GEOGRAPHICAL VARIATION OF ELYTRAL SPOT PATTERNS IN THE PHYTOPHAGOUS LADYBIRD, *EPILACHNA VIGINTIOCTOPUNCTATA* (COLEOPTERA: COCCINELLIDAE) IN THE PROVINCE OF SUMATERA BARAT, INDONESIA

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INTRODUCTION

In a previous article, we recorded four common species of epilachnine ladybirds that were abundant as crop pests in the Province of Sumatera Barat, Indonesia (KATAKURA et al., 1988). Among them, *Epilachna vigintioctopunctata* (FABRICIUS) was a serious pest of solanaceous crops such as eggplants and potatoes over a wide range from Japan (NAKAMURA, 1976) to South Asia and Australia (RICHARDS, 1983; KALSHOVEN, 1981).

E. vigintioctopunctata in Sumatera Barat exhibited extreme intraspecific variations in body size, shape and size of pronotal and elytral spots, color tone of elytra, melanization of ventral parts, etc. The purpose of the present article is to document the geographical diversity of elytral spot pattern that was most conspicuous among these variations. Analyses of other characters and multivariate analyses among characters will be reported elesewhere. Rich intraspecific variations in elytral spot patterns have been reported in tropical and subtropical epilachnines (e.g. DIEKE, 1947; KALSHOVEN, 1981), but rarely documented in detail except by RICHARDS (1983) for Australian E. viginticctopunctata populations and by ABBAS and NAKAMURA (1985) for E. septima in Padang, Sumatera Barat. Detailed documentation of the spot pattern variations is a necessary step for further taxonomic, genetical, and ecological studies of the beetles in these regions.

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MATERIALS AND METHODS

Description of elytral spot patterns

Figure 1 shows the standard elytral spot pattern of epilachnine beetles: The basic pattern consists of six black "persistent" spots (1-6) on each elytron. This pattern may be modified by the addition of one to eight "non-persistent" spots (a-h) on each elytron, or by the enlargement and confluence of several spots (DIEKE, 1947). In the present study, individuals were classified according to the position of non-persistent spots, to their total number on each elytron, and to the type of confluences. For example, an individual with non-persistent spots "agh" is counted independently in columns a, g, and h, and is also arranged in the "3 non-persistent spots" class.

Collection of specimens

The Province of Sumatera Barat occupies the western part of Central Sumatra, ranging from 0°54'N to 3°30'S, and 98°36'E to 101°53'W. The coastal plain is very narrow, only a few to 25 km in width, and soon gives way to the steep western slope of the Barisan Mountains (Bukit Barisan), which run parallel to the coast with numerous peaks higher than 1000 m. The eastern slope of the mountains is far less steep, forming plateaux 200 to 1000 m high, which are important for the highland agriculture (see the relief map in Fig. 2 here and Fig. 1 in KATAKURA et al., 1988). Specimens used in the present study were collected at various localities in the Province of Sumatera Barat during October 1980 to March 1985 by I. ABBAS, K. NAKAMURA, A. HASYIM, S. SALMAH, AWALUDDIN and T. INOUE (Fig. 2).

Clustering of collection sites

Figure 2 shows the distribution of collection sites, each of which included one or more (usually several) samples from adjacent fields and/or different host plants. These adjacent collection sites were further clustered into 24 "localities" based on their topography and altitudes (Fig. 2). Hereafter, each locality is cited by the code given



Fig. 1. Left: Standard elytral spot pattern of *Epilachna*, showing codes for persistent (1-6) and non-persistent (a-h) spots according to DIEKE (1947). Right: The confluence of spots, exemplified by 4+3+5.



Fig. 2. Geographical distribution of the two forms of *E. vigintioctopunctata* in Sumatera Barat. Filled circles indicate collection sites. Adjacent collection sites are clustered into "localities" that are denoted by larger circles identified by codes. Inset map indicates elevations of Sumatera Barat. See the text for the detail.

in Fig. 2. The collection sites in the coastal and inland lowlands were easily grouped into localities. On the other hand, highland habitats are physiographically and topographically so complicated that distinct grouping of the sampling sites was rather difficult.

Host plants

Among host plants, the eggplant (Solanum melongena) was cultivated at any altitude

surveyed, whereas the potato (S. tuberosum) was confined to the plateaux 700 to 1500 m high. "Rimbang" (S. torvum) is a perennial semi-shrub weed growing on roadsides, and also in fields and gardens located 0 to 1500 m high. Besides these three widespread and common hosts, "kecubung" (Datura metel) and several other unidentified solanaceous plants were infested by E. vigintioctopunctata, but these were either rare or localized and were not important as hosts (KATAKURA et al., 1988).

RESULTS AND DISCUSSION

Comparison of the elytral spot patterns between sexes and among different host plants
 Sex ratio

A total of 4320 individuals (2740 males and 1580 females) were collected for the present analysis. Males outnumbered females in all 24 localities. The sex ratio calculated for all specimens collected in Sumatera Barat was significantly biased toward the male at 0.1% level ($\chi^2=311.5$). The sex ratio was again significantly male-biased in 19 out of 24 localities (at 0.1-5% level, Chi-square test). The cause of this male-biased sex ratio is unknown, but may be an artifact due to behavioral difference between the sexes. This is considered possible because no significant deviation from the expected 1:1 ratio was detected ($\chi^2=3.56$, N.S. at 5% level) in the sex ratio of marked adults (2437 males and 2309 females) in a rimbang dependent field population in Ulu Gadut Padang (Fig. 5), which was continuously studied by mark-recapture of adult beetles for three successive years (NAKAMURA et al., in preparation). Before describing the geographical variations, variations of elytral spot patterns are compared between sexes and between the specimens from different host plants.

1.2. Elytral spot variations and sexes

Non-persistent spot: Among all the specimens collected in Sumatera Barat,

Collection site	Elevation, m	Locality	Host plant*
PS-1-1	50	PS-A	E (41) vs. S (104)
AG-7, 8, 9, 10	470	AG-B	E (38) vs. S (145)
AG-11, 12	465	AG-C	E (52) vs. S (26)
AG-18	650	AG-D	E (39) vs. S (40)
TD-3-1	1100	TD-A	E (52) vs. P (119)
TD-9, 10, 11, 12, 13	550-620	TD-C	E (64) vs. S (74)
SL-2, 3, 4, 5	365	SL-A	E (142) vs. S (81)
SW-6, 7	95-120	SW-C	E (73) vs. S (47)
PD-1, 2, 3, 4, 5, 6, 7	2-150	PD	E (302) vs. S (301)

 Table 1.
 List of collection sites, where sufficient number of beetles were sampled from different kinds of host plants.

* E: Eggplant, S: Solanum torvum (Rimbang), P: Potato. Numbers of beetles examined are parenthesized. females appeared to have more non-persistent spots than males. The average number of non-persistent spots per elytron was 1.308 in male and 1.657 in female (significantly different at 0.1% level by KOLMOGOROV-SMIRNOV two sample test) (Table 3, see Section 2.1 for explanation of "group"). When the patterns were analyzed separately for each locality, females again tended to have more non-persistent spots than males in 19 out of 24 localities, but the differences were not statistically significant except for PD and LK (in both localities female-biased at 0.1% level by KOLMOGOROV-SMIRNOV two sample test). The combinations of non-persistent spots (*a-h*) appearing on each individual were not significantly different between the sexes either for the whole Sumatera Barat or for each locality (Chi-square test).

Confluence: The average number of confluences per elytron calculated for Sumatera Barat overall was 0.164 in male and 0.173 in female (N.S. by KOLMOGOROV-

0		Ι		II		I	11	IV		To		
Group			No.2)	%	No.	%	No.	%	No.	%	No.	%
		а	0	0.0	0	0.0	0	0.0	0	0.0	0	0.0
		b	21	2.2	34	3.9	76	11.0	160	29.7	387	9.0
		с	38	4.0	58	6.6	86	12.4	203	37.7	510	11.8
	Positional	d	47	5.0	83	9.5	117	16.9	231	42.9	643	14.9
	code	e	60	6.4	118	13.5	143	20.6	222	41.2	713	16.5
		f	58	6.2	109	12.4	144	20.8	217	40.3	719	16.6
		g	100	10.6	161	18.4	258	37.2	363	67.3	1185	27.4
Non-		ĥ	180	19.1	353	40.3	442	63.8	478	88.7	2011	46.6
persistent		0	725	77.0	510	58.2	244	35.2	63	11.7	2248	52.0
spot		1	117	12.4	192	21.9	184	26.6	109	20.2	833	19.3
		2	30	3.2	49	5.6	89	12.8	84	15.6	338	7.8
	Total	3	17	1.8	21	2.4	31	4.5	53	9.8	174	4.0
	1 Otal 110.	4	18	1.9	26	3.0	37	5.3	30	5.6	159	3.7
	per crytron	5	14	1.5	41	4.7	30	4.3	32	5.9	157	3.6
		6	5	0.5	11	1.3	31	4.5	48	8.9	126	2.9
		7	15	1.6	27	3.1	47	6.8	125	23.2	285	6.6
		8	0	0.0	0	0.0	0	0.0	0	0.0	0	0.0
		1+2	1	0.1	10	1.1	6	0.9	40	7.4	66	1.5
		3+4	0	0.0	19	2.2	27	3,9	99	18.4	185	4.3
		3+5	2	0.2	35	4.0	24	3.5	23	4.3	112	2.6
	Tune	4+3+5	1	0.1	124	14.1	13	1.9	86	16.0	260	6.0
	^{1 ypc} 4+	3+5+6	0	0.0	44	5.0	1	0.1	28	5.2	88	2.0
Confluence		4+6	0	0.0	1	0.1	0	0.0	0	0.0	3	0.1
Connuence		5+6	0	0.0	5	0.6	0	0.0	0	0.0	5	0.1
		Others ³⁾	0	0.0	0	0.0	0	0.0	3	0.6	3	0.1
	Total no.	0	937	99.6	644	73.4	620	89.5	285	52.9	3625	83.9
		1	4	0.4	228	26.0	72	10.4	239	44.3	668	15.5
	per eivtron	2	0	0.0	5	0.6	1	0.1	20	3.7	27	0.6
Total no. of beetles examined			941	100	877	100	693	100	539	100	4320	100

Table 2. Variation of elytral spot patterns in *E. vigintioctopunctata* in Sumatera Barat. Spot patterns are given for each group.

¹⁾ Groups I, II, III, IV and all intermediate groups are combined.

²⁾ Sexes are combined.

³⁾ 2+1+3+4, 5+3+4+6, and $1+2+3<\frac{4}{5}$.



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Fig. 3



Fig. 4. Variation groups, denoted by numerals I-IV, of elytral spot patterns in *E. vigintioctopunctata* in Sumatera Barat. Dotted circles indicate the "localities" shown in Fig. 2. For simplification, intermediate groups such as I/II and III/IV are not denoted.

Fig. 3. Geographical variation of elytral spot patterns in *E. vigintioctopunctata* in Sumatera Barat. Frequency graphs, drawn for each locality, follow the geographical arrangement of the localities. Frequencies of individuals with each non-persistent spot (*a-h*, closed column), with respective number of non-persistent spots (1-8, open column) and with confluent spots (*j*, shaded column) are separately presented. Numerals I-IV shown in the graphs signify variation groups explained in the text and shown in Fig. 4. SMIRNOV two sample test) (Table 3). Type of confluences was not significantly different between the sexes (Chi-square test). On an individual locality basis, the difference was detected neither in the average number nor in the type of confluences.

1.3. Comparison of the elytral spot patterns among different host plants

The elytral spot patterns between the beetles collected from different host plants were compared in collection sites where a sufficient number of specimens were sampled from two kinds of plants (Table 1). Beetles collected from eggplants and rimbangs and from eggplants and potatoes showed no difference in the variation patterns of nonpersistent spots and confluences. Since eggplants and rimbangs were frequently found proximately in lowland and potatoes were mixed with them at higher elevations, it is not surprising that the beetles feeding on these different host plants showed no difference in their elytral spot variations.

2. Geographical variations of elytral spot patterns

2.1. Geographical variation trend

Figure 3 shows the geographical variations of elytral spot patterns in Sumatera Barat. In calculating the elytral spot frequency in the figure, specimens of the two



Fig. 5. Local variation of elytral spot patterns in *E. vigintioctopunctata* in and near Padang (PD in Fig. 2). At Ulu Gadut an intensive field study was made from 1982 to 1985 using the method of mark-recapture of adults. See Fig. 3 for further explanation.

sexes and the specimens collected from different host plants were combined, because the sex ratios were uniformly male-biased in most localities and no difference in elytral spot pattern variations among the specimens collected from different hosts was detected as described above. Variation trends of the spot patterns are similar in adjacent localities as exemplified in Fig. 5 which shows variations in and near Padang.

By grouping the data of adjacent localities with similar variation trends, four major groups (I-IV) were recognized (Fig. 4). Tables 2 and 3 summarize the elytral spot pattern variation of specimens found in each group and in Sumatera Barat as a whole. Some comments on the variation trends are given as follows:

(1) Relative frequency of non-persistent spots shows the following descending order in all four groups: h>g>f=e>...>a.

(2) Among the various types of confluences, 4+3+5 was most common, followed by 1+2, 3+5, 3+4, and 4+3+5+6.

(3) The two extreme groups, Groups I and IV were distinctly different in spot pattern frequencies (Fig. 3 and Tables 2 and 3). In Group I, confluences were almost absent, and 70-80% of the specimens lacked a non-persistent spot. On the other hand, in Group IV, the ratio of specimens bearing non-persistent spots and confluences was much higher everywhere; especially in TD-A, 92% of individuals possessed at least one non-persistent spot, 9% and 25% had 6 and 7 non-persistent spots, respectively, and 47% had one and 8% two confluences.

(4) However, all groups were connected with intermediate groups that are designated as I/II, III/IV in Fig. 3. Therefore, Groups I and IV ard considered as the two extremes of a series of continuous variations.

(5) Besides the spot patterns indicated in Fig. 3, other features also seemed to change gradually from Groups I to IV. The body size, size of each elytral spot, and melanism of both elytra and abdomen increased in the following order: I < II < III < IV (the variation trends of these characters will be presented elsewhere).

(6) Group III/IV resembled Group IV in the adjacent areas (e.g., LK and SL-B in Fig. 3) in spot frequency but not in the advanced melanism found in the latter.

(7) The western coastal plains were divided by the three groups into four regions: from the north, Group I (PM-A), Group II (PP-A, B and PD), Group III (PS-E), and Group I (PS-C, PS-B, PS-A). These lowland forms were isolated neither physiographically nor by the absence of food plants. The rimbang was found sparsely but continuously along roadsides even in mountainous areas without crop fields.

2.2. Variations along the elevational gradient

From the results given above, it was considered that the spot variation of E. vigintioctopunctata might be linked to altitude. Then the relationships betwen spot variations and the elevations were analyzed. In this, only the male was used to make the relationships more distinct.

Non-persistent spot: Figure 6 shows the relationships between the elevations of

	I able 5.	Average	number p	er eiytron	od-uou 10	ersistent s	pots and	of confiue	nces in <i>L</i> .	orginiticio	punctata ac	ic m sinc	ımatera I	sarat.	
Group		I			Ш			II			N			Total ¹⁾	
Sex	Male	Female	Total	Male	Female	Total	Male	female	Total	Male	Female	Total	Male	Female	Total
Ncn				*	2)						*		*	*	
persistent	0.510	0.581	0.537	0.906	1.282	1.046	1.764	1.939	1.830	3.196	3.972	3.454	1.308	1.657	1.436
Confluence	0.002	0.009	0.004	0.265	0.282	0.277	0.093	0.130	0.107	0.501	0.536	0.513	0.164	0.173	0.167
Total no. beetles examined	588	353	941	551	326	877	432	261	693	358	181	539	2740	1580	4320
1) Groups 2) Signific	I, II, III, antly differ	IV and all ent at 5%	l intermed (*) and 0.	iate group .1% (**)	s are con level by k	ibined. Col.MoGOR	OV-SMIRN	iov two s	ample test						

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Fig. 6. Relationships between the elevations of sample sites and the average number of nonpersistent spots per elytron (male beetles only). Different symbols are used to indicate the variation groups shown in Figs. 3 and 4: (●), Group I; (▲), I/II; (■), II; (○), III; (△), III/IV, (□), IV.

sample sites and the average number of non-persistent spots per elytron. Each dot in the figure refers to the site where 20 or more males were sampled. Applying Ken-DALL's rank test, the following conclusions were obtained:

(1) Combining all groups, relation between spot pattern trends and elevations was positive (n=55, $\tau=0.43$, at 0.1% level).

(2) As to the group level, no clear-cut relation was detected in any of the four groups by KENDALL's rank test. In particular, Group I, the least melanic group, included localities of widely different altitudes, namely, localities in the southern coastal plains (PS-A to PS-C, 10-50 m) and those in the northern highlands (PM-A to PM-C, 25-830 m). Localities of Group II (PP-A, B and PD, 2-150 m), situated on the coasts, showed a considerable variation.

(3) The relationships were variable within each locality, e.g. positive relationships in PM-B, LK-A and AG-A, negative in PM-C, and no clear relationship in most other localities.

Confluence: Relations between elevation and average number of confluences per elytron were examined again by *Kendall*'s rank test: Significant relation was detected neither for the entire group combined, nor for individual groups.

CONCLUDING REMARKS

In a previous paper we recognized two forms, formae A and B, in *E. vigintioctopunc*tata in the Province of Sumatera Barat (KATAKURA et al., 1988). These two forms were identical in structural characters but were different in body coloration, pronotal and elyral spot variations and vertical distribution. The two forms are also slightly different in body size, and in the conditions of pubescence on the body surface. We withheld a decision on whether they are two dinstinct but closely related species, or whether they merely represent altitude-linked variations of one and the same species (KATAKURA et al., op. cit.). Here, however, we have recognized four major groups on the basis of variation trends of elytral spot patterns alone. The characteristics of these four groups are summarized in Table 4. Groups I, II and most of III correspond to forma A in our previous paper and Group IV (and a part of III/IV and III) to forma B. As discussed above, there is no positive evidence to unite Groups I to III into a single taxon and separate it from Group IV (= forma B) as far as the variation trends of elytral spot patterns are concerned. In other words, the present result favors the view that the two previously recognized forms of E. vigintioctopunctata represent complicated intraspecific variations rather than two distinct sibling species. However, the two forms are different in some characters other than elytral spot patterns. The final solution to this problem will be obtained by multivariate analyses of these characters, rearing of various groups under regulated conditions, and crossing experiments between groups and between forms. Such analyses would also be indispensable to elucidate the effect of genetical and environmental factors on the spot pattern variations in E. vigintioctopunctata.

In many species of coccinellids including Epilachna elaterii (=E. chrysomelina) and E. varivestis the elytral pattern shows a considerable variation (see reviews e.g. KOMAI, 1956; HONĚK, 1973; SASAJI, 1983). This depends to some extent on environmental conditions under which the larvae and pupae develop: Elytral color often becomes

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Group	Forma*	Distribution (Locality, elevation)	Non-persistent spots	Confluences
I	Α	Southernmost coastal plains (PS- A to C, 10-50 m), nothernmost areas (PM-A to C, 25-830 m), and southernmost areas of interior (SW-C, 95-240 m)	Rare, absent in 70-80%	Nearly absent
II	A	Central coastal plains (Padang to Pariaman, PD, 2–150 m, PP-A & B, 5–50 m)	More frequent than in Group I, but absent in 60% though some individuals had many non- persistent spots	20-30%
III	A and B (partim)	Interior plateaux at middle eleva- tions (AG-C, 465m, TD-C, 480- 1130 m & SL-A, 360-370m) and coastal plain (PS-E, 50-100 m)	More frequent than Group II, individuals without non- persistent spot less than 30-40%	10-20%
IV	В	Interior plateaux at higher elevations (AG-D & TD-A, 850– 1140 m)	Most frequent, individuals without non-persistent spots 10-15%	40-50%
* 17.		1 (1007)		

Table 4. Four major groups in the elytral spot variations of E. vigintioctopunctata in Sumatera Barat.

paler under high temperatures and low humidities, whereas melanic under reversed conditions. Decrease of elytral non-persistent spots under high temperatures was also reported by YASUE (1956) for Japanese populations of *E. vigintioctopunctata*. However, the elytral pattern is also genetically controlled (the reviews above). Crossing experiments using adults of *E. vigintioctopunctata* collected in Padang showed that the frequency of offspring adults with the same spot pattern as their parents distinctly increased by selective mating, suggesting the participation of genetic factors in the spot determination (ABBAS and NAKAMURA,. in preparation). Undoubtedly not only environmental factors, but also genetical factors must be responsible for determination of elytral spot variations and colors of *E. vigintioctopunctata* (as confirmed in other coccinellids), but no more detailed analyses have been published so far.

Summary

Geographical variation in the elytral spot patterns of a phytophagous ladybird, Epilachna vigintioctopunctata, was studied in the Province of Sumatera Barat, Indonesia. Populations of E. vigintioctopunctata were divided into four major groups (I-IV) by the incidence of spot pattern variations. Group I, occurring in the coastal plains and inland lowlands, and Group IV, confined to the highlands, were the extremes of the spot pattern variations, the latter had many more non-persistent spots and confluences with larger body size and advanced melanism than the former. These two groups were connected with each other via the intermediate groups. A positive relationship was detected between the elevations of sample sites and the average number of non-persistent spots per elytron. Consequently, present results favor the view that the two previously recognized forms of E. vigintioctopunctata (formae A and B in KATAKURA et al., 1988) represent a complicated intraspecific variation rather than two distinct sibling species. Elytral spot pattern variations were not different between the sexes or between the beetles collected from different kinds of host plants.

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スマトラ西部州におけるニジュウヤホシテントウムシの鞘翅斑紋の地理変異

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スマトラ西部州(インドネシア共和国スマトラ島)の多数の地点で、ニジュウヤホシテントウ (Epilachna vigintioctopunctata FABRICIUS) を採集し、鞘翅斑紋の地理変異を調査した.本種は、片翅 (elytron) あたり6個の「常在斑紋 persistent spot」を必ずもつが、「非常在斑紋 non-persistent spot」 の個数(0-8)や「斑紋結合 confluence」の有無については非常に変異に富んでいた.スマトラ西 部州の本種個体群は、これらの変異の出現頻度をもとに、グループIとNを両極端とする4グルー ブ(I-N)に大別された.グループIは、海岸沿いの平野と内陸部の低地に分布し、「非常在斑紋」 も「斑紋結合」もほとんど持たず、淡色で小型であった.これに対して、グループNは、高地にの み局在し、多数の「非常在斑紋」と「斑紋結合」を持ち、体色は黒化し大型であった.この両グルー プは、不連続ではなく中間グループ(II、II)によって連続していた.生息地の海抜高度と「非常 在斑紋数(片翅あたり)」には正の相関関係があった.ここに述べた結果だけからみると、前報 (KATAKURA et al., 1988)で述べた本種の2型(form A と form B)は、2つの姉妹種(sibling species)に相 当するのではなく、同一種内の変異とみなすほうが妥当であろう.斑紋パターンには、雌雄差や食 草の種類による差はみられなかった.