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STRONG NATURAL SELECTION IN A WARNING-COLOR HYBRID ZONE

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Abstract.—Frequency-dependent selection on warning color can maintain narrow hybrid zones between unpalatable prey taxa. To measure such selection, we transferred marked *Heliconius erato* (Lepidoptera: Nymphalidae) in both directions across a 10-km-wide hybrid zone between Peruvian races differing in color pattern. These experimental *H. erato* were released at four sites, along with control *H. erato* of the phenotype native to each site. Survival of experimental butterflies was significantly lower than that of controls at two sites and overall. Most selection, measured as differences in survival, occurred soon after release. Selection against foreign morphs was 52% (confidence limits: 25–71%) and was probably due to bird attacks on unusual warning-color morphs (more than 10% of the recaptures had beak marks). Since only three major loci determine the color-pattern differences, this suggests an average selection coefficient of 0.17 per locus, sufficient to maintain the narrow clines in *H. erato.*

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Hybrid zones and clines are areas where genetically distinct populations, subspecies, or partially interfertile species meet. Clines repay investigation because they are stabilized by selective factors similar to those constraining the initial evolution of differences between the interacting forms. Two sorts of stable clines are generally recognized: clines between forms that are differently adapted to abutting environments (Haldane, 1948; Fisher, 1950; Jain and Bradshaw, 1966; Slatkin, 1973; Endler, 1977; Moore, 1977) and clines maintained by heterozygote disadvantage or selection against recombinants, as in clines of chromosomal rearrangements (Bazykin, 1969; Barton, 1979). However, a third class of clines can be maintained by frequency-dependent selection favoring the most common form (the opposite, "apostatic" selection favoring rare forms, causes global polymorphism rather than a stable cline). These clines have stable widths because selection acts against individuals that disperse across the cline to areas where their own phenotype is rare. Although selection is quite different at the genotypic level, clines maintained by frequency-dependent selection and those stabilized by selection against hybrids are both frequency-dependent at the gene level; in fact, linear frequency-dependent selection on genotypes (see Mallet, 1986*a*) leads to gene-frequency recursion equations that are identical to those for the case of heterozygote disadvantage (Barton, 1979). Therefore, underdominance and frequency dependence favoring the more common form give equal postmating isolation for any given selection pressure. Since selection depends on the genetic composition of the clinal populations, rather than on adaptations to the local environment, a cline that is maintained by frequency-dependent selection or heterozygote disadvantage is mobile (Bazykin, 1969; Barton, 1979; Mallet, 1986*a*). Such clines are termed "tension zones" (Key, 1981; Barton and Hewitt, 1985), because their maintenance is by intrinsic, rather than extrinsic, environmental sources of selection. In practice, tension zones are hard to distinguish from environmentally stabilized clines, because they can be trapped at ecological discontinuities which inhibit dispersal.

Clines maintained by frequency dependence may exist in warningly colored butterflies (Mallet, 1986*a*), as well as between races under some kinds of sexual selection (Moore, 1979, 1987; Johnson et al., 1987). Warning colors serve as protection from predation because predators associate a color pattern with an unpleasant experience. Prey of each color pattern are sampled by predators during learning. If a common prey form and a rare prey form are equally con-

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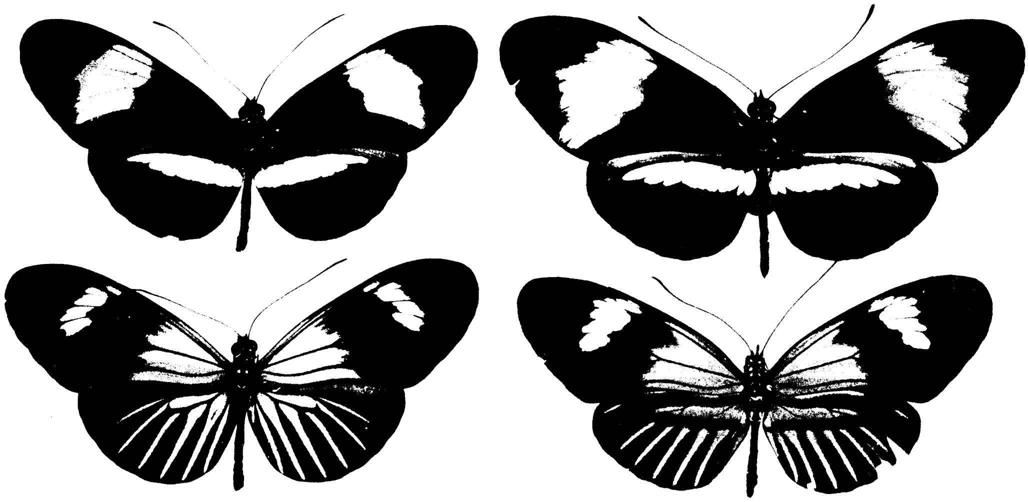

FIG. 1. "Postman" and "rayed" *Heliconius* near Tarapoto, Peru. Top row: postman pattern (black with crimson forewing band and yellow hindwing bar); bottom row: rayed pattern (black with yellow forewing band, orange forewing proximal patch, and orange hind-wing rays); left: *Heliconius erato*; right: *H. melpomene.*

spicuous, memorable, and unpalatable, naive predators should take about the same number of each type of prey to learn the patterns and thus take a higher proportion of the rare form. Thus, frequency-dependent selection favors the most common form (Greenwood et al., 1989). This selection results in monomorphism within populations (Clarke, 1962; Mallet and Singer, 1987; Endler, 1988), leads to Müllerian mimicry within communities (Müller, 1879; Turner, 1977), and helps to maintain clines (Mallet, 1986*a*).

The South American butterflies *Heliconius erato* and *H. melpomene* (Lepidoptera: Nymphalidae) are unpalatable to birds (Brower et al., 1963; Boyden, 1976; Chai, 1986), and each species has divergent color-pattern races separated by narrow clines (Turner, 1971*a*; Brown et al., 1974; Lamas, 1976; Mallet, 1986*a*). Except in regions where one species is absent, races of the two species mimic each other; *H. melpomene* has probably tracked the divergence of the more common *H. erato* by Müllerian mimicry, although some coevolution may have occurred (Eltringham, 1916; Gilbert, 1983). The existence of this striking mimicry, the narrowness of many of the clines (as narrow as 10 km), the concordance of clines between the species, and cage experiments demonstrating the effectiveness of *Heliconius* mimicry (Brower et al., 1963; Boyden, 1976) all point to a strong warning function for the bright color patterns of *Heliconius.*

In this paper, we present a direct estimate of the strength of natural selection on *H. erato* at a hybrid zone in Peru. We transferred live adult butterflies across the hybrid zone and compared their survival with that of controls. We argue that the strong selection we measured against "foreign" butterflies is due to warning-color differences, rather than to differing ecological adaptations of the races. However, we were not able to confirm directly that the differences in survival of experimental and control butterflies were due to predation. Our protocol improves on experiments that use paint to create or alter mimetic patterns, since the patterns in our butterflies are natural. A possible drawback of our method is that, by using variants taken from different populations, we may find differences in survival due to genes affecting traits other than color pattern. However, selection due to differences in habitat requirements of the two races could also help maintain the hybrid zone; as far as the structure of the zone is concerned, the mechanism of selection is not crucial (Barton and Hewitt, 1985).

MATERIALS AND METHODS

In the Department of San Martín, Peru, there is a hybrid zone between two divergently patterned races of *H. erato.* These are

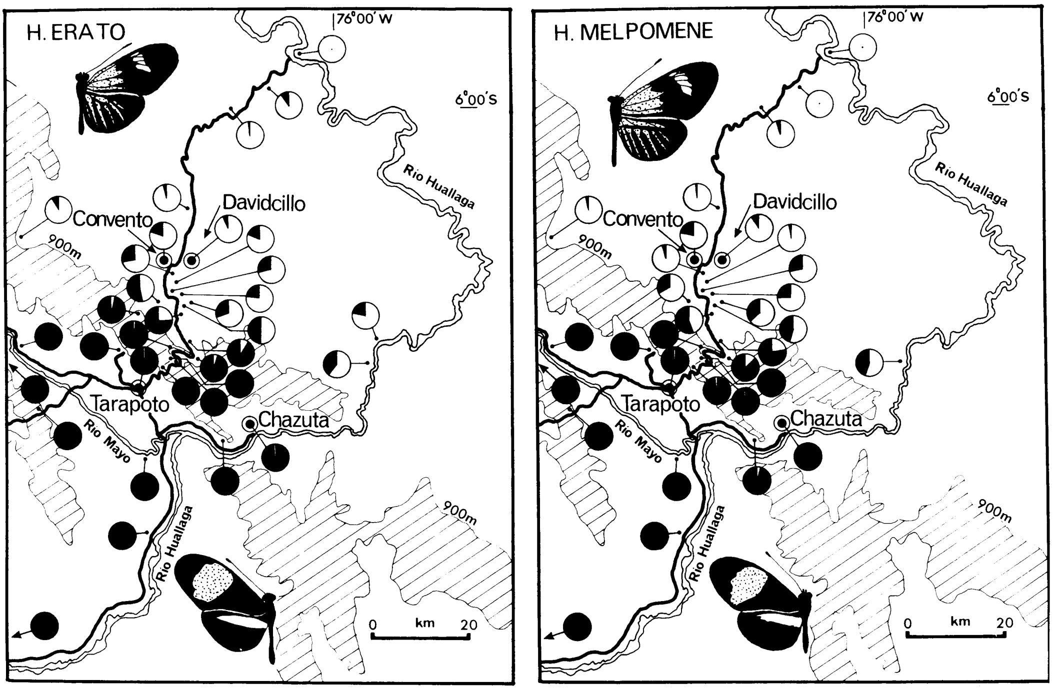

FIG. 2. Maps of the hybrid zones of *Heliconius erato* and *H. melpomene.* Release sites are circled and named; broad lines represent roads. Pie diagrams show the average gene frequencies at three major loci determining color pattern in *H. erato* and four major loci in *H. melpomene* (see Sheppard et al. [1985] and Mallet [1989] for genetic details). Because of gene dominance, only the Convento release site was visually polymorphic (though mainly "rayed" in phenotype), and even there only one wild "postman" *H. erato* (i.e., a triple homozygote) was ever seen.

the "postman"-patterned race (*H. e. favorinus*) and the "rayed" race (*H. e. emma*) (Lamas, 1976). These races, together with those of the mimetic *H. melpomene,* which has an identically placed hybrid zone, are shown in Figure 1. These concordant zones are each about 10 km wide and are centered on the eastern foothills of the easternmost range of the Andes (Fig. 2).

H. erato individuals set up small home ranges (Turner, 1971*b*; Mallet, 1986*b*) that are little affected by capture and marking (Mallet et al., 1987). Individuals transferred out of their normal home ranges to other sites set up new home ranges that include gregarious roots with native *H. erato* (Mallet, 1986*b*). We exploited these features of *Heliconius* behavior to perform our release experiment.

Four release sites, all within 40 km of each other, were selected in the Department of San Martín: Chazuta, Tarapoto, Convento, and Davidcillo (Fig. 2, Table 1). The sites were approximately matched for altitude. At Chazuta and Tarapoto, as well as in the Río Mayo and upper Río Huallaga drainages at 220–1,500 m above sea level, the postman pattern is fixed in *H. melpomene* and *H. erato,* though mimetic rayed heliconiines of other species (*Eueides tales, Laparus doris, Neruda aoede, Heliconius demeter, H. xanthocles, H. burneyi,* and *H. elevatus*) are sometimes present. Rayed heliconiines, including those species mentioned above are typical of the lower Huallaga and Ucayali drainages (150–250 m above sea level), as well as the Andean Pozuzo valley (up to 1,500 m above sea level); the rayed pattern in *H. erato* and *H. melpomene* is nearly fixed at Convento and Davidcillo, but occasional hybrids and even some pure postman patterns have been seen in these sites.

Fresh, undamaged *H. erato* were captured in a variety of locations including the release sites (old butterflies were not used so as to reduce variability in survival). They were marked individually on the undersides of their forewings with a felt pen, fed, and transported in hanging cages to different release sites (see Table 3 for distances moved). Butterflies were released at flowers frequented by native *H. erato.* Two kinds of transfers were made: "experimentals" with foreign color patterns were transported across the hybrid zone and released at sites on the other side; "controls" of the same race and pattern as those native to the release site were transferred from other sites on the same side of the hybrid zone as the release site in question. Controls were kept in the same way and for the same time as the experimental butterflies, and they were released at exactly the same flowers as the experimentals. In addition to transfers, mark-recapture studies of native *H. erato* were initiated at each release site. "Native controls" (fresh, undamaged *H. erato* that were captured, marked, and immediately released at the same site) were used to check for the effects of captivity and transport. Convento, at the edge of the hybrid zone (Fig. 2), had a fourth class of butterflies, known as "native experimentals." This class consisted of hybridized native individuals that clearly differed in at least one component from the locally predominant rayed pattern. One suitable native experimental was found at Davidcillo and one at Tarapoto (the latter had an F_1 hybrid phenotype and was probably an offspring of an experimental); these have been excluded from the analysis for lack of sufficient comparable data.

At each site, 1–4 batches of butterflies, both of experimental and of controls, were released. Sites were visited about every two weeks (when the roads were open) for 1–9 weeks before the first release and for 18–22 weeks after the last release. During visits, all possible rayed and postman heliconiines were captured. Their identities and any wing damage were recorded before release. Two types of wing damage were recorded: definite impressions of a bird's beak and major wing damage, the latter using Benson's (1972) definition of a straight tear passing through at least one major wing vein. All sightings of jacamars (*Galbula* spp.) were recorded as an indication of likely butterfly predation (jacamars are probably the most important predators influencing *Heliconius* mimicry [Chai, 1986]).

Statistical Analysis of Mark-Recapture Data

We used estimates of life expectancy to measure the fitness differences between experimentals and controls. *Heliconius* breed throughout the year and have a long reproductive life. After an initial refractory period immediately following eclosion, they reproduce at an approximately constant rate for up to six months (Dunlap-Pianka et al., 1977). The reduction in lifespan of experimentals is therefore approximately equal to the selection pressure against immigrants into the release sites.

Most methods of analyzing mark-recapture data such as ours assume that survivorship decays exponentially. However, our results suggest that disappearances are most likely to occur shortly after release. More than half of the released butterflies were never recaptured; however, butterflies seen once were often seen again. The simplest model that can describe such a pattern is that each butterfly has a probability P_E of "establishment" in the area and once established it has a probability $e^{-\lambda t}$ of survival for t days or longer, where λ is the exponential rate parameter. In the model, P_E represents establishment, but the estimated P_E will also include any components of early mortality after establishment with rate parameters much greater than λ. The overall life expectancy of a butterfly is then given by the probability (P_E) of establishment multiplied by the expected lifetime after establishment ($1/\lambda$).

The plausibility of different combinations of the parameters P_E and λ is judged for each treatment and site by their likelihood (the probability of a set of observed results given particular hypotheses of P_E and λ). Likelihood is extremely useful in this context because it is usually simple to calculate the probabilities involved in nonstandard models. Using likelihood, the hypotheses to be tested can be partitioned and combined in a variety of ways; one is not limited to a single null hypothesis.

The method used is explained in the Appendix. It depends on the usual biological assumption of mark-recapture analyses, that each available individual is equally likely to be recaptured. This is not strictly true for

TABLE 1. Ecological data for release sites and results. Treatments: C = controls, E = experimentals, NC = native controls, NE = native experimentals; see text for definitions.

				Numbers of butterflies				Mean known lifespan of recaptured butterflies (days)
Site	Altitude (m)	Jacamars seen	Treatments	Total released or captured	Total recaptured	With beak marks	With other damage	
Chazuta	280	*Galbula tombacea*	C (postman)	14	7	0	3	53.1
6°35'S			E (rayed)	16	2	1	0	29.5
76°09'W			NC (postman)	20	12	1	4	56.6
Tarapoto	350	none	C (postman)	26	16	1	5	36.2
6°29'S			E (rayed)	26	9	1	2	70.4
76°21'W			NC (postman)	41	18	0	4	63.3
Convento	270	*Galbula albirostris*	C (rayed)	18	6	0	4	54.2
6°16'S		(once only)	E (postman)	15	4	2	1	61.8
76°18'W			NC (rayed)	39	19	2	10	54.8
			NE (hybrids)	21	5	1	3	103.0
Davidcillo	250	*Galbula albirostris,*	C (rayed)	25	11	1	5	56.0
6°15'S		*Jacamerops aureus*	E (postman)	23	3	1	2	57.7
76°16'W			NC (rayed)	27	15	1	7	54.7
Overall:			C	83	40	2	17	47.3
			E	80	18	5	5	61.8
			NC	127	64	4	25	58.3

Heliconius over periods of a few days, since they learn to avoid areas or situations in which they have recently been captured; however, the experience of capture and release does not apparently cause much dispersal from the home range (Mallet et al., 1987). Since captures were made in all available subsites within each home range and since the experiments were performed over periods of months, it seems likely that all individuals in this study are equally likely to be captured. Provided that experimentals and controls do not differ in average likelihood of recapture, individual differences should have little effect on the relative measurement of survival.

The likelihood surfaces shown in Figure 3 were constructed separately for each site and each butterfly treatment by calculating likelihood values for a 40 × 40 grid of combinations of P_E and λ. The reliability of the estimates can be gauged from the "support" limits (contours where the $\log_e$-likelihood is two units below the maximum). Along any one axis, the support limits approximate the 95% confidence limits (Edwards, 1972). The plausibility of different hypotheses was compared using the difference in their $\log_e$-likelihoods (δL). In large samples, the G statistics ($G = 2\ \delta L$) is approximately distributed as chi-square.

RESULTS

Overall, experimental butterflies were recaptured less often than controls ($G_{[1]} = 11.97$, $P < 0.01$, Table 1). However, butterflies seen once were often seen again, and the known-residence time of recaptured experimentals (mean ± SE = 61.8 ± 11.3 days) did not differ significantly from that of controls (47.3 ± 5.4 days). At Convento, there was no significant difference between capture rates of native experimental and native control butterflies ($G_{[1]} = 3.67$, $P = 0.055$; Table 1). The percentage of butterflies with beak marks or presumed predator-caused wing damage was 47.5% overall, and was lowest at Tarapoto (30.2%), where no jacamars were sighted. No predatory attacks on butterflies were observed directly.

The estimated probabilities of establishment (P_E) and exponential death rates after establishment (λ) are shown in Table 2. The likelihood surfaces for each treatment and site are shown in Figure 3a–d. Using the method of likelihood, a large number of hypotheses can be tested. The likelihood surfaces for all sites can be summed to give overall estimates of P_E and λ in each experimental category (Fig. 3e, Table 2). These overall estimates of P_E and λ differ strongly between experimentals and controls (Fig. 3e;

TABLE 2. Maximum-likelihood estimates of life expectancy. (P_E to within 0.025; λ to within 0.0015). Two-dimensional support limits (~95% confidence limits) for these estimates are shown in Figure 3.

Site	Treatment	Estimates		
		P_E	λ	Life expectancy (days)
Chazuta	C (postman)	0.700	0.0090	78
	E (rayed)	0.325	0.0315	10
	NC (postman)	0.925	0.0075	123
Tarapoto	C (postman)	0.875	0.0210	42
	E (rayed)	0.450	0.0105	43
	NC (postman)	0.525	0.0060	88
Convento	C (rayed)	0.550	0.0120	46
	E (postman)	0.350	0.0075	47
	NC (rayed)	0.700	0.0075	93
	NE (hybrids)	0.300	0.0000	—[a]
Davidcillo	C (rayed)	0.975	0.0180	54
	E (postman)	0.300	0.0180	17
	NC (rayed)	1.000	0.0120	83
Overall (Summed *L*)	C	0.800	0.0165	48
	E	0.350	0.0120	29
	NC	0.700	0.0075	93

[a] Estimated life expectancy (P/λ) undefined.

$G_{[2]} = 12.43, P < 0.01$). Setting λ to its most likely joint value of 0.015 (assuming experimentals and controls have equal λ), estimates of P_E are significantly lower in experimentals than in controls ($G_{[1]} = 11.59$, $P < 0.01$). However, estimates of λ do not differ when P_E is held constant ($G_{[1]} = 1.00$). Performing the same analysis for each site, P_E is significantly higher in controls than experimentals at Chazuta ($G_{[1]} = 4.60, P < 0.05$) and Davidcillo ($G_{[1]} = 5.78, P < 0.05$), but not at Convento ($G_{[1]} = 0.31$) or Tarapoto ($G_{[1]} = 3.16$; Fig. 3a–d). On the other hand, estimates of P_E and λ differ little between controls and native controls (Fig. 3e; $G_{[2]} = 5.93$). The main effects of capitivity and transfer seem to be to reduce life expectancy after establishment ($1/\lambda$) by about 60% ($G_{[1]} = 5.25, P < 0.05$), rather than to affect P_E ($G_{[1]} = 0.29$).

Native experimentals were available only at Convento, where the difference between joint estimates of P_E and λ for native controls and native experimentals was marginally significant ($G_{[2]} = 6.04, P < 0.05$). Native experimentals had a low estimated P_E, but their life expectancy could not be determined because their estimated λ was 0, due to the high survival of the few native experimentals that were recaptured. However, holding λ constant, differences in P_E were not significant ($G_{[1]} = 2.53$), nor were differences in λ significant when P_E was held constant ($G_{[1]} = 1.30$). The native experimentals seem to have a low probability of establishment, (P_E) similar to that of the introduced experimentals, although not significantly lower than native controls. In view of the lack of significant differences between native experimentals and native controls, they will not be discussed further.

A simple way to express confidence limits in a two-parameter system is to reduce the dimensionality. Since the estimated λ for experimentals is not significantly different from the estimate of λ for controls, $\lambda_{\text{experimentals}} = \lambda_{\text{controls}} = \lambda$ can be taken as part of the statistical model. The sum of log-likelihoods over all sites (i.e., the log-probability of the data given a hypothesis of particular parameter values) is then maximized over all possible values of P_E for experimentals, P_E for controls), and λ. This gives $\lambda = 0.015$, P_E (for controls) = 0.775, P_E (for experimentals) = 0.375. This corresponds to a life expectancy for controls of 52 days with support limits (approximate 95% confidence limits) of 42–63 days. The corresponding life expectancy of experimentals is 25 (17–35) days. These maximum-likelihood values are slightly different from those in Table 2 because, in producing Table 2, λ

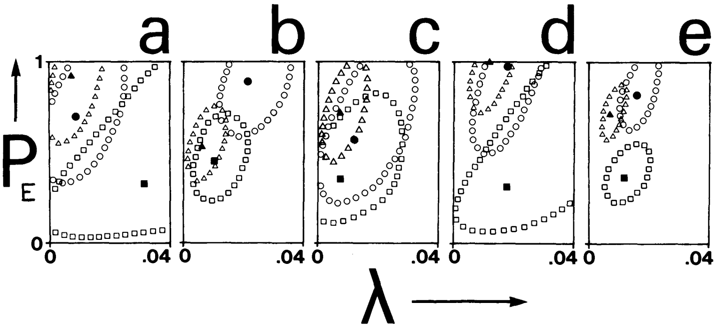

FIG. 3. Probabilities of establishment (P_E) and exponential death rates (λ) of released butterflies. These diagrams show the $\log_e$-likelihood (L) surface for the parameters P_E and λ for each site. In these graphs, estimates shown at the top left indicate high life expectancies, those at the bottom right represent low life expectancies. a) Chazuta; b) Tarapoto; c) Convento (native experimentals are not shown); d) Davidcillo; e) sums of L over all sites. Closed symbols mark the maximum-likelihood estimate for each treatment (● = controls, ■ = experimentals, ▲ = native controls), and the surrounding open symbols give the contour two L-units below this maximum. Along any one axis, this contour corresponds to the 95% confidence interval.

was allowed to differ between experimentals and controls. Compared with controls, the reduction in lifespan for experimental butterflies is 52% (25–71%). Since this is assumed to measure the selection in the experiment (see above), experimental butterflies have about half the fitness of controls.

DISCUSSION

Evidence for Selection

Different rates of loss of experimental and control butterflies probably represent selection by predators against foreign morphs of *H. erato.* However, differential dispersal could also produce the observed difference in residence times. The reciprocal-transfer method controlled for a greater intrinsic dispersal tendency of one of the races; but if the experimentals are less well adapted to the release sites, they might leave the release sites more rapidly than controls on both sides of the hybrid zone. This differential movement would presumably also indicate selection, since it would be due to a lack of suitable resources for the foreign morphs.

Strong gametic associations in the hybrid zones between loci determining color pattern give independent evidence for strong selection: in *H. erato* the average standardized pairwise disequilibrium ($D/D_{\max}$) is approximately 0.27, all genes unlinked; in *H. melpomene* the average $D/D_{\max}$ is approximately 1.00 for linked genes, and the average $D/D_{\max}$ is approximately 0.39 for unlinked genes (Mallet and Barton, unpubl.).

Local Adaptation or Selection for Warning Color?

Since there is a significant reduction of the establishment probability (P_E) of experimentals and no difference between control and experimental butterflies in subsequent survival (λ), selection seems to act soon after release. This might be expected if there are different adaptations to each race's habitat, since experimentals would be expected to die or leave soon after transplanting.

The problem with this local-adaptation hypothesis is that the most noticeable difference between races is in warning pattern. There are no fixed or strong frequency differences at soluble enzyme loci (Mallet and King, unpubl.; see also Turner et al. [1979]). Behaviorally, the two races are also similar. In insectaries, both races and their hybrids laid eggs preferentially on one species (*Passiflora trifasciata*) of a number of host plants made available (Mallet, 1989). At Tarapoto, introduced controls and experimentals were repeatedly seen roosting gregariously with native controls.

We controlled for local (within-race) adaptations to temperature as closely as possible by choosing release sites of similar altitude (Table 1). We controlled for local

TABLE 3. Recapture rates broken down by distance transferred. The null hypothesis tested is that recapture frequencies are not affected by distance transferred within each treatment. Homogeneity tests: $G_{[3]} = 1.75$, ns (controls); $G_{[3]} = 5.38$, ns (experimentals).

Statistic	Distance transferred (km) 5-10	11–20	21–30	31–40	≥41	Total
Controls:						
Total number released	33	14	33	3	0	83
Number recaptured	14	7	18	1	—	40
Percentage recaptured	42	50	55	33	—	48
Experimentals:						
Total number released	0	6	35	31	8	80
Number recaptured	—	2	11	3	2	18
Percentage recaptured	—	33	31	10	25	22

adaptation to biotic conditions by transferring both experimentals and controls from some distance away, but we could not exactly match transfer distances of experimental and control butterflies, because of limited access by roads. However, survival rates did not decline with distance moved for either experimental or control butterflies (Table 3).

Since the only known difference between the races of *H. erato* is the mimetic color pattern, it would be perverse to assume that selection due to predation is unimportant. The tight concordance of the color-pattern clines in the two species supports the predator explanation (Fig. 2). Although the lack of differences in λ between controls and experimentals might seem to argue against the importance of predation, accurate estimation of λ requires large sample sizes of survivors, and the lack of significant differences may be due to insufficient data. Also, relatively few individual predators may have been the selective agents (our observations with binoculars suggest that there are at most only 2–3 pairs of jacamars at each site). These predators may have quickly learned to avoid experimental butterflies, leading to significant differences in establishment (P_E) and no differences in later mortality (λ). Other experimenters have used similar explanations for a lack of survival differences between mimics and controls late in their experiments (Brower et al., 1967; Benson, 1972). Our results suggest that the most reliable estimates of life expectancy are those obtained from recapture rates within the first few days of release.

In general, it has been difficult to verify that natural selection acts on mimicry in the field. Most previous field studies used paint to alter butterfly and moth color patterns (Brower et al., 1964; Brower et al., 1967; Cook et al., 1969; Benson, 1972; Sternburg et al., 1977; Jeffords et al., 1979; Jeffords et al., 1980); thus the selection demonstrated was not "natural" (Endler, 1986 p. 128). However, see Edmunds (1966), Bowers et al. (1985), and Gordon (1987) for examples of natural selection possibly due to warning color and mimicry. Another difficulty with field studies of mimicry (including ours) is that the evidence for predation as the actual selective agent is weak. To demonstrate that predation is the cause of differential survival in a mark-recapture study, one should ideally know the fates of a random subsample of disappearing butterflies, both controls and experimentals. Of the above-mentioned experiments, only Cook et al. (1969) observed many instances of predation in the field, but in that experiment, the mimics and nonmimics did not differ in survival, perhaps because the nonmimics were themselves accidental mimics of a different model (Waldbauer and Sternburg, 1975).

Other experimental studies showing a survival advantage to painted mimics have used beak marks or wing damage on recaptured individuals as an index of predation. However, to use this index, one must assume that the probability of receiving a beak mark and surviving is correlated with the risk of being killed. Such correlations are not expected in general (Schoener, 1978). Indeed Carpenter (1941) found that beak marks occurred more often on unpalatable than on palatable species and used this as evidence that aposematic butterflies were better protected from predators. Within an unpalatable species, beak marks could be correlated with predation if the probabilities of rejection are the same for different morphs (Edmunds, 1974), but there is no experimental evidence for this view.

In our study, about 10% of all individuals were obviously beak-marked (we did not record whether beak marks were of the narrow form made by jacamars [see Benson, 1972], but these seemed to be the most common type), and 40% of individuals had major wing damage (Table 1). More experimental than control butterflies were beak-marked (Fisher's exact test, $P = 0.02$), but there was no difference between native experimentals and native controls ($P = 0.28$). Major wing damage overall did not differ significantly between treatments. While this evidence shows that birds attacked butterflies, it does not prove that the *differences* in disappearance rates are due to predation.

Jacamars are probably the most important visual predators of heliconiines (Chai, 1986). No jacamars were seen at the Tarapoto site, and only one pair of jacamars was sighted at Convento (and these were only seen more than a kilometer away from where most of the transferred butterflies were released). In these two sites, experimentals and controls did not differ significantly in their survival. In contrast, at Chazuta and Davidcillo, where experimentals survived less well than controls, jacamars were seen or heard calling on almost every visit. These observations suggest that there is an association between jacamar presence and poor establishment of experimentals. However, the percentages of recaptured butterflies with beak marks differed little between sites.

If the selection is primarily due to predators, then the total selection of 52% will be spread among the three major loci involved in color-pattern change (Sheppard et al., 1985; Mallet, 1989 [each locus may consist of linked blocks of genes]), so selection coefficients (s) are approximately 0.17 per locus (ignoring disequilibria). This high estimate for per-locus selection on mimicry seems to contradict the neo-Darwinian principle that selection tends to act weakly on many loci of small individual effect. However, while occasional foreign color patterns may occur naturally at the sites, selection will normally be lower within hybrid-zone populations. This study has measured the maximal selection, i.e., that against homozygous foreign individuals at the edge of the cline, where they are rarest. It is possible to estimate selection against nonmimics in other experiments on mimicry by assuming that the reduction in lifespan is equal to the selection coefficient. Under this assumption, published experiments using painted insects give similarly high selection pressures: for *H. erato*, $s = 0.22$ (Benson, 1972); for *Callosamia* moths, $s = 0.35$ (Sternburg et al., 1977); $s = 0.62$–0.69 (Jeffords et al., 1979), or $s = 0.70$ (Jeffords et al., 1980). The selective value of mimicry is often high.

The transfer experiments revealed strong selection against foreign morphs. Predator attacks seem to be the most likely explanation, for the following reasons: 1) there is no evidence for any local adaptation, whereas the races do obviously differ in mimetic color pattern; 2) beak marks show that predators (especially jacamars) do handle the butterflies; 3) selection was detected only where jacamars were common; and 4) the two mimetic species have highly concordant interracial hybrid zones, suggesting that selection acts directly on the color patterns. However, differing racial adaptations to local ecological conditions may also be involved; further experiments are planned to test between these hypotheses.

Strong selection could maintain a narrow hybrid zone. If warning-color differences cause selection with approximately linear frequency dependence, a balance between gene flow (σ) and selection (s, here approximately 0.17) will maintain a single-locus cline of width $3.55\sigma/\sqrt{s}$ (Mallet, 1986*a*). The 10-km-wide Peruvian hybrid zone of *H. erato* could therefore be maintained if gene flow, σ, is approximately 1.2 km, which seems reasonably similar to the direct estimate of dispersal (less than 0.3 km; Mallet, 1986*c*). Similarly, calculations based on selection caused purely by a stepped environmental difference (Endler, 1977) yield 1.7 km as an approximation of σ. Further studies should concentrate on determining the cause of selection, which seems to occur within the first few days of release.

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Appendix

Using Likelihood to Estimate Life Expectancy

In order to calculate the likelihood for each individual's capture record, we need some estimate of the probability that a butterfly will be captured on each day the site was visited, given that it is present. Since butterfly activity and sampling effort varied between days, the probability of recapture also varied. However, it was not feasible to estimate that recapture probability separately for each day. Instead, we have assumed that the recapture probability (θ_i) of a living butterfly on a day (i) when the site was visited is proportional to the sampling effort measured by the total number, E_i, of rayed and postman heliconiines captured on day i: $\theta_i = \alpha E_i$. The parameter α was estimated by Jolly's (1965) method, as follows. For each day, consider the number, N_i, of *H. erato* that are available for capture (the number released or captured before day i and last recaptured after day i). Then αE_i is estimated by the number, R_i, of these available butterflies that were actually recaptured on day i. Overall, the log-likelihood of α is

$$\sum_i \{R_i \log \alpha E_i + (N_i - R_i)\log(1 - \alpha E_i)\}.$$

This gives maximum-likelihood estimates for α of between 0.0095 and 0.0123, depending on the site. Clearly, the assumption that recapture probability is linearly related to total numbers caught is only an approximation. However, the method is accurate when average recapture rates on any day are less than about 40%. The average R_i/N_i was 0.22 for Chazuta, 0.40 for Tarapoto, and 0.23 for Convento and Davidcillo.

Now, consider the probability that some individual butterfly, having been released on day 0 is recaptured on, for example, sample days 2, 5, . . . , and L. Assuming that *H. erato* do not emigrate temporarily, the individual must have been present at least until day L. Therefore, the probability contributed by this period is

$$(\theta_2\theta_5 \ldots \theta_L)(\tilde{\theta}_1\tilde{\theta}_3\tilde{\theta}_4 \ldots \theta_{L-1})P_{\mathrm{E}}e^{-\lambda(t_L - t_0)}$$

where $\theta_i = 1 - \tilde{\theta}_i$ is the probability of capture on day number i given that the butterfly has survived, and $t_j - t_i$ is the actual number of days between days i and j. The butterfly may not have been seen after t_L, either because it was not present, or because it was not recaptured. The probability that it was present from t_L until some time between t_j and t_{j+1} ($j \geq L$) is

$$e^{-\lambda(t_j - t_L)}\{1 - e^{-\lambda(t_j + 1 - t_j)}\}$$

and the probability that it was missed on days ($L + 1$, $L + 2, \ldots, j$) is $\tilde{\theta}_{L+1}\theta_{L+2} \ldots \tilde{\theta}_j$. Therefore, the net probability over all days is given by the expression at the bottom of the page, where U is the last day on which the site was visited. For individuals that were never recaptured, this probability is instead

$(1 - P_{\mathrm{E}})$ +

P_{E}\{expression below in braces, setting $t_L = t_0$\}

The product of these terms over all butterflies gives the likelihood of (P_{E}, λ).

$$\begin{aligned}(\theta_2\theta_5 \ldots \theta_L)(\tilde{\theta}_1\tilde{\theta}_3\tilde{\theta}_4 \ldots \tilde{\theta}_{L-1})P_{\mathrm{E}}e^{-\lambda(t_L - t_0)}\, \{[1 - e^{-\lambda(t_{L+1} - t_L)}] + [e^{-\lambda(t_{L+1} - t_L)} - e^{-\lambda(t_{L+2} - t_L)}]\tilde{\theta}_{L+1} \\ \ldots + [e^{-\lambda(t_j - t_L)} - e^{-\lambda(t_{j+1} - t_L)}]\tilde{\theta}_{L+1}\tilde{\theta}_{L+2} \ldots \tilde{\theta}_j \\ \ldots + [e^{-\lambda(t_U - t_L)}]\tilde{\theta}_{L+1}\tilde{\theta}_{L+2} \ldots \tilde{\theta}_U\}\end{aligned}$$