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Individual variation in two host plants of the ladybird beetle, *Epilachna pustulosa* (Coleoptera: Coccinellidae)

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Individual variation in two species of host plants (thistle, *Cirsium kamtschaticum*, and blue cohosh, *Caulophyllum robustum*) of the herbivorous ladybird beetle *Epilachna pustulosa* was examined under laboratory conditions for their acceptability to adult beetles as a food resource, for adult preference and for larval performance. When clones of these plants were subjected to non-choice tests using posthibernating female beetles, there was found to be significant intraspecific variation among clones in terms of their acceptability, but interspecific variation was not detected. Significant intraspecific as well as interspecific variation were frequently detected in the two host plants when clones of these plants were subjected to choice tests using posthibernating female beetles; the magnitude of interspecific plant variation for beetle preference is not necessarily larger than that of intraspecific plant variation. Individual variation across plant species with respect to beetle larval performance was also significant. A positive correlation between adult preference and larval performance is suggested across the two taxonomically remote host plant species, thistle and blue cohosh, although this needs further investigation.

Key words: adult preference; *Caulophyllum robustum*; *Cirsium kamtschaticum*; *Epilachna pustulosa*; larval performance.

INTRODUCTION

In recent years, it has gradually become evident that individuals belonging to one plant species often act as a heterogeneous resource for insect herbivores (e.g. Edmunds & Alstad 1978; Whitham 1983; Karban 1989; Horner & Abrahamson 1992; Suomela & Nilson 1994). There may be intraspecific or intrapopulational variation in various attributes of plants. Hence, studies of insect herbivore host specificity need to investigate not only differences between plant species but also variations within a single plant species. The present paper aims to assess intraspecific variation and interspecific differences between two species of host plants (thistle, *Cirsium kamtschaticum* Ledeb., Asteraceae, and blue cohosh, *Caulophyllum robustum* Maxim., Berberidaceae) of the herbivorous ladybird beetle *Epilachna pustulosa* Kôno in terms of their acceptability to adults as a food resource, adult preference, and larval performance.

Epilachna pustulosa is univoltine and hibernates as an adult. Females lay eggs on the leaves of food plants. Due to low vagility, larval development is usually restricted to the plant or the cluster of plants on which the eggs were originally laid. Thus, the choice of larval food plant by ovipositing females is of critical importance to the fate of their offspring.

In the present paper, we ask two questions: (i) does a difference in adult acceptability, adult preference and larval performance exist between the two species of plants and between different individuals of the same plant species; and (ii) if it does exist, do adult females prefer particular plants that are more suitable for larval performance?

METHODS

Insect material

Posthibernating adult females (and their offspring) of the so-called Sapporo form of *E. pustulosa*, known to occur on two major host plants, thistle and blue cohosh, were used (Katakura 1976; Hoshikawa 1983; Kimura & Katakura 1986).

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Posthibernating *E. pustulosa* adult females were collected from thistle plants in three places (Maruyama, Sankakuyama and Miyagaoka) in the western suburbs of Sapporo, Hokkaido, northern Japan. Since the three places are within a radius of only 2 km, and there is no distinct difference in host preference between beetles from the former two sampling places (T. Kimura, unpubl. data), we assumed that there is no distinct difference in the host preference of beetles from these three sampling places. All collected females were sexually mature, and had probably copulated and stored sperm before collection (Katakura 1982).

The collected beetles were fed on alternative food plants, *Scopolia japonica* Maxim. (Solanaceae) and/or *Solanum japonense* Nakai (Solanaceae; cf. Katakura 1981), for more than 1 week before the experiments started in order to diminish the possible influence of feeding experience before collecting. When the same beetles were used repeatedly in different feeding tests, they were fed on *S. japonica* and/or *S. japonense* for more than 24 h prior to each experiment to minimize the influence of preceding tests.

Plant material

Both thistle and blue cohosh are perennial wild herbs. Thistle usually grows at the margin of forests as an assemblage of a few to several clonal shoots (henceforth called 'clone'), and blue cohosh is an undergrowth plant that grows amid forests as an assemblage of several clonal shoots (= clone) or an assemblage of clones (= patch). Thistle and blue cohosh grow up to about 200 cm and 60 cm in height, respectively. A detailed description of the phenologies of thistle and blue cohosh in the Sapporo area can be found in Hinomizu *et al.* (1981) and Kimura and Katakura (1986).

All plant materials were obtained in Sapporo, Hokkaido, northern Japan. Thistle leaves were obtained from six sites in three places, and those of blue cohosh from four sites in three places as shown in Table 1. *Epilachna pustulosa* occurs at Hoshioki, Sankakuyama and Kobetsuzawa but it does not occur on the campus of Hokkaido University.

Plant clones were marked with tags to avoid mix-up. For each clone, sampled leaves were kept separately in a plastic bag at 5°C and were used within 2 days after they were picked. It is well known that leaves of thistle and blue cohosh can be kept fresh for at least several days under such conditions (cf. Shirai 1994). Thistle leaves were sampled at mid-height from shoots approximately 80–150 cm above the ground to avoid using leaves that were too young or too old.

Table 1 Plant material sampling sites

Sampling place	Sampling site codes	
	Thistle	Blue cohosh
Hokkaido University campus	U_1, U_2	
Hoshioki	H_1, H_2, H_5	H_3, H_4
Sankakuyama	S_1	S_2
Kobetsuzawa		K_1

Hereafter, each plant clone is referred to by a combination of the following codes. Plant species are indicated by the capital letter T for thistle and C for blue cohosh. The Arabic numeral following the plant code is the clone number. The sampling site code as shown in Table 1 is also attached. For example, thistle leaves sampled from the clone number 4 growing at site H_2 are abbreviated to $T4H_2$.

Experiments

We examined host acceptability of adult female beetles by non-choice tests, and the degree of female beetle food preference by choice tests. In addition, we examined larval performance under four different food conditions to investigate whether suitability varies among plant clones and between plant species for *E. pustulosa*, and whether ovipositing females prefer particular plants suitable for larval performance.

It is virtually impossible to distinguish between oviposition and adult food preference in *E. pustulosa*. Unlike insects such as lepidopterans in which larval foods are distinctly different from those of adults, both adults and larvae of epilachnine beetles feed on the same part of the same species of plant. Furthermore, in captivity in small cages, females often lay eggs not only on host plants, but also on non-host plants and other material such as paper sheets and cages, although they usually lay eggs on the leaves of food plants in natural conditions. Hence, it is very difficult to quantitate oviposition preference under laboratory conditions. For this reason, in the present study, we treated the food choice of ovipositing females as equivalent to the choice of food plant for larvae.

Non-choice tests and the rearing of larvae were carried out under a controlled regime of 16:8 h LD cycle at 20°C in 1992. Choice tests were carried out in 1992 and 1993 under the same regime. To examine the temporal change in the relationship between clones/species, some pairs of clones were examined more than once in different seasons and years in choice tests.

Non-choice tests

A piece of a leaf (about 20 cm^2) was placed in a transparent styrene cage (8.0 cm × 15.5 cm × 3.0 cm), the bottom of which was covered with moist filter paper. A female beetle was released into the cage and allowed to feed on the leaf piece for 24 h, after which the consumed area was measured using a portable area meter (KP-82N, Uchida-Youkou Co., Tokyo, Japan).

Four clones each of thistle ($T1U_1$, $T3H_5$, $T4H_2$, $T5S_1$) and blue cohosh ($C1H_3$, $C2H_4$, $C3H_3$, $C5S_2$) were selected at random and examined. Nine to 15 replicates of feeding tests, using different females, were carried out simultaneously for each plant.

The thickness and water content of thistle and blue cohosh leaves differs. The dry weight of the consumed area per unit area of blue cohosh measured in the present study was 0.29 of that of thistle. Hence, we adjusted the consumed area of blue cohosh leaves by multiplying by 0.29 to make possible a comparison with those of thistle clones.

Choice tests

One female beetle and two pieces of leaves (about 20 cm^2, each from different clones or species) were placed in a transparent styrene cage (8.0 cm × 15.5 cm × 3.0 cm) as in the non-choice tests described above. After 24 h from the start of the test, the consumed area of each leaf piece was classified into one of the following five ranks: 0, no leaf material was eaten or only a nibbled scar could be observed on the leaf; +1, the consumed area was less than 0.25 cm^2; +2, the consumed area was 0.25–1.00 cm^2; +3, the consumed area was 1.00–4.00 cm^2; +4, the consumed area was more than 4.00 cm^2. The rating was carried out by measuring the consumed area with a square template. Ten to 15 replicates were used in each food choice test as in the non-choice tests.

One particular leaf piece was judged to be preferred to another leaf piece by a single female when: (i) only one piece was consumed; or (ii) both pieces were consumed but the consumed areas of the two pieces differed by two ranks or more. We compared two categories of leaves by this method between: (i) clones of the same plant species; and (ii) clones of different plant species.

Another square template, each square being 3.45 times larger than that used for the intraspecific comparison, was used for the rating of blue cohosh when the consumed areas were compared between the two plant species, since the dry weight of the consumed area per unit area is different between thistle and blue cohosh (see above).

Larval performance

A more-preferred and a less-preferred thistle clone were chosen on the basis of the results of food choice tests made in early June when the oviposition activity of the beetles was high. In the case of blue cohosh, two patches each containing a more-preferred and a less-preferred clone, respectively, were chosen as a single clone of blue cohosh was too small to rear all of the larvae being tested (50 individuals for each food condition, see below). Larvae were reared on freshly picked leaves of these clones (thistle) and patches (blue cohosh).

The detailed procedure of rearing larvae was as follows. Egg masses were gathered from posthibernating females that were collected and kept for the above-mentioned experiments. In order to equalize the genetic background of the beetles between food conditions, four sets of newly hatched larvae were made each containing five individuals from a single egg mass, and were reared on the four food conditions. Rearing was carried out in transparent styrene cages (8.0 cm × 15.5 cm × 3.0 cm), the bottoms of which were covered with moist filter paper. A total of 50 larvae from 10 egg masses derived from different females were reared under each food condition. Throughout the experimental period, a sufficient amount of food plant leaves picked from the field every second or third day were provided. Larvae were reared until emergence. The eclosion rate (i.e. the percentage of individuals reaching adulthood), the developmental duration (the number of days required from hatching to adult eclosion) and the pronotum width (a body size index) of newly emerged adults were recorded.

RESULTS

Non-choice tests

The means and standard errors of the consumed areas of each clone are shown in Fig. 1. Mean consumed areas varied significantly among clones within host species (nested ANOVA, $P < 0.001$; Table 2). There was no significant difference between the two host species ($P = 0.189$).

Choice tests

Choices between the 21 pairs of different thistle clones were investigated (Table 3). A statistically significant preference was observed in nine pairs. Beetle preference for $T1U_1$ and $T4H_2$ was examined three times in different seasons and years and yielded the same results (G-test, $G = 2.25$, $P > 0.05$). Likewise, choices between $T1U_1$ and $T2U_2$ were examined twice in different seasons and yielded the same results (G-test, $G = 0.82$, $P > 0.05$). Choices between blue cohosh clones were tested for 17 pairs, and a statistically significant preference was observed in three pairs (Table 3). No interspecific combination of blue cohosh was examined more than once.

Table 2 Mixed model nested ANOVA of mean consumed areas obtained by non-choice tests using adult females of *Epilachna pustulosa*

Source	d.f.	SS	MS	F
Host species	1	2.284	2.284	2.192
Clones within host species	6	6.167	1.028	10.654***
Error	97	9.357	0.096	

SS, sum of squares; MS, mean square. ***$P < 0.001$.

Clones were nested for each host species. Host species and clones of each host species were set as fixed and random effect, respectively. The data were *log (consumed area + 1)* transformed to achieve normality and homogeneity of variance before the analysis. The F-values were calculated with modification for unequal sample size (Sokal & Rohlf 1995).

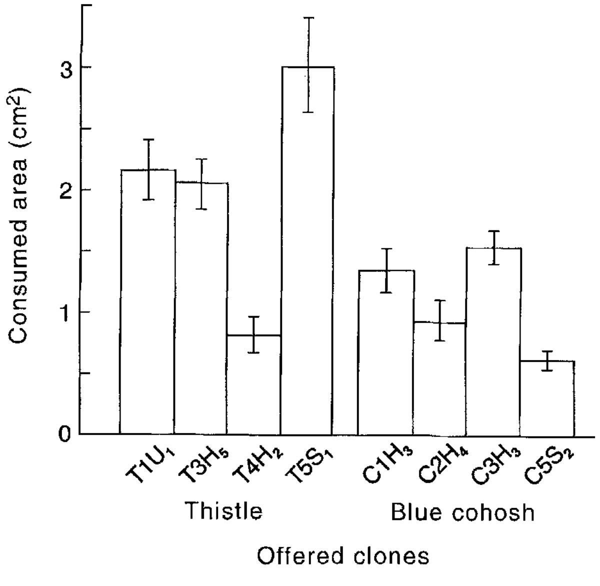

Fig. 1. Mean leaf areas of four thistle and four blue cohosh clones consumed by posthibernating female beetles of *Epilachna* in non-choice tests. Vertical lines represent ± SE. Consumed areas of blue cohosh clones are given in modified values (multiplied by 0.29; see text) so that they can be compared to those of thistle clones.

A total of 31 interspecific pairs of plant clones were subjected to choice tests (Table 4). Statistically significant preference was observed in 10 pairs. There was no definite trend that a particular plant species was always preferred, although concerning the plants sampled at sites in Hoshioki (i.e. H_1, H_2, H_3 and H_4), blue cohosh was always equally or more preferred to thistle.

Choices between $T4H_2$ and $C1H_3$ were examined three times in the first half of June and July 1992 and the second half of May 1993. The order of preference altered seasonally (G-test, $G = 21.81$, $P < 0.005$): in the first half of July 1992, $C1H_3$ was significantly preferred (binomial test, $P < 0.005$), although no clear preference for a particular clone was detected in the other two replications ($P > 0.05$).

Larval performance

On the basis of the results obtained by the choice test carried out in the first half of June 1992, $T1U_1$ and $T4H_2$ were chosen as the more-preferred and less-preferred thistle clone, respectively. Likewise, two patches (henceforth abbreviated as C-H_4 and C-H_3) were chosen as patches containing a more-preferred clone ($C2H_4$) and a less-preferred clone ($C1H_3$) of blue cohosh, respectively (see Table 3).

Table 3 Results of choice tests between intraspecific pairs of plant clones by ovipositing females of *Epilachna pustulosa**

Spatial relations between clones	Offered clones and preference
Thistle	
Between places	$T1U_1 > > > T3H_5$, $T1U_1 > > > T4H_2$*, $T1U_1 > > > T5S_1$, $T3H_5 < < < T5S_1$, $T4H_2 < < < T5S_1$, $T5S_1 > T6U_1$
Within place, between sites	$T1U_1 > > > T2U_2$*, $T2U_2 < < T6U_1$, $T3H_5 < < T4H_2$, $T4H_2 = T7H_1$, $T4H_2 = T20H_1$
Within site	$T1U_1 = T6U1$, $T8H_1 = T14H_1$, $T8H_1 = T15H_1$, $T10H_1 = T24H_1$, $T12H_1 = T13H_1$, $T12H_1 = T21H_1$, $T14H_1 = T15H_1$, $T17H_1 = T18H_1$, $T17H_1 = T19H_1$, $T21H_1 = T23H_1$
Blue cohosh	
Between places	$C7S_2 = C9K_1$, $C8S_2 = C10K_1$
Within place, between sites	$C1H_3 < < < C2H_4$, $C1H_3 = C11H_4$, $C3H_3 = C6H_4$, $C3H_3 > C18H_4$, $C6H_4 = C21H_3$, $C13H_3 = C20H_4$, $C17H_4 = C26H_3$
Within site	$C3H_3 = C4H_3$, $C6H_4 = C14H_4$, $C9K_1 < C10K_1$, $C11H_4 = C15H_4$, $C12H_3 = C13H_3$, $C12H_3 = C19H_3$, $C23H_4 = C24H_4$, $C27H_3 = C28H_3$

*Analyzed by binomial test.

The inequality sign opens towards more preferred clone: triple, $P < 0.005$; double, $0.005 < P < 0.01$; single, $0.01 < P < 0.05$. An equal mark means no significant difference ($P > 0.05$). Pairs of clones with asterisk were examined more than once in different seasons or years, and yielded the same results (see text).

Table 4 Results of choice tests between interspecific pairs of plant clones by ovipositing females of *Epilachna pustulosa*

Spatial relations between clones	Offered clones and preference
Between places	$T1U_1 = C1H_3$, $T1U_1 = C4H_3$, $T1U_1 > C13H_3$, $T1U_1 = C2H_4$, $T1U_1 < < < C6H_4$, $T1U_1 < C20H_4$, $T1U_1 = C5S_2$, $T6U_1 = C5S_2$
Within place, between sites	$T5S_1 > > > C5S\ _2$, $T8H_1 = C20H_4$, $T9H_1 = C16H_4$, $T10H_1 = C17H_4$, $T11H_1' = C18H_4$, $T12H_1 = C12H_3$, $T13H_1 = C19H_3$, $T16H_1 = C21H_3$, $T16H_1 = C22H_3$, $T18H_1 = C25H_3$, $T20H_1 < C1H_3$, $T22H_1 = C1H_3$, $T4H_2 =/< < < C1H_3$*, $T4H_2 = C3H_3$, $T4H_2 < C13H_3$, $T4H_2 = C26H_3$, $T4H_2 < < < C2H_4$, $T4H_2 = C6H_4$, $T4H_2 = C11H_4$, $T4H_2 = C17H_4$, $T4H_2 < C18H_4$, $T4H_2 = C20H4$, $T25H_2 < < < C29H_3$

Pairs of clones with asterisk were examined more than once in different seasons and years, and yielded different results (see text). Other explanations are the same as for Table 3.

The results of the rearing tests are summarized in Table 5. All measured traits of beetles adopted as indices of larval performance were significantly influenced by food conditions (eclosion rate, $H = 14.690$, d.f. = 3, $P = 0.002$; developmental duration, $F = 104.904$, d.f. = 3, $P < 0.001$; pronotum width, female, $F = 6.182$, d.f. = 3, $P < 0.001$, male, $F = 7.793$, d.f. = 3, $P < 0.001$).

In all three measured traits, performance was worst in larvae reared on $T4H_2$ (i.e. low eclosion rate, long developmental duration and small body size), and the differences in these traits were often significant in pair-wise comparisons (Table 5). With regard to the other three food conditions, the results were not consistent among traits: $C\text{-}H_3$ was the best with respect to the eclosion rate and developmental duration but worst with respect to pronotal width, whereas the reverse was true for $C\text{-}H_4$; rearing larvae on $T1U_1$ resulted in an intermediate degree of performance between two patches of blue cohosh in all the measured traits.

DISCUSSION

Individual variation in the two host plants of *Epilachna pustulosa*

As described above, considerable individual variation exists in the two host plants of *E. pustulosa* in

Table 5 Mean ± SE for developmental traits of *Epilachna pustulosa* reared on two thistle clones and two blue cohosh patches

Clone/patch	Eclosion rate**[1]	Developmental duration (days)***[2]	Pronotum width (mm)	
			Female***[2]	Male***[2]
$T1U_1$	92.0 ± 4.42 (10)^a	34.85 ± 0.342 (46)^{ab}	3.50 ± 0.031 (15)^{ab}	3.36 ± 0.023 (31)^a
$T4H_2$	64.0 ± 8.84 (10)^b	42.69 ± 0.642 (32)^c	3.42 ± 0.049 (12)^a	3.25 ± 0.032 (20)^b
$C\text{-}H_3$	98.0 ± 2.00 (10)^a	33.94 ± 0.271 (49)^a	3.45 ± 0.019 (28)^a	3.33 ± 0.024 (21)^{ab}
$C\text{-}H_4$	88.0 ± 5.33 (10)^{ab}	35.34 ± 0.225 (44)^b	3.57 ± 0.023 (22)^{bc}	3.43 ± 0.023 (22)^a

**$0.01 > P > 0.001$, ***$P < 0.001$. [1]Analyzed by Kruskal–Wallis test. [2]Analyzed by one-way ANOVA.

Sample sizes (*n*) are given in parentheses. The mean and SE of eclosion rates was calculated from the rates of 10 replicates for each food condition. Figures with the same letters are not significantly different at the 0.05 significance level (percentage of eclosion, non-parametric test for multiple comparisons using the Mann–Whitney–Wilcoxon *U*-statistic [Sokal & Rohlf 1995]; developmental duration and pronotum widths, Scheffé method [Sokal & Rohlf 1995]). Data were log-transformed before analyses in ANOVA.

terms of their acceptability to adult beetles, adult preference and larval performance. The degree of variation seems to be larger in thistle than blue cohosh in all respects studied (Fig. 1, Tables 3 and 5). It was reported that the Sapporo form of *E. pustulosa* preferred blue cohosh to thistle under laboratory conditions (Hoshikawa 1984; Kimura & Katakura 1986); however, such a tendency was suggested only in a portion of the results of the interspecific choice test (Table 4). There was no clear tendency to show that one plant species was generally always more accepted than and preferred to the other (Tables 2 and 4). Probably due to the heterogeneity of thistle clones, adult *E. pustulosa* females seemed to recognize thistle and blue cohosh as an array of variable but continuous food resource, at least under laboratory conditions. Similarly, there was no clear tendency to show that a particular plant species was always superior to the other species in regard to larval performance; larval performance on the thistle $T1U_1$ was intermediate between those on the two blue cohosh patches, and that on $T4H_2$ was the worst among all the four clones/patches (Table 5).

These results may relate to differences in the habitats of host plants. Thistle grows in a wide range of habitats from sunny grassland to shaded forest margin, that vary in water and sunlight conditions, whereas blue cohosh is found only in shaded and mesic forest floor. Since the physical environment of a plant can influence plant chemical composition in a fairly predictable manner (e.g. Bryant *et al.* 1983; Waring *et al.* 1985), it is expected that chemical composition is more variable in thistle than blue cohosh.

Concerning the heterogeneity of thistle clones, the existence of between-place difference was suggested through intraspecific choice test (Tables 3 and 6). A significant difference was more frequent in pairs of clones from different sites than in those from the same site, and in pairs of clones from different places than in those from the same place (Table 6; *G*-test, $G = 9.02$ for within site *vs* between sites, $G = 14.24$ for within place *vs* between places, $P < 0.005$ in both cases), although data were biased due to the prevalence of clonal pairs from Hoshioki (12/24, 50.0%). If such a difference between thistle plants concerning spatial scale really exists, it suggests a possibility that each local population of *E. pustulosa* may be more adapted to thistle clones growing at the birth place. Further intensive studies are needed to ascertain this possibility.

Table 6 Results of intraspecific choice tests rearranged with respect to sampling place and site

	Difference	Within place		Between places
		Within site	Between sites	
Thistle	Significant	0	4	8
	Not significant	10	2	0
Blue cohosh	Significant	1	2	0
	Not significant	7	5	2

When tests were repeated using the same pair of clones, the result of each test was counted separately.

A between-place difference was not suggested for blue cohosh.

Correlation between adult preference and larval performance

Understanding the correlation between oviposition preference of females and larval performance has been considered the crux of understanding coevolution between host plant and insect herbivores. The correlation should be strongest in insects in which offspring development is restricted to the host plant chosen by the adult females. It is often assumed that ovipositing females will show a strong preference for plants on which the performance of their offspring will be good, although existing data range from strong positive correlations to very weak ones (reviewed in Thompson 1988; Thompson & Pellmyr 1991).

In the present study, we tried to detect such a correlation using two clones of thistle ($T1U_1$ and $T4H_2$) and two patches of blue cohosh ($C\text{-}H_3$ and $C\text{-}H_4$). Adult beetle preference and larval performance are well correlated in the two thistle clones; the larval performance was worse (i.e. lower eclosion rate, longer developmental duration and smaller adult body sizes; Table 5) on $T4H_2$, the less-preferred thistle clone by adult beetles. Even including two patches of blue cohosh, the larval performance was worst on $T4H_2$.

However, relationships between preference and performance across two taxonomically remote species are difficult to interpret for the following four reasons.

(1) As for the three clone/patches except $T4H_2$, there seemed to be a trade-off between eclosion rates and body sizes, both of which seemed to be functions of developmental duration. Longer developmental duration resulted in lower eclosion rates and larger adult sizes (e.g. $C\text{-}H_4$, the more-preferred patch), whereas shorter developmental duration resulted in higher survival rates and smaller adult sizes (e.g. $C\text{-}H_3$, the less-preferred patch). In other words, the three plant clone/patches affected the larval performance in different ways. A field study showed that the mortality of *E. pustulosa* on blue cohosh was mainly caused by food shortage (Kimura & Katakura 1986). Hence, it seems adaptive for this beetle to choose $C\text{-}H_3$, the clone on which the larvae could complete the growth in shorter periods. However, small adult body size may result in higher mortality during hibernation. Thus, it is difficult to detect a positive correlation even if adult female *E. pustulosa* beetles did select particular clones/patches bearing higher fitness among all the examined clones/patches.

(2) Blue cohosh is a supplement host plant of *E. pustulosa*; no population of this beetle species depending exclusively on blue cohosh has been reported (Katakura 1981). It is possible, therefore, that *E. pustulosa* has been adapted to effectively utilize its main host plant, thistle. If so, a positive correlation may be observed only among thistle clones as detected in the present study.

(3) We used two patches of blue cohosh instead of two clones in larval rearing. Although the two patches were chosen based on the result of a choice test using two clones ($C1H_3 <<< C2H_4$), choice tests subsequently made between different clones from these two patches failed to detect such a definite preference for particular clones or patches (Table 3). A possibility remains that $C1H_3$ was an exceptionally less-preferred blue cohosh clone; the two blue cohosh patches might not be so different with respect to beetle preference.

(4) Alternatively, the quality of blue cohosh patches affecting adult preference may have changed temporally. If this temporal change really exists, a greater preference for $C\text{-}H_4$ (requiring longer developmental duration and higher mortality but yielding larger body size) is relatively adaptive early in the season, but a greater preference for $C\text{-}H_3$ (resulting in smaller body size but yielding lower mortality in shorter developmental duration) may be more adaptive late in the season. Such temporal change of plant quality was also suggested from the results of the interspecific choice test (Table 4, asterisked).

For these reasons, it cannot be concluded whether a positive correlation exists between adult preference and larval performance in *E. pustulosa*. Although still not conclusive, the results obtained in the laboratory are however at least not negative about the positive correlation.

It is not known how the observed qualitative variations in host plants affect the entire process of host selection by *E. pustulosa* under natural conditions. Host selection by a herbivorous insect is a complex process. It involves detection and location of its host from a distance, and confirmation of the appropriateness of that plant in terms of its species and quality (Bernays & Chapman 1994) managed by chemical characteristics of the plants (i.e. attractant, repellent, feeding or oviposition stimulant and deterrent). The roles of temporal and spatial distribution of plants, environmental difference in plant habitats, interactions with competitors, effects of predators, etc., are also important. All of these factors need to be clarified in order to understand the entire process of host selection. It is emphasized that the observed physiological ability of *E. pustulosa* in response to the qualitative difference in host plants should be taken as a basis for further studies on the *E. pustulosa* host selection.

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