Patterns of Phenotypic Plasticity in Common and Rare Environments: A Study of Host Use and Color Learning in the Cabbage White Butterfly *Pieris rapae*

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ABSTRACT: Phenotypic plasticity is adaptive in variable environments but, given its costs, may be disfavored if only one environment is commonly encountered. Yet species in relatively constant environments often adjust phenotypes successfully in rare or novel environments. Developmental biases may reduce the costs of plasticity in common environments, favoring the maintenance of plasticity. We explored this proposition by studying the flexibility of visually guided host-selection behavior in cabbage white butterflies (*Pieris rapae*), wherein common and rare environments consisted of green and red host types, respectively. We demonstrated in greenhouse assays that adult females display an innate bias toward green color during host search but alter that bias through learning in red-host assemblages such that, after several hours of experience, red hosts are located as efficiently as green hosts. Full-sib analyses suggested there was genetic variation in host and color choice that was more pronounced in the red-host environment. We found no evidence of genetic correlations in behavior across host environments or of fitness costs of plasticity in color choice. Our results support the idea that learning may persist in less variable environments through the evolution of innate biases that reduce operating costs in common environments.

Keywords: phenotypic plasticity, variable environment, learning, host use, *Pieris rapae*.

Introduction

Phenotypic plasticity, the ability of an organism to vary its phenotype, is an important means by which organisms cope with variation in environments over space and time (Schlichting and Pigliucci 1998). Theory suggests that phenotypic plasticity will be most useful when alternative types of environments occur with similar frequencies (Lynch and Gabriel 1987; Moran 1992; Sultan and Spencer 2002; Ernande and Dieckmann 2004). Yet environments often vary in frequency, with some being common and some rare. Given that plasticity has costs (e.g., Agrawal et al. 2002; Weinig et al. 2006; reviewed by DeWitt et al. [1998]), the commonness of a particular environment is expected to select against plastic phenotypes in favor of a fixed phenotype that is specialized for high performance in that environment (Moran 1992; Schlichting and Pigliucci 1998). Nevertheless, many species from relatively constant environments show a capacity to vary their phenotypes in other environments (Karban and Nagasaka 2004; Nicotra et al. 2007). Such species may even adjust appropriately to entirely novel conditions, such as those accompanying habitat shifts, species introductions, and climate change (Aubret et al. 2007; Muth and Pigliucci 2007; reviewed by Agrawal [2001]). For instance, some avian species have advanced laying date, possibly through learning, in response to phenological changes in prey availability caused by global warming (reviewed by Visser and Both [2005]).

Such observations raise the possibility that phenotypic plasticity is maintained in relatively constant environments because its overall cost is low. Can selection in such circumstances give rise to and maintain mechanisms of plasticity that are effective yet cost little? The answer to this question may depend on the nature of the cost (DeWitt et al. 1998; Sultan and Spencer 2002; Ernande and Dieckmann 2004). Some costs of plasticity are global, meaning that they are sustained in all environments. For instance, a global cost of learning (e.g., Mery and Kawecki 2003) may arise from the requirement that a brain be congenitally large in order for an animal to learn well (Johnston 1982; Dukas 1998; Laughlin et al. 1998). Because global costs are paid in all environments, including the common

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one, even modest global costs may favor fixed phenotypes adapted to the common environment. For example, selection may result in a brain of modest size whose neural circuits specify innate behavior suited to the common environment.

In contrast, some costs are not global in nature but are induced, expressed only in certain environments, such as under stressful conditions (e.g., Dorn et al. 2000; van Kleunen et al. 2000; Steinger et al. 2003). The process of learning involves certain "operating costs" of the learning process itself (e.g., Mery and Kawecki 2004), such as the addition of neural tissue with experience (Clayton and Krebs 1994; Snell-Rood et al., forthcoming; reviewed by van Praag et al. [2000]). Operating costs in a common environment will figure more in the maintenance of plasticity than will operating costs in a rare one (Ernande and Dieckmann 2004). If operating costs are low enough in the common environment, plasticity may be favored over a fixed phenotype, even if operating costs in the rare environment are substantial. Thus, we may expect selection to decrease operating costs in a common environment, such that operating costs of the learning process itself are specific to, or induced in, the rare environment.

One means of reducing operating costs in a common environment is a bias, or "educated guess," in development that predisposes an organism to perform well in that environment (e.g., Garcia and Koelling 1966; Smith 1977; Åbjörnsson et al. 2004). The organism may possess a capacity to change this "default setting" if and when a rare environment is encountered (e.g., flexible exploration; Mettke-Hofmann 2007), thus paying an operating cost only in the rare environment (fig. B1 in the online edition of the *American Naturalist*). For example, an animal may show congenital expression of foraging behavior suited to the common environment (i.e., innately biased behavior) but possess the capacity to learn a more appropriate behavior if and when the rare environment is experienced. Under this scenario, the forager pays an operating cost of learning only in the rare environment, since the innate bias precludes the need for learning in the common environment. Biases in development for high performance in common environments may explain why adaptive plastic responses to rare or even novel environments persist in populations under strong selection to adapt to common environments.

This study examined patterns of phenotypic plasticity in common versus rare environments, with a focus on innate versus learned behavior. We tested the prediction that, since plasticity costs are expected to be higher in rare environments than in common ones, performance should be higher in common environments than in rare ones. We additionally sought to determine whether a difference in performance was due to an innate bias predisposing high performance in the common environment, with learning playing a critical role in the rare environment. We next addressed the extent to which phenotypic plasticity could evolve if common environments became rare and rare environments common. The potential of a population to adapt to such change will depend in part on the extent of genetic variation in phenotype in each environment.

To address these objectives, we studied aspects of host selection behavior in the cabbage white butterfly (*Pieris rapae*), for which common and rare resource environments consisted of common and rare host types. Host selection behavior in this butterfly species is a useful model for studies in phenotypic plasticity. Oviposition behavior, in particular, has been well described. The cabbage white uses only plants in the family Brassicaceae as larval hosts (Shapiro 1975; Scott 1986). Gravid females use visual and olfactory cues to find host plants; after alighting, egg laying on host plants is elicited by contact with glucosinolates (Renwick and Chew 1994; reviewed by Courtney [1986] and Hern et al. [1996]). Naive *Pieris* females display a bias during host search toward green colors (Kolb and Scherer 1982; reviewed by Hern et al. [1996]). However, *P. rapae* females in laboratory assays using paper targets have been shown to learn colors other than green in the context of oviposition (Traynier 1984, 1986; reviewed by Hern et al. [1996]).

We focused our study on the female cabbage white's response to variation in host leaf color. Our putatively common and rare host environments consisted of known hosts of *P. rapae* whose foliage was either green (the putatively common host type) or red (the putatively rare host type). While most butterfly hosts bear green foliage, red foliage is not uncommon; and while larval pierids are well adapted to feed on common, visually apparent hosts (Chew and Courtney 1991), red hosts are often just as nutritious (Slansky and Feeny 1977). In fact, the distribution and function of red leaf coloration has drawn interest recently in the context of insect herbivory (Lev-Yadun et al. 2004; Schaefer and Rolshausen 2006). Of special interest with respect to host use in butterflies is the possible use of red as a cue in host selection. The distribution of red-sensitive opsins in the order Lepidoptera is variable (Briscoe and Chittka 2001; Briscoe 2008). Where they occur, sensitivity to long wavelengths has been proposed to play a role in host selection. In *Lycaena* butterflies, it has been suggested that a red-sensitive photopigment permits females to find their conspicuously reddish *Eriogonum* and *Rumex* host species (Bernard and Remington 1991). In citrus-feeding swallowtail butterflies, by contrast, it has been proposed that a red-sensitive opsin enables females to distinguish more preferred young yellow-green foliage from less preferred mature green foliage (Kelber 1999). The cabbage white has a long-wavelength

(LW) opsin with peak response at 563 nm (Briscoe 2008) that could potentially permit butterflies to detect plants with red foliage. *Pieris rapae* also possesses red-filtering pigments that could potentially improve color vision at longer wavelengths (Qiu and Arikawa 2003). The possibility that *P. rapae* females can discriminate red foliage from green foliage in host selection has not been evaluated in natural contexts.

We asked the following questions in our experiments: (1) In terms of success in finding hosts in an assemblage of hosts and nonhosts, is performance in a green-host assemblage higher than performance in a red-host assemblage? (2) Is any performance difference linked to a phenotypic trade-off in which color orientation responses are matched better to a green-host environment than to a red-host environment? (3) Does genetic (family-level) variation exist in the ability to find green hosts and red hosts and in a butterfly's propensity to orient to green versus red colors? (4) At the genetic (family) level, has strong selection in the common environment reduced the costs of plasticity and any trade-offs in performance across environments? Before addressing these questions, we surveyed the visual properties of cabbage white hosts, seeking to confirm that green hosts constitute a common resource environment and red hosts a rare resource environment.

Methods

Overview of Experiment

Female butterflies were reared to adulthood in a common-garden environment, allowed to mate, and then split among four treatment groups in a 2 × 2 factorial design. Females were assigned to one of two host-plant environments (green cabbage or red cabbage) that differed in foliar color and one of two nonhost contexts (simple or complex) that differed in diversity of nonhost plants (fig. B2 in the online edition of the *American Naturalist*). We tested host-search behavior in both simple and complex nonhost environments to increase the generality of our results for various plant communities (Agrawal et al. 2006) and to perform later tests on exploration as a cost of learning. In greenhouse arrays of each of these four treatment combinations, gravid females were permitted to search for hosts. Two aspects of host selection behavior were quantified: host-finding efficiency (a measure of the effectiveness with which females discriminated hosts from nonhosts before landing) and color choice (a measure of preference for green vs. red foliage). Possible genetic variability in these traits was assessed by analyzing variation among full-sib families tested across the four treatment combinations.

Insect Care and Establishment of Sibling Groups

Egg Collection from Females. Adult female *Pieris rapae* were obtained from field populations, and eggs were collected individually from these females. Females were held in 2-L plastic cages in full sunlight (south-facing window or greenhouse). Females were provided ad lib. access to 15% honey water and were also hand-fed daily. Various host plants (*Brassica oleracea* var. *viridis*, *acephala*, and *capitata*, *Lepidium* sp., or *Hirschfeldia incana*; all green) were presented to females each day, either in pots or as cuttings in water.

First-generation offspring from these females were assumed to be full siblings on the basis of published reports of mating frequency and sperm allocation. Dissections of field-caught females indicate that females have, on average, mated one or two times (E. C. Snell-Rood, unpublished data; Suzuki 1979). Even when females mate twice, fertilization is strongly biased toward one or the other male (>90% of brood sired by one mate; Shapiro 1970 and references therein; Wedell and Cook 1998). In this experiment, we assayed seven families of first-generation full siblings from females collected in Stony Brook, New York ($N = 2$), and San Diego, California ($N = 5$). A full-sib design does not strictly control for maternal effects; however, maternal effects in *P. rapae* appear to apply mostly to early stages of larval development (Rotem et al. 2003). Full-sibling families were included in analyses of genetic variation and family-level correlations. However, in analyses of overall patterns of learning, we also included individuals from "population lines" to roughly double our sample size. These butterflies consisted of four groups of third-generation butterflies derived from females collected in Hollister, California ($N = 2$), and Sedona, Arizona ($N = 2$). These lines were maintained in the lab through partial inbreeding (one generation of outbreeding, one of inbreeding) to supplement full-sibling groups when local field collections of *Pieris* became difficult.

Larval Rearing. Rearing methods were modified from Webb and Shelton (1988). Larvae were reared on an artificial diet containing green cabbage plant material (Troetschler et al. 1985): in grams per liter of water, 2 cabbage flour, 8 wheat germ, 4 casein, 3 sucrose, 1 Wesson salt mix, 2 torula yeast, 0.3 cholesterol, 0.2 sorbic acid, 0.1 methyl paraben, 0.4 ascorbic acid, 1 Vanderzandt vitamin mix, 0.02 streptomycin sulfate, and 2 agar, plus 3 mL linseed oil. As first or early-second instars, larvae were transferred with paint brushes from plant material to artificial diets. Larvae were reared in groups of three in 4-oz plastic cups (~1,500 total cups). Larvae were reared in a 12L : 12D incubator maintained at 23°C and 50% relative humidity.

Adult Care. Upon eclosion, adult butterflies were marked (black fine-tipped permanent marker) on the ventral hindwing with a unique identification number (independent of their family). Butterflies were held in glassine envelopes at 5°C until transfer to the greenhouse for mating and behavioral assays; individuals held longer than 2 days were hand-fed 15% honey water every other day. Efforts were made to hold constant across families the amount of time butterflies were held in the cold before testing. Before testing, butterflies were held in 1-m^2 cages in the greenhouse for at least 2 days. Cages contained three different flowering plants (e.g., phlox, penta, salvia, lantana) as natural nectar sources; in addition, females were provided with ad lib. access to brightly colored sponges soaked in 15% honey solution. In each cage, 10–20 females were placed with at least 10 reproductively mature males. The genetic background of females was randomized with respect to their greenhouse holding cage.

Experimental Arrays of Hosts and Nonhosts

Experimental plant arrays were generated for each of four treatments: (green/simple, green/complex, red/simple, and red/complex; fig. B2). Hosts in a given array consisted of either a green or a red variety of cabbage (*Brassica oleracea* var. *capitata*: Brassicaceae). The two varieties differed in leaf hue and luminance (fig. B3 in the online edition of the *American Naturalist*) but were similar in leaf shape and plant architecture. Reportedly, nutritional quality for *P. rapae* caterpillars is similar for both varieties (Slansky and Feeny 1977).

Nonhosts in a given array ($N = 40$) consisted of red and green plants in equal numbers. The nonhost assemblage varied in complexity as follows. The simple nonhost assemblage consisted of red and green varieties of a single species, the geranium *Ligularia dentate* (Asteraceae). The complex assemblage consisted of red and green varieties of geranium, red and green varieties of basil (*Ocimum basilicum*: Lamiaceae), red and green varieties of swiss chard (*Beta vulgaris* var. *cicla*: Amaranthaceae), red and green species of oxalis (*Oxalis stricta* and *O. rubra*: Oxalidaceae), and red and green variants of an *Ipomea* vine (*Ipomea batatas*: Convolvulaceae). Only vegetative parts of plants were present; any flowers were removed from the plants before the experiment. Spectral differences between red and green nonhost plants of a given species or genus were confirmed by using an Ocean Optics USB2000 spectrophotometer with a halogen-deuterium light source (fig. B3; E. C. Snell-Rood, unpublished data).

Plants were distributed in a 2 × 2-m area within a large nylon-screen flight cage (4 m × 4 m × 2 m) located inside a temperature-controlled glass-walled greenhouse. Plants were arranged by placing eight host plants in a Cartesian grid arrangement and then haphazardly filling in the remaining space with 40 nonhost plants. The locations of all plants were changed haphazardly at least once daily to prevent position effects, including spatial learning by the butterflies.

Behavioral Observations

Observation Conditions. Behavioral observations took place under sunny conditions between 0800 and 1330 hours. Temperatures ranged between 25° and 32°C (mean = 28.8°C, SD = 1.08°C) and relative humidity ranged between 25% and 70% (mean = 51.1%, SD = 11.1%). Butterflies from a given family were haphazardly assigned to treatment and observation groups on the day of their emergence. An observation group of butterflies consisted of 10–20 butterflies assigned to a particular treatment (e.g., green host, complex nonhost) and brought together to the experimental array for testing. Observation groups contained multiple families and were housed in the same cage in the greenhouse until testing (see rearing information above). Two or three observation groups were tested each day (at least 1 h each). Each observation group was observed for two successive days (thus, butterflies in each group were allowed to search for hosts for at least 2 h total).

Observation Methods. A butterfly was introduced into the plant array by placing it on a host plant, which generally induced the female to initiate host search. Host search is characterized by a slow, fluttering stereotypical flight, with frequent turning and hovering near plants. About three-quarters of the butterflies searched for hosts shortly after placement on a host; a small number of butterflies never engaged in host search and were excluded from analysis. Landings on foliage were considered to be part of host search when accompanied by tarsal drumming and occurring in the context of slow, fluttering flight. In contrast, landings were considered to be related to basking, resting, or foraging when accompanied by probing with the proboscis and/or resting with wings at a 45° angle.

During host search, females periodically alight on leaves, both hosts and nonhosts, drumming the leaves with foretarsi. Tarsal drumming is thought to be associated with chemoreception using tarsal receptors (reviewed by Renwick and Chew [1994]). If the plant is a nonhost plant, the female will usually take flight immediately and resume search. Eggs were never laid on nonhost plants. If the plant is a host plant, the female might leave, or she might curl her abdomen and, having curled her abdomen, might lay a single egg on the plant. We recorded the following data: landings on host and nonhost foliage, the color and species of the plant on which landings were made, and the oc-

currence of tarsal drumming, abdomen curling, and oviposition. Revisits to a given plant were counted as independent landings if butterflies landed on a different plant between a visit and a revisit.

The observer (E.S.-R.), blind to the family of each individual butterfly, was able to observe several butterflies from different families on an array simultaneously. Each butterfly in an observation group was relabeled (with a fine-tipped permanent marker) with a uniquely color-coded number in at least six locations on the wings, allowing for easy identification of individuals during behavioral observations from an observation seat at the edge of the array. During an observation session, butterflies were released in small groups of two to four into the host-plant array, thereby staggering search effort among group members in time. If butterflies made more than 30–40 landings, they were replaced on the array with the next butterfly. All behavior for all butterflies was recorded using The Observer software (Noldus) installed on a laptop computer. The use of computerized behavioral coding and color-coded identifiers, combined with the fact that butterflies often took frequent breaks during host searching (generally following oviposition), allowed the observer to record the behavior of several butterflies released simultaneously.

Behavioral Analyses

To be included in analyses of host-finding efficiency and color choice, an individual had to have made at least 10 landings during host search. We analyzed a female's behavior in two ways. First, to control for the time point in an individual's learning sequence, we arbitrarily divided a female's search record into successive bins of 10 landings each and analyzed data variously by bins and across bins. Second, we analyzed a female's behavior over the entire search record to get an idea of total host-finding efficiency and total color choice.

Host-Finding Efficiency. Host-finding efficiency, a measure of performance in finding hosts, was calculated as the proportion of all landings that were made on a host. For instance, if a female landed on four hosts and six nonhosts during landings 1–10, her host-finding efficiency score during this bin of landings would be 0.4 (4 of 10). Host-finding efficiency was calculated for two bins, landings 1–10 and 11–20, on each of two successive days of observations for a given female. Changes in host-finding efficiency were calculated as the difference in choice between landings 1–10 on day 1 and landings 1–10 on day 2.

Host-finding efficiency in our assays was highly predictive of a butterfly's reproductive output. Because host acceptance was high on both host types (landings on green and red host plants resulted in an egg being laid 91.9% and 92.2% of the time, respectively; $N_{\text{green landings}} = 3{,}402$; $N_{\text{red landings}} = 1{,}404$), the number of hosts found in an oviposition bout translates directly to the number of ovipositions in that bout. To the extent that landing "mistakes" on nonhosts take time, host-finding efficiency should be correlated strongly with the rate at which females lay eggs. As predicted, across all females on both host types, host-finding efficiency during the first 10 landings on day 1 was significantly positively correlated with oviposition rate ($N = 4{,}806$ host landings; $R^2 = 0.78$, $F_{1,422} = 1{,}539$, $P < .0001$).

Color Choice. Color choice, a measure of a female's response to green versus red color, was estimated in terms of "mistake" landings on nonhost plants bearing green versus red foliage. Color choice was scored arbitrarily as the proportion of all nonhost landings that were on green nonhosts. For instance, an individual that made four green nonhost and two red nonhost landings during landings 11–20 would receive a color choice score of 0.67 (four of six). Color choice scores were calculated during the same landing bins as host-finding efficiency: landings 1–10 and 11–20 on each of two successive days. Changes in color choice were calculated as the difference in choice between landings 1–10 on day 1 and landings 1–10 on day 2. Measures of host-finding efficiency and color choice were also quantified over an individual's entire search record in two ways. First, we measured "overall host-finding efficiency" (and overall color choice) as the proportion of total landings (or nonhost landings) on hosts (or green nonhosts). Second, in family-level analyses, we quantified "total hosts located" (and total color choice) over the entire search record as the total number of hosts (or green nonhosts) located while controlling for total landings (or total nonhost landings).

Statistical Analyses

All analyses were performed with JMP 7.0 (SAS Institute). Analyses of overall host finding behavior and learning were performed in several ways. First, we used ANOVA to test for effects of host color, nonhost complexity, and the interaction between these factors on host and color choice for binned landings over the 2 days of learning (all factors were treated as fixed effects). In this analysis, we included only those individuals that participated for at least 20 landings on day 1 of host finding and 10 landings on day 2 of host finding to ensure that changes in behavior over time attributed to learning were not due to "population processes," such as poorly performing individuals losing motivation over time (in general, analyses repeated with stringent participation criteria did not change the main

results). We used one-sample *t*-tests to test whether least squares means were significantly different from null expectations for each time point (20/40 = 0.5 for color choice; 8/48 = 0.167 for host-finding efficiency). Second, we used *t*-tests to determine whether butterflies showed significant changes in overall behavior over time. For instance, we tested whether the difference in host-finding efficiency between landings 11–20 and landings 1–10 was significantly greater than 0, which would suggest that butterflies were learning to locate hosts. Finally, we used ANOVA to test whether the color choice of individual butterflies (independent variable) was significantly related to their overall host-finding efficiency (dependent variable). In these overall analyses of host-finding behavior, we were interested primarily in determining general patterns of host learning. We included butterflies from both population lines and full-sibling lines to increase sample size, and thus we did not control for genetic background; when analyses were repeated with "family" (either full-sibling group or population line) as a factor, results reported here did not change qualitatively (Snell-Rood 2007).

Family-level analyses were performed using only data on groups of full-sibling butterflies. We tested for genetic variation in host-finding behavior and plasticity in this behavior, using mixed-model ANOVA ("traditional estimated mean square model"; in JMP, this model is handled like the SAS procedure GLM with a random statement). In these models, host color and nonhost complexity were treated as fixed effects, while family and the "family × host color" interaction were treated as random effects. We used this model to estimate family means (as least squares means) in each host-color environment while controlling for nonhost complexity and, in analyses of overall behavior, total landings. We repeated our tests for genetic variation using a linear mixed model (in JMP with restricted maximum likelihood [REML] estimation) because such REML methods can better accommodate unbalanced designs (Shaw 1987; Fry 1992; Windig 1997; Astles et al. 2006). Some of these analyses did not reach maximum likelihood convergence criteria in JMP (two of eight). We tested for significance of the family × host color effects by using log-likelihood tests, $X^2 = -2(L_i - L_0)$, where L_i is the log likelihood of the model including the term of interest and L_0 is the log likelihood of the null model that does not include this term. This statistic is distributed as χ^2, with degrees of freedom as the number of terms in the null model (Shaw 1987). These results are presented in appendix A in the online edition of the *American Naturalist*, but they generally confirm the significance of tests using mixed-model ANOVA.

We tested for genetic correlations in behavior across host environments, using Pearson product-moment correlations (r_m; Via 1984) with family means generated from mixed-model ANOVAs (above). Analyses repeated with least squares means generated from best linear unbiased predictors (BLUPs) in the linear mixed model (REML) did not differ (data not shown). While these correlations can sometimes underestimate genetic correlations, we used this method instead of calculating r_G from variance components (e.g., Fry 1992; Windig 1997; Astles et al. 2006) because variation was significantly lower in the green-host environment than the red-host environment (some of these variance components were negative, as estimated with REML methods), violating the assumptions of r_G calculation.

Finally, we tested for evidence of costs of plasticity, where plasticity was measured as a family's absolute difference in overall color choice (total green nonhosts chosen) between the two environments. First, we tested whether plasticity was related to a family's overall fitness (total hosts located) in each host environment. Next, we tested for costs of plasticity, using methods of DeWitt et al. (1998). Family-level color choice (independent variable) was correlated with family-level host-finding efficiency (dependent variable) in each host environment by means of linear regression. The residuals of the color choice–host-finding efficiency regression were plotted against plasticity: costs would be indicated by lower residual fitness of more plastic families in the environment in which fixed genotypes are specialized (the green host).

Natural Distribution of Host-Plant Color

To evaluate the notion that green host plants are common and red host plants are rare, we sought to quantify foliar color in naturally occurring *P. rapae* host plants. A complete list of host plants was obtained from Scott (1986). Scientific names were entered into Google image search to obtain one to three JPEG images of each of the 58 species living in natural ecosystems (an average of 2.7 images per species). The first three photographs of live plants (medium to high resolution, in good light with visible foliage, not simply flowers, stems, and/or seed pods) were chosen for subsequent analyses. The observer (E. S.-R.) first classified the foliage of each plant as "green," "green-red" (green with some red), "red-green" (red with some green), or red. Then, ImageJ (NIH) was used to quantify the red and green brightness of the foliage of these plants (using the "RGB measure" plug-in). The relative color of an image was quantified with an index equal to (green brightness minus red brightness)/(green brightness + red brightness). Finally, measurements were also made of JPEG images of hosts and nonhosts used in this experiment. The color indices computed from these measurements were compared with the distribution of host-plant color indices.

The above procedure is an imperfect method of quantifying foliar color from a butterfly's perspective. First, the RGB system is tuned to human color vision, which differs from butterfly color vision in multiple ways. Second, the use of random photographic images drawn from the internet is subject to many sources of variation, including type of camera and nature of lighting conditions. Nevertheless, after comparing RGB-based color indices of photos of the hosts and nonhosts used in our experiment with their actual reflectance spectra, we are convinced that the method is a useful first step in characterizing gross differences in foliar color among *Pieris* hosts that are of possible significance to the butterflies.

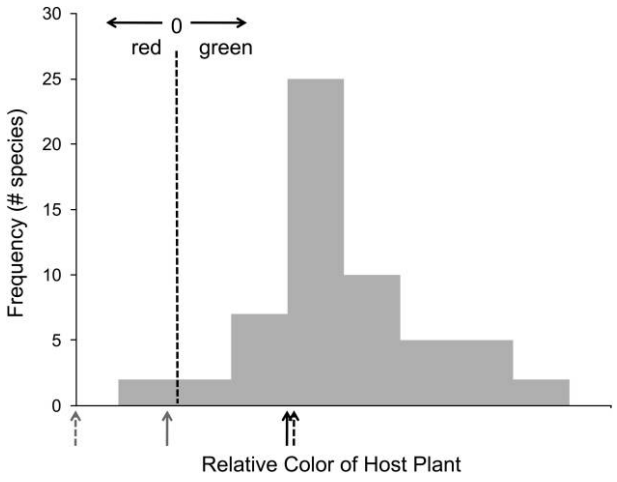

Figure 1: Color distribution of naturally occurring host plants of *Pieris rapae* ($N = 165$ photographs of 58 species in 35 genera). Relative color was measured as the difference between the green and red color channels (measured with ImageJ) corrected for luminance (the sum of the two channels). The dashed line indicates a relative color value of 0. To the left of the line, plants are red; to the right of the line, plants are green. Hosts (*solid arrows*) and nonhosts (geranium in the simple treatment; *dashed arrows*) used in this study were measured in the same way; green and red experimental plants are indicated by black and gray arrows, respectively. The overall frequency of red hosts was 4.4% for plants bearing at least some red foliage and 1.9% for plants bearing entirely red foliage.

Results

Commonness of Green and Red Hosts

In an RGB-based analysis of internet images, species of naturally occurring host plants ($N = 165$ measurements of 58 species in 35 genera) were highly biased toward green (fig. 1; $t_{57} = 14.5$, $P < .0001$) but included red forms as well. Categorical classifications of colors of individual plants by an observer (green, green-red, red-green, or red) were consistent with quantification of color using ImageJ ($F_{3,162} = 29.7$, $P < .0001$). Under this classification system, the frequency of naturally occurring individual host plants that consist of some and entirely red foliage was 4.4% and 1.9%, respectively. The color indices of host plants used in this experiment and nonhost plants used in the simple nonhost treatment were also measured by using this method: the green-host (and green-nonhost) indices fell within the range for the more common naturally occurring green hosts, while those of the red hosts (and red nonhosts) fell within the range for the rarer naturally occurring red hosts (fig. 1). The actual reflectance spectra of these hosts and nonhosts indicates that the RGB-based index captured a difference between those red and green plants in green reflectance centered at ~550 nm.

Phenotypic Pattern in Host-Finding Efficiency

Overall, individual performance in green-host arrays (mean = 0.47 [SE = 0.03]), as estimated by overall host-finding efficiency, measured over the entire period of search (proportion of total landings on hosts), was significantly greater than performance in red-host arrays (0.37 [0.03]; table 1). The performance difference was due primarily to a difference early in host search: the success with which females found hosts at the outset of host search (bins 1 and 2 on day 1) was significantly higher in the green-host array than in the red-host array (fig. 2*A*). By day 2, performance in the red-host array was not significantly different from that in the green-host array (table 1; fig. 2*A*).

In both host environments, the butterflies' ability to distinguish hosts from nonhosts improved over time (fig. 2*A*; tables 1, 2), although it improved more conspicuously in the red-host environment. In the green-host environment, host-finding efficiency was lowest in the first 10 landings on day 1 and relatively high thereafter (fig. 2*A*); at all time points, host-finding efficiency was consistently greater than that expected if butterflies were choosing plants randomly (random host-finding efficiency = 0.167; table A1 in the online edition of the *American Naturalist*). In the red-host environment, host-finding efficiency was lowest in the first 10 landings on day 1 and then improved progressively until late in day 2, when it became comparable to performance of individuals in the green-host environment (fig. 2*A*; tables 1, 2). In fact, host-finding efficiency in the red-host environment was not statistically distinguishable from random landing during the first 10 landings on the first day of host search (table A1).

For both host-finding efficiency and color choice, nonhost complexity generally had no significant (or a very minor) influence on behavior (table 1), suggesting that variation in the diversity of nonhosts was not nearly as

Table 1: Variation in host-finding efficiency and color choice with host color and nonhost complexity (ANOVA)

Landings	*F*-test df	Host color	Nonhost complexity	Color × complexity
Host-finding efficiency:				
1–10, day 1	1, 57	19.7***	3.72	2.16
11–20, day 1	1, 57	13.6***	4.18*	.82
1–10, day 2	1, 57	2.03	.03	.01
11–20, day 2	1, 27	.02	1.66	.77
All landings	1, 57	7.31**	1.65	.38
Color choice:				
1–10, day 1	1, 57	10.8**	.36	6.91**
11–20, day 1	1, 57	17.0***	.001	.04
1–10, day 2	1, 57	37.8***	.17	2.36
11–20, day 2	1, 27	27.3***	5.36*	21.9***
All landings	1, 57	34.82***	.07	1.23

Note: Shown are *F* values from ANOVAs that include host color (green or red), nonhost complexity (simple or complex), and the interaction between the two variables, all treated as fixed effects. This test included only individuals that participated in at least the first three bins shown ($N = 61$).

* $P < .05$.

** $P < .01$.

*** $P < .001$.

important in determining host finding as variation in color characteristics of the hosts. Thus, in subsequent analyses, we controlled for nonhost complexity, but we do not address its effects explicitly.

Phenotypic Pattern in Color Choice

The difference in host-finding efficiency was linked directly to a difference in color choice in the two host environments. We estimated color choice as the proportion of all nonhost landings that were on green nonhosts; a value of 1 indicates perfect green preference, and a value of 0 indicates perfect red preference (null hypothesis = 0.50). In the green-host environment, females displayed a very strong, consistent preference for green from the outset of host search, significantly greater than null expectations (fig. 2*B*; table A1). In the red-host environment, color choice changed significantly over time (table 2); the initial strong green bias declined until color choice across all butterflies was not statistically distinguishable from random (fig. 2*B*; table A1). Thus, even after two days of experience, color choice in the red-host environment was not nearly as biased toward red as color choice in the green-host environment was biased toward green (fig. 2*B*; table A1). Finally, color choice in the green-host environment was significantly more green biased than choice in the red-host environment for all four sampling bins (fig. 2*B*; table 1).

Association Between Host-Finding Efficiency and Color Choice

At the level of individual butterflies, overall host-finding efficiency (proportion of total landings on hosts) was correlated with color choice in the red-host environment (fig. 3*A*, 3*B*) but not in the green-host environment (fig. 3*C*, 3*D*; $N = 39$, $P > .35$). In the red-host environment, butterflies whose initial color choice (in the first 10 landings) was less green biased had more success in terms of overall host-finding efficiency (fig. 3*A*). Likewise, butterflies in the red-host environment that shifted to search for fewer green nonhosts across two days of host search had more success in terms of host-finding efficiency (fig. 3*B*; color choice in landings 1–10, day 1: $F_{1,16} = 48.4$, $P < .0001$; between-day change in color choice: $F_{1,16} = 19.4$, $P < .0001$; model also controlled for within-day change in color choice and for nonhost complexity).

Genetic (Family-Level) Variation in Host-Finding Efficiency and Color Choice

Full-sibling families ($N = 7$) reared in a common-garden environment showed significant variation in host-finding efficiency and color choice. When total nonhost landings were controlled for, there was a significant family × host color effect in a mixed-model ANOVA predicting total green nonhosts chosen during host search (table 3; fig. 4*B*). Similarly, with total landings controlled for, there were significant family × host color and family × host color × total landings effects in a model predicting total hosts located during host search (table 3; fig. 4*A*), indicating that families

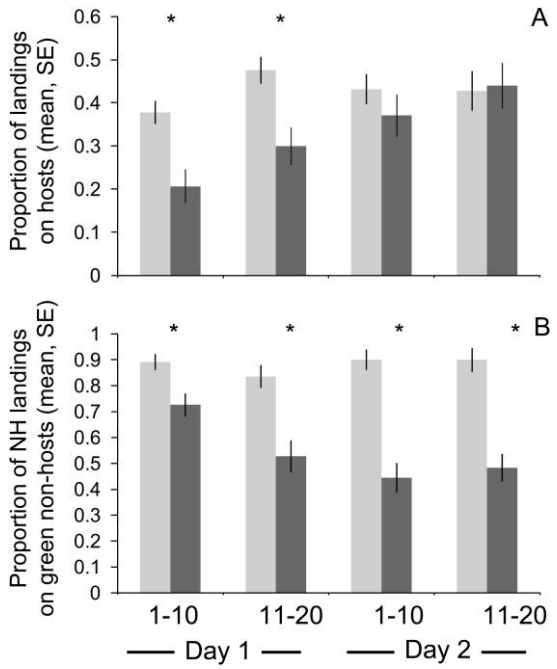

Figure 2: Pattern of host-finding performance and color choice in green (*light gray*; common host) and red (*dark gray*; rare host) host assemblages. *A*, Pattern of host-finding efficiency over time. Host-finding efficiency was measured as the proportion of landings on hosts. Host-finding efficiency was measured in bins of 10 landings on the first and second days of host searching. Asterisks indicate significant differences in host-finding efficiency between green and red host environments when we controlled for nonhost complexity and the interactions between host color and nonhost complexity. *B*, Pattern of color choice over time. Color choice was measured as the proportion of nonhost landings on green nonhosts. Color choice was measured in bins of 10 landings on the first and second days of host searching. Asterisks indicate significant differences in color choice between the red and green host environments when we controlled for nonhost complexity and the interactions between host color and nonhost complexity. This analysis included only butterflies (from both full-sibling families and population lines) that participated for at least 20 landings on day 1 and at least 10 landings on day 2. We also used *t*-tests to determine whether behavior differed significantly from null expectations (host-finding efficiency = 0.167, color choice = 0.50; see table A1 in the online edition of the *American Naturalist*).

differed in total hosts located and overall host-finding efficiency in the two environments. The supplemental linear mixed model (REML method) confirmed a significant family × host color interaction for overall host-finding efficiency but not for overall color choice (table A2 in the online edition of the *American Naturalist*). These patterns of genetic variation in overall host-finding efficiency and color choice were driven by the variable responses of families to the red- (but not the green-) host environment: when mixed-model ANOVAs were repeated for each host color, family effects were significant only in the red-host environment (table A3 in the online edition of the *American Naturalist*; fig. 4). This observation was further supported by the variance components analysis in a supplemental linear mixed model performed for each host environment (REML method): the family term accounted for 47.9% and 26.9% of the total variance in hosts and green nonhosts, respectively, located in the red-host environment but only 0.03% and 3.5% of the total hosts and green nonhosts, respectively, located in the green-host environment.

Full-sibling families showed genetic variation in host-finding efficiency and, to some extent, color choice when these measures were quantified specific to bins of landings during the search record (tables 4, A2, A3; fig. 5). In particular, mixed-model ANOVA revealed significant family × host color effects for host-finding efficiency in each search record considered (table 4; fig. 5*A*–5*C*); these interactions were also significant in the supplemental mixed-model (REML method) analysis (A2). Crossing reaction norms were observed for color choice but were not significant (tables 4, A2; fig. 5*D*–5*F*; table A2). Similar to patterns of overall behavior, genetic variation in behavior specific to search bins was driven by the response of families to the red-host environment (table A3); the family effect was a significant determinant of binned host-finding efficiency and, to some extent, color choice in the red-host environment.

Visual inspection of reaction norms (figs. 4, 5) revealed several trends. First, reaction norms changed over time (increasing variability), likely as a result of learning (fig. 5). Second, some individual families appeared to decrease color choice in the red-host environment to values significantly biased toward preference for red (<0.50), even though pooled butterflies appeared to choose nonhost colors randomly later in host search (fig. 2; table A1). For instance, by the first 10 landings on the second day of host searching in the red-host environment, one family was choosing significantly more red nonhosts than green nonhosts (least squares untransformed mean = 0.15 [SE = 0.09], $t_7 = -3.88$, $P = .006$; fig. 5*C*).

Trade-Offs in Host Performance and Costs of Plasticity

There was no evidence of a genetic correlation in a full-sib family's host-finding ability in either environment; total hosts located by a family in the red-host environment was not correlated with that family's total hosts located in the green-host environment (fig. B4*A* in the online edition of the *American Naturalist*; total hosts located over all landings, $N = 7$ families: $r = -0.27$, $P = .59$). Similarly, there was no evidence of a trade-off between a full-sib family's total color choice in either host environment: total green nonhosts chosen by a family in the red-host environment was not correlated with the same measure in the

Table 2: Changes in host-finding efficiency and color choice with time

	Green host			Red host		
	t	df	*P*	*t*	df	*P*
Host-finding efficiency:						
Change within day 1	4.14	106	<.0001	4.75	73	<.0001
Change between days 1 and 2	2.16	59	.03	2.95	40	.005
Color choice:						
Change within day 1	−.44	106	.66	−3.93	73	.0002
Change between days 1 and 2	1.11	59	.27	3.87	40	.0004

Note: Host-finding efficiency was measured as the proportion of landings on hosts for bins of 10 landings. Color choice was measured as the proportion of nonhost landings on green nonhosts for bins of 10 (host and nonhost) landings. Changes in host-finding efficiency and color choice over time were transformed and analyzed with a *t*-test to determine whether the difference between behavior at two points of time—either within day 1 (landings 11–20 minus landings 1–10) or between the two days of learning (landings 1–10 [day 2] minus landings 1–10 [day 1])—was significantly different from 0.

green-host environment (fig. B4*B*; $N = 7$, $r = 0.087$, $P = .87$).

We found that the plasticity of a family (the absolute difference in total color choice between host environments) was positively related to total hosts located in the red-host environment (fig. 6*B*; $F_{1,5} = 6.68$, $P = .04$, β [slope] $= 1.21$) but not that in the green-host environment (fig. 6*A*; $F_{1,5} = 0.002$, $P = .97$, $\beta = -0.01$). We also used the methodology of DeWitt et al. (1998) to estimate costs. A family's total color choice in an environment was regressed against their total hosts located in that environment (green environment: $F_{1,5} = 2.35$, $P = .18$, $\beta = 0.49$; red environment: $F_{1,5} = 12.23$, $P = .02$, $\beta = -1.45$). The residuals from this regression were then

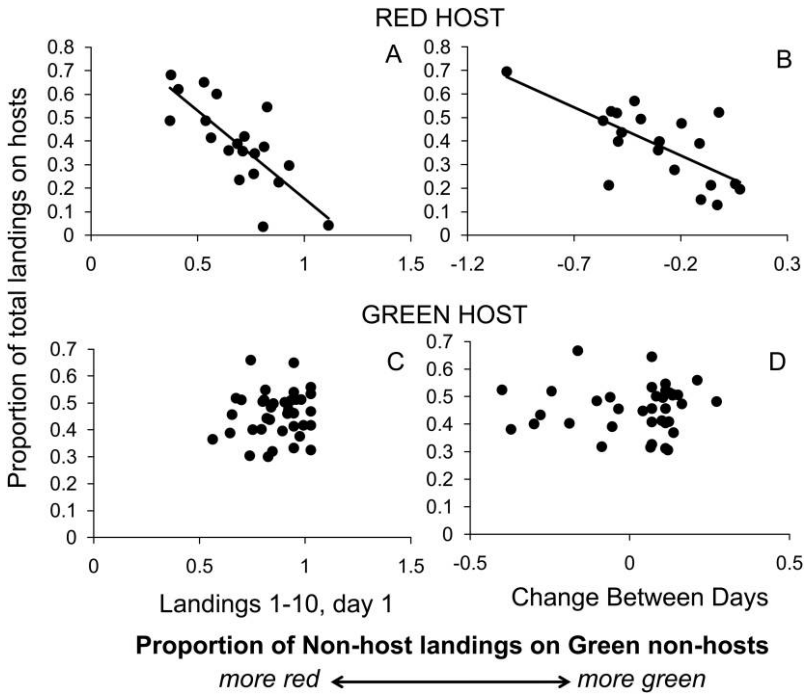

Figure 3: Host-finding efficiency as a function of color choice. Shown are leverage plots from a model that included nonhost complexity, initial color choice (landings 1–10 on day 1), and change in color choice within and between days of host learning. Change in color choice is measured such that more negative values represent a shift to searching for more red colors. Overall host-finding efficiency was measured as the proportion of total landings (over both days of learning) that were made on hosts. Color choice was measured as the proportion of all nonhost landings that were made on green nonhosts for the first 10 landings on day 1 of learning and as changes within and between the two days of host search.

Table 3: Genetic variation in total hosts located and total color choice (mixed-model ANOVA)

	Total hosts located				Total color choice			
	df	MS	*F*	*P*	df	MS	*F*	*P*
Host color (HC)	1	817.9	7.82	.02	1	78.3	5.21	.03
Nonhost complexity	1	44.5	2.82	.09	1	2.58	.29	.59
Family	6	98.2	.71	.65	6	32.2	1.05	.47
Landings[a]	1	823.2	52.1	<.0001	1	504.0	56.4	<.0001
Family × landings	6	49.0	.68	.67	6	25.6	1.61	.29
Family × HC	6	139.3	8.65	<.0001	6	30.4	3.34	.005
Landings × HC	1	81.9	2.64	.13	1	1.73	.17	.67
Landings × HC × family	6	71.9	4.55	.0004	6	15.8	1.76	.11
Error	91	15.8			91	8.93		

Note: Results from a mixed-model ANOVA (JMP). Total hosts located (or color choice) was measured as the total number of hosts (or green nonhosts) an individual located when we controlled for host color, nonhost complexity, total landings (or total nonhost landings), full-sibling family, and several interaction terms. Family and all interaction terms including family were treated as random effects.

[a] Total landings for host-finding efficiency; total nonhost landings for total color choice.

plotted against plasticity, the absolute difference in total color choice between the two environments. By this method, there was no trade-off between plasticity and host-finding ability in the common (green) environment ($F_{1,5} = 0.71$, $P = .43$, $\beta = -0.17$) or the rare (red) environment ($F_{1,5} = 0.06$, $P = .81$, $\beta = 0.10$).

Discussion

The Evolution of Performance Differences

How phenotypic plasticity arises and is maintained in common versus rare environments has been a focus of theory (Lynch and Gabriel 1987; Moran 1992; Sultan and Spencer 2002; Ernande and Dieckmann 2004), but tests of predictions derived from theory are relatively few. Also scarce are studies of sensory and cognitive traits within a phenotypic-plasticity framework. Yet our results suggest that visual systems and associated cognitive processing, including learning, are appropriate to treat within this context.

Our prediction that performance in a common environment would be greater than that in a rare environment was met with respect to host-finding efficiency in cabbage white butterflies (table 1; fig. 2). Specifically, the effectiveness with which females discriminated hosts from nonhosts before landing was better in the more typical green-host environment than in the less typical red-host environment. A female's capacity to discriminate hosts from nonhosts is related directly to her oviposition rate (see "Methods"). If *Pieris* butterflies are time limited, an increase in oviposition efficiency should increase a female's fitness directly. In contrast, if butterflies are egg limited, an increase in oviposition efficiency may permit females to be choosier about the quality of the plants on which eggs are laid, which would increase female fitness indirectly through an increase in offspring fitness. The high accep-

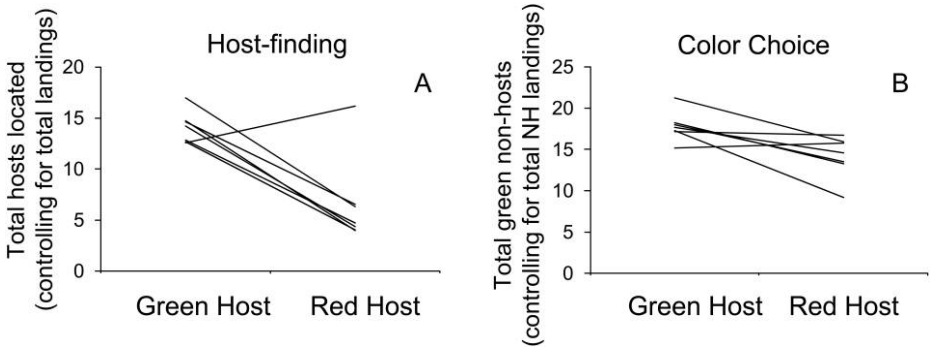

Figure 4: *A*, Overall host-finding efficiency measured as total hosts located (with total landings controlled for). *B*, Overall color choice measured as total green nonhosts chosen (with total nonhost landings controlled for). Shown are least squares means from mixed-model ANOVAs in which family and family × host color are treated as random effects.

Table 4: Genetic variation in host-finding efficiency and color choice (mixed-model ANOVA)

	Host color		Complexity		Family		Family × host color	
Landings	*F*	df	*F*	df	*F*	df	*F*	df
Host-finding efficiency:								
1–10, day 1	17.8**	1, 102	.00	1, 102	.98	6, 102	4.81***	6, 102
11–20, day 1	6.93*	1, 54	.94	1, 54	1.19	6, 54	4.93***	6, 54
1–10, day 2	.51	1, 26	.01	1, 26	.49	4, 26	3.11*	4, 26
Color choice:								
1–10, day 1	13.5**	1, 102	.21	1, 102	1.48	6, 102	1.28	6, 102
11–20, day 1	11.0**	1, 54	.18	1, 54	1.63	6, 54	1.29	6, 54
1–10, day 2	32.7**	1, 25	.01	1, 25	4.15	4, 25	1.37	4, 25

Note: *F* values from a mixed-model ANOVA (JMP) that included host color, nonhost complexity, family, and the family × host color interaction. Family and the family × host color interaction were treated as random effects. Host-finding efficiency (the proportion of landings on nonhosts) and color choice (the proportion of nonhost landings on green nonhosts) were the dependent variables and were measured in bins of 10 landings.

* $P < .05$.

** $P < .01$.

*** $P < .001$.

tance of host plants in our assays suggests that females were behaving as though time limited under those conditions; whether *Pieris rapae* females are time limited in nature is not known.

Also uncertain is the spatial and temporal distribution of green and red hosts in nature. Our preliminary survey of North American *P. rapae* host species suggests that red host types do occur (fig. 1). However, we do not know the extent to which a female encounters just a single host type in her lifetime, as opposed to a mixture. Field data suggest that at least some female pierids oviposit on a variety of host species, but individual sequences of female oviposition may be biased toward one species because of host aggregations or female choice (Chew 1977; Ives 1978; Stanton 1982, 1984). In our survey of plant visual characteristics, we characterized a given host species in terms of a mean value, but the variance around those means is also of interest. Some plant species are polymorphic in leaf coloration over space and time. The precise distribution of host foliar coloration is likely to be as crucial

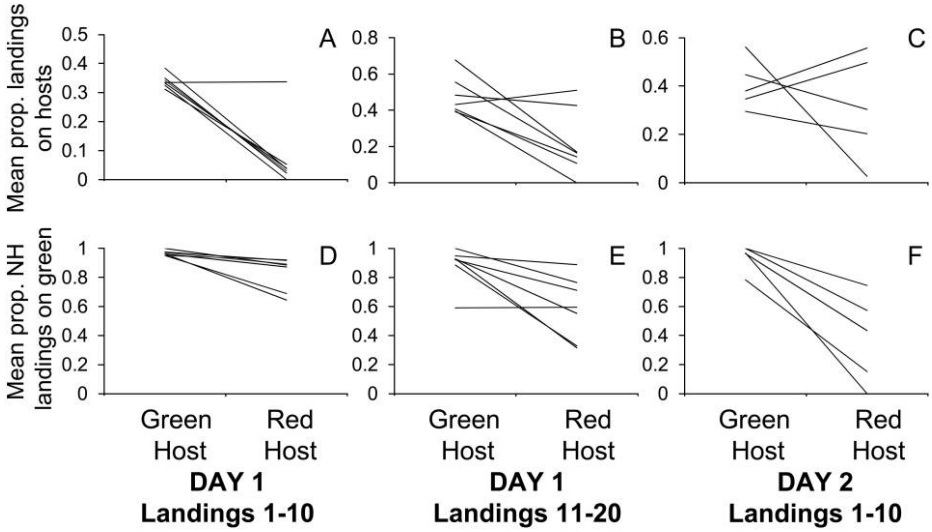

Figure 5: Reaction norms of full-sib families for host-finding efficiency and color choice in green (common) and red (rare) host arrays, arranged by period of search. *A–C*, Host-finding efficiency, measured as the proportion of all landings during a host search period that were made on hosts (vs. nonhosts). *D–F*, Color choice, measured as the proportion of all nonhost landings that were made on green nonhosts. Shown are least squares means from mixed-model ANOVAs where family and family × host color are treated as random effects.

in shaping plasticity in host selection behavior as are the costs of plasticity.

Color Vision and Learning as Elements of Plasticity in Host Use

We suggested in the "Introduction" that selection in common and rare environments should result in the evolution of foraging behavior innately biased to function in the common environment but capable of modification through learning in a rare environment. In that case, the forager pays an operating cost of learning only in the rare environment, leading to lower performance in that environment. Our data are consistent with this mechanism. Specifically, host selection in *P. rapae* females appears to be biased innately toward green-leaved host plants; however, females given contact with red-leaved hosts learn to orient toward the red color.

We believe that patterns in color choice (figs. 2, 3) are evidence of the use of true color vision in host selection. However, because green and red foliage differ not only in color but in other visual properties such as luminance (fig. B3), our results do not prove that butterflies were using true color vision to distinguish red and green foliage. It is conceivable that butterflies were learning that red cabbage was dark against background, not red in terms of long wavelength. Nevertheless, our results are consistent with physiological evidence of an LW opsin and red-filtering pigments in *P. rapae* (Shimohigashi and Tominaga 1991; Qiu and Arikawa 2003; Briscoe 2008) and with behavioral evidence that *Pieris* butterflies are sensitive to wavelength during host search (Kolb and Scherer 1982).

Color responses appear to be matched by default to the common green-host environment, resulting in high host-finding efficiency from the outset of host search in the green-host arrays. We interpret the improvement in host-finding efficiency over time in the red-host assemblage (table 2; figs. 2, 3) as evidence of learning. There is abundant evidence of associative learning in insects, including learning during nectar foraging and host selection in butterflies (Papaj and Prokopy 1989; Weiss and Papaj 2003 and references therein). Our experiments provide further evidence that the improvement in host-finding efficiency is due at least in part to color learning; evidence of color learning in host selection in *P. rapae* butterflies has been reported previously (Traynier 1984, 1986; reviewed by Hern et al. [1996]).

The difference in performance in the two host environments appears to be due at least in part to the time required to learn red color in the red-host environment. This time requirement can be considered an operating cost of the learning process (e.g., Mery and Kawecki 2004), at least as measured relative to a hypothetical genotype that had an innate bias toward searching for red colors.

While butterflies showed significant changes in color choice over time, responses to red foliage never became as strong in the red-host environment as responses to green foliage were in the green-host environment (fig. 2; table A1). The best that most females achieved in the red-host environment was nonpreference with respect to color. The fact that red preference never became as strong in the red-host environment as green preference was in the green-host environment suggests that learning cannot completely offset the innate bias toward green. Similar patterns have been observed in the pipevine swallowtail butterfly *Battus philenor*, where innate biases toward green persisted even after other colors were learned (Weiss and Papaj 2003). Innate biases of bumblebees (*Bombus terrestris*) also continue to influence flower choice after training to novel colors (Gumbert 2000; Lynn et al. 2005).

The occurrence of an innate green bias and a capacity to learn red is an example of an "open program" of behavior (Mayr 1974), consisting of useful combinations of innate and learned components. Such programs may be the rule rather than the exception, yet our study is one of the few that explores the functional consequences of mixtures of innate and learned behavior in a variable environment. Our work suggests that the commonness of an environment can drive the evolution of an innate bias, while the persistence of rare environments can maintain the capacity to modify the innate bias through learning. It is possible that learning may even buffer further selection to increase innate biases toward green.

Genetic Variation in Use of Rare Environments

Our research is the first to estimate the degree of genetic variation in a behavior with both learned and innate components in relation to variation in the resource environment. In our study, butterfly families showed significant family × host color interactions for measures of behavior. A number of generalizations can be drawn from the reaction norms (tables 3, 4, A2, A3; figs. 4, 5). First, there were no "supergenotypes" that performed better than any other genotypes in both red- and green-host environments ("jack-master" genotypes; Richards et al. 2006). Second, reaction norms for color choice and host-finding efficiency tended to become more variable over time, likely because of learning processes (fig. 5).

Third, family-level variation in the red-host environment was greater than that in the green-host environment (table A3; figs. 4, 5). This pattern is somewhat reminiscent of the findings of studies in which genetic variation is revealed or more pronounced under novel or stressful conditions (e.g., Etterson 2004; Donohue et al. 2005; Sangster

et al. 2007). These results may reflect a difference in the intensity of selection in the two environments. Strong selection to find the more commonly encountered green hosts may have driven effective use of green hosts and depleted genetic variation in host selection traits, leading to host-finding efficiency and color choice values that are uncorrelated with each other and "bunched up" at high values in the green-host assemblage (figs. 4, 5). In contrast, relatively weak selection for finding red hosts, in conjunction with the generation of variation through mutation and gene flow, has possibly resulted in maintenance of the significant genetic variation in red-host use observed in our assays. In the red-host environment, moreover, there was a significant family-level correlation between host-finding efficiency and color choice. Given an increase in the frequency of the red-host environment, such as widespread plastic response to rising photic stress (Chalker-Scott 1999; Gould 2004), one might expect a relatively rapid response of butterfly populations to selection.

Costs and Trade-Offs and the Maintenance of Variation in Plasticity

We predicted that strong selection in common environments and the evolution of innate biases for such environments would diminish the overall costs associated with plasticity. Consistent with this prediction, we found no fitness costs of plasticity in terms of host-finding efficiency (fig. 6), similar to the findings of some other studies on the costs of plasticity (e.g., DeWitt 1998; Scheiner and Berrigan 1998; Steiner and van Buskirk 2008): families able to vary color choice across host environments, relative to less plastic families, suffered no host-finding cost in the common, green-host environment, as measured by absolute and residual host-finding ability (fig. 6; methods of DeWitt et al. 1998). In other words, we found no evidence for a true "cost of naivete" (e.g., Greenberg 1983; Laverty and Plowright 1988), whereby plastic genotypes have to take the time and energy to learn regardless of the environment encountered (Johnston 1982; Dukas 1998; Mery and Kawecki 2004): good learners in this system appear to avoid these costs in the common environment through the use of an innate preference for green. Furthermore, although different phenotypes were favored in the use of the common versus rare host types (figs. 2, 3), there was no evidence of a genetic trade-off at the level of full-sib families (fig. B4). Families that were relatively more effective in host-finding efficiency and color choice in the green-host environment were not necessarily less effective in the red-host environment. This lack of a trade-off suggests that selection on host use in the one environment will not be constrained by selection in the other environment.

Taken together, these results are consistent with our prediction of cost reduction in the common environment. However, the complete lack of fitness costs and trade-offs in performance across environments, together with the finding of at least some fitness benefits of plasticity in the rare environment (fig. 6*B*, although not when estimated with the methods of DeWitt et al. 1998), raise the question of why variation in plasticity is maintained in this system. It is likely that plastic genotypes suffer other fitness costs that we did not measure in this study. For instance, one of the hypothesized costs of learning is a delay in reproduction (Johnston 1982; Dukas 1998) as good learners invest more in "production costs" of developing the neural tissues necessary for the ability to learn (DeWitt et al. 1998; Mery and Kawecki 2004). We may have missed behavioral signatures of this cost by measuring learning in butterflies that were at least 2 days old. It is also possible that actual differences in host-finding ability would be amplified in

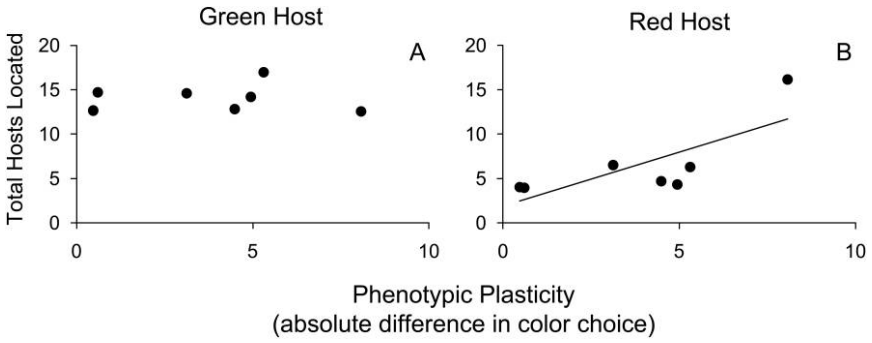

Figure 6: Costs of plasticity. Plasticity was measured as the absolute difference in total color choice across environments. In this analysis, costs were measured as total hosts located (fitness) in the common (green) host environment (*A*), according to the methods of DeWitt et al. (1998). Shown are least squares means for each full-sibling family.

the field: green-specialized genotypes may be better able to use olfactory cues to locate patches of hosts from long distances (e.g., Stanton 1982) or quickly disperse from patches of rare, less apparent hosts (E. C. Snell-Rood unpublished data).

While our results support our overall hypothesis, there are several alternative hypotheses and points to consider. First, selection on learning in other contexts may facilitate the maintenance of the ability to learn rare hosts. For instance, butterflies commonly learn motor patterns and color associations during nectar foraging (e.g., Lewis 1986, 1989; Weiss and Papaj 2003), and nectar plants are highly variable over space and time. It is possible that selection on plasticity in contexts such as learning nectar plants results in increases in plasticity in other behaviors; this hypothesis has been put forward to explain the lack of variation in host-learning ability between butterfly populations that use one or multiple hosts (Papaj 1986). Second, we were able to measure the host-finding behavior of only seven full-sibling families in this experiment. We may have been able to detect small genetic correlations and costs of plasticity if we had been able to include more families. Finally, our hypothesis speaks to the maintenance of plasticity in common versus rare environments; however, because of the difficulties of measuring host-finding behavior in a range of environments (which required rearing several thousand butterflies), we considered only one pair of common and rare environments, green and red hosts. It would be ideal to extend this experiment to consider other contrasts of common and rare environments: not only other rare-colored hosts (e.g., blue-tinged leaves, such as are found in kale) but also rare host shapes or chemistries, rare nonhost resources, such as rare nectar plants, or rare mates. Given our consideration of only one pair of environments, the generality of our results to the evolution of plasticity should be interpreted with caution until tests in other environments and other systems have been conducted.

Predicting Population Responses to Environmental Change

Given substantive family-level variation in host use in the red-host environment, *Pieris* populations ought to respond relatively rapidly, in an evolutionary sense, if red hosts were to become more common. Such change is not inconceivable. Photic stress and high temperatures are thought to play a role in the production of red pigments in plants (Chalker-Scott 1999; Steyn et al. 2002; Gould 2004). It is not inconceivable that climate change could increase production of red host foliage in many parts of the butterfly's range.

Host range in cabbage whites has been shown to be dynamic in nature. After being introduced into North America, *P. rapae* incorporated novel native hosts into its host range (Graves and Shapiro 2003). Similarly, other pierids have adopted novel nonnative hosts and subsequently evolved increased larval performance (Keeler and Chew 2008). The focus of our study was a host resource that is presumed to be rare, though not necessarily novel. However, we maintain that the maintenance of plasticity in host use in relation to rare host resources may result in rapid evolution of populations in response to novel resources. By extension, adult (or even larval) butterflies may have the ability to respond adaptively to rare or novel environments with respect to other cues involved in host search or other behavior, such as flower choice and handling. In general, the evolution of developmental biases may reduce the costs of plasticity in common environments such that the ability to respond adaptively to rare environments is retained in populations. This research adds to a rapidly growing body of information about how populations might respond developmentally and genetically to novel and changing environments.

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