

Puddling behavior by Bay checkerspot butterflies (*Euphydryas editha bayensis*)

A. E. Launer, D. D. Murphy, C. L. Boggs, J. F. Baughman, S. B. Weiss, and P. R. Ehrlich

Center for Conservation Biology, Department of Biological Sciences,
Stanford University, Stanford, CA 94305

Abstract. Large numbers of male and female Bay checkerspot butterflies, *Euphydryas editha bayensis*, were observed taking moisture from the banks of a seasonal creek in 1990. Previous observations of this long-studied subspecies imply that taking moisture from saturated substrates is not a common behavior for males, and is certainly not the norm for females. This study found that during the early and mid-portions of the adult flight season in 1990, a large number of older individuals visited the creek site, and that many butterflies were apparently flying more than one kilometer round-trip to visit the creek site. This behavior may have been induced by drought conditions.

This communication reports numerous observations of a butterfly, the Bay checkerspot (*Euphydryas editha bayensis*), not typically thought to be a "puddling species." Although adults, and males in particular, of many species of Lepidoptera have been observed extracting fluids from moist soils (Adler 1982, Berger and Lederhouse 1985, Boggs and Jackson 1991, Collenette 1934, Downes 1973, Norris 1936, Scott 1986), such behavior has not been reported previously for the long-studied Bay checkerspot butterfly. We also report on the comparatively large number of females and older individuals observed visiting puddles. This is in apparent contradiction to most previous field observations that suggest that puddling behavior is much more likely to be exhibited by males than by females, and particularly by young males (Adler 1982, Adler and Pearson 1982, Boggs and Jackson 1991, Collenette 1934). We discuss our observations in an effort to further an understanding of the reasons Lepidoptera visit moist soils, and to introduce the question of the importance of such areas for conservation planning.

STUDY SYSTEM

Bay checkerspot butterflies are restricted to serpentine soil-based grasslands of the San Francisco Bay area, and have been studied intensively since the early 1960s (e.g., Ehrlich and Murphy 1981, Harrison *et al.* 1988, Launer and Murphy 1994, Weiss *et al.* 1988). Previous research has addressed the effects of adult diet on fecundity and longevity for female Bay checkerspot butterflies (Boggs 1996, Murphy *et al.* 1983). Nectar, considered to be the primary source of adult-derived nutrients, contains water, sugars, amino acids, along with trace amounts of other compounds (reviewed by Boggs 1987). Under laboratory conditions, water increases adult longevity, but has

no influence on fecundity, while sugar increases fecundity and longevity (Murphy *et al.* 1983). Sugars acquired by adult butterflies are used in increasingly greater amounts in egg production as individuals age, and amino acids available from nectar are also used in egg production (Boggs 1996). On average, 18% of the oocytes carried by female Bay checkerspot butterflies are fully yolked at adult eclosion (Labine 1968). These eggs are by necessity composed of material derived from larval feeding and do not benefit from adult-derived nutrients. The remaining 82% of oocytes may receive at least some benefit from adult-derived nutrients.

Nutrients acquired directly from males may also be used by female checkerspot butterflies in egg production. Use of sugars and amino acids received by females from males at the time of mating follows a common pattern: an initial increase with age in incorporation of male-derived nutrients in eggs, followed by a decrease in incorporation (Boggs 1996).

METHODS

In late March 1990 numerous Bay checkerspot butterflies were observed apparently drawing moisture — that is, individual butterflies were seen with proboscides extended into water-saturated substrate (mud, sand, and gravel) — along a seasonal creek in the East Hills, near Morgan Hill (Santa Clara County, California). This site had been visited on four occasions during February and early March, during which no Bay checkerspot butterflies were observed to be associated with moist soils. Subsequent to the initial observations of butterflies exhibiting puddling behavior, the creek was visited on 12 occasions between 25 March and 20 April. During these visits, 209 Bay checkerspot butterflies (66 male and 143 female) were captured, scored for age (wing condition), identified by sex, and released. These butterflies were captured either as they took moisture along the creek bank or while they were flying along the creek, either after taking flight at our approach or as they were apparently settling down to the creek bank. In addition, four surveys on the hillsides and ridges within 200 meters of the creek recorded fewer than ten Bay checkerspot butterflies.

Concurrent with activities at the creek site, samples of adult butterflies were taken from a 500 meter by 100 meter study area straddling the main ridgeline. The ridge-straddling site was located 750 meters up-slope and somewhat north of the creek site. In 1990, the Bay checkerspot butterfly population was apparently centered along the northeast side of that ridge, and butterflies at the ridge-straddling site were considered phenologically representative of the butterflies in the vicinity. While it is well documented that male *Euphydryas editha* tend to congregate along hilltops or ridgelines (Baughman *et al.* 1988, Ehrlich *et al.* 1990, Ehrlich and Wheye 1988), samples from the broad ridge-straddling site should provide sex ratios that are representative of the population as a whole. Out of a large pool of butterflies present at the ridge-straddling site, 576 butterflies (402 male and 174 female) were captured at random, scored for condition, identified by sex, and released.

For statistical analyses the butterfly flight season was divided into five sampling periods, each with a duration of approximately five days. Mean wing conditions were compared across sites using Wilcoxon two-sample tests, and sex ratios (as % of male butterflies) were compared similarly using χ^2 tests.

Table 1. Summary statistics for butterflies captured at each study site during the five sampling periods.

sampling period	puddling site				% males
	males		females		
	number	mean condition (s)	number	mean condition (s)	
1	0	na	0	na	na
2	1	2.5 (na)	25	2.0 (0.53)	4%
3	20	2.8 (0.54)	83	2.1 (0.62)	19%
4	39	3.0 (0.48)	30	2.1 (0.60)	57%
5	6	3.2 (0.52)	5	2.4 (0.42)	55%

sampling period	ridge-straddling site				% males
	males		females		
	number	mean condition (s)	number	mean condition (s)	
1	55	1.3 (0.29)	5	1.2 (0.27)	92%
2	59	1.9 (0.58)	22	1.5 (0.36)	73%
3	100	2.2 (0.61)	60	1.6 (0.45)	63%
4	106	2.8 (0.45)	58	2.3 (0.51)	65%
5	82	3.2 (0.37)	29	2.6 (0.44)	74%

RESULTS

Substantial numbers of both male and female Bay checkerspot butterflies were observed exhibiting puddling behavior at the creek site during most of the adult butterfly flight season (Table 1). The sex ratio (as % male butterflies) varied through the season. At the creek site, sex ratio increased from a low of 4% males in the second sampling period (no Bay checkerspot butterflies were observed at the creek site during the first sampling period), to 57% and 55% during fourth and fifth sampling periods respectively. At the ridge-straddling site the percentage of male butterflies ranged from 92% (first sampling period) to 63% and 65% (third and fourth sampling periods, respectively). Sex ratios at the creek site and ridge-straddling site differed significantly during sampling periods 2 and 3, with relatively more female butterflies being found at the creek site (in both cases, $P < 0.001$). In sampling periods 4 and 5, sex ratios at the creek site and ridge-straddling site did not differ significantly.

At both the creek site and at the ridge-straddling site, mean condition of butterflies increased over the course of the season (Table 1). Mean condition of both male and female butterflies differed between the study sites in earlier samples, but not in later samples. For female butterflies, during sampling periods 2 and 3, mean condition was significantly higher for individuals at the creek site as compared to the ridge-straddling site (Table 1; sampling period 2, mean condition at the creek site was 2.0, while at the ridge-

straddling site it was 1.5, $P < 0.01$; sampling period 3, mean condition at the creek site was 2.1, while at the ridge-straddling site it was 1.6, $P < 0.001$). In sampling periods 4 and 5, the mean condition of female butterflies did not differ between the sites. These data indicate that during sampling periods 2 and 3 the condition of females at the creek site was not representative of the population of female butterflies as a whole — relatively older female butterflies were visiting the creek site. During the last two sampling periods, female butterflies visiting the creek site were also generally older individuals, but they did not differ from the aging population as a whole.

Mean condition of male butterflies showed similar differences between the creek site and the ridge-straddling site. In the middle of the flight season (sampling periods 3 and 4), the mean condition of male butterflies at the creek site was significantly greater than mean condition of male butterflies at the ridge-straddling site (mean condition of 2.5 vs. 2.2 in sample period 3, $P < 0.001$; and 3.0 vs. 2.8 in sample period 4, $P < 0.01$). At the end of the season the mean condition of males observed in the two study areas were similar. As was noted with female butterflies, these data indicate that comparatively older butterflies were exhibiting puddling behavior during early portions of the flight season, while older butterflies representative of the population as a whole visited the site late in the season.

DISCUSSION

These observations merit discussion for several reasons, all concerning a vexing question — why were large numbers of *Euphydryas editha bayensis*, and females in particular, observed exhibiting puddling behavior in 1990, but not in other years? Unfortunately this question has no clear-cut answer, although at least five possible explanations exist which center on 1990 as the third year of a severe drought and on the importance of adult-derived nutrients to butterfly reproduction.

First, Bay checkerspot butterflies may visit moist areas on a regular basis, but at smaller and more cryptic sources. If Bay checkerspot butterflies routinely benefit from adult-derived water or some essential mineral (e.g., sodium), then puddling behavior may be typical of their behavioral repertoire. Indeed, a number of the other twenty-odd subspecies of *Euphydryas editha* have been observed to exhibit puddling behavior. The 1990 observations could reflect loss of numerous small sources of water or minerals due to the drought — concentrating the behavior into the few remaining sites providing saturated substrates. The lack of observations of large numbers of Bay checkerspot butterflies visiting moist areas during previous droughts could reflect observer bias because we have only infrequently visited seeps, creeks, and puddles in and adjacent to serpentine soil-based grasslands.

In the four field seasons since 1990, however, including several moderately dry years, we have visited the creek site on many occasions and have observed few *Euphydryas editha bayensis* associated with the creek, and even fewer have been observed feeding on water-saturated soils there or at other sites in the serpentine soil-based grasslands. It is likely true that under non-drought

conditions Bay checkerspot butterflies occasionally visit moist soils, but the rarity of observations implies that this behavior is comparatively uncommon and probably does not provide butterflies with a significant source of water or nutrients during most years.

The second explanation concerns the possibility that resources typically available to adult butterflies were relatively scarce in 1990. While large numbers of flowers were present at the study site, the relative volume of nectar in these flowers may have been reduced compared to more typical, non-drought years. The butterflies may have altered their typical foraging behavior in response to a widespread shortage of either water or sugars, and were seeking water from the creek site (water, sugars, and amino acids are typical components of nectar, salt is not). Water from the creek site could serve to increase an individual's lifespan, but would not necessarily increase an individual's reproductive fitness (especially when viewed in light of the decreasing chance of survival of eggs laid later in the season — see Cushman *et al.* 1994, Murphy *et al.* 1983). This explanation fits the observations that individuals of both sexes were visiting the creek site. It also provides a partial explanation as to the comparatively large numbers of older butterflies observed at the site. If nectar availability was reduced due to the drought, individual butterflies that had depleted stores of water might be expected to search for alternate sources.

The remaining three explanations concern sodium, and center on likely increased butterfly demand for sodium, on the potential for reduced levels of sodium available from larval feeding, and on possible elevated levels of sodium present at the creek site. As pointed out by Arms *et al.* (1974), sodium is necessary for many physiological functions, including flight. Adult-derived sodium may also be beneficial for both sexes in terms of reproduction (Adler and Pearson 1982, Berger and Lederhouse 1985, Boggs and Jackson 1991, Collette 1934, Lederhouse *et al.* 1990, Pivnick and McNeil 1987, Scully and Boggs unpublished).

The first explanation connected with sodium availability concerns the possibility that the drought may have affected the rates at which butterflies were using sodium. It is possible that butterflies in 1990 were forced to fly greater distances to find mates, oviposition sites, or nectar, thereby depleting their stores of sodium at atypically high rates. Dispersal by *Euphydryas editha* has been demonstrated to vary considerably from one year to the next (Gilbert and Singer 1973, Murphy and White 1984, White and Levin 1981). In 1990, it was apparent that butterflies visiting the creek site were regularly traveling distances farther than are considered typical for *Euphydryas editha bayensis* (Baughman *et al.* 1988, Ehrlich 1965, Ehrlich *et al.* 1984, 1990, Harrison *et al.* 1988, Launer unpublished data, Sisk unpublished data). This could account for both sexes visiting the creek site and for the relative abundance of older individuals — individuals of both sexes need sodium, but only after they have depleted larval-derived stores.

The drought also may have affected the sodium content of larval hostplants, such that older butterflies depleted their larval-derived salt stores earlier

than is typical, thus forcing older butterflies to seek supplementary sources of sodium. Unfortunately, no data concerning the effect of drought on local serpentine-soil dwelling plants are available, and this explanation is untested.

Lastly, the creek could have been abnormally enriched for salts in 1990, thereby attracting butterflies not typically found at sites with lower salt concentrations. Again, we have no way to test this possibility; soil and water samples were not taken in 1990, and Bay checkerspot butterflies have been observed to visit the site on a very infrequent basis since. Aside from a likely reduction of stream flow in response to the drought, the creek site in 1990 did not appear different than it had in the previous two years or during the subsequent four years. Land use at the East Hills study site, winter-spring cattle grazing, has remained relatively constant for more than a decade.

Overall, our observations are generally consistent with hypotheses that butterflies are visiting moist areas in order to replenish essential nutrients or water expended during mating, gamete production, or general metabolism. As is predicted by such replacement hypotheses, the mean condition of butterflies at the creek site was relatively high, implying that older butterflies, those individuals likely to have used up nutrients acquired during larval feeding, were over-represented. For Bay checkerspot butterflies, it may be that nutrients that are acquired from puddles are not limiting during most years, but during periods of extreme drought nutrient reserves derived from larval feeding may be so low in older individuals as to trigger searches for alternate sources.

While these observations do not settle the issue as to what benefit, if any, the butterflies are receiving from areas that provide moisture, they do introduce the question of the importance of such areas to the persistence of populations of Bay checkerspot butterflies. The general lack of observations of puddling behavior suggests that during most years visits to puddles or other sources of water-saturated soils are infrequent, and of minimal importance to butterfly reproduction or survival. However, since our observations were taken during a period of extreme drought, it is possible that the resources provided at the creek site may aid individuals during harsh environmental conditions — possibly to the extent that the chance of local persistence is enhanced by moisture-providing areas. If true, this could be a problem as site specific conservation planning for this butterfly has focused on preservation of comparatively dry grasslands (Murphy and Weiss 1988, 1992, Weiss *et al.* 1988, 1994). Canyon bottoms and other moist areas have historically received little, if any, protection. If puddling behavior plays a role in sustaining populations during protracted periods of dry weather, such areas that are within or adjacent to butterfly population centers need to be incorporated into conservation planning for this subspecies.

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