The oviposition sites were also adequate to avoid cannibalism by the larvae and may be heated easily by sunrays.

On the contrary, *H. axyridis* laid their eggs mainly on the alfalfa leaves, when the adults and the hatched larvae could feed on a large number of aphids on the plant.

The distribution of the larvae and adults of *C. septempunctata brucki* showed a negative correlation with the distribution of aphids in early May, presumably due to the effect of the predation of aphids by *C. septempunctata brucki*. The 3rd instar larvae dispersed rapidly to other alfalfa plants when there were enough aphids around them.

Intra- and inter-specific predation of lady beetles was found in spring alfalfa fields[13]. All the egg predations consisted of sibling cannibalism. Besides

Table 1. Life table of *C. septempunctata brucki* on alfalfa

Age interval (x)	No. of individuals alive at the beginning of $x(1x)$	Factors of dx (dxF)	No. of individuals dying during $x(dx)$	dx %
(1992)				
Egg	280	Cannibalism, Unknown	74	26.4
1st instar	206	Unknown	117	56.8
2nd instar	89	Unknown	20	22.5
3rd instar	69	Unknown	13	18.8
4th instar	56	Hail, Unknown	54	96.4
Pupa	2	Hail	2	100
Adult	0	–	–	–
(1993)				
Egg	280	Cannibalism	59	21.1
1st instar	221	Unknown	107	48.4
2nd instar	104	Unknown	15	14.4
3rd instar	89	Unknown	3	3.4
4th instar	86	Unknown	20	23.3
Pupa	66	Unknown	20	30.3
Adult	56	–	–	–

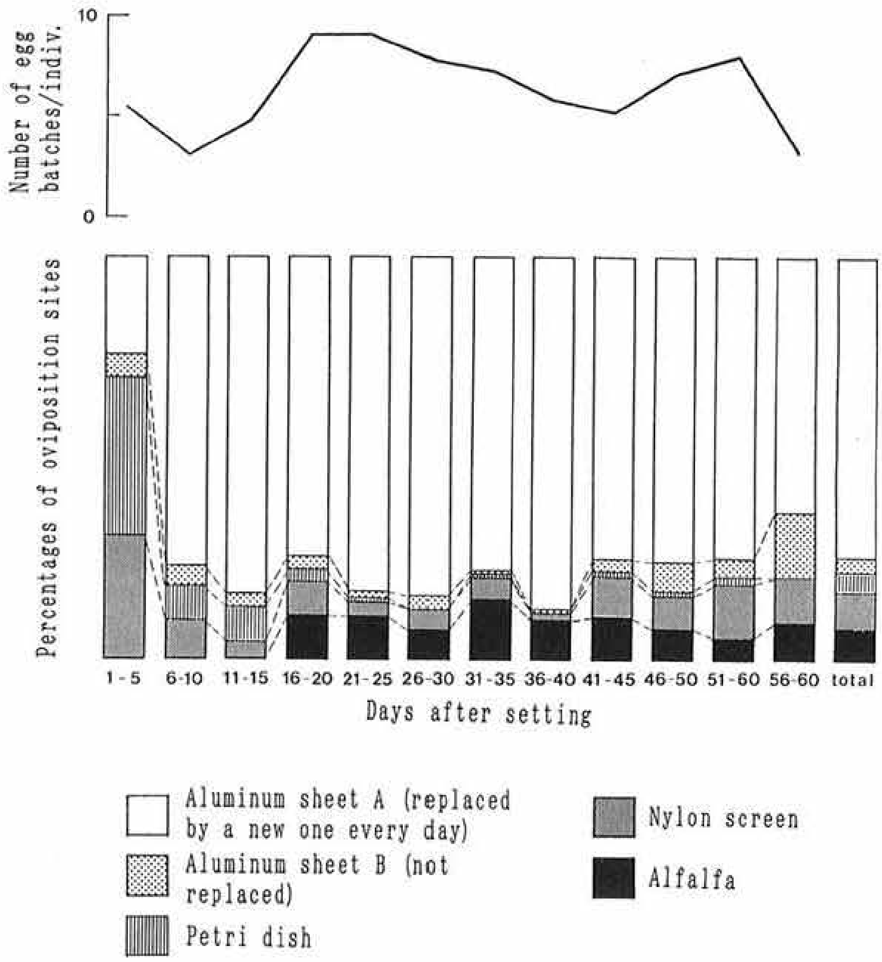

Fig. 4. Selection of oviposition sites of *C. septempunctata brucki* in the laboratory

egg cannibalism, intra- and inter-specific pupal and prepupal predation was observed as the larvae of *H. axyridis* which were detected most frequently attacked the pupae or prepupae of *C. septempunctata brucki*. The inter-specific predation between *H. axyridis* and *C. septempunctata brucki* can be attributed to the difference in the oviposition period of both species and the lack of aphids.

Life tables of *C. septempunctata brucki* were constructed in the spring of 1992 and 1993 (Table 1). The rate of sibling cannibalism was around 20, 60% of the 1st instar larvae were likely to die of starvation and about 20% of the egg population developed into adults successfully. It was assumed that the main predator for *C. septempunctata brucki* was *H. axyridis*.

Mass production of *C. septempunctata brucki*

Many kinds of artificial diets have been tested for lady beetles. Although the adults of *C. septempunctata brucki* eat powder of drone honeybee, the powder is not adequate for their oviposition[6]. Only aphids have been found to be the optimum diet for all the development stages of *C. septempunctata brucki* so far. The adult beetles ate frozen and freeze-dried aphids, but egg production was lower than when live aphids were used.

As a prey for the adult, *A. pisum* population was reared on *Vicia faba* under laboratory conditions at the temperatures of 20–25°C. The biomass of the aphid population reached a maximum value 2 weeks after the inoculation of 1 vivipara on the seedling. About 61.5 eggs of *C. septempunctata brucki* can be produced with the aphids obtained from 1 stem. If an artificial diet of *C. septempunctata brucki* could be developed, it would contribute significantly to mass production.

In the laboratory test, *H. axyridis* laid eggs mainly on alfalfa[10]. On the contrary, the females of *C. septempunctata brucki* did not lay eggs on alfalfa nor conditioned sites with their excretions, including aluminum sheet B, petri dish, nylon screen and alfalfa as shown in Fig. 4. Therefore, it was easy to obtain eggs on an aluminium sheet put into a

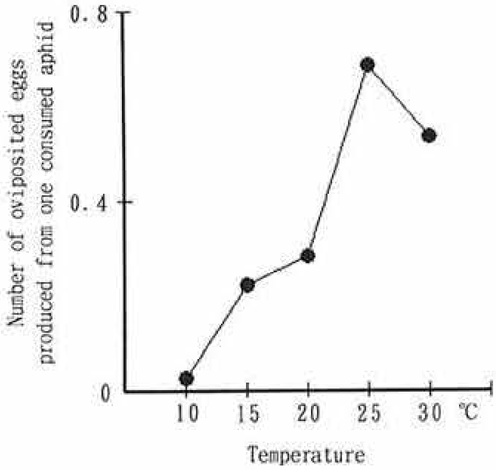

Fig. 5. Number of oviposited eggs produced from 1 aphid consumed by an adult beetle

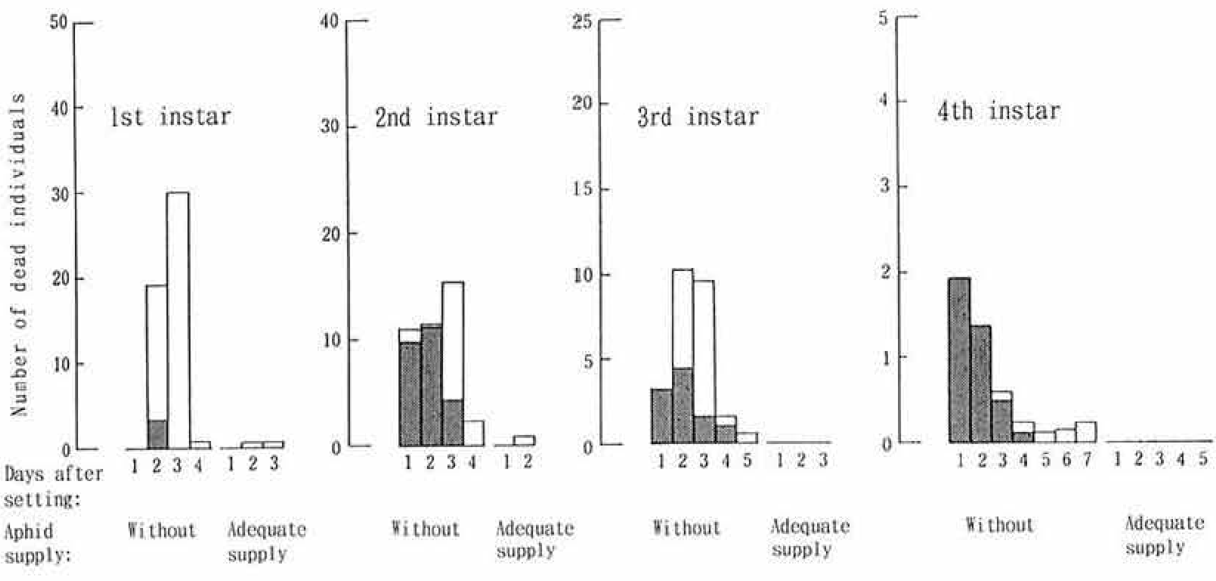

Fig. 6. Larval cannibalism among individuals at each development stage with and without supply of aphids

■ : Number of larvae killed by cannibalism.

rearing container replaced by a new one every day. The optimum temperature to obtain eggs was 25°C and 0.68 eggs could be obtained when 1 adult beetle consumed one 4th instar larva of *A. pisum* (Fig. 5).

The mortality of the eggs and larvae at low temperatures, 5 - 10°C, was high and the adult stage was the optimum stage for storage. It was easy to use the overwintering females to obtain eggs for mass production since the overwintering adults could be kept alive for more than 40 days at 5°C without feeding on aphids. All the females after overwintering were able to oviposit fertilized eggs without mating and no males were needed for mass production.

When no aphids were supplied under the mass production conditions, the eggs were easily attacked by larvae in each instar, and most of the 1st and the 2nd instar larvae molted to the next stages only with eggs[11]. When a sufficient number of aphids was supplied, only the 1st and the 2nd instar larvae attacked a few eggs, while the 3rd and the 4th instar larvae never attacked eggs.

When no aphids were supplied, larval cannibalism occurred frequently in the 2nd, the 3rd and the 4th instar larvae, whereas, when an adequate number of aphids was supplied, larval cannibalism was minimal (Fig. 6). Pupal cannibalism by the larvae occurred only once when no aphids were supplied.

Release of *C. septempunctata brucki* in the alfalfa field and its establishment

In the laboratory test, proper development stages

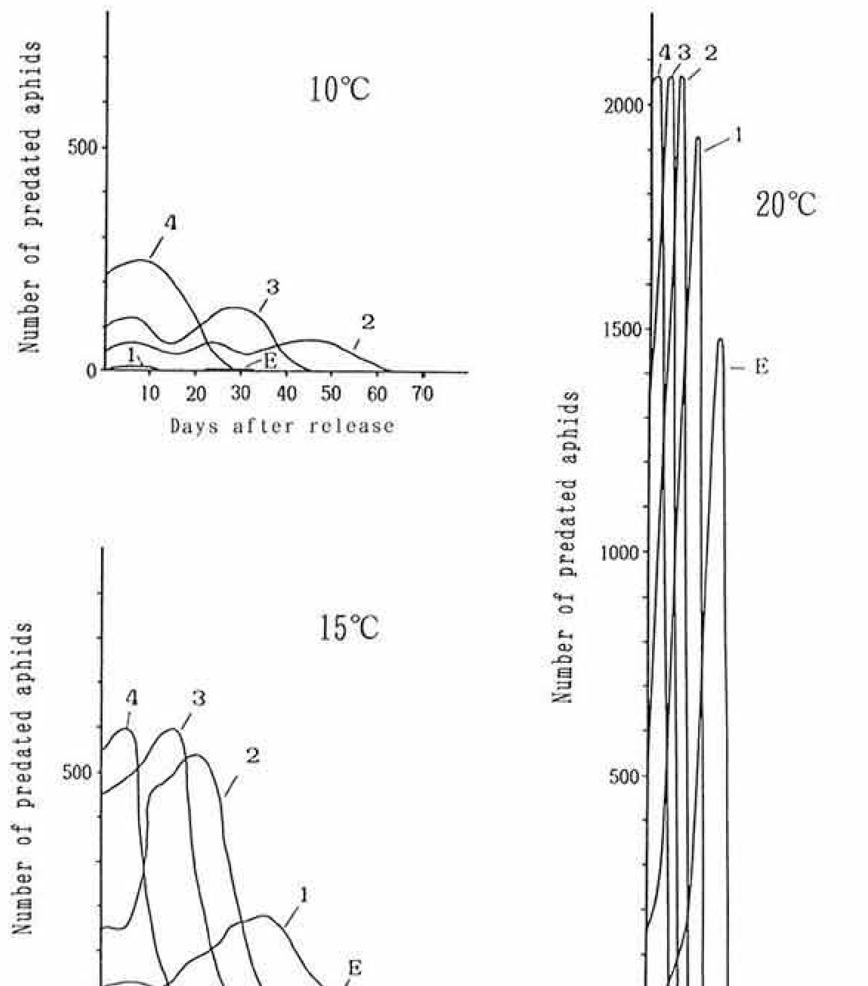

Fig. 7. Amount of predated aphids by the release of each stage of *C. septempunctata brucki* at 10, 15, 20°C
E: Eggs, 1: 1st-instar, 2: 2nd-instar, 3: 3rd-instar, 4: 4th-instar.

of the lady beetle for release in the alfalfa fields at 10–15°C were the 2nd, the 3rd and the 4th instar larvae. Since the mortality of the eggs and the 1st instar larvae at low temperatures, around 10°C in early March was high, it was assumed that the release of these stages was not effective for mass release (Fig. 7).

The 2nd, the 3rd and the 4th instar larvae which were released in the cages located in the alfalfa field, provided an effective control of the aphids. The higher density of release of *C. septempunctata brucki* larvae led to a lower density of aphids and a higher yield of alfalfa. The optimum time for the release was early March when the maximum temperature was around 10°C.

The release of the 2nd larvae of *C. septempunctata brucki* in the open field was effective for aphid control. However, there were no significant differences in yield because of the low population density of the aphids at the time of release.

Japanese pampas grass was planted around an alfalfa field. Many overwintering and estivating adults were observed in the year following planting.

In the field, it is preferable to use IPM for permanent pest management. The combination of the use of *C. septempunctata brucki*, other natural enemies and resistant strains should enable to develop more reliable IPM for aphid management in the future.

References

1) Fukushiro, K. & Furuta, C. (1985): Biology and control of spring black stem and leaf spot of alfalfa caused by *Phoma medicaginis*. *Bull. Chugoku Natl. Agric. Exp. Stn.*, **E22**, 21–53 [In Japanese with English summary].
2) Lehman, W. F. et al. (1982): Registration of CUF 101 alfalfa. *Crop. Sci.*, **23**, 398.
3) Lessen, A. W. et al. (1991): Depression of forage quality of alfalfa leaves and stems by *Acyrthosiphon kondoi* (Homoptera: Aphididae). *Environ. Entomol.*, **20**, 71–76.
4) Milne, W. M. (1988): The effectiveness of aphidophagous insects in lucern. *In* Ecology and effectiveness of aphidophaga. SPB Academic Publishing, Hague, 203–210.
5) Naito, A. (1967): Pest insects of alfalfa. *Proc. Kanto Plant Prot. Soc.*, **14**, 121–122 [In Japanese].
6) Niijima, K., Matsuka, M. & Okada, I. (1986): Artificial diets for an aphidophagous coccinellid, *Harmonia axyridis*, and its nutrition. *In* Ecology of aphidophaga. ed. Hodek, I., 37–50.
7) Sharma, R. & Stern, V. (1980): Blue alfalfa aphid: Economic threshold levels in southern California. *In* California agriculture, February 16–17.
8) Suzuki, S. (1992): Alfalfa. Yukijirushi Shubyo Co. Ltd., pp. 251 [In Japanese].
9) Takahashi, K. & Naito, A. (1984): Seasonal occurrence of aphids and their predators (Col. Coccinellidae) in alfalfa fields. *Bull. Natl. Grassl. Res. Inst.*, **29**, 62–66 [In Japanese with English summary].
10) Takahashi, K. (1986): An experiment on the collecting method of eggs of the lady beetles, *Coccinella septempunctata brucki* Mulsant and *Harmonia axyridis* (Pallas) (Coleoptera: Coccinellidae). *Jpn. J. Appl. Ent. Zool.*, **30**, 62–64 [In Japanese].
11) Takahashi, K. (1987): Cannibalism by the larvae of *Coccinella septempunctata brucki* Mulsant (Coleoptera: Coccinellidae) in mass-rearing experiments. *Jpn. J. Appl. Ent. Zool.*, **31**, 201–205 [In Japanese with English summary].
12) Takahashi, K. (1987): Differences in oviposition initiation and sites of lady beetles, *Coccinella septempunctata brucki* Mulsant and *Harmonia axyridis* (Pallas) (Coleoptera: Coccinellidae) in the field. *Jpn. J. Appl. Ent. Zool.*, **31**, 253–254 [In Japanese].
13) Takahashi, K. (1989): Intra- and inter-specific predations of lady beetles in spring alfalfa fields. *Jpn. J. Ent.*, **57**, 199–203.
14) Takahashi, K. (1993): Overwintering and aestivation sites of *Coccinella septempunctata brucki* Mulsant (Coleoptera: Coccinellidae) and its migration to alfalfa fields. *Appl. Entomol. Zool.*, 28, 473–478.
15) Takahashi, K. (1993): Daily behavior of adult *Coccinella septempunctata brucki* (Coleoptera: Coccinellidae). *Jpn. J. Ent.*, **61**, 839–845.

(Received for publication, June 21, 1996)