Should I stay or should I go? Complex environments influence the developmental plasticity of flight capacity and flight-related trade-offs

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Complex environments, characterized by co-varying factors (e.g. temperature and food availability) may cause animals to invest resources differentially into fitness-related traits. Thus, experiments manipulating multiple environmental factors concurrently provide valuable insight into the role of the environment in shaping not only important traits (e.g. dispersal capacity or reproduction), but also trait–trait interactions (e.g. trade-offs between traits). We used a multi-factorial design to manipulate variation in temperature (constant 28 °C vs. 28 ± 5 °C daily cycle) and food availability (unlimited vs. intermittent access) throughout development in the sand field cricket (*Gryllus firmus*). Using a univariate approach, we found that temperature variability and unlimited food availability promoted survival, development, growth, body size and/or reproductive investment. Using principal components as indices of resource allocation strategy, we found that temperature variability and unlimited food reduced investment into flight capacity in females. Thus, we detected a sex-specific trade-off between flight and other life-history traits that was developmentally plastic in response to variation in temperature and food availability. We develop an experimental and statistical framework to reveal shifts in correlative patterns of investment into different life-history traits. This approach can be applied to a range of biological systems to investigate how environmental complexity influences traits and trait trade-offs.

ADDITIONAL KEYWORDS: cricket – dispersal – flight capability – food availability – *Gryllus* – life-history – temperature variation.

INTRODUCTION

Animals can employ a range of resource allocation strategies for important life-history traits, such as investment into somatic and reproductive tissues (Stearns, 1992; Zera & Harshman, 2001; Roff, 2002). However, trait–trait trade-offs may occur wherein the allocation of limited resources toward one life-history trait obligates reduced allocation toward another trait (van Noordwijk & de Jong, 1986; Kaitala, 1987; Chippindale *et al.*, 1993; Nijhout & Emlen, 1998; Zera *et al.*, 1998; Zera & Brink, 2000). Although trait–trait trade-offs can occur regardless of environmental conditions (e.g. King *et al.*, 2011), developmental environments are characterized by variation in both biotic and abiotic factors, which may profoundly affect

an animal's resource allocation during ontogeny (Kaitala, 1987; Sibly & Atkinson, 1994). Thus, when faced with challenging conditions, the allocation of resources among these traits may change – that is, environmental variation may influence both individual traits and trait–trait trade-offs.

Environmental factors, such as variation in temperature and food availability, can affect investment into somatic and reproductive tissues via variation in resource acquisition (e.g. increased food intake promotes reproductive investment: King *et al.*, 2011) or resource allocation (e.g. higher temperatures increase growth rates at the expense of adult body size: reviewed by Angilletta, 2009). Environmental temperature is important because it influences a range of biological processes, including behaviour, energy use, locomotion and reproduction (Angilletta, 2009). This sensitivity to environmental temperature leaves ectothermic animals particularly susceptible to the ongoing effects

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of climate change (Parmesan *et al.*, 1999; Root *et al.*, 2003; Sinervo *et al.*, 2010). Environments are expected to continue to exhibit greater mean temperatures, as well as greater thermal variation (IPCC, 2014). This variation in temperature may pose a greater risk to species and biodiversity than the gradual warming characterized by shifts in mean temperature (Vasseur *et al.*, 2014). However, variation in temperature is not the only challenge many animals encounter. Food availability, like temperature, is variable across space and time, and it can have large effects on animal growth and development (Dunham, 1978; Jones, 1986; Shafiei *et al.*, 2001). Additionally, food availability and temperature can vary simultaneously (Mattson, 1980; Stamp, 1993) and together indicate the quality of a given environment (e.g. a high-quality environment may be characterized by high food availability and a stable temperature). Thus, co-variation of food availability and temperature can affect the perceived quality and distribution of habitats (Roff, 1990, 1994).

Animals have adapted to environmental heterogeneity (e.g. variation in temperature and food availability) by employing a variety of locomotor strategies (reviewed by Aidley, 1981). Flight, in particular, gives some animals the ability to travel greater distances than walking, and it allows flying animals to quickly leave low-quality environments in search of better habitats (Roff, 1994). However, the act of flight is energetically costly (Schmidt-Nielsen, 1972; Thomas & Suthers, 1972; Bartholomew & Casey, 1978; Norberg, 2012). Furthermore, building and maintaining flight musculature, and synthesizing flight fuels (e.g. triglycerides) can greatly increase metabolic demands (Zera & Mole, 1994; Zera *et al.*, 1994; Zera & Denno, 1997). This energetic investment into flight can obligate trade-offs with other life-history traits, such as reduced investment into reproduction (reviewed by Zera & Harshman, 2001; Guerra, 2011; Kalberer & Kölliker, 2017). Researchers have gained a better understanding of how animals navigate flight-related trade-offs by typically manipulating a single environmental variable (e.g. effects of food availability on a trade-off between flight and fecundity: Mole & Zera, 1993, 1994; Zera & Mole, 1994; King *et al.*, 2011), but it is still unclear how animals navigate these trade-offs when experiencing shifts in complex environments characterized by multiple co-varying environmental factors (e.g. variation in temperature and food availability).

The quality of complex environments may mediate the prioritization of flight capability relative to other important life-history traits (Mole & Zera, 1993, 1994; Zera & Mole, 1994). In the wing-dimorphic sand field cricket (*Gryllus firmus* Scudder), females of each discrete wing morph adopt a different strategy to a flight–fecundity trade-off. During early adulthood,

long-winged (LW) females specialize in dispersal by investing into functional flight musculature, which comes at a cost to reproduction. Short-winged (SW) females sacrifice their ability to fly in return for greater reproductive abilities (i.e. greater investment into ovary mass) during early adulthood (reviewed by Zera, 2005). Males also exhibit trade-offs between investment into flight capability and reproduction (mating-call duration: Crnokrak & Roff, 1998; testes size: Saglam *et al.*, 2008). In sum, the established flight-related trade-offs in *G. firmus* provide a unique opportunity to examine the role of complex environments in the plasticity of traits and trait–trait interactions.

Thus, we examined several dynamics of developmental plasticity in both sexes and morphs of *G. firmus* due to multiple, co-varying environmental factors. Specifically, we used a multi-factorial design to manipulate variation in temperature (constant 28 °C vs. 28 ± 5 °C daily cycle) and food availability (unlimited vs. intermittent access) throughout development. The factorial design allowed us to test two hypotheses related to the role of complex environments in the developmental plasticity of not only individual traits, but also trait–trait interactions. First, we hypothesized that variability in temperature and food availability influences the developmental plasticity of individual life-history traits, such as growth and reproduction. We predicted an additive effect of food and temperature where high food availability and stable temperatures would enhance these fitness-related traits (e.g. abundant food and constant temperature would lead to faster development and larger gonads). Second, we hypothesized that variability in temperature and food influences the developmental plasticity of trait–trait interactions, including trade-offs associated with investment into flight capability. Here, we predicted that fluctuating temperatures and intermittent food availability would be non-ideal developmental conditions that would promote the prioritization of flight capacity over other life-history traits, such as reproductive investment. By factorially manipulating two ubiquitous environmental factors, this study will inform our understanding of how dynamic environments influence important traits (i.e. survival, developmental rate, and investment into reproductive and flight tissues) and trait trade-offs in animals.

MATERIAL AND METHODS

STUDY ANIMALS

The wing dimorphic sand field cricket (*Gryllus firmus*) is endemic to the south-eastern USA and ranges from Connecticut to Texas (Capinera *et al.*, 2004). Crickets used in the study were acquired from two

artificially selected LW and two artificially selected SW populations that have been previously described (Zera, 2005). Newly emerged hatchlings (1st instar; <5 days post-hatching; $N = 540$) from eggs laid by ≥ 80 female *G. firmus* (i.e. ≥ 20 females from each of four selected blocks) were randomly distributed among the treatment groups described below.

EXPERIMENTAL DESIGN

To investigate the developmental plasticity of traits and trait–trait interactions, a 2 × 2 factorial design was employed on crickets ($N = 540$) reared under a 16-h photoperiod, which were housed individually in translucent plastic containers (473 mL) with *ad libitum* access to water. Half of the crickets were reared in an incubator (model I-36, Percival Scientific, Inc., Perry, IA, USA) at a constant 28 °C ('constant' temperature treatment), the temperature regime at which this stock is typically reared (reviewed by Zera, 2005). The remaining crickets were reared in an incubator (model I-36, Percival Scientific), which created a sinusoidal diel temperature cycle (28 ± 5 °C; 'fluctuating' temperature treatment) that approximates the average diel temperature variation in Gainesville, Florida (where the founders of the stock were collected), during the crickets' active season (May–September: National Weather Service). Crickets experienced one of two food treatment levels: *ad libitum* access to food (commercial dry cat food) ('high' food treatment) or access to food for two 24-h periods each week ('low' food treatment, which is ecologically relevant due to crickets' intermittent feeding habits: Gangwere, 1961).

At 5 days of adulthood (i.e. when the flight–fecundity trade-off peaks: Zera & Larsen, 2001), wing morphology (i.e. SW or LW) was verified, and crickets were weighed (a measure of food assimilation: Zera & Mole, 1994), killed and stored at −20 °C. Crickets were later dissected, and their flight musculature [dorsolongitudinal muscles (DLM)] was scored from 0 (DLM absent) to 1 (white, histolysed and non-functional DLM) to 2 (pink and functional DLM) (King *et al.*, 2011). Each cricket's femur length (a proxy for body size: Simmons, 1986) was measured. Although an animal's body size and mass are typically correlated, they can become uncoupled in animals with relatively few energy (fat and protein) reserves (*sensu* low body condition: reviewed by Peig & Green 2009, 2010), which is of interest for our study given our manipulation of the animals' food availability. Thus, body mass and size can provide fundamentally different metrics of body quality. Last, each cricket's gonads were removed and dried to a constant mass to determine gonad mass. Scoring wing musculature of *G. firmus* allowed for an estimate of investment into flight, while determining gonad dry mass provided an estimate of investment into reproduction (Roff & Fairbairn, 1991; Crnokrak & Roff, 1998). Growth rate (mm/day) was determined by dividing femur length by the time required for newly hatched crickets to reach the adult phase (i.e. development time).

PRINCIPAL COMPONENT ANALYSES

A multivariate statistical method was used to reveal correlative patterns of investment into different life-history traits, and to generate an index of resource allocation strategy (*sensu* Stahlschmidt & Adamo, 2015; Bertram *et al.*, 2017a, b; Nguyen & Stahlschmidt, 2019). These indices were subsequently analysed (see 'Statistical analyses' below) similar to other variables (e.g. body size and mass: see above). For example, some females in our study allocated more toward flight capability relative to reproduction and other life-history traits (see below). Specifically, two principal component analyses (PCAs) were performed on initial dependent variables (developmental time, body mass, femur length, gonad mass and DLM status). Male and female data were analysed independently because examinations of individual life-history traits indicated female-biased sexual dimorphisms for some traits (body mass, body size and gonad mass) but not for others (growth rate and DLM: described below). Because this a priori information suggested males and females used different allocation strategies, performing a PCA on the entire data set (i.e. both sexes) would obscure sex-specific correlative patterns of investment into life-history traits. Two principal components (PCs) accounted for significant percentages of the variation found in the data (see below), and they were each used as a dependent variable in subsequent analyses (i.e. linear mixed models, see 'Statistical analyses' below).

In the PCAs, the initial dependent variables were generally significantly correlated with one another, with the exceptions of weaker correlations between DLM and other traits – particularly, in males (Table S1). Bartlett's measure, which determines whether there is a significant pattern of correlations in a given data set, for our data sets was highly significant (< 0.001 for both data sets). Yet, our data sets did not exhibit extreme multicollinearity (overly correlated variables) because they each had an adequately large determinant of the correlation matrix value (≥ 0.03 for both data sets). The Kaiser–Meyer–Olkin (KMO) measure of sampling adequacy ranges from 0 (diffuse pattern of correlations) to 1 (compact pattern of correlations), and the KMO values for our data sets were 0.62 and 0.73 for females and males, respectively, which are acceptable (Kaiser, 1974). Therefore, our male and female data sets satisfied the assumptions of having significant and compact patterns of correlations.

The first principal component extracted from our female data (PC1f) accounted for 57% of the variation in the female data. PC1f loaded positively onto growth rate, body mass, body size and reproduction, and negatively onto development time and flight muscle (Fig. 1). Thus, an individual with a high PC1f value had bias against dispersal, which means it had a lower investment into maintenance of flight musculature and capability, and a higher investment into development, body size and reproduction (Fig. 1). The first principal component extracted from our male data (PC1m) accounted for 58% of the variation in the male data. Similar to PC1f, PC1m loaded onto body mass, body size and gonad mass in the same direction, and onto development time in the opposite direction (Fig. 1). However, unlike PC1f, PC1m loaded only weakly (0.1) onto flight muscle (Fig. 1). Thus, PC1m scores from our data signify a different metric than PC1f, as we found no evidence of a negative correlation between flight capability and gonadal investment in males (Roff & Fairbairn, 1993; but see Saglam *et al.*, 2008) (Fig. 1). Rather, PC1m represented a quality index where a higher value reflected greater investment into nearly all fitness-related traits (Fig. 1).

STATISTICAL ANALYSES

Data were tested for normality, transformed (e.g. natural log, log base ten or square root) when
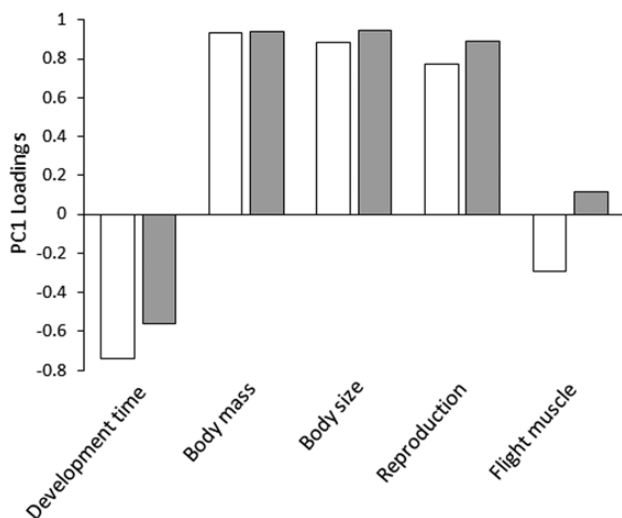

Figure 1. PC1 loadings representing investment of female (white) and male (grey) *G. firmus* into development, body mass, body size, reproduction and flight muscle. Females appeared to exhibit a trade-off between investment into flight capability and all other traits [i.e. PC1 for females (PC1f) represented an index of dispersal bias). Males did not appear to exhibit this trade-off; rather, PC1 for males (PC1m) indicated an index of overall quality.

necessary, and analysed using R (v.3.3.2; R Foundation for Statistical Computing, Vienna, Austria). Two-tailed significance was determined at α = 0.05. To examine the independent effects of temperature and food treatments, a linear mixed model analysis was performed on each of the following traits across all individuals (i.e. both sexes were analysed together): development time, growth rate and body mass. Both body size (femur length) and gonad mass data were analysed independently for each sex because female-biased dimorphisms that exist in *G. firmus* [e.g. in our study, sexes varied in femur length (*t* test: $t_{246} = 3.9$, $P < 0.001$) and gonad mass (*t* test: $t_{246} = 7.1, P < 0.001$)] made residuals of our data irrevocably non-normally distributed. For each mixed model, temperature and food treatments, wing morphology (herein, 'morph': SW or LW), and sex were included as main effects.

An ordinal logistic generalized linear mixed model was performed on the categorical DLM scores (scored from 0 to 2, see above), and temperature and food treatments, morph, and sex were included as main effects. Similarly, a binary logistic generalized linear model was used on data from each cricket to determine the main and interactive effects of temperature and food treatments on survivorship (0: did not survive to adulthood; 1: survived to adulthood). A linear mixed model was performed on the PC1 scores for each sex independently to determine if variation in temperature and food availability had a significant effect on the trade-off between flight and fecundity in females (i.e. PC1f, an index of dispersal bias: Fig. 1) or on the overall quality of males (i.e. PC1m, an index of quality: Fig. 1). All models included selected block as a random effect. They also tested for interactions between and among temperature treatment, food treatment, sex and morph. There was then a stepwise removal of insignificant variables until no additional independent variable could be eliminated without leading to an increase in Akaike's information criterion (*sensu* Pagano & Arnold, 2009; Arnold, 2010). Only significant results from these final, parsimonious models are reported below.

RESULTS

Food availability was the only factor influencing survival, which increased with the availability of food ($Z = 22$, $P < 0.001$), and all other factors were removed from the final model as described above. For this analysis, morph and sex were not considered as sex and wing morphology could not be determined in immature crickets. Crickets reared in a thermally fluctuating environment developed faster (mean: 72 vs. 82 days spent in development; $F_{1,246} = 13, P < 0.001$), and so did those given high food (mean: 74 vs. 82 days;

$F_{1,246} = 13$, $P < 0.001$) and those that were LW (mean: 74 days vs. 82 days; $F_{1,246} = 4.6$, $P = 0.033$) (Supporting Information, Figs S1A, S2A). Similarly, crickets reared in a thermally fluctuating environment or high food availability grew faster (temperature: mean: 0.19 vs. 0.16 mm/day; $F_{1,244} = 13$, $P < 0.001$, food: mean: 0.19 vs. 0.16 mm/day; $F_{1,244} = 14$, $P < 0.001$) (Figs S1B, S2B). LW crickets also grew faster than SW crickets (morph: mean: 0.19 vs. 0.17 mm/day; $F_{1,244} = 6.1$, $P = 0.014$) (Figs S1B, S2B).

Individuals reared in a thermally fluctuating environment and those with high food had greater body mass at adulthood (temperature: mean: 768.8 vs. 682 mg; $F_{1,231} = 4.3$, $P = 0.039$, food: 762.7 vs. 663.5 mg; $F_{1,231} = 16$, $P < 0.001$) (Supporting Information, Figs S1C, S2C). Female crickets were significantly heavier than male crickets (sex: mean: 781.7 vs. 664.4 mg; $F_{1,231} = 9.0$, $P = 0.003$), and LW individuals were heavier than SW individuals (mean: 765.6 vs. 662.1 mg; $F_{1,231} = 7.1$, $P = 0.008$) (Figs S1C, S2C). Female body mass was more sensitive to food treatment than male body mass (food × sex: $F_{1,231} = 10$, $P = 0.001$, Figs S1C, S2C). Crickets given unlimited food access during development were larger (i.e. had longer femurs) than those with limited access (mean: 12.8 vs. 12.5 mm; $F_{1,232} = 5.4$, $P = 0.021$; Figs S1D, S2D). Females and LW crickets were significantly larger than males and SW crickets (sex: 13.0 vs. 12.4 mm; $F_{1,232} = 6.4$, $P = 0.012$; morph: 13.0 vs. 12.2 mm; $F_{1,232} = 14$, $P < 0.001$) (Figs S1D, S2D).

Female crickets, SW crickets and crickets in the high food treatment had heavier gonads (sex: mean: 22.1 vs. 8.7 mg; $F_{1,230} = 57$, $P < 0.001$; morph: mean: 16.4 vs. 14.9 mg; $F_{1,230} = 5.2$, $P = 0.023$; food: 19.5 vs. 9.3 mg; $F_{1,230} = 36$, $P < 0.001$) (Fig. 2A, B). The effect of sex on gonad mass was greater in the high food treatment (food × sex: $F_{1,230} = 33$, $P < 0.001$) and in SW individuals (morph × sex: $F_{1,230} = 6.8$, $P = 0.010$) (Fig. 2A, B). Crickets given high food and those exposed to fluctuating temperatures exhibited reduced investment into flight muscle (food: $\chi^2 = 33$, $P < 0.001$; temperature: $\chi^2 = 4.1$, $P = 0.043$), and the effect of temperature on DLM was greater in the high food treatment (food × temperature: $\chi^2 = 5.7$, $P = 0.017$) (Fig. 2C, D). Male and SW crickets also had reduced DLM status (sex: $\chi^2 = 4.4$, $P = 0.035$; morph: $\chi^2 = 69$, $P < 0.001$) (Fig. 2C, D).

Females reared in high food environments exhibited greater investment in development, growth, body size, and reproduction, but reduced investment into flight capability ($F_{1,122} = 17.3$, $P < 0.001$) with effects approaching significance on females reared in the fluctuating thermal treatments ($F_{1,122} = 3.6$, $P = 0.060$) (Fig. 3A). Males reared in a fluctuating thermal environment had slightly higher quality indices than those reared in a constant temperature, although this

effect was not signficant ($F_{1,122} = 3.2$, $P = 0.064$) (Fig. 3B). LW males were also of higher quality than SW males ($F_{1,122} = 5.6$, $P = 0.020$) (Fig. 3B).

DISCUSSION

Typically, studies examining the plasticity of traits and trait trade-offs do so by manipulating (at most) a single environmental variable, such as food availability or temperature (Kaitala, 1991; Nunney & Cheung, 1997; Clark *et al.*, 2015). Conducting single-variable experiments has improved our understanding of how animals invest resources into traits and navigate trade-offs, but our study further clarified how animals deal with *multiple* environmental pressures in concert. Understanding the effects of complex environmental variability is important when considering how animals respond to environmental shifts associated with climate change (e.g. increasing frequency of concurrent drought and heat wave conditions: Mazdiyasni & AghaKouchak, 2015).

In support of our hypotheses, we demonstrate that fitness-related, life-history traits (e.g. flight capacity, growth and reproduction) and trait trade-offs can be developmentally plastic in response to variation in temperature and food availability (Figs 2, 3; Supporting Information, Figs S1, S2). For example, high food availability and fluctuating temperature promoted investment into key components of somatic and reproductive tissues (Figs S1, S2, Figs 2A, B) while reducing investment into flight capacity in females (Figs 2C, D, 3). However, contrary to our first prediction, thermally fluctuating (not stable) environments promoted somatic growth (Figs 3, S1, S2). Although we found limited interactive effects of food and temperature treatments on life-history traits (Fig. 2C, D), these environmental factors did have additive effects on several traits (Figs S1A–C, S2A–C). Because temperature and food availability can naturally co-vary (Mattson, 1980; Stamp, 1993), experiments manipulating multiple environmental factors improve our understanding of how the environment shapes important traits (Fig. 2; Todgham & Stillman, 2013; Kaunisto *et al.*, 2016) and trait–trait interactions (Fig. 3).

Fitness is strongly linked to survival, and food availability during development influences survival across taxa (e.g. insects: Boggs & Freeman, 2005; amphibians: Scott *et al.*, 2007; reptiles: Garnett, 1986; fish: Wilkins, 1967, birds: Davis *et al.*, 2005). In support of this, individuals reared in the high food treatment in our study exhibited a 60% increase in survival to adulthood relative to those in the low food treatment. Food availability also promotes shorter development time, faster growth rate, and/or larger
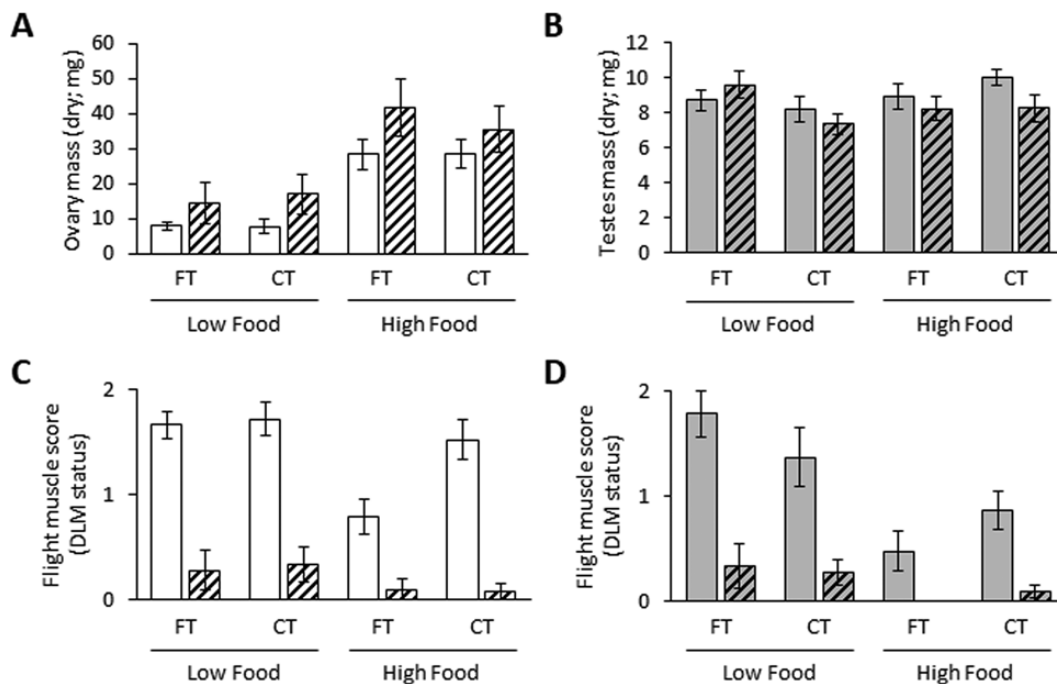
Figure 2. Effects of temperature and food treatments during development on gonad mass of (A) females and (B) males, and flight muscle of (C) female and (D) male *G. firmus*. Data for both sexes were analysed together but are displayed separately to improve visualization and comparison. Note: none of the short-winged males that experienced high food and fluctuating temperature exhibited any flight muscle. Long-winged morphs are depicted by open columns, and short-winged morphs are depicted by diagonal cross-hatched columns. Fluctuating and constant temperature regimes are denoted by FT and CT, respectively. Values are displayed as mean ± SEM.

adult size in both invertebrates and vertebrates (e.g. insects: Richardson, 1991; amphibians: Scott & Fore, 1995; reptiles: Dunham, 1978; Ballinger & Congdon, 1980). Similarly, we observed that crickets reared on high-food diets were larger and heavier than those given limited access to food (Supporting Information, Fig. S1C, D). However, high food availability did not increase investment into all somatic traits – the construction and maintenance of flight musculature was significantly reduced (not enhanced) in high food conditions (Fig. 2C, D). Presumably, individuals were more likely to invest in dispersal when developing in low food (low quality) environments, which would allow them to leave to find a higher quality environment. Thus, understanding the adaptive function of a given trait is important when determining its response to environmental variation.

Temperature, like food, can affect many biological processes, such as development, growth and reproduction (reviewed by Angilletta, 2009). In particular, mean temperature strongly influences development time, growth rate and body mass in ectotherms (Ratte, 1985; Atkinson, 1994; Kingsolver *et al.*, 2009; Williams *et al.*, 2012). However, independent of differences in mean temperature, we found that variation in temperature experienced during development also leads to

decreased developmental time and increased growth rate and body mass (Supporting Information, Figs S1A–C, S2A–C), consistent with other invertebrates (Ratte, 1985; Atkinson, 1994; Kingsolver *et al.*, 2009; but see Kjærsgaard *et al.*, 2013), as well as vertebrates (Pepin, 1991; Shine & Harlow, 1996; Booth, 1998; Wijethunga *et al.*, 2016). Many studies have shown that ectotherms typically benefit from experiencing variable temperatures during their life-histories, as long as the rearing temperatures are within physiological limits (Worner, 1992; Shine & Harlow, 1996; Elphick & Shine, 1998; Angilletta, 2009; Fischer *et al.*, 2011; Colinet *et al.*, 2015). The benefits of temperature variability may be explained by the asymmetric effects of high and low temperatures on physiological processes (i.e. benefits of experiencing higher temperatures outweigh the costs of exposure to lower temperatures: reviewed by Colinet *et al.*, 2015). For example, digestion may occur predominately during the photophase and could benefit from higher temperatures; by contrast, energy savings may be possible during the cooler scotophase when digestive processes are minimal. However, we do caution the interpretation of our results on the effects of temperature fluctuations, as we were unable to separate the effects of temperature fluctuation from the effects of degree-days (Allen,
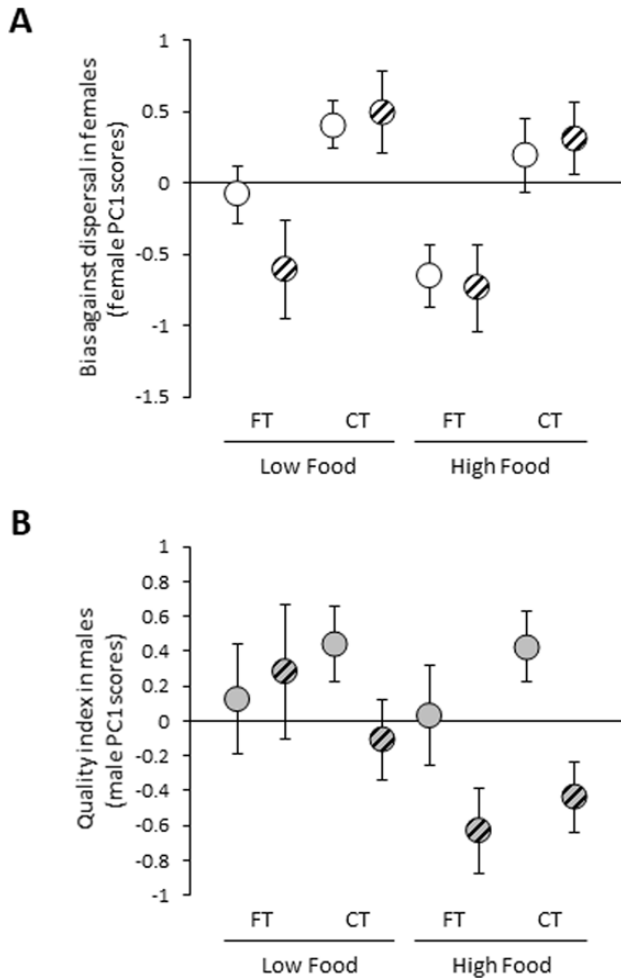
Figure 3. Effects of temperature and food treatments during development on (A) the bias against investing in flight musculature in favour of investing in other fitness-related traits in female *G. firmus*, and (B) the quality of male *G. firmus* (see text and Fig. 1 for details). Long-winged morphs are depicted by open circles, and short-winged morphs are depicted by diagonal cross-hatched circles. Fluctuating and constant temperature regimes are denoted by FT and CT, respectively. Values are displayed as mean ± SEM.

1976). Calculating degree-days requires determination of the temperature at which development ceases (i.e. threshold temperature for growth) to understand the effect of temperature on rates of development and growth. For example, a treatment wherein temperatures fluctuate between two values may have the same mean temperature as one that is constant, but the fluctuating treatment would have fewer degree-days per day if the temperature drops below the threshold for growth. Our data do not indicate this effect, however, because growth and development were enhanced in the fluctuating (not constant)

temperature regime. Other work on temperature fluctuations and insect development has yielded similar results, and caution should be used when incorporating degree-days for quantitative predictions of development (Wu *et al.*, 2015). Nonetheless, future work should determine and incorporate the threshold temperature for growth to better understand the effects of temperature variability on life-history traits and strategies.

Given enough time (i.e. generations), the optimal temperature for physiological performance for animals is expected to match their environmental temperature (Leroi *et al.*, 1994; Gilchrist *et al.*, 1997). We provide insight into this concept of thermal matching because our experimental crickets were obtained from genetically isolated, true-breeding lines that had been cultured at a constant temperature of 28 °C for several decades (see Mole & Zera, 1994; Zera *et al.*, 1998, 2018; Zera, 2005). We found that crickets did not perform better at the temperature regime under which they have been selected (i.e. constant 28 °C: Figs 2, 3; Supporting Information, S1, S2). Thus, the optimal temperature regime for development in our study system did not appear to be strongly influenced by laboratory selection. In sum, our study (and others) indicates that some traits can exhibit significant plasticity while also exhibiting limited sensitivity to environmental selection, such as exposure to a homogeneous thermal environment over many generations (Lee & Baust, 1982; Brakefield & Kesbeke, 1997; Oikawa *et al.*, 2006; Alton *et al.*, 2017; but see Kingsolver *et al.*, 2009).

For decades, life-history evolution and trade-offs have been rich sources of investigation (e.g. Pianka, 1981; Ricklefs, 1996; Roff, 2002). In this context, *Gryllus* crickets have been frequently investigated due to the trade-off they exhibit between investment into flight capability and female reproduction during early adulthood that is mediated by wing dimorphism (e.g. Roff, 1984, 1990; Mole & Zera, 1994; Zera & Brink, 2000; Zera & Zhao, 2003). However, the association of wing morphology with other fitness-related traits is still unclear (but see Rantala & Roff, 2005). Here, we found that LW individuals developed and grew faster, had greater adult body mass, and invested more resources into the development and maintenance of flight muscle than their SW counterparts (Figs 2C, D; Supporting Information, Figs S1A–C, S2A–C; *sensu* Roff, 1995). Although Mole and Zera (1994) observed the elimination of the flight–fecundity trade-off when LW female crickets were given *ad libitum* access to food, we did not observe this in our study (Fig. 2A, C). However, the flight–fecundity trade-off may be eliminated in stressful conditions because both morphs exhibit similar-sized ovaries when food is less abundant, or when they are immune- or oxidatively challenged

(ZRS & JRG, unpubl. data). This morph-specific variation in life-history traits may be attributed to genes underlying wing morphology, which may be linked to other non-wing morphology genes, exhibiting pleiotropic effects (Stirling *et al.*, 1999, 2001; Roff & Fairbairn, 2012).

The present study found that fitness-related traits and trait trade-offs (i.e. between flight capability and reproduction) can be developmentally plastic in response to variation in temperature and food availability. Our results emphasize the importance of manipulating multiple environmental factors when studying the environmental effects on animals (e.g. Todgham & Stillman, 2013; Kaunisto *et al.*, 2016). Furthermore, we encourage work examining the effects of mean (rather than variability in) temperature on the developmental plasticity of trait interactions. For example, the flight-related trade-offs may be obviated or enhanced at higher temperatures. Future work can build upon our approach by examining the developmental plasticity of trait–trait interactions across developmental stages. For example, frog eggs and larvae (i.e. tadpoles) exhibit varying developmental plasticities in response to differences in mean temperature with regard to fitness-related traits of morphology, physiology and locomotion (Seebacher & Grigaltchik, 2014). We also develop an experimental and statistical framework (i.e. factorial manipulations of environmental factors combined with PCA to reveal shifts in correlative patterns of investment into different life-history traits) that can be applied to a range of animal systems to investigate how environmental complexity influences traits and trait trade-offs.

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AUTHOR CONTRIBUTIONS

J.R.G. and Z.R.S. conceived this study. J.R.G. performed the experiment and collected the data. J.R.G. and Z.R.S. conducted statistical analyses. J.R.G. drafted the manuscript, and both J.R.G. and Z.R.S. edited and approved the final manuscript.

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SUPPORTING INFORMATION

Additional Supporting Information may be found in the online version of this article at the publisher's website.

Figure S1. Effects of variation in temperature and food availability during development on (A) development time, (B) growth rate, (C) adult body mass and (D) femur length in female *G. firmus*. Note: data for both sexes were analysed together but are displayed separately (see Fig. S2 for male results) to improve visualization and comparison. Long-winged morphs are depicted by open columns, and short-winged morphs are depicted by diagonal cross-hatched columns. Fluctuating and constant temperature regimes are denoted by FT and CT, respectively. Values are displayed as mean ± SEM.

Figure S2. Effects of variation in temperature and food availability during development on (A) development time, (B) growth rate, (C) adult body mass and (D) femur length in male *G. firmus*. Note: data for both sexes were analysed together but are displayed separately (see Fig. S1 for female results) to improve visualization and comparison. Long-winged morphs are depicted by open columns, and short-winged morphs are depicted by diagonal cross-hatched columns. Fluctuating and constant temperature regimes are denoted by FT and CT, respectively. Values are displayed as mean ± SEM.

Table S1. Correlation coefficients of initial variables entered into principal components analyses for female ($N = 125$) and male ($N = 121$) *G. firmus* surviving a multi-factorial design that manipulated variation in temperature and food availability throughout development. Correlations are denoted by no asterisks ($P > 0.1$), * ($P < 0.1$), ** ($P < 0.05$), or *** ($P < 0.01$).