Res. Popul. Ecol. (1963) V, 107—116

A MARKING-AND-RECAPTURE ANALYSIS OF THE ADULT POPULATION OF A PHYTOPHAGOUS LADY-BEETLE, *EPILACHNA SPARSA ORIENTALIS*[1]

Syun'iti IWAO and Akisato MACHIDA[2]

Entomological Laboratory, College of Agriculture, Kyoto University, Kyoto

INTRODUCTION

As shown in a previous paper (IWAO *et al.*, 1963) the marking-and-recapture method is proved to be effective for estimating the absolute numbers and other population parameters of adults of *Epilachna vigintioctomaculata*, in which direct count of individuals on the host plants does not provide a reliable estimate of the population, because of their dropping response to slight disturbance.

In the present paper, the closely related species, *Epilachna sparsa orientalis* is subjected to the marking-and-recapture analysis in order to obtain the informations on the density, survival and movement of the overwintered adults in spring.

We are deeply indebted to Professor Syunro UTIDA for his invaluable advice. Thanks are also due to Messrs. K. MIZUTA, H. NAKAMURA, T. ODA, Y. SATO, T. OKAUTI and T. SUGIMOTO for their co-operation in the field work.

MATERIALS AND METHODS

Life history of the species:

E. sparsa orientalis, which is largely allopatric to *E. vigintioctomaculata*, is distributed in the areas where annual mean temprature is higher than *ca.* 14°C (TAKAHASHI, 1932). It has two generations a year in the central part of Japan. Overwintered adults breed on potato, egg-plant or some wild Solanaceae in May-June. The new generation emerges as adults from late June to middle July and within a week or so the females lay eggs for another brood, mainly on egg-plant.

Census area and field method:

The experiment was conducted at Itihara, the north-eastern part of Kyoto City, during the period 23 May to 27 June in 1960. The area is situated in a northern fringe of distribution range of this species (IWAO, 1954).

Two stations were selected for the experiment. Station 1, a field involving 485 potato plants and 63 egg-plants, was mainly used for the analysis. On 23 and 24 May, total 152 adults were captured, paint-marked and returned to the respective

1. Contribution from the Entomological Laboratory, Kyoto University, No. 371.
2. Present address: Hamada Branch, Shimane Agricultural Experimental Station, Shimane.

plants on which they were caught. Each of marked individuals could be identified by its specific mark when recaptured. On five subsequent samplings during 30 May to 23 June, no additional marking was operated, and the beetles captured or recaptured on each occasion were immediately released to the same sites after record was taken. On 27 June, potato plants had been harvested and no marked insect was recovered on egg-plants. To detect possible migration of the marked individuals, nearby fields were also examined during the sampling period.

Station 2 was composed of 600 potato plants where 78 adults were marked and released on 24 May, and recapture samplings were made on only two occasions, 30 May and 13 June. Data obtained in this station is used for the assesment of individual movement within field.

Statistical method of analysis:

Several statistical methods based on re-recapture data are available for estimating the population parameters on successive occasions, from data of the experiment involving single release and multiple recapture samplings. Here, the analysis is made by LESLIE's (1952) method in which re-recaptures are grouped according to the time since they were last recaptured. Only a brief account of the method is mentioned below. For details of mathematical theory and application of the method, the reader is referred to the papers by LESLIE (1952) and LESLIE, CHITTY and CHITTY (1953).

If individuals had been marked and released at $t=0$ and recapture samplings were made at $t=1, 2, \cdots\cdots, T$, the number of marked insects surviving at a particular time x $(1 \leqq x \leqq T-1)$ can be estimated by the following formula,

$$\check{\Psi}_{t=x} = s_x\left(\sum_{x+1}^{T} s_t + 1\right) \Big/ \left(\sum_{t=x+1}^{T} k'_{xt} + 1\right)$$

with the variance

$$V(\check{\Psi}_x) = \check{\Psi}_x^2 \left[\frac{\sum s_t - k'_{xt}}{(\sum s_t + 1)(\sum k'_{xt} + 2)} \right],$$

where s_t is total recaptures at time t, hence s_x the recaptures at a particular time x, and k'_{xt} the expected number of re-recaptures at t $(t > x)$ who had previously recaptured at time x.

The survival factor or the fraction of marked population surviving over the interval t to $t+1$ is then simply estimated as

$$\hat{P}_t = \check{\Psi}_{t+1} / \check{\Psi}_t \ ,$$

and

$$V(\hat{P}_t) = \hat{P}_t^2 \left[\frac{V(\check{\Psi}_t)}{\check{\Psi}_t^2} + \frac{V(\check{\Psi}_{t+1})}{\check{\Psi}_{t+1}^2} \right].$$

In this expression, the fraction $1-\hat{P}_t$ would include the losses due to both death and emigration. Since the estimate $\hat{P}_t$ is based on the marked population released on a single occasion, it is valid as a parameter of total population only when no dilution through birth or immigration was occurring into the population.

The size of the total population at time t is also calculated by

$$\check{N}_t = \check{\Psi}_t (C_t+1)/(s_t+1),$$

and the variance of this estimate is being

$$V(\check{N}_t) = \check{N}_t^2 \left[\frac{C_t - s_t}{(s_t+2)(C_t+1)} + \frac{V(\check{\Psi}_t)}{\check{\Psi}_t^2} \right]$$

where C_t is the total catch at time t.

Then, the dilution factor over the interval t to $t+1$ is estimated as $B_t = \check{N}_{t+1}/\check{N}_t - \hat{P}_t$, and therefore the number of new members alive at $t+1$ is $B_t\check{N}_t$. In the present case, however, it is noticed that if dilution was taking place over two or more intervals of time into the population, B_t value for a later interval would be affected by the cumulative number of new entries up to that time.

Results

Estimation of total numbers and rates of survival and emigration:

In station 1, 44 and 108 adults were marked and released on 23 and 24 May, respectively. Now, we treat both groups of marked insects as if they were released on a single occasion. The validity of such pooling of the data will be discussed later.

The distribution of various classes of re-recaptures, together with total numbers of marked and unmarked insects in the sample, are shown in Table 1. For example, 35 marked individuals were recaptured on 13 June, of which 19 were last recaptured on 6 June, 7 were last recaptured on 30 May and remaining 8 were first recovered after their initial release on 23~4 May.

Table 1. Summary of the sampling data.
Re-recaptures are grouped according to the time since they were last recaptured.

		Date of re-recapture (or capture) (t)					
		23-4 May (0)	30 May (1)	6 June (2)	13 June (3)	18 June (4)	23 June (5)
Date of last recapture (x)	30 May (1)			25	7	1	1
	6 June (2)				19	5 (3)*	2 (1)
	13 June (3)					4 (2)	3 (2)
	18 June (4)						3 (2)
Total No. recaptured (s_t)			54	75	35 (1)	14 (8)	9 (4)
Total catch (C_t)		152	97	119	67 (3)	40(31)	31(15)
Recapture rate (s_t/152)%			35.5	49.3	23.0	9.2	5.9

* Numerals in the parentheses indicate the numbers discovered on egg-plants out of total numbers captured.

As indicated in the table, some individuals moved from potato to egg-plant in the latter half of the sampling period. Since the number of recaptures on egg-plants was small, the estimation of population parameters is made by using the sampling data from potato field alone and those from whole field including potatoes and egg-plants (Table 2).

Small values of dilution factors (B_t) indicate that no appreciable immigration was occurring during the period of this study, though small amount of dilution into egg-plant field might occur in the latter part of the census period. It becomes more apparent when the dilution test given by LESLIE (1952) is applied to the present data; it is based on the assumption that if no dilution through birth or immigration is occurring into the population, the proportion of marked individuals in the sample is expected to be remained constant throughout the sampling period. Comparison of expected and observed numbers of marked and unmarked individuals at successive sampling dates are tabulated in Table 3. With regard to the subpopulation on potato field no significant dilution detected, while the occurrence of dilution is suggested by the χ^2-value for the whole population. The deviation in the latter is apparently attributable to the high proportion of unmarked individuals in the sample taken at the last two occasions.

Since the emergence of new generation adults did not begin by 23 June, this dilution of the population that occurred in the latter half of June might be due to

Table 2. Estimation of population parameters for overwintered adults of *E. sparsa orientalis*.
A : Whole population living in potato and egg-plant fields;
B : Subpopulation living in potato field alone.

Parameter* \ Sampling date (t)		23–4 May (0)	30 May (1)	6 June (2)	13 June (3)	18 June (4)	23 June (5)
$\check{\Psi}_t$	A	—	153.1±13.5	137.5±12.3	95.0±18.3	35.0±9.2	21.3
	B	—	148.9±14.4	136.7±13.6	94.0±24.6	18.0±6.4	11.4
$\hat{P}_t$	A	1.008±0.089	0.898±0.113	0.691±0.147	0.368±0.120	0.608	
	B	0.980±0.094	0.918±0.127	0.688±0.193	0.191±0.085	0.634	
	A−B	0.028	−0.020	0.003	0.177	−0.026	
$\check{N}_t$	A	301.8±43.9**	272.9±27.6	217.1±22.4	179.5±38.1	95.7±29.1	
	B	301.8±43.9**	265.4±28.6	215.8±24.2	174.6±48.2	30.0±11.6	
	A−B	—	7.5	1.3	4.9	65.7	
B_t	A	−0.104	−0.102	+0.136	+0.161		
	B	−0.101	−0.105	+0.121	−0.019		

* Because of the finite nature of the population, the standard error of each estimate is calculated as $V(1-\bar{f}_t)$, where V is the estimated variance and $\bar{f}_t=\frac{(\Sigma s_t)}{\Sigma\check{\Psi}_t}$ the average fraction of the population being sampled (see LESLIE, 1952, p. 369). The values of $\bar{f}_t$ are 0.42 for the whole population and 0.45 for the subpopulation on potatoes.

** Calculated by JACKSON's positive method improved by BAILEY (1951).

Table 3. Comparison of observed and expected numbers of marked and unmarked individuals in the sample.

Sampling date (t)	Whole population		Subpopulation on potatoes	
	No. marked	No. unmarked	No. marked	No. unmarked
1	54 (50.9)	43 (46.1)	54 (55.0)	43 (42.0)
2	75 (62.5)	44 (56.5)	75 (67.5)	44 (51.5)
3	35 (35.2)	32 (31.8)	34 (36.3)	30 (27.7)
4	14 (21.0)	26 (19.0)	5 (5.1)	4 (3.9)
5	9 (16.3)	22 (14.7)	5 (9.1)	11 (6.9)
	$\chi^2=17.429$ ($P<0.01$)		$\chi^2=6.584$ ($0.20>P>0.10$)	

Expected values are given in parentheses.

the immigration of overwintered adults from elsewhere, probably after destruction of the plant attacked.

The absence of dilution for the most part of the census period enables us to assume that the survival factor calculated for the marked population would also be valid as the parameter of total population.

It can be seen from Table 2 that the survival factor of the population was maintained as high as 90 per cent or more until early June, and thereafter it was decreased rapidly. Such decline of the survival rate is especially remarkable when we consider the population on potato field alone; it is partly attributable to the emigration of some individuals from potatoes to egg-plants. In this case, the rate of emigration can not be estimated by the formulae described by Iwao (1963), since the number of insects caught on egg-plants was small. However, the emigration rate would be indicated by the difference in the survival factors between the whole population and the subpopulation on potatoes alone (A-B in Table 2), indicating that one half of the population on 13 June migrated from potatoes to egg-plants during the next 5 days.

On the other hand, no marked individual could not be recovered from other field throughout the sampling period. It is, therefore, assumed that the survival factors calculated for the whole population may be little affected by emigration out of the field, so that $1-\hat{P}_t$ represents the death rate *per se*.

The total number of individuals was maximum at the beginning of the experiment (23 June) where the population density was 0.62 per potato plant. From census data on egg and larval populations, it is indicated that the oviposition by overwintered females had begun only few days before that time and it reached a peak in early June. Therfore, overwintered adults in this field seemed not to be more abundant at an earlier date.

The decreasing trend of the total numbers from 23 May onward was similar to that of marked individuals, except for the latter half of June. On 18 June, there was a considerable difference in the total numbers between whole population and subpopulation on potatoes, which was brought about by both migration from potatoes to egg-plants within the field and the immigration into egg-plants from outside of the field; the former is estimated as 179.5×0.177=31.8 and the latter as 179.5×0.161=28.9.

Movement of overwintered adults within potato field:

For each marked individual, the distance between the release point on a given day and its recovery point on any subsequent sampling day was measured on the map. The frequency distribution of the distances moved by individuals living in station 1 is shown in Fig. 1.

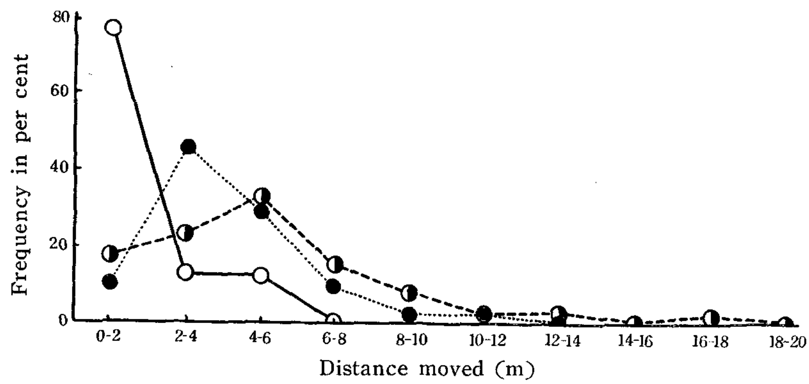

Fig. 1. Frequency distributions of the distances moved by marked individuals in a potato field (Station 1).
—○— : 1-day interval; ---◐--- : 1-week interval; ···●··· : 2-week interval.

On the next day of release, almost 80 per cent of the individuals recovered did move only 2 m or less from their original sites (Av. 1.71 m). The distance travelled by them increased to 4.84 m on avarage after a week, but it did not further increase after another 1 week (Av. 4.09 m), indicating that majority of the overwintered adults lived within a radius of less than 3 m as far as the habitat condition remained favourable.

In station 2, 600 potato plants were planted uniformly over a rectangular space of 27×11 m. Then we can compare the observed frequencies of distances moved by individuals with the expected frequencies from random movement of individuals calculated by MORISITA's formula (see MORISITA, 1950 ; IWAO *et al.*, 1963). It is apparent from Table 4 that observed frequencies of the distances less than 5 m are

Table 4. Comparison of observed and expected frequencies of the distance moved by marked individuals (Station 2).
Expected values are calculated based on the assumption of random movement of individuals.

	Distance moved in a week					
	0–1 m	1–2 m	2–3 m	3–4 m	4–5 m	5 m or more
Expected	0.34	1.02	1.36	1.70	2.04	27.54
Observed	3	2	7	2	3	17

much exceeded the expected ones. Thus, the extent of movement of the overwintered adults was very limited even within a single field.

DISCUSSION

Validity of the population estimates:

The non-significant result of the dilution test for the population on potato field provides an evidence of the valid application of the marking-and-recapture method, because it indicates the fulfilment of random sampling of marked and unmarked individuals and no detachment of the mark.

A further proof of random sampling is obtained by analysing the recapture frequency of marked individuals. For example, out of 35 marked insects who were recaptured on 13 June, 16 had previously been recovered on 30 May and 19 on 6 June. Under the assumption of purely random sampling, the probabilities that an individual of the group had recovered 0, 1 and 2 times during these intervening occasions are then calculated as 0.248, 0.503 and 0.248 respectively, so that expected frequencies are 8.69, 17.62 and 8.69. It is obvious that the observed frequencies 9, 17 and 9 agree well with the expected.

If we treat two groups of marked insects released on 23 and 24 May separately, two sets of successive estimates of population parameters, which are independent from each other, can be obtained. Table 5 shows the successive estimates of survival factors calculated from two sets of the recapture data. In spite of small amount of the data in each series, two estimates of corresponding time intervals agree well. Also, a modified method of BAILEY's triple catch (IWAO *et al.*, 1963) which involves another method of grouping the data gives rise similar values. These results indicate that the underlying assumptions of the marking-and-recapture method are well satisfied in this experiment.

Table 5. Comparison of estimated values of survival factors calculated by different sets of data.

Estimates based on:	Sampling date (t)			
	0	1	2	3
44 marked insects released on 23 May	0.952	1.042	0.641	0.214
108 marked insects released on 24 May	*1.019*	*0.845*	*0.637*	*0.450*
Pooled data by modified method of BAILEY's triple catch	1.025	1.111	0.653	0.400

The pooling of the data from two release groups of marked individuals is justified by the fact that high survival rate near unity was maintained during the first interval of time. It is further confirmed when the population sizes on 23 and 24 May are estimated by LINCOLN index improved by BAILEY (1951). Fourty-four marked insects were released on 23 May and total 126 insects captured on 24 May, of which 18 were recaptures. Then, estimated population on the first day is 294.1±60.6. Similarly, out of 108 marked insects released on 24 May 35 were recovered in the total 97 insects captured on 30 May. The total numbers on 24 May is therefore estimated as 294.0±38.4. These values are consistent with the estmate by JACKSON's positive method using pooled data (see Table 2).

On the basis of these considerations, it may be concluded that the marking-and-recapture method can be applied successfully to the population of adult *Epilachna sparsa orientalis*, if the indivduals captured or recaptured return to their respective sites to secure random mingling of marked and unmarked individuals.

Comparison of population characteristics of E. sparsa orientalis with those of E. vigintioctomaculata:

A similar marking-and-recapture experiment was also carried out with *E. vigintioctomaculata* at Kurama and Ninose, only 2 km north of Itihara, in the same year (IWAO *et al.*, 1963). It enables us to make ready comparisons between the population characteristics of overwintered adults of the two species.

Survivorship curve for the overwintered adults of *E. sparsa orientalis*, constructed from successive estimates of survival factors, is shown in Fig. 2. As mentioned earlier, the curve for the whole population may reflect the true trend of survival of the overwintered adults. The curve is concave in its shape, which is similar to that observed in *E. vigintioctomaculata* population, indicating that in both species the overwintered adults surviving at their reproductive season seemed not to be severely

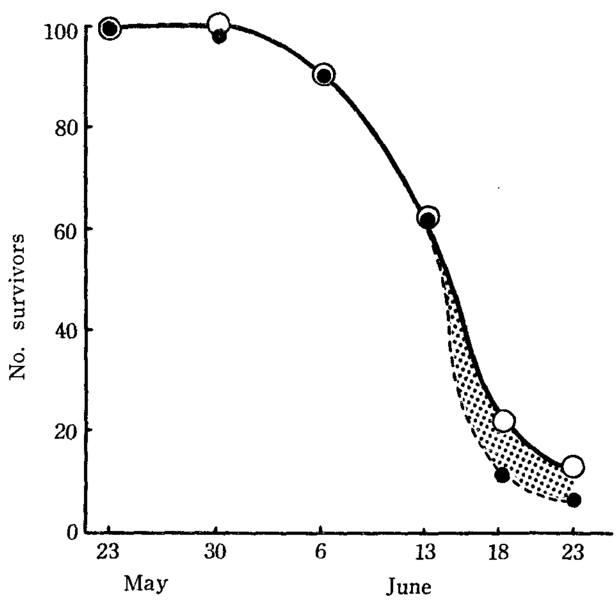

Fig. 2. Surviorship curve of the overwintered adult population in spring.
—○— : whole population ; ···●··· : subpopulation in potato field.
Dotted area indicates the amount of emigration from potato to egg-plant.

affected by environmental mortality factors but they were dying off by early July as approaching their physiological senescence.

Although mobility of the overwintered adults is proved to be small in both species, we have gained an impression that *E. sparsa orientalis* was more mobile than *E. vigintioctomaculata*, because the former was frequently flying about on the field. The average distance moved by marked insects within a potato field was 4.84~6.42 m per week for *E. sparsa orientalis* and 1.57~2.77 m for *E. vigintioctomaculata*. Since the fields used for the measurements were larger in size in the former species, however, these values can not be compared directly. In Fig. 3, the cumulative frequency curves of the distance moved by marked insects in different fields are shown for both species. It can be seen that about half of the individuals moved less than 5 m in *E. sparsa orientalis* and less than 2 m in *E. vigintioctomaculata*, irrespective of size of the field. And, in each of the species, the divergence of the cumulative frequency curves for different fields becomes marked at the larger distances moved, which shows the influence of size of the field on the movement. It seems, therefore, plausible that the difference in the initial part of the curves, below the 50 per cent line, may reflect the specific difference in the mobility.

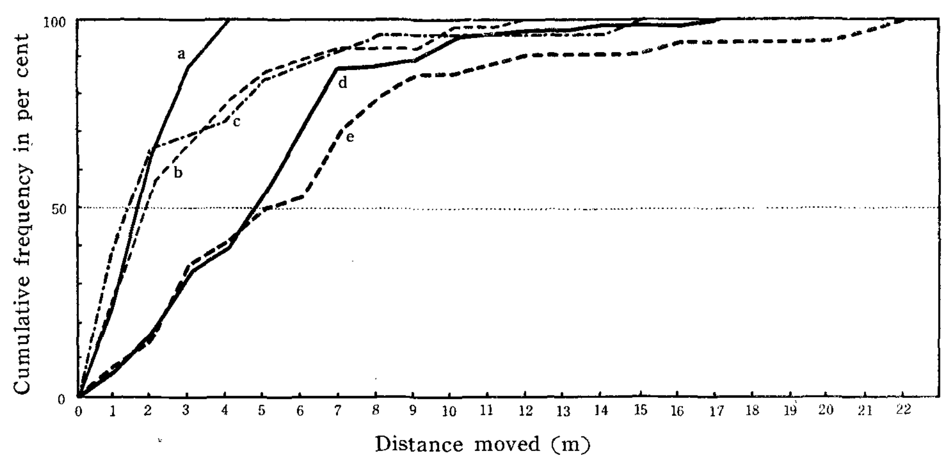

Fig. 3. Cumulative frequehcy curves of the distances moved by adults of *E. vigintioctomaculata* (a, b, c) and *E. sparsa orientalis* (d, e). Sizes of fields are as follows: a, 13 m^2; b, 48 m^2; c, 35 m^2; d, 168 m^2; e, 297 m^2.

An important difference of their behaviour is that some adults of *E. sparsa orientalis* moved to egg-plant even when potato plant was still growing, while *E. vigintioctomaculata* adults rarely migrated to this plant as far as potato leaves were available as food (Iwao *et al.*, 1963). It is not only a simple reflection of different mobility of the two species, but also related to their differential preference to these host plants (Iwao, 1954).

Summary

A marking-and-recapture study of a population of overwintered adults of *Epilachna* *sparsa orientalis* was carried out in a small farm containing potatoes and egg-plants. The experiment involved a single release and five recapture samplings during May-June. The data were analysed by LESLIE's re-recapture method. Assumption of randomness of sampling proved to be well satisfied.

Survival rate of the overwintered adult population was maintained as high as 90 per cent per week or more until early June, and then it declined rapidly towards the end of June. Initially overwintered adults exclusively lived on potato plants, but half of the population alive in mid June migrated to nearby egg-plants. Emigration out of the experimental field, however, seemed not to be occurring.

Recapture data of marked individuals in the potato field indicate that majority of the individuals stayed within a radius of 3 m, at least for 2 weeks. Similar data taken from another field also suggest that their mobility was far restricted as compared with random movement within the field.

Some comparisons were made between the population behaviours of overwintered adults of *E. sparsa orientalis* and *E. vigintioctomaculata.*

REFERENCES

BAILEY, N. T. J. (1951) On estimating the size of mobile population from recapture data. *Biometrika* **38**: 293–306.

IWAO, S. (1954) On the distribution of *Epilachna sparsa orientalis* DIEKE and *E. vigintioctomaculata* MOTSCHULSKY at the boundary of their geographical distributions, I. *Ōyō-Kontyū* **9**: 135–141. (In Japanese with English summary).

IWAO, S. (1963) On a method for estimating the rate of population interchange between two areas. *Res. Popul. Ecol.* **5**: 44–50.

IWAO, S. *et al.* (1963) Studies on a natural population of the large 28-spotted lady beetle, *Epilachna vigintioctomaculata* MOTSCHULSKY. I. Preliminary analysis of the overwintered adult population by means of the marking-and-recapture method. *Jap. J. Ecol.* **13**: 109–117.

LESLIE, P. H. (1952) The estimation of population parameters from data obtained by means of the capture-recapture method. II. The estimation of total numbers. *Biometrika* **39**: 363–388.

LESLIE P. H., D. CHITTY and H. CHITTY (1953) Ditto III. An example of the practical applications of the method. *Biometrika* **40**: 137–169.

MORISITA, M. (1950) Dispersal and population density of a waterstrider, *Gerris lacustris* L. *Contrib. Physiol. Ecol. Coll. Sci., Kyoto Univ.* **65**: 1–149. (In Japanese).

TAKAHASHI, S. (1932) Studies on *Epilachna* lady beetle in Japan. *J. Tokyo Agric. Coll.* **3**: 1–115.

ニジュウヤホシテントウの記号放逐実験

巌　俊　一・町　田　明　哲

ニジュウヤホシテントウの越冬後成虫に個体識別のマークをほどこして放虫し，以後5回の再捕調査のみを行なった実験のデータから，個体数，生存率，ジャガイモからナスへの移動率，および畑内での移動距離などを推定した。またこの結果を先に報告したオオニジュウホシテントウの場合（巌ほか，1963）と比較し異同を論じた。