



SYMPOSIUM

Complex Life Cycles and the Responses of Insects to Climate Change

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Synopsis Many organisms have complex life cycles with distinct life stages that experience different environmental conditions. How does the complexity of life cycles affect the ecological and evolutionary responses of organisms to climate change? We address this question by exploring several recent case studies and synthetic analyses of insects. First, different life stages may inhabit different microhabitats, and may differ in their thermal sensitivities and other traits that are important for responses to climate. For example, the life stages of *Manduca* experience different patterns of thermal and hydric variability, and differ in tolerance to high temperatures. Second, life stages may differ in their mechanisms for adaptation to local climatic conditions. For example, in *Colias*, larvae in different geographic populations and species adapt to local climate via differences in optimal and maximal temperatures for feeding and growth, whereas adults adapt via differences in melanin of the wings and in other morphological traits. Third, we extend a recent analysis of the temperature-dependence of insect population growth to demonstrate how changes in temperature can differently impact juvenile survival and adult reproduction. In both temperate and tropical regions, high rates of adult reproduction in a given environment may not be realized if occasional, high temperatures prevent survival to maturity. This suggests that considering the differing responses of multiple life stages is essential to understand the ecological and evolutionary consequences of climate change.

Introduction

Climate change is not simply global warming. General circulation models (GCMs) predict that the magnitude of recent and future warming will vary with latitude, continent, season, and the diel cycle (IPCC 2007). The increasing frequency of extreme temperature and precipitation events will vary seasonally and regionally (Battisti and Naylor 2009) and may pose a greater challenge to the survival and fecundity of organisms than do changes in mean conditions (Helmuth et al. 2002; Hoffmann et al. 2003; Stillman 2003). The spatial and temporal heterogeneity of climate change will generate novel climates in many geographic regions—seasonal patterns of temperature and precipitation that do not currently exist (Williams and Jackson 2007; Williams et al. 2007).

The appearance of novel climates poses an important challenge for scientists trying to predict the ecological and evolutionary responses of organisms to climate change. To predict the geographic ranges of species, environmental niche models (ENMs) depend critically on how different seasonal and diurnal components of temperature and precipitation are correlated (Meynard and Quinn 2007). Anticipated changes in this correlation structure during climate change, and the need to extrapolate to new environmental conditions, will limit the utility of ENMs for predicting changes in geographic range or abundance (Williams and Jackson 2007; Williams et al. 2007). More mechanistic models that incorporate phenotypic and ecological information are now being developed to predict ecological responses to climate

changes (Kearney and Porter 2009; Morin and Thuiller 2009; Buckley et al. 2010); the present symposium provides several excellent examples of this approach (A. L. Angert et al., submitted for publication). These models strive to translate environmental conditions into physiological impacts on organisms (Bakken et al. 1985) and to incorporate organismal responses to temporal and spatial climate variations that are often averaged in ENMs (Helmuth et al. 2005).

Our paper explores an issue rarely addressed by either ENMs or mechanistic models: life cycles often are complex. Most multicellular organisms have distinct life stages that vary in size, morphology, physiology, and other traits. Different life stages of an organism often experience different seasonal environments; inhabit different habitats and microclimates; and have different physiological sensitivities and responses (Coyne et al. 1983). As a result, different life stages contribute differentially to total lifetime fitness, and their relative contributions can vary markedly with weather and climate (Dempster 1983; Kingsolver 1989; Crozier 2003; Both and Visser 2005). How are different life stages specifically adapted to the microclimatic conditions they experience? How might this alter ecological and evolutionary responses to climate change?

Here, we address these questions in two ways. First, we explore the connections among life stage, microclimate and climatic adaptation in two well-known and well-studied insects: the sphingid moth, *Manduca sexta*, and *Colias* butterflies. Second, we consider recent models that predict the fitness consequences of climate change for tropical and temperate insects, based on the integrated measures of lifetime fitness (Deutsch et al. 2008). We explore how decomposing lifetime fitness into the thermal sensitivities of immature and adult stages may alter these predictions, and highlight the potential importance of survival at extreme high temperatures. Our analyses suggest that differences among life stages may be critical in understanding ecological and evolutionary responses of insects to variation and extremes in climate.

Life stage, microclimate, and adaptation in *M. sexta*

Manduca sexta inhabit parts of North America and South America (Rothschild and Jordan 1903) and is common across the southern United States. It occurs in smaller, more scattered populations further north in the United States, especially along the East Coast (Opler et al. 2010). Like all holometabolous insects, *M. sexta* has distinct egg, larval, pupal and

adult stages. The adults (hawkmoths) are nocturnal nectar-feeders and pollinators. Females lay eggs on leaves of host plants, which are primarily in the nightshade family (Solanaceae). Typical host plants in the southeastern United States include domesticated tobacco (*Nicotiana tabacum*) and tomato (*Lycopersicon esculentum*), and native *Datura* (*D. stramonium*); in the southwestern United States, typical hosts include native tobacco (*N. attenuata*), chilies (*Capsicum* sp.), and *Datura* (*D. wrightii* and *D. discolor*). Larvae (tobacco hornworms) feed and grow through five larval instars, sometimes more. Toward the end of the final instar, larvae wander off the host plant and burrow into nearby soil, creating a chamber in which pupation occurs (Madden and Chamberlin 1945). A facultative winter diapause, mediated by photoperiod during the larval stage, occurs during the pupal stage (Rabb 1966, 1969). At the end of the pupal stage, adults eclose and crawl to the surface before expanding and drying their wings. The number of generations (voltinism) varies with latitude; in the southern United States, there are typically two or more generations per year. As a result, each life stage of *M. sexta* experiences distinctly different climatic and microclimatic conditions, with important consequences for stage-specific thermal and hydric adaptations.

Adults

Hawkmoths are large, powerful flyers, mostly active at dusk and at night. They hover during nectar-feeding, and are capable of sustained, long-distance flights covering many kilometers in a single night. Generating enough power for flight is supported in part by high thoracic temperatures, the heat for which comes from muscle contractions (a form of periodic endothermy) (Heinrich 1970). Before taking off, moths intensively shiver their thoracic flight muscles, which generates heat and raises body temperatures well above ambient air temperatures. During flight, moths maintain thoracic temperatures of 38–42°C over air temperatures from 10°C to 35°C (Heinrich 1970).

As a result, hawkmoth flight probably is not strongly limited, in a proximate sense, by local temperatures, although ambient temperatures do affect the nectar (energy) resources required for flight. Indeed, adult *M. sexta* are frequently captured in the United States well north of areas that support year-round populations (Opler et al. 2010).

Eggs

Female moths lay eggs singly on the undersides of host-plant leaves. As is typical for most eggs of

insects, those of *M. sexta* are small (~1.5 mm diameter), sessile, and cannot thermoregulate. Unlike adults, which have the advantage of mobility and functional organs, embryos must rely on cellular mechanisms to survive thermal stress (Feder 1997). Development and survival of eggs therefore depend strongly on local temperature (Potter et al. 2009; Woods 2010). Interactions of temperature with other factors, including oxygen and water, may be important as well; e.g., low humidity may help eggs survive short heat shocks if they, or their host's leaves, can cool themselves significantly via evaporation.

Potter et al. (2009) have recently explored the performance of eggs, of *M. sexta*, in the extreme thermal and hydric conditions of southeastern Arizona. Maximum daily air temperatures in locations where *M. sexta* lives in Arizona, during the summer, routinely exceed 40°C, with extremes up to 45°C (Potter et al. 2009). Daytime humidities in summer are typically lower than 25%, increasing the potential for evaporative water loss from eggs. Since the eggs are small, they are nearly always isothermal with nearby microenvironments. However, eggs lie within the boundary layer of the leaf's surface (Woods 2010): within this layer, transpiration from the leaf can reduce temperatures and increase humidities relative to ambient conditions (Smith 1978). For *D. wrightii*, the major host plant in Arizona, maximum leaf temperatures during the day is 4–8°C below ambient air temperatures, so that eggs on this host plant rarely experience temperatures >40°C (Potter et al. 2009). Laboratory studies show that development times and mortality rates of eggs increase dramatically with exposure to cyclical temperatures that peak >40°C.

Furthermore, the conditions that eggs experience affect the larvae that they produce; hatchlings from warmer eggs are smaller and have slower initial growth rates (Potter et al. 2010). More generally, eggs of *M. sexta* do not develop successfully at mean (constant) temperatures >32°C (Woods and Bonnecaze 2006). Poor performance of eggs at high temperatures may be driven, in part, by interactions between temperature and oxygen status—eggs at higher temperatures appear to be increasingly limited by the availability of oxygen (Woods and Hill 2004). In the field, microclimates ameliorate extremes of ambient temperature, increasing egg survival, and developmental rates (Potter et al. 2009). Since evaporative cooling is effective only at low humidities, it is unlikely to be as important in the humid, southeastern United States. However, maximum daily air temperatures in the southeastern United States rarely exceed 38°C, so that fried eggs may be less likely anyway.

Although loss of water by eggs varies inversely with ambient humidity, eggs of *M. sexta* can develop to hatching in essentially any humidity (0–100%) (Woods et al. 2009) (K. A. Potter and H. A. Woods, unpublished data). Ecologically relevant rates of water loss do not affect rates of embryonic mortality or how well they survive subsequent heat shock (K. A. Potter and H. A. Woods, unpublished data), suggesting that in nature extreme heat is a more serious risk to eggs than is desiccation. Moreover, eggs appear to have well-developed physiological mechanisms for dealing with a wide range of potential water losses (Zrubek and Woods 2006).

Larvae

Manduca larvae weigh ~1 mg at hatching, when they begin to feed. Under favorable conditions, larvae can grow to 8–12 g in <3 weeks—a 10,000-fold increase in size. Larvae rapidly outgrow the leaf boundary layer (usually less than few millimeters thick) (Woods 2010), and forage actively among different leaves. As a result, the temperature of a larva changes during ontogeny (see Reavey 1993) from being governed by leaf temperature (early instars) to being governed by ambient air temperature and solar radiation (later instars).

Because of their larger size, *M. sexta* larvae, compared to eggs, experience a somewhat wider range of temperatures in the field. During the daytime in the southwestern United States (Mojave Desert, CA), larvae are ~1–4°C above ambient air temperatures (Casey 1976). Measurements with physical models of larvae in tobacco gardens in the southeastern United States (Piedmont, NC) suggest a similar pattern (Kingsolver 2000) (J. G. Kingsolver, unpublished data). When temperatures permit, larvae feed throughout the diel cycle (Bernays and Woods 2000). Thus, larvae of *M. sexta* do not actively regulate body temperature (except to avoid deleteriously high temperatures) and can feed over a wide range of body temperatures (10–37°C) (Casey 1976).

The thermal niche—the range of temperatures over which larvae survive to pupation—is somewhat narrower than the range of temperatures over which larvae feed (Reynolds and Nottingham 1985; Petersen et al. 2000; Kingsolver and Nagle 2007): survival to pupation exceeds 80% at mean rearing temperatures between 20°C and 30°C, but declines rapidly below 18°C and above 35°C. Both body size and time to pupation decline with increasing temperatures between 20°C and 35°C (Davidowitz and Nijhout 2004), although this pattern for body size can be reversed if larvae eat low-quality host plants

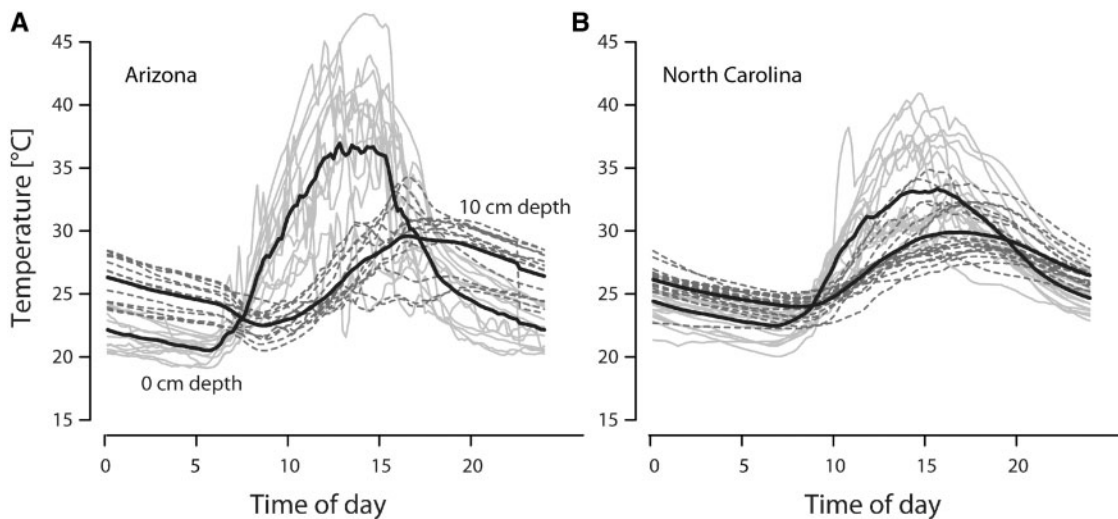


Fig. 1 Soil temperatures over 24 h at depths of 0 and 10 cm, measured using calibrated thermocouples, at *M. sexta* sites near Portal, Arizona (A) and Chapel Hill, North Carolina (B). Black lines are means over multiple days; background lines are individual traces (solid gray for 0 cm; dotted gray for 10 cm). Measurements were made over ~2 weeks in early August 2010 at both sites. Data from J. C. Sprague and H. A. Woods (unpublished data) and J. G. Kingsolver (unpublished data).

(Diamond and Kingsolver 2010). Pupal (and adult) mass is strongly correlated with production of eggs (Diamond and Kingsolver 2010).

Pupae

Toward the end of the final instar, *M. sexta* larvae stop feeding, wander from their host plant, and burrow into the soil to pupate. Burrowing depth varies from 5 to 30 cm, depending on density and composition of the soil (Madden and Chamberlin 1945) (J. C. Sprague, unpublished data). At depth, each larva spends several days constructing a pupal chamber, consisting of walls of compacted soil held together by oral secretions (Joesten et al. 1982). Chambers have total volumes ~5× larger than the pupae. The functions of chambers are currently unknown but probably include improvement of either the physical or biotic conditions around the pupae (J. C. Sprague, unpublished data).

Variability in temperature and moisture of the soil declines rapidly with depth into the soil (Campbell and Norman 1998). For example, in Arizona, temperatures at the soil surface often swing 25–30°C over 24 h, with maximum surface temperatures of 35–50°C (Fig. 1). In contrast, diurnal fluctuations at 10 cm depth are only 5–10°C (J. C. Sprague, unpublished data). Maximum surface temperatures in NC are somewhat lower (Fig. 1), but we see comparable reductions in diurnal temperature fluctuations at typical pupal depths of 10–20 cm (J. G. Kingsolver, unpublished data). As a result, nondiapausing pupae of *M. sexta* experience a much

narrower range of temperatures than do other life stages, and they rarely experience temperatures exceeding 30°C. Diurnal temperature fluctuations for pupae are quite small, and temperatures of diapausing pupae over the winter probably track mean monthly ambient air temperatures quite closely (Campbell and Norman 1998).

Data on thermal sensitivity of pupae are more limited than for other life stages (Fittinghoff and Riddiford 1990; Kingsolver et al. 2007). At constant temperatures, pupae survive well at temperatures between 20°C and 30°C, but very poorly at temperatures of 15°C or 35°C (J. G. Kingsolver and A. M. Nagle, unpublished data). Similarly, rate of development increases rapidly with temperature from 20°C to 30°C but declines at higher temperatures, as mortality increases (J. G. Kingsolver and J. K. Higgins, unpublished data).

In summary, egg, larval, pupal, and adult stages of *Manduca* inhabit different microhabitats, with distinct microclimates, none of which directly reflect macroclimatic conditions. For example, although larvae and eggs both live on plants, their body temperatures may differ by $\geq 10^\circ\text{C}$, even on the same leaf. Accordingly, individual insects exhibit different thermal responses, and perhaps climatic adaptations, in each stage. At higher temperatures, in particular, rates of development and survival depend strongly on life stage (Fig. 2): larvae develop successfully and rapidly at constant temperatures ($\geq 32^\circ\text{C}$) that cause complete mortality in eggs. Our preliminary data suggest that pupae and eggs respond similarly

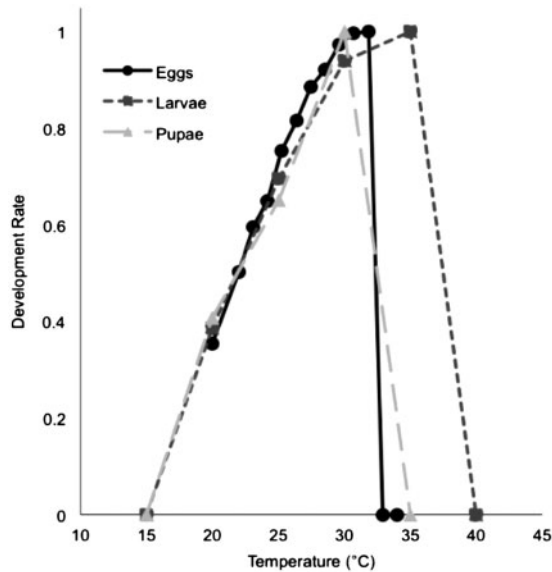


Fig. 2 Development rate (fraction of maximum) as a function of (constant) rearing temperature for different life stages of *M. sexta*. Eggs: Solid line and circles (data from Woods and Bonnacaze 2006). Larvae: Dark gray dashed line and squares (data from Kingsolver and Naylor 2007). Pupae: Light gray dashed line and triangles (data from J. G. Kingsolver and A. M. Nagle, unpublished data).

to high temperature (J. G. Kingsolver and J. K. Higgins, submitted for publication). Differences in sensitivity at lower temperatures are not apparent. These differences in physiological responses among eggs, pupae, and larvae reflect the different microclimates experienced by each life stage, particularly at higher temperatures. Flight in adult *Manduca* typically occurs at even higher body temperatures—temperatures that are deleterious or fatal to eggs.

Because of their sheltered microclimates, eggs and pupae are less exposed to extreme temperatures than larvae. However, larvae can use behavior to thermoregulate or to seek thermal refuges (Coyne et al. 1983; Feder 1997). In addition, because they have the longest duration of any stage, pupae must cope with increasing seasonality, including greater variation in both temperature and precipitation. The relative importance of microclimatic buffering (eggs and pupae) and behavioral movements (larvae) for reducing the negative fitness consequences of future climate warming remains to be explored.

Adaptation to local climate across life stages of *Colias*

Colias is a diverse, largely Holarctic genus of butterflies (family Pieridae). It has been an important insect model system for studying wing color, thermal biology, and interactions with host plants (Boggs et al. 2003; Watt 2004). In particular, different

Colias species and populations exhibit adaptation to local climatic and host-plant conditions across a range of elevations and latitudes in western North America (Watt 2003). Here, we summarize differences in local adaptation of thermal sensitivity and thermoregulatory traits between larval and adult stages, and the potential implications of these differences for responding to climate change. Larvae are cryptically colored (with no known relevance to thermal biology) and thermoregulate only in extreme temperatures. The thermal sensitivity of development and growth differs among populations (Sherman and Watt 1973). In contrast, thermal limits for adult flight are similar across populations, but populations differ in their thermoregulatory traits (wing melanism; thickness of thoracic setae (“fur”).

Shifts in climatic means and extremes are likely to be most significant for *Colias* in montane and alpine species (e.g., *Colias meadii* ~3200–3700 m) (Watt et al. 1977; Watt 2003). Models of climate project that typical summer temperatures in 2050 in Colorado, a focal location for research on *Colias*, will be as warm as, or warmer than, the hottest 10% of summers that occurred between 1950 and 1999 (based on projections averaged over 22 climate models and three emissions scenarios) (Ray et al. 2008).

Larvae

Colias larvae are cryptic green and have four larval instars, with a facultative diapause during the third instar. For most *Colias* species, host plants for larvae are in the legume (Fabaceae), willow (Salicaceae), or heath (Ericaceae) families. Field studies with several species of *Colias* in Colorado show that survival to pupation is often <4%, due to a combination of weather, host-plant quality, host-plant senescence, and natural enemies (Hayes 1981, 1984; Stanton 1984). Feeding and growth of larvae are strongly limited by temperature, especially at higher elevations and latitudes. Larvae do not actively regulate their body temperatures except to avoid temperatures >35°C (Sherman and Watt 1973), which are deleteriously high. However, some evidence suggests that *Colias* from different regions differ in their thermal sensitivities [thermal performance curves (TPCs)] for feeding. For example, larvae of *Colias eurytheme* from lowland California have maximal feeding rates at body temperatures of 26–29°C, and can feed effectively at temperatures up to 31°C (Fig. 3) (Sherman and Watt 1973). In contrast, *Colias eriphyle* sensu (Wheat and Watt 2008) from montane Colorado (elevation ~2800 m) experience

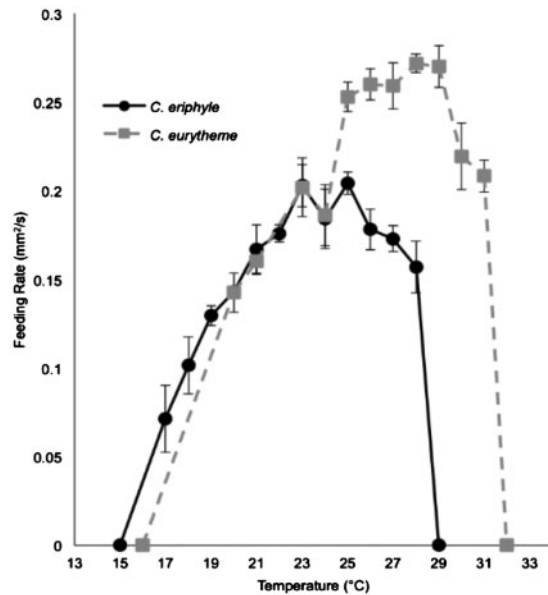


Fig. 3 Feeding rate (mean \pm 1 SE) as a function of body temperature in fifth-instar *Colias* larvae from low-elevation in California (*C. eurytheme*, gray dashed line and squares) and montane Colorado (*C. eriphyle*, black solid line and circles). Data from Sherman and Watt (1973).

cooler ambient conditions, have maximal feeding rates at 23–25°C, and do not feed at temperatures $>28^{\circ}\text{C}$ (Fig. 3). Differences in feeding rates at low temperatures ($<20^{\circ}\text{C}$) are not as great (Fig. 3). These data suggest that *Colias* larvae have adapted to local climatic conditions via shifts in their optimal and maximal temperatures for feeding and growth (Sherman and Watt 1973). The generality of this pattern for other *Colias* species or populations has not been established.

Adults

Colias butterflies are strong flyers, and all important adult activities—feeding on nectar, locating mates, courtship and mating, oviposition—require active flight. Field studies show that adults typically live for <7 days (Watt et al. 1977; Watt et al. 1979; Tabashnik 1980), so that time available for flight can strongly influence reproductive success (Kingsolver 1983a; Springer and Boggs 1986).

To take off and fly, *Colias* require thoracic temperatures of 30–40°C (Watt 1968). Unlike *Manduca*, *Colias* warm up by behavioral rather than by physiological mechanisms. Under cool, sunny conditions, *Colias* bask by closing their wings and orienting their ventral wing surfaces perpendicular to the sun, increasing absorption of solar radiation (Watt 1968). At deleteriously high body temperatures ($>40^{\circ}\text{C}$), they stop flying and orient parallel to the sun, thereby reducing solar heating. Amount of melanization

(percent of scales that are melanic) in the posterior region of the ventral hindwing (pVHW) determines how much solar radiation they absorb and, therefore, how far above ambient temperature their thoracic temperature goes (Watt 1968; Kingsolver 1983b).

Unlike larvae, adults from different populations and species do not have different TPCs: flight activity is consistently highest at body temperatures of 34–38°C (Watt 1968). Instead, adults have adapted to local climates via two morphological traits: wing melanin and the thickness of setae (“fur”) on the ventral thorax (Watt 1968; Watt 1969; Kingsolver 1983b; Kingsolver and Watt 1984; Roland 1999). In *Colias*, wing colors arise from yellow and orange pteridine pigments and black melanic pigments (Watt 1968). White “*alba*” wings are also common in females of some species (Watt 1974). Individual wing scales each have a single pigment type and color, so that the overall wing pattern is a pointillistic amalgam of differently colored scales (Nijhout 1986). For example, populations of *C. eriphyle* along an elevation gradient (1.8–2.9 km) in Colorado show significant increases in VHW melanin with increasing elevation (Ellers and Boggs 2004). In *C. eriphyle*, these population-specific differences affect the ability and propensity of adults to fly and, therefore, also affect egg production (Kingsolver 1983a; Springer and Boggs 1986).

Colias from high elevations and latitudes are particularly challenged by the thermal requirements for flight. For example, *C. meadii* is restricted to habitats >3000 m in Colorado, and adults have extensive VHW melanin and thoracic fur. Analyses of museum specimens of *C. meadii*, together with climatic data from the past 30 years (1980–2005), suggest that both maximum temperatures in July in Colorado have increased during the past several decades and that higher maximum temperatures are correlated with lower VHW melanin (Stamberger 2006). It has not been established whether these patterns reflect evolutionary changes in wing melanin in response to climate change. However, the well-known linkages between thermoregulatory traits and fitness suggest that selection may act to decrease melanism and fur thickness in response to climate change.

Understanding how an organism’s phenotype determines the fitness consequences of shifts—both in mean temperatures and in the incidence of extreme temperatures—is central to forecasting ecological and evolutionary responses to climate change. *Colias* provides an ideal system for linking phenotypes to fitness. Biophysical models enable us to predict flight times from thermoregulatory traits (melanism, fur thickness, and environmental data)

(Kingsolver 1983b). Since a *Colias* female typically oviposits a single egg on a host-plant leaf before flying to the next hostplant, flight time directly affects female fecundity (Kingsolver 1983a). Besides flight-related effects of temperature, *Colias* also experience mortality of eggs in response to acute thermal stress (Kingsolver and Watt 1983). Mechanistic models of responses to acute and chronic thermal stress can predict the ecological and evolutionary consequences of climate change.

In summary, larvae and adults of *Colias* have quite different responses and adaptations to climate. Adults use behavior to achieve and regulate the elevated body temperatures required for flight; larvae do not thermoregulate, and maximal rates of feeding and growth occur at much lower temperatures. Larvae and adults adapt to local conditions via quite different mechanisms: larvae via physiology (e.g., optimal and maximal temperatures for feeding and growth) and adults via morphology (e.g., wing melanin and thoracic fur).

These results raise an intriguing question: in response to climate change, are physiological or morphological traits more likely to evolve? (Ruibal 1961) There is abundant evidence for directional selection and rapid evolution of morphological traits in natural populations (Kingsolver et al. 2001a; Reznick and Ghalambor 2001), including selection on wing melanin in pierid butterflies (Kingsolver 1995a, 1995b). Unfortunately, few field studies have examined rapid evolution of physiological traits, except in response to pesticides or other environmental toxins (Dalziel et al. 2009; Kitano et al. 2010; Barrett et al. 2011). The answer to the above question has important implications for how rapidly different life stages will adapt to climate change.

Temperature, fitness components, and climate change

The impacts of climate change on populations will depend both on the magnitude and patterns of climate change and on the thermal sensitivity of the organisms in question (Helmuth et al. 2005; Tewksbury et al. 2008). Janzen (1967) predicted tropical ectotherms to be thermal specialists compared with higher-latitude relatives, because they live now, and have evolved in, relatively seasonal thermal environments. This prediction has been supported for ectotherms in several taxa (Ghalambor et al. 2006; Sunday et al. 2011), but exceptions have also been reported (Brattstrom 1968).

Several recent analyses have explored the consequences of this difference, by combining different

scenarios of climate change with data on thermal sensitivity of ectotherms, to predict consequences of climate warming (Deutsch et al. 2008; Huey et al. 2009). For example, Deutsch et al. (2008) used data and models for 38 insect species across a range of latitudes to predict the fitness consequences of climate warming, using the intrinsic rate of population increase (r) as a metric of fitness. Their analyses suggest that climate warming in the next century will increase mean fitness of species at temperate and higher latitudes but decrease it for tropical and low-latitude species. This prediction stems from two factors. First, as predicted by Janzen (1967), tropical species have narrower thermal tolerances, and thus are more sensitive to given changes in temperature. Second, many tropical species now live at environmental temperatures close to their optimal temperatures, such that even small increases in environmental temperature may strongly depress fitness. In contrast, high-latitude species are currently living at environmental temperatures cooler than optimal, such that climate warming may enhance fitness (Deutsch et al. 2008). These important analyses suggest that climate warming may have greater negative consequences for tropical than for temperate organisms, even if the magnitude of warming is smaller in the tropics than in temperate regions.

The intrinsic rate of increase (r) integrates age-specific patterns of survival and reproduction into a single metric. Its use as a measure of total fitness is most appropriate for populations with overlapping generations in constant environments once a stable age distribution has been achieved (Roff 2002). To apply this approach to variable thermal environments, Deutsch et al. (2008) used experimental data to estimate $r(T)$ (r at different constant or mean temperatures T) and then compute the (arithmetic) mean r for a population based on temporal variation in ambient temperature at a site. This method is reasonable for limited environmental variation in T , but may be misleading under conditions when $r(T)$ approaches zero. One key issue is how different components of fitness contribute to total fitness in variable environments (Levins 1962; Tuljapurkar 1990; Roff 2002). Here we illustrate this issue for organisms with distinct immature and adult life stages.

For simplicity, consider a semelparous organism with an immature (prereproductive) stage that then reproduces once as an adult at age G . In a constant environment, the overall fitness r is:

$$r = \ln[R_0]/G = \ln[SF]/G \quad (1)$$

where R_0 = net reproductive rate; S = probability of survival to reproduction; F = adult fecundity; and G = generation time. Note that in the case of semelparity, adult fecundity $F = R_0/S$. In general, each of these parameters varies with T —i.e., $S(T)$, $R_0(T)$, $F(T)$, and $G(T)$. Suppose environmental temperature varies temporally over a wide range, including extreme high (or low) temperatures at which $S(T)$ and/or $R_0(T)$ approach zero. Under extreme fluctuating temperatures, reproduction may stop temporarily, then resume when temperatures allow. In this case, the (arithmetic) mean reproduction over time and temperature will properly account for the contributions of reproduction to overall fitness. In contrast, if extreme temperatures force immature survival to zero, the population will go extinct, regardless of future thermal conditions and potential fecundity (Levene 1953; Gilchrist 1995, 2000). Thus, in fluctuating environments, survival of juveniles, and reproduction by adults may affect total fitness in quite distinct ways.

These considerations suggest that knowing both $S(T)$ and $R_0(T)$ is essential for predicting the fitness consequences of fluctuating environments and climate change. For example, suppose that for some high-temperature anomaly T_h , $S(T_h) > 0$ when $R_0(T_h) = 0$. In this case, the population may survive temporarily at temperatures that prevent reproduction—i.e., when $r(T_h) = 0$. How, then, for insects do the survival of immature and reproduction by adults vary with temperature? How does rates change with latitude, and with natural thermal variation, observed in the field?

To address these questions, we examined the set of insect species analyzed by Deutsch et al. (2008) and Frazier et al. (2006). From the original data (Frazier et al. 2006; Supplementary Material), we extracted data on survival of immatures (to adulthood or first reproduction), generation time (G), net reproduction rate (R_0) and r at different temperatures. We obtained useful data on survival, reproduction and r at four or more temperatures for 12 species. We also used the critical thermal limit (CT_{max}) estimated for each species (Frazier et al. 2006; Deutsch et al. 2008). Note that these fitness and CT_{max} values are based on studies using constant or mean temperatures experienced throughout the life cycle for each species: CT_{max} values based on acute, short-term exposure would undoubtedly be higher. Accordingly, we used weather data for the monthly mean of mean daily ambient temperatures at each site to compute the overall mean temperature (Te_{mean}) and the maximum monthly mean temperature (Te_{max} , mean temperature of the warmest month) for the past 50 years

(1960–2009) at each site. We used data for monthly rather than daily mean temperatures because the generation times of most of these species is on the order of several weeks to several months and, therefore, similar to the time scales at which fitness and CT_{max} values were measured.

Several interesting results emerge (Fig. 4). First, r (intrinsic rate of increase) generally has a higher optimal temperature (T_{opt}) than does R_0 (net reproductive rate) (Huey and Berrigan 2001). This occurs because increased temperature reduces the generation time (G), and G has a strong effect on r but not on R_0 [see Equation (1)].

Second, at high temperatures (above T_{opt}), the decline in survival and reproduction vary widely among species (Fig. 4 and Supplementary Figure). In some cases, S and R_0 decline toward zero at similar rates (e.g., Fig. 4, upper left and lower left); in other cases R_0 declines more rapidly than S at high temperatures (e.g., Fig. 4, lower right). In general, however, insects had high rates of mortality at the same high mean temperatures that also preclude reproduction; as a result, (arithmetic) mean r does not fully capture the fitness consequences of high temperatures in fluctuating climates—because extreme temperatures cause mortality.

Third, data for fitness and its components at higher temperatures are quite limited for some species; in many cases the value of r at the highest measured temperature was $>80\%$ of the maximum r (at the optimal temperature) (Fig. 4 and Supplementary Figure). These limits temper our confidence in predicting fitness at extreme temperatures. For example, for *Acyrtosiphon pisum* (Supplementary Figure), the estimated value of CT_{max} (based on the measured values of r) is $>6^\circ\text{C}$, higher than the highest measured temperature.

The fourth result concerns the relationship of the upper thermal limits of a species (CT_{max} : Fig. 4, plus) and maximum monthly temperatures experienced in the field (Te_{max} : Fig. 4, asterisks). For some species, $Te_{max} > CT_{max}$, and this occurs for both high-latitude (e.g., Fig. 4, upper left) and tropical (Fig. 4, lower right) species. Differences between Te_{max} and CT_{max} can vary substantially at similar latitudes, and there is no simple relationship between latitude and $(Te_{max} - CT_{max})$ (Fig. 4 and Supplementary Figure). We have full data for only a single species (Fig. 4D) from latitudes $<20^\circ$, and $Te_{max} > CT_{max}$ by $>5^\circ\text{C}$ in this case. This is consistent with previous analyses showing that many tropical ectotherms already experience environmental temperatures near their upper thermal limits for

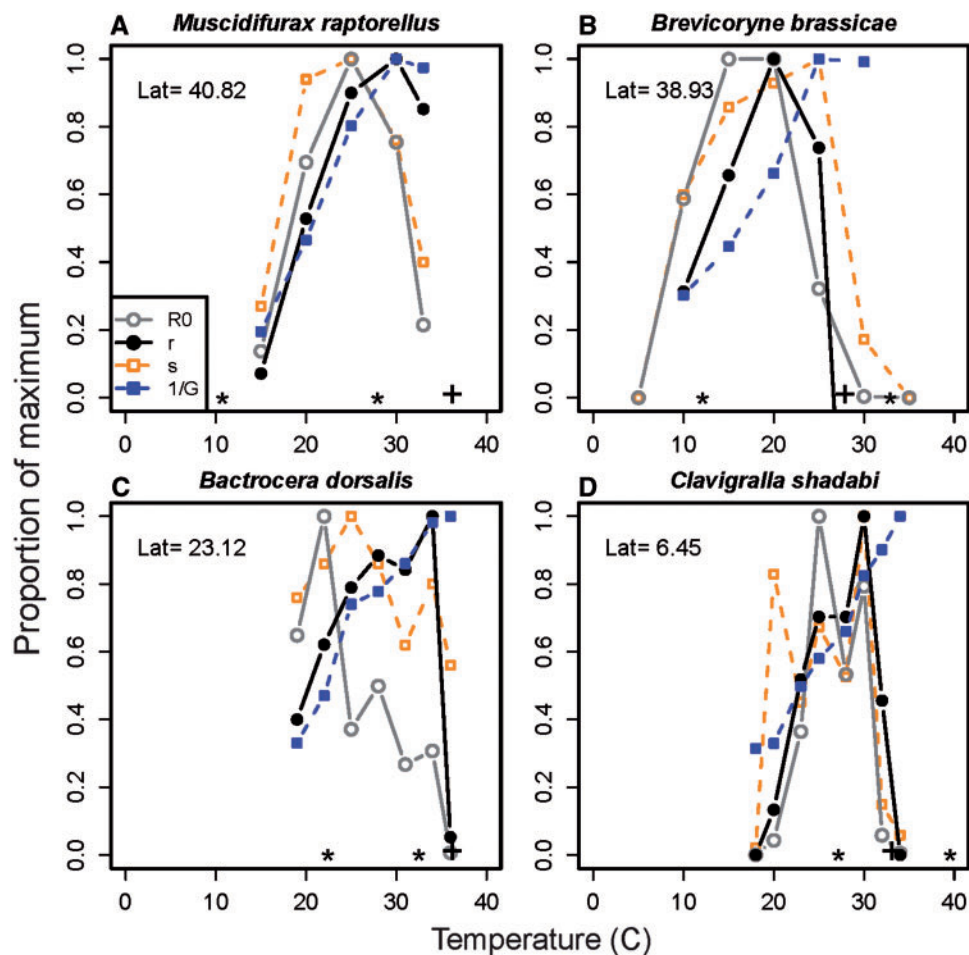


Fig. 4 Components of fitness (proportion of maximum) as functions of mean rearing temperature for four insect species from different latitudes, in relation to climate conditions during 1960–2009. Net reproductive rate, R_0 : Solid gray line and open circles. Intrinsic rate of population increase, r : Solid black line and filled circles. Survival of juveniles (to adulthood or first reproduction), s : Dashed orange line and open squares. Generation time (reciprocal), $1/G$: Dashed blue line and filled squares. Critical thermal maximum (CT_{max}) for each species (from Frazier et al. 2006) is indicated as a plus sign. Mean annual temperature and the mean temperature for the hottest month at each site over the period 1960–2009 are also indicated as asterisks. Data for fitness components are derived from source papers by Frazier et al. (2006) (Supplementary Material).

survival and reproduction (Deutsch et al. 2008; Tewksbury et al. 2008).

More data on responses to high temperatures by tropical species are urgently needed (Brattstrom 1968; Deutsch et al. 2008). However, our limited data support the hypothesis that TPCs and thermal limits of ectotherms reflect extremely high environmental temperatures rather than mean or “typical” conditions (Heinrich 1977; Huey and Kingsolver 1993). Our results also imply that the thermal “safety margins” of ectotherms at higher latitudes may be smaller than suggested by previous analyses (Deutsch et al. 2008; Tewksbury et al. 2008). As a consequence, future climate warming—particularly increase in the frequency of extreme high temperatures—may have negative

consequences for fitness of both temperate and tropical species.

Summary and prospects: stage-specific environmental responses and climate change

A major theme of this article is that different life stages can experience very different climatic conditions. Life stages often occur in different seasons and live in different habitats and microhabitats. In addition, changes in size and shape across life stages alter the exchanges of mass and energy that determine patterns of body temperature and water balance. These factors may be particularly important in understanding responses of different life stages to variation and extremes in environmental temperature

and water. For example, in *Manduca*, larvae are much more likely to experience extremely high temperatures and low humidities (see above) than are either eggs or pupae, due to a combination of microhabitat and large size (Fig. 2). These issues are likely to be even more important for organisms whose life cycles include both terrestrial and aquatic stages. For example, many semi-aquatic insects spend egg and juvenile stages in water but adulthood on land. Because water has much higher heat capacity than does air, the water-dwelling stages are likely, in general, to experience much lower mean temperatures and substantially reduced variability. Vannote and Sweeney (1980) compiled data on diurnal and seasonal temperature variation at White Clay Creek (39°53' N 75°47' W; southeastern Pennsylvania). Although the seasonal variation was substantial (range 16°C), diurnal variation in water temperature was only 3–5°C (Vannote and Sweeney 1980). Clearly, aquatic eggs and larvae will experience much moister, colder, more thermally stable conditions than would emergent, terrestrial adults.

Environmental differences among life stages have several important consequences for responses to climate change. First, changes in seasonal climate, and in climate variability, may strongly affect some life stages but not others; demographic analyses will be critical for predicting the consequences of these effects for abundances and geographic distributions. For example, although studies of insect demography frequently identify specific life stages and fitness components that most strongly influence population fluctuations (Dempster 1983; Kingsolver 1989; Crozier 2003; Doak et al. 2006), changes in seasonal patterns of climate may alter which life stages are most important in determining overall fitness of a population.

Life stages can evolve different physiological responses to temperature, water and other environmental factors. For large, holometabolous insects, differences in thermal sensitivity between larvae and adults are well established, and stem primarily from the substantial thermal and energetic requirements for flight (Heinrich 1993). Our results for *Manduca* indicate stage-specific differences in developmental rates and thermal tolerance among eggs, larvae, and pupae that reflect microclimatic differences in exposure to temperature fluctuations and extremely high temperatures (Fig. 2). We emphasize that these results are from animals reared under constant conditions; we expect even greater differences among stages under more realistic, fluctuating conditions (Kingsolver et al. 2009; Potter et al. 2009). To our knowledge, there are remarkably few data available

on physiological responses to temperature and other climatic factors across life stages (Tucic 1979).

The existence of differences among life stages raises an important issue: are physiological responses phenotypically or genetically correlated across life stages? For example, if changes in climate generate selection and evolution for increased heat tolerance in larvae, will larval evolution lead to correlated evolutionary responses in heat tolerance (or other traits) in eggs or pupae? (Lande and Arnold 1983) We are not aware of data that addresses this issue; studies in quantitative genetics or experimental evolution would be particularly valuable.

Numerous studies document that populations and species adapt to local climatic conditions by a variety of mechanisms. Our discussion emphasizes that life stages of an individual can adapt to local conditions via different mechanisms. For example, *Colias* larvae under different climatic conditions differ in their TPCs for feeding and development rates but not in morphological traits; conversely, *Colias* adults differ in morphological traits (wing melanin and thoracic fur) but not in TPCs for flight. As climate changes, are morphological or physiological traits more likely to show evolutionary responses? There is abundant evidence for genetic variation, directional phenotypic selection and microevolution for morphological traits, including size and color pattern (Roff 1997; Kingsolver et al. 2001b; Reznick and Ghalambor 2001; Siepielski et al. 2009), but far less information on rapid evolution of physiological traits in nature (Dalziel et al. 2009; Kitano et al. 2010; Barrett et al. 2011). Adaptive plasticity in response to climate changes may also affect selection and evolutionary responses in morphological and physiological traits. In *Colias* (and other butterflies), adaptive plasticity of wing melanin in response to photoperiod (and in some cases, pupal temperature) has been documented (Watt 1969; Hoffman 1973, 1978). Comparable plasticity (beneficial acclimation) of TPCs for larval growth or development has not been reported, although thermal acclimation has sometimes been reported for other insects (Huey et al. 1999).

Finally, recent analyses predict that the increasing frequency of high temperatures during future climate warming is likely to decrease mean fitness of insects in tropical regions but increase it in temperate regions (Deutsch et al. 2008). These studies properly emphasize that responses to climate change involve both the magnitude and the patterns of climate warming and the thermal sensitivities (TPCs) of different species (Deutsch et al. 2008; Tewksbury et al. 2008). A key challenge is to define and estimate

“mean” fitness in fluctuating environments (Tuljapurkar 1990; Roff 2002) and to parameterize such models using laboratory measurements in simplified (often constant) environmental conditions. We suggest differences in sensitivity and tolerance to high temperatures for different life stages and components of fitness could alter our predictions about the fitness consequences of climate change. Our initial analyses of how changing temperature affects mortality of juveniles, and reproduction of adults, suggest that insect species in both temperate and tropical regions have probably experienced elevated mortality of juveniles in response to extremely high temperatures during past decades—conditions close to their upper thermal limits (CT_{max}) for survival. Such conditions, which could greatly increase the likelihood of population extinction, will probably be more frequent in the near future (Battisti and Naylor 2009). These considerations, while preliminary, suggest that thermal safety margins for insects may be smaller than indicated by previous analyses; as a result, future climate warming may decrease fitness and the persistence of populations in both tropical and temperate insects. Exploring how seasonal patterns of temperature variability under climate changes will affect both survival of juveniles and reproduction by adults would be particularly valuable for improving our predictions of the consequences of global warming for fitness.

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Supplementary Data

Supplementary data are available at *ICB* online.

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