Adaptive and incidental consequences of the alba polymorphism in an agricultural population of *Colias* butterflies: female size, fecundity, and differential dispersion

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**Summary.** A sex-limited color dimorphism occurs in many populations of *Colias eurytheme*. Alba females exhibit different patterns of resource allocation and are less attractive than orange females. This study examines some of the consequences of these differences in terms of reproductive success and population structure in a high density agricultural population.

Alba females produced more eggs than orange females on a daily basis, but the morphs did not differ in three measures of size. Spermatophore counts revealed that fresh and worn females did not differ in mating frequency between the two morphs, but very worn alba females gained more matings than very worn orange females.

In two mark-release-recapture experiments, alba females exhibited longer residence times than orange females. Changes in population structure over time suggest that this was due to dispersal of orange females from the mature field. Evidence is presented that orange females emigrate in response to male harassment at high density while alba females, exposed to less harassment, remain behind. We suggest that the persistence of the polymorphism in this agricultural population is at least partially facilitated by the cyclic cutting of the alfalfa.

In the genus *Colias*, the sulphur butterflies, at least 35 of the 57 known species exhibit a sex-limited color dimorphism (Remington 1954). In *Colias eurytheme* and *C. philodice*, females (the heterogametic sex in butterflies) that carry the dominant autosomal allele A are white, or alba, in wing coloration. Their homozygous recessive aa sisters are typically a shade of yellow or orange and resemble conspecific males (Gerould 1911, 1923; Hovanitz 1944c). Males may pass on the dominant alba allele but it is expressed only in females. The mode of inheritance appears similar in several *Colias* species (Remington 1954).

*Colias eurytheme* was formerly restricted to the grasslands of western North America, where vetches and clovers served as larval hosts. New habitats were opened to these butterflies in the 1850's with the planting and irrigation of alfalfa, *Medicago sativa*, in California. As alfalfa production spread eastward across the continent, *C. eurytheme* followed, invading the range of its congener, *C. philodice* (Hovanitz 1944a). Today both species and their hybrids are sympatric in alfalfa fields across the U.S. (Gerould 1943; Hovanitz 1944b; Taylor 1970).

The frequency of the alba genotype varies geographically in most *Colias* species. Alba phenotypes are most common in the northern part of the range and at high altitudes (Gerould 1923; Hovanitz 1944a; 1944b; 1950; Remington 1954). A basic assumption in explanations of the maintenance of persistent polymorphisms is that unique adaptive trade-offs have evolved among the different morphs. Much of the research on *Colias* has examined the nature of these trade-offs and the selective pressures that have shaped them. There is a redirection of nitrogenous precursors from pteridine pigment synthesis in the white phenotype to alternative metabolic pathways during pupal development (Watt 1973). White females might use these resources freed from wing scale coloration for more rapid development. Graham and his colleagues (1980) found that alba *C. eurytheme* females reach adulthood sooner and with significantly more mature eggs than their orange sisters. They also found that white females from natural populations of *C. alexandra* and *C. scudder* have significantly more fat bodies than yellow females.

The metabolic advantages of the alba phenotype may be offset by reduced mating attractiveness. White females are approached less frequently than are yellow *C. alexandra* and *C. scudder* (Graham et al. 1980) and *C. eurytheme* (Taylor 1970). Males also show less courtship persistence with white females in *C. philodice* and *C. eurytheme* (Marshall 1982).

The purpose of this study was to examine how these differences in resource allocation and mating attractiveness affect the reproductive success and population structure of *C. eurytheme*. The high density agricultural population examined in this study provides a contrast to the low density *Colias* populations studied by Graham et al. (1980). The adaptive trade-offs at low density need not have the same relative values at high density, especially in terms of female attractiveness. How do these trade-offs assist in maintaining polymorphism under these circumstances?

First, we measured females to see if albas might use their freed metabolites to increase size or fecundity or both. In many insect orders, large females produce more eggs (references in Thornhill and Alcock 1983). Second, we examined the potential costs incurred by alba females as a result of decreased attractiveness. Spermatophore counts...
in females of each phenotype were used to assess mating frequency and a mark-release-recapture experiment was executed to study behavioral and ecological effects of differential attractiveness. Based on results of the first mark-release-recapture, a second experiment was conducted to confirm and amplify those results. To test our interpretation of the release experiments, field samples were taken to document temporal changes in population structure and a focal animal survey was used to study key factors influencing the behavior of the two morphs.

Materials and methods

Field work for this study was conducted at the Arizona State University Experimental Farm in Tempe, Arizona from May through August 1984. Laboratory strains of *C. eurytheme* were derived from adults captured at this site. Larvae were reared on alfalfa cuttings at 27 ± 1°C; light and humidity were not controlled. In field samples taken for some of the following experiments, *C. eurytheme* and *C. philodice* were considered members of a single species complex. This was necessary as alba females of the two species are not readily distinguished. Less than 10% of the sampled individuals were identifiably *C. philodice*.

**Pupal and adult weights, forewing lengths**

Pupae and adult females of both orange and alba phenotypes of lab-reared *C. eurytheme* were weighed 24 h after molting. Adult forewing length was measured to the nearest mm on both lab-reared and field-collected females of each phenotype.

**Fecundity**

Virgin females of each phenotype were mated in the field, then returned to the lab to monitor oviposition. They were held in individual cages on a 12 h light/12 h dark regime. Each cage contained a sprig of alfalfa in water and a sponge soaked in 10% sucrose solution; both were renewed daily. Eggs were collected between 0800 and 0900 each day.

**Spermatophore counts**

In the Lepidoptera, the male deposits a cuticular spermatophore in the female's bursa copulatrix at each mating. Counts of spermatophores in individual females provide an index of mating frequency (Burns 1968; Pliske 1973). Females of both phenotypes were collected throughout the study period, killed by freezing, classed by wing wear, and dissected as in Rutowski et al. (1981).

**Mark-release-recapture experiments**

Two replicates of a mark-release-recapture experiment were performed using lab-reared stock. Pupae were held under refrigeration for up to 10 days, then placed in plastic shoe boxes for eclosion. All animals emerged the day prior to release. They were marked with numbers on the ventral hindwings using waterproof markers. Both releases were performed in the fourth week after cutting of the alfalfa. On 2 July at 1030, 113 orange and 107 white females were released in the center of the study site. In the second experiment, we released a ratio of alba and orange females that corresponded to the ratio observed in the field at the time as well as a large number of males. On 13 August at 1100, 319 orange females, 96 alba females, and 300 males were released at the same location. Recapture was by hand-netting; two workers spent 3–4 man-hours on each of the 12 days following a release searching for marked individuals. Recaptures were killed by freezing.

**Temporal changes in population structure**

On 11 days between 1 August and 20 August, samples of the field population were collected and analyzed to obtain relative measurements of density, sex ratio, and phenotypic composition. Two workers netted as many butterflies as possible in a 5 min period. Each was identified with respect to phenotype and sex and then released.

**Percentage of time in courtship**

A focal animal survey was conducted to determine the amount of time females of each morph spent in courtship-related activities. Free-flying females were followed at approximately 10 m and their activities were recorded using a hand-held tape recorder.

**Statistics**

Parametric statistics are reported as mean ± 1 standard deviation and are compared using Student's *t*-test (2-tailed) unless stated otherwise. Data were normalized by transformation as noted. Non-parametric tests were used as reported. All statistical comparisons were made at the 0.05 level of significance.

**Results**

**Pupal and adult weights: forewing length**

Measures of size for orange and alba females are summarized in Table 1. There were no significant differences between the two phenotypes in pupal weight (*t* = 0.87, 142 df, 0.4 > *P* > 0.2), adult weight (*t* = 0.64, 94 df, 0.9 > *P* > 0.5), or forewing length (lab-reared: *t* = 1.76, 142 df, 0.1 > *P* > 0.05; field-collected: *t* = 1.01, 197 df, 0.4 > *P* > 0.2).

**Fecundity**

The number of eggs laid is shown as a function of days after mating in Fig. 1 for each of the two morphs. Alba females laid significantly more eggs per day than orange females. Fecundity per performed using lab-reared stock. Pupae were held under refrigeration in the female's bursa copulatrix at each mating. Virgin females of each phenotype were mated in the field, phenotype.

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