How butterflies keep their cool: physical and ecological traits influence thermoregulatory ability and population trends.

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Abstract

1. Understanding which factors influence the ability of individuals to respond to changing temperatures is fundamental to species conservation under climate change.
2. We investigated how a community of butterflies responded to fine-scale changes in air temperature, and whether species-specific responses were predicted by ecological or morphological traits.
3. Using data collected across a UK reserve network, we investigated the ability of 29 butterfly species to buffer thoracic temperature against changes in air temperature. First, we tested whether differences were attributable to taxonomic family, morphology or habitat association. We then investigated the relative importance of two buffering mechanisms: behavioural thermoregulation versus fine-scale microclimate selection. Finally, we tested whether species' responses to changing temperatures predicted their population trends from a UK-wide dataset.
4. We found significant interspecific variation in buffering ability, which varied between families and increased with wing length. We also found interspecific differences in the relative importance of the two buffering mechanisms, with species relying on microclimate selection suffering larger population declines over the last 40 years than those that could alter their temperature behaviourally.
5. Our results highlight the importance of understanding how different species respond to fine-scale temperature variation, and the value of taking microclimate into account in conservation management to ensure favourable conditions are maintained for temperature-sensitive species.

Keywords: Behavioural thermoregulation, butterflies, climate change, generalist, microclimate, population trends, specialist, temperature.

Introduction

Climate change affects the distribution, abundance and phenology of species. These changes can comprise range shifts, with increases in abundance in the cooler parts of species' ranges and declines in abundance in warmer parts (Fox et al., 2015; Lehikoinen et al., 2013; Parmesan et al., 1999), and altered activity patterns, with species emerging or becoming active earlier in the year or in warmer conditions (Sparks & Yates, 1997; Thackeray et al., 2010). Research on the impacts of climate change is now well-advanced, with many studies predicting accelerating effects on the natural world as warming progresses (Dennis & Shreeve, 1991; Devictor, Julliard, Couvet, & Jiguet, 2008).

Changing temperatures can have a particularly marked effect on butterflies, with species at the edge of their distribution showing the most dramatic shifts (Dennis & Shreeve, 1991; Hill, Thomas, & Huntley, 1999; Menéndez et al., 2006; Parmesan et al., 1999). In the UK, where the butterfly fauna is dominated by species at their poleward range limit, climate change is generally predicted to drive range expansions and increases in abundance of butterflies (Thomas & Lewington, 2016; Warren et al., 2001; C. J. Wheatley, unpublished data), although the negative impacts of habitat fragmentation and degradation could counteract these effects (Oliver, Thomas, Hill, Brereton, & Roy, 2012; Warren et al., 2001). In contrast, for a handful of cold-adapted northern or montane species, climate change will likely result in declines (Franco et al., 2006). Given these effects, many habitats are predicted to experience turnover of species and altered species richness as time goes on (González-Megías, Menéndez, Roy, Brereton, & Thomas, 2008; Menéndez et al., 2007). Such changes may be linked to the direct effects of temperature on individuals (Bladon et al., 2019; Calosi, Bilton, & Spicer, 2008), temperature-mediated impacts on water balance (Smit & McKechnie, 2015) and oxygen availability (Pörtner & Knust, 2007), or through the impacts of changing temperatures on species interactions (Diamond et al., 2017). For example, temperature can directly affect the speed of insect life cycles (Rebaudo & Rabhi, 2018; Wilson & Maclean, 2011) and reproductive rates, affecting population growth rates (Mills et al., 2017). Higher temperatures can also change butterfly activity patterns and facilitate more frequent, longer or more effective territorial and mate-locating behaviours, potentially increasing breeding success (Advani, Parmesan, & Singer, 2019; Hayes, Hitchcock, Knock, Lucas, & Turner, 2019; Ide, 2010; Rutowski, Demlong, & Leffingwell, 1994).

In general, species can respond to changing temperatures in three main ways: by directly buffering their temperature through physiological or metabolic means (which we term “physiological thermoregulation”), by changing their behaviour in situ and therefore increasing warming or cooling (“behavioural thermoregulation”), or by shifting their distribution to more favourable microclimatic conditions (“microclimate selection”). Behavioural thermoregulation is widespread in ectotherms (Abram, Boivin, Moiroux, & Brodeur, 2017) and taxa such as butterflies display a number of behavioural mechanisms to control their temperatures. In cooler conditions, these include angling their body and wings so that the surface is perpendicular to the sun and absorbs more energy (Kemp & Krockenberger, 2002; Pivnick & McNeil, 1986; Wasserthal, 1975) or even acts to concentrate sunlight (Shanks, Senthilarasu, ffrench-Constant, & Mallick, 2015), thereby increasing body temperature. In contrast, in hotter conditions, butterflies can adopt postures that reduce the surface area exposed to the sun or that reflect more sunlight (Dreisig, 1995; Rutowski et al., 1994), thus reducing body temperature. Microclimate selection is also common, with individuals selecting sunny locations to warm up or shady locations to cool down (Hayes et al., 2019; Ide, 2010; Kleckova & Klecka, 2016; Kleckova, Konvicka, & Klecka, 2014). Over slightly larger spatial scales, individuals can also select a microhabitat with the preferred temperature (Dreisig, 1995; Rutowski et al., 1994), enabling individuals to maintain their body temperature under shifting climatic conditions (Kleckova, Konvicka, & Klecka, 2014). This can result in individuals within a species preferring more northerly slopes at the southern end of their range, but more southerly slopes at the northern end of their range (Oliver, Hill, Thomas, Brereton, & Roy, 2009; Suggitt et al., 2012). Understanding the ability of species to buffer against changing temperatures, and the means by which they do this, is therefore important for predicting the future impact of climate change on whole communities.

Models exploring the long-term effects of climate change on species distributions and population trends are well-advanced, although these are typically based upon weather station data collected from standardised environments (Bramer et al., 2018) and therefore do not take into account the fine-scale impacts of local habitat structure and topography on temperature (“microclimatic temperature”). Microclimatic temperature can differ dramatically from the climatic mean, and varies with topography, vegetation cover and altitude (Suggitt et al., 2011). Microclimatic temperature is also variable over short time-frames, with extremes often exceeding meteorological means in exposed areas, but being much more stable in sheltered areas (Maclean, Suggitt, Wilson, Duffy, & Bennie, 2017). It is also microclimatic temperatures that individual organisms experience on a day-to-day basis and which can affect the distribution and abundance of species at the local scale. Therefore, information on how individuals respond to fine-scale microclimatic temperature variation over short time-periods (Bladon et al., 2019; Ide, 2002; Kelly, Godley, & Furness, 2004) may need to be incorporated into models to accurately predict species’ responses to climate change (Kearney, Shine, & Porter, 2009; Lembrechts et al., 2019). For example, it is likely that a diversity of microclimates at the local scale could protect species from wider temperature change, by providing pockets of favourable microclimate for temperature-sensitive species (Thomas & Simcox, 2005). Indeed, recent research has found that areas with more diverse microclimates have lower levels of extinction for insect and plant populations than areas with more homogenous microclimates (Suggitt et al., 2018).

Previous studies have quantified the thermoregulatory capacity of several lepidopteran species using direct measurements of body temperatures from live individuals (Bryant, Thomas, & Bale, 2000; Casey, 1976; Ide, 2010; Kleckova & Klecka, 2016; Kleckova et al., 2014; Rutowski et al., 1994). These have used a regression of body temperature against concurrent free air temperature to parameterise the thermal biology of each species (Bryant et al., 2000), and to classify species as “behavioural thermoregulators” or “thermal conformers” (Knapp & Casey, 1986). However, comparisons of thermoregulatory capacity have never been made across entire communities, nor assessed against a broad range of traits. For example, a species’ thermoregulatory capacity may be determined by physical traits, such as wing size and colour, which affect an individual butterfly’s ability to cool or warm itself, or by some underlying physiological characteristics related to its evolutionary history. In addition, no studies have yet taken the temperature of the immediate environment of the individual into account, although this may differ dramatically from free air (Bramer et al., 2018), and be explicitly selected by individuals.

Here, we explore how 29 UK butterfly species differ in their ability to respond to local temperature variation. We test whether the ability to buffer body temperature against changes in air temperature varies between species, according to species’ taxonomic affiliation, wingspan, wing colouration and habitat association. We also use direct field measures to partition each species’ thermoregulatory capacity into microclimate selection and behavioural thermoregulation. We then compare differences in thermoregulatory ability between species to recent population trends and range changes across the UK. Our results have important implications for predicting which species are most at risk from warming temperatures, and for informing habitat management in the face of climate change.

Materials and methods

Data collection

We collected data from four calcareous grassland sites in Bedfordshire, UK, owned and managed by The Wildlife Trust for Bedfordshire, Cambridgeshire & Northamptonshire: Totternhoe Quarry [-0.56836, 51.89199], Totternhoe Knolls [-0.58039, 51.88989], Pegsdon Hills [-0.37020, 51.95354] and Blows Downs [-0.49580, 51.88321] (Fig. S1). Between April and September 2009, and between May and September 2018, the entire area of each reserve was searched systematically (to within 20 metres) for adult butterflies each month. After recording the behaviour of each butterfly when first encountered, we attempted to catch as many individuals as possible using a butterfly net. We did not chase individuals, to ensure that the temperature recorded reflected the activity of the butterfly prior to capture. Immediately after capture, we used a fine (0.25 mm) mineral-insulated type K thermocouple and hand-held indicator (Tecpel Thermometer 305B) to record external thoracic temperatures (T_{body}). Only three individual devices were used for data collection, and were calibrated to the same readings prior to use. The thermocouple was pressed gently onto an exposed area of each butterfly's thorax, while the butterfly was held securely in the net, away from the hands of the recorder to avoid artificially elevating the recording or causing any damage to the butterfly. Butterflies were then released. Previous comparisons between external and internal body temperature readings in both large and small moths have found no significant difference in the temperatures recorded (Casey, 1976; Knapp & Casey, 1986), indicating that our readings are a realistic measure of the internal thoracic temperature of butterflies of a range of different sizes. A second temperature recording was then taken at the same location in free air, in the shade, at waist height (T_{air}). In 2018, for butterflies perching on vegetation at the time of capture, a third temperature reading was taken by holding the thermocouple a centimetre above the perch location (T_{perch}), providing a measure of the thermal properties of the butterfly's chosen microhabitat.

In 2018, additional fieldwork was conducted at two sites in Cumbria (Irton Fell [-3.34000, 54.40672] and Haweswater [-2.84598, 54.50756], May-June) and one site in Scotland (Ben Lawers [-4.27326, 56.53287], July), specifically to collect data on the Mountain Ringlet *Erebia epiphron*, a montane specialist. In August 2018, additional data were also collected from a chalk grassland at Winterbourne Downs [-1.68500, 51.14963] in Wiltshire, UK, a site owned and managed by the Royal Society for the Protection of Birds (RSPB), to gain further high-temperature measurements for a range of species (Fig. S1). At these sites, systematic site searches were not conducted, and fieldwork was dedicated to catching butterflies to take temperature recordings. The number of individual butterflies for each species caught in each location and each year is presented in Table S1.

Statistical analyses – buffering ability

All statistical analyses were performed in R version 3.5.3 (R Core Team, 2019). For the 29 species (Table S2) for which at least ten thoracic temperature measurements were collected, we fitted simple linear regression models of T_{body} against T_{air}, and extracted the slope of this relationship. This slope represents a measure of the ability of individuals within each species to "buffer" their body temperature against changes in air temperature. A species with a shallow slope exhibits a relatively narrow range of T_{body} over a large range of T_{air} (at the extreme a slope of 0 would indicate thermal independence of T_{body} with respect to T_{air}), while a species with a steep slope exhibits a wider range of T_{body} (for example a slope of 1 would indicate complete dependence of T_{body} on T_{air}) (Bryant et al., 2000). For ease of interpretation, we subtracted the regression slope for each species from one, so that higher values represent a better buffering ability, and lower values represent a poorer buffering ability. Put another way, a higher value indicates that as air temperature increases, the difference between thoracic temperature and air temperature decreases. We define this as the species' "overall buffering ability".

To investigate which traits affect species' buffering ability, we fitted a hierarchical, or mixed effects, model in which individual thoracic temperature was regressed against air temperature, taxonomic family, mean wingspan, wing colour category, habitat association, and each two-way interaction of air temperature with the other variables. In this model structure, any term which interacts with air temperature effectively modifies the slope of the regression of T_{body} on T_{air} (Fig. 1). Random coefficients were fitted by species, to account for differences in both the intercept and slope of each species' relationship between thoracic temperature and air temperature. Mean wingspan was taken from Eeles (2020) and, in cases of sexual size dimorphism, the mean was taken from the estimate for males and females. Colour category was determined by assigning each species a score from 1 (white) to 6 (black) on the basis of how pale/dark wing colours appeared by eye (Table S2). This represents a robust and repeatable method, especially given the wide range of colours represented across the species studied. Habitat association was based on Asher et al. (2001), where our 29 species were classified as habitat generalists (21 species), habitat specialists (six species) or migrant species (two species, Painted Lady *Vanessa cardui* and Red Admiral *Vanessa atalanta*) (Table S2). A backwards step-wise procedure was used to remove non-significant terms until we arrived at a minimal model, in which all remaining terms were significant (Table S3). In this model structure, the significance of any two-way interaction between air temperature and another predictor variable in the minimal model indicates that the variable is important in explaining the difference in buffering ability between species. We tested for the presence of any collinearity between our predictor variables by regressing them on one another (Dormann et al., 2013). Where correlations existed, we did not change the model structure, but considered their effects in our interpretation of the results in the discussion.

Statistical analyses – population trends and range shifts

Next, we tested whether overall buffering ability explained population trends or changes in northern range margins across species. We obtained long-term (series trend; 39-42 years depending on species) and short-term (10-year) population data for butterflies across the UK from Butterfly Conservation's UK Butterfly Monitoring Scheme (Brereton et al., 2018). We used published estimates of species' northern range margins for two time periods (1966-1975 and 2001-2010) (Mason et al., 2015), and calculated the distance between them, in kilometres, as the change in northern range margin between these two time periods. We then fitted three linear regressions using the long- and short-term population trends and the change in northern range margin as response variables, with each maximal model containing species-specific buffering ability, taxonomic family, habitat association (generalist, specialist or migrant), and the interaction between buffering ability and family as predictors. The other possible interaction terms were not included due to the many missing factor levels caused by both the monospecific Riodinidae and by having only two migrant species in the data. A backwards step-wise procedure was applied to each model to remove non-significant terms until we arrived at a minimal model, in which all remaining terms were significant (Tables S4). Mountain Ringlet *Erebia epiphron* was excluded from these models, as no population trend estimates were available, owing to the paucity of regularly sampled squares which contain the species in the UKBMS data (Brereton et al., 2018), and because latitudinal range margin changes for montane species are confounded by altitudinal shifts (Mason et al., 2015). Additionally, range margin estimates were not available for migrant species, ubiquitous species (found in more than 90% of mainland Britain) or species whose northern range margins were already within 100 km of the north coast of mainland Britain in the earlier time period (Mason et al., 2015), resulting in a further 13 species being omitted from the range change analyses (Table S2).

Since the Duke of Burgundy *Hamearis lucina* represents a monospecific family in the UK (Riodinidae), and because its buffering ability is an outlier relative to all other species, we refitted each of the above models with and without the Duke of Burgundy included, to check that the results obtained were not dependent upon this species being included. All results presented were robust to excluding the Duke of Burgundy from the dataset.

Statistical analyses – buffering mechanisms

For the 16 species with at least ten T_{perch} records, we tested the extent to which their overall buffering ability was driven by their choice of microclimate, or by alternative behavioural mechanisms. To do this, we calculated "microclimate selection" as the difference between T_{air} and T_{perch}, and "behavioural thermoregulation" as the difference between T_{perch} and T_{body} for each individual butterfly (Table S2). We used this approach, rather than a slope-based approach similar to the analysis for overall buffering ability, because the response of interest was the extent to which individual butterflies were able to utilise either microclimate temperatures or behavioural mechanisms to adjust their thoracic temperature relative to air temperature. Species which use behavioural mechanisms to control their thoracic temperature are more likely to be able to respond to larger-scale changes in temperature because they have more thermal independence from their environment than species which rely upon the thermal buffering provided by their choice of microhabitat. These robust species may therefore be expected to have more positive population trends or shifting ranges under climate change. To test this, we again fitted each of three variables (short-term and long-term population trend, and change in northern range margin) as response variables, with the average difference in magnitude between "behavioural thermoregulation" and "microclimate selection" for each species, taxonomic family, and the interaction between these terms, as predictor variables (Table S5). We were unable to include habitat association in these models, as we only had sufficient data from two specialist species.

Results

Temperatures sampled

Air temperature during sampling ranged from 10.0–32.4°C in 2009, and 11.3–34.8°C in 2018. This represented what is likely to be a normal range of daytime temperatures experienced from May to September in our study locations. While the majority of species were sampled over much of this range some, such as those which only fly in early spring (Dingy Skipper *Erynnis tages*: 10.0–26.3°C; Duke of Burgundy *Hamearis lucina*: 13.0–24.7°C; Orange-tip *Anthocharis cardamines*: 13.1–22.3°C) and one montane specialist (Mountain Ringlet *Erebia epiphron*: 13.9–26.1°C), were only recorded at a lower range of air temperatures (Fig. 1).

Buffering ability

The 29 species differed markedly in their response to changes in temperature: overall buffering ability estimates ranged from -0.404 (Duke of Burgundy *Hamearis lucina*) to 0.717 (Orange-tip *Anthocharis cardamines*) (mean ± 1 standard error = 0.234 ± 0.038) (Table S2, Fig. 1). The mean adjusted R-squared value for these models was 0.49, indicating that the models were a good fit to the data. Taxonomic family was the strongest predictor of buffering ability ($\chi^2 = 26.11$, d.f. = 4, $P < 0.001$), and this result held when the Duke of Burgundy was excluded from modelling ($\chi^2 = 23.43$, d.f. = 3, $P < 0.001$). Larger species also had a better buffering ability ($\chi^2 = 4.88$, d.f. = 1, P = 0.027, Table S3). Pierids were best at buffering their thoracic temperature against air temperature changes (mean ± 1 standard error = 0.460 ± 0.075), followed by hesperiids (0.269 ± 0.082), lycaenids (0.209 ± 0.030) and nymphalids (0.198 ± 0.041), which had the lowest buffering ability (Fig. 2). The pattern across families also reflects a general pattern in colouration, and a univariate regression of buffering ability against wing colour category returned a significant negative response (F = 4.58, d.f. = 1, P = 0.042), with paler butterflies showing greater buffering ability. Across species, mean wingspan and wing colour category were not related (F = 0.254, d.f. = 1, P = 0.619), but butterflies in the family Pieridae were paler than other families (F = 13.80, d.f. = 4, $P < 0.001$), generalist species were marginally paler than specialist and migrant species (F = 3.31, d.f. = 2, P = 0.052), butterflies in Pieridae and Nymphalidae were larger than other families (F = 10.54, d.f. = 4, $P < 0.001$) and migrant species were larger than resident species (F = 7.12, d.f. = 1, P = 0.013).

<u>Population trends and range shifts</u>

Neither species' long-term (F = 0.28, d.f. = 1, P = 0.602) nor short-term (F = 0.59, d.f. = 1, P = 0.450) population trends in the UK were predicted by species' overall buffering ability. There was also no effect of taxonomic family (long-term: F = 0.11, d.f. = 5, P = 0.989; short-term: F = 1.00, d.f. = 4, P = 0.426) on either population trend. Migrant species had more positive short-term population trends than either habitat generalists or habitat specialists (F = 5.11, d.f. = 2, P = 0.009), although there was no difference between these groups' long-term population trends (F = 1.96, d.f. = 2, P = 0.150, Table S4).

There were no significant associations between species' overall buffering ability and changes in their northern range margins (F = 1.11, d.f. = 1, P = 0.313). Rather, the northwards advance of species' ranges was predicted by their habitat association, with generalists (mean ± 1 standard error: 131.9 ± 19.3 km) moving northwards more than specialists (mean ± 1 standard deviation: 46.3 ± 21.7 km, F = 11.32, d.f. = 1, P = 0.002). There was no difference between taxonomic families in extent of range margin change (F = 0.62, d.f. = 4, P = 0.651, Table S4).

Buffering mechanisms

For the reduced set of 16 species for which we were able to estimate the thermoregulatory value of microclimate selection and behavioural thermoregulation (Fig. 3, Fig. S2), species for which the magnitude of behavioural thermoregulation was greater than the magnitude of thermoregulation via microclimate selection had more positive long-term population trends (F = 10.30, d.f. = 1, P = 0.009, Fig. 4) than species which were more reliant on microclimate selection. Taxonomic family (F = 4.95, d.f. = 3, P = 0.023) also predicted long-term population trends amongst this reduced set of species, but there was no interaction between the difference in buffering mechanism and family (F = 1.16, d.f. = 3, P = 0.389, Table S5). There was no effect of the difference between behavioural thermoregulation and microclimate selection on either species' short-term population trends (F = 1.35, d.f. = 1, P = 0.272) or changes in species' northern range margins (F = 0.14, d.f. = 1, P = 0.726, Table S5).

Discussion

The 29 butterfly species differed markedly in their ability to buffer thoracic temperature against air temperature. Interspecific differences in buffering ability were related to wingspan and taxonomic family, but not to species' UK population trends or northern range expansion. Instead, migrant species exhibited more positive short-term population trends than resident species, and habitat generalists advanced their range margins further north than habitat specialists (corroborating a result found previously (Menéndez et al., 2006; Warren et al., 2001)). Species which were more reliant on the selection of a suitable microclimate for thermoregulation experienced more negative long-term population trends than species which used other behavioural thermoregulatory mechanisms.

Differences between taxonomic families explained most of the variation in overall buffering ability, with pierids showing the greatest capacity for thoracic temperature buffering. It is possible that this is, in part, driven by colouration. In a univariate model, wing colour category significantly predicted buffering ability, with paler species exhibiting better buffering ability. However, this result was swamped in the multivariate model by the effect of family, but with families containing generally paler species (Pieridae and Hesperiidae) better at buffering than darker families (Lycaenidae and Nymphalidae). Pale butterflies, such as the Large White *Pieris brassicae* and Brimstone *Gonepteryx rhamni*, may be better able to harness the high reflectance of their wings to increase both heat loss and heat gain. By spreading their wings, high reflectance will dissipate excess radiation at high temperatures, enabling them to remain cooler. Meanwhile, by angling their wings upwards, the high reflectance can also be used to concentrate heat onto their thorax, enabling them to warm themselves up more efficiently at low temperatures (Shanks et al., 2015). Our results therefore corroborate the findings of Shanks *et al.* (2015) using field data. By contrast, while darker butterflies, such as the Gatekeeper *Pyronia tithonus* and Dingy Skipper *Erynnis tages*, should be able to warm themselves up at low air temperatures through increased absorption of incoming radiation, they may have no easy mechanism of behavioural thermoregulation for heat loss at higher temperatures.

Within families, mean wingspan was also a significant predictor of buffering ability, with larger species (such as the Peacock *Aglais io* and Large White *Pieris brassicae*) better at buffering than their smaller relatives (such as the Small Heath *Coenonympha pamphilus* and Green-veined White *Pieris napi*). Again this is intuitive, as larger species have a larger wing surface area, affording them greater temperature control from basking (Gilchrist, 1990; Shanks et al., 2015; Wasserthal, 1975). Conversely, a larger wing area may also help butterflies to reduce body temperatures during flight, when rapid muscle contraction will otherwise raise body temperature (Advani et al., 2019). Thus, larger butterflies may be better at both raising and lowering their thoracic temperature using behavioural thermoregulation.

We found that two species, Mountain Ringlet *Erebia epiphron* and Duke of Burgundy *Hamearis lucina*, had particularly poor buffering ability, exhibiting a greater range of thoracic temperature than the range of air temperature to which they are exposed. The Mountain Ringlet *Erebia epiphron* is a montane specialist, only found at altitudes over 200 m (with most populations over 500 m in the UK) (Thomas & Lewington, 2016), which appears to be tracking its suitable climate to higher altitudes (Franco et al., 2006). As a cold-adapted species, it would be expected to be better adapted to warming itself up at cooler temperatures than cooling down at higher temperatures. The Duke of Burgundy *Hamearis lucina* has an early spring flight season (late April to late May) (Thomas & Lewington, 2016), but is reliant upon warm, sheltered habitat patches for male territory defence (Hayes et al., 2019; Turner et al., 2009). However, beyond its choice of microhabitat, our data indicates that the Duke of Burgundy *Hamearis lucina* may have very poor buffering ability, which could explain why populations of this species seem particularly sensitive to habitat management changes that could influence the local microclimate (Hayes et al., 2018; Turner et al., 2009).

However, it is possible that the results for the Mountain Ringlet *Erebia epiphron* and the Duke of Burgundy *Hamearis lucina* are in part a function of the air temperatures at which they were recorded. As high-altitude and early spring species (respectively), it was difficult to obtain data at air temperatures above 20-25°C. At these temperatures, it is likely that most butterflies would still be attempting to warm themselves up, rather than cool down, and thus we currently lack data on how these species respond to air temperatures above their optimum. It is notable, however, that other early spring species (e.g. Orange-tip *Anthocharis cardamines* and Dingy Skipper *Erynnis tages*), for which data were collected at the same time and over the same air temperature range as the Duke of Burgundy *Hamearis lucina*, exhibited a much better buffering ability. Further work to understand the responses of these and other early spring and high-altitude species to a wider range of air temperatures is vital in order to draw robust conclusions about their overall thermal tolerance.

We found a clear difference in the degree to which different species used microclimate selection versus behavioural thermoregulation to buffer their thoracic temperature. Butterflies show a number of mechanisms for behavioural thermoregulation (Kemp & Krockenberger, 2002; Pivnick & McNeil, 1986; Rutowski et al., 1994; Wasserthal, 1975), and an ability to select cooler or warmer microhabitats (Hayes et al., 2019; Kleckova & Klecka, 2016; Kleckova et al., 2014; Rutowski et al., 1994; Suggitt et al., 2012). Our results demonstrate that the relative importance of these two mechanisms varies between species, with some (such as Brown Argus *Aricia agestis*, Small Copper *Lycaena phlaeas* and Small Heath *Coenonympha pamphilus*) being much more reliant upon the thermal environment provided by their choice of microhabitat, whilst others (such as Large Skipper *Ochlodes sylvanus*, Ringlet *Aphantopus hyperantus* and Small Skipper *Thymelicus sylvestris*) exhibit thoracic temperatures much higher than their immediate surroundings, suggesting effective behavioural thermoregulatory mechanisms and a greater degree of thermal independence from their environment.

This range of responses, from dependence on the thermal environment of their microhabitat through to effective behavioural thermoregulation, was found among species currently classified as habitat generalists. Species' classification as generalist or specialist is currently based solely on their habitat preferences (Asher et al., 2001). However, our results suggest that some species, with generalist habitat requirements, may in fact have quite specialist thermal requirements. In the context of climate change, it is important to consider both species' habitat and thermal requirements, and it may be necessary to re-classify some generalists as "thermal specialists", on which future conservation attention should be focused. These species may be at particular risk from landscape homogenisation, and conservation actions to increase structural and thermal heterogeneity may be particularly important for these species' persistence (Kleckova et al., 2014; Suggitt et al., 2018). In addition, we were only able to include two habitat specialists (Chalkhill Blue *Polyommatus coridon* and Mountain Ringlet *Erebia epiphron*) in this analysis. It is important to collect further data on the buffering mechanisms used by other habitat specialists, to understand the extent to which they are also threatened by climate change.

The effectiveness of behavioural thermoregulation compared to microclimate selection predicted species' long-term population trends, with those relying more on microclimate selection experiencing more negative population trends over the last 40 years. Recent research into species' responses to ongoing global change has begun to focus on local microclimatic conditions as well as on habitat availability (Bramer et al., 2018; Curtis & Isaac, 2015). Landscapes with diverse microclimatic environments experience higher persistence of insect (including butterflies) and plant species (Suggitt et al., 2015, 2018), possibly because such environments offer pockets of cooler conditions for climate-sensitive species. However, the exact mechanisms underpinning these landscape-level effects are not yet fully understood. Our results demonstrate that links between individual-level responses to temperature and species-level responses to climate must be more thoroughly investigated (Briscoe et al., 2019). Developing understanding of the mechanisms underlying species-specific buffering ability is crucial to predicting species' responses to climate change and designing mitigation strategies to conserve them (Greenwood, Mossman, Suggitt, Curtis, & Maclean, 2016). This study represents an important step towards understanding how reserve management can provide not only suitable habitat heterogeneity, but also thermal heterogeneity, for example through the provision of diverse topography, to protect a broad community of species in the face of climatic change (Curtis & Isaac, 2015; Suggitt et al., 2018).

This study has focussed on adult butterflies, but these represent just one of the four life stages of lepidopterans. The adult is the stage most able to disperse and may, therefore, be the least restricted in terms of habitat (Thomas & Lewington, 2016) and the best able to buffer temperature through microclimate selection. The eggs, larvae and pupae are more sedentary and may have a more limited ability to thermoregulate and be more sensitive to temperature change. Alternatively, they may have evolved greater robustness to the fluctuating temperatures of their small, local environment, rendering them less sensitive to environmental changes. So far few studies have focussed on the impacts of temperature on life stages other than adults (although see (Bryant et al., 2000; Casey, 1976; Knapp & Casey, 1986; Turlure, Radchuk, Baguette, Van Dyck, & Schtickzelle, 2011)). It is therefore important that more work is carried out to assess how these different stages differ, to gain a better idea of the temperature buffering ability of butterflies as a whole.

The different buffering abilities of adult butterflies identified in this study emphasises the variation in species' vulnerability to climate change. By identifying which characteristics of butterflies predict buffering ability and the importance of different mechanisms of thermoregulation in determining long-term trends in butterfly populations, we pave the way for more in-depth studies to predict species' responses to long-term climate change and to inform the design of reserves that provide suitable microclimates to protect vulnerable species against the future impacts of global warming.

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Authors' contributions: AJB co-designed the study, and led the 2018 data collection and analyses. ML assisted with data collection, led the data entry, and assisted with analyses. SJB, AM and JMW assisted with data collection and data entry. EKB, SC, MPH, GEH, RK, CL assisted with data collection. SRE and RM assisted with data collection and analysis. TMF assisted with data collection and conceived some analyses. ECT co-designed the study, led the 2009 data collection, and assisted with analyses. AJB wrote the first draft of the manuscript, and all authors contributed to the revisions.

Data Availability Statement: Should this manuscript be accepted, the data supporting the results will be archived in an appropriate public repository (Dryad, Figshare or Hal) and the data DOI will be included at the end of the article.

References

Abram, P. K., Boivin, G., Moiroux, J., & Brodeur, J. (2017). Behavioural effects of temperature on ectothermic animals: unifying thermal physiology and behavioural plasticity. *Biological Reviews*, *92*(4), 1859–1876. doi: 10.1111/brv.12312

Advani, N. K., Parmesan, C., & Singer, M. C. (2019). Takeoff temperatures in *Melitaea cinxia* butterflies from latitudinal and elevational range limits: a potential adaptation to solar irradiance. *Ecological Entomology*, *44*(3), 389–396. doi: 10.1111/een.12714

Asher, J., Warren, M. S., Fox, R., Harding, P., Jeffcoate, G., & Jeffcoate, S. (2001). *The Millennium Atlas of Butterflies in Britain and Ireland*. Oxford: Oxford University Press.

Bladon, A. J., Donald, P. F., Jones, S. E. I., Collar, N. J., Deng, J., Dadacha, G., … Green, R. E. (2019). Behavioural thermoregulation and climatic range restriction in the globally threatened Ethiopian Bush-crow *Zavattariornis stresemanni*. *Ibis*, *161*, 546–558. doi: 10.1111/ibi.12660

Bramer, I., Anderson, B. J., Bennie, J., Bladon, A. J., De Frenne, P., Hemming, D., … Gillingham, P. K. (2018). Advances in Monitoring and Modelling Climate at Ecologically Relevant Scales. In D. A. Bohan, A. J. Dumbrell, G. Woodward, & M. Jackson (Eds.), *Advances in Ecological Research* (pp. 101–161). doi: 10.1016/bs.aecr.2017.12.005

Brereton, T. M., Botham, M. S., Middlebrook, I., Randle, Z., Noble, D., Harris, S., … Roy, D. B. (2018). *United Kingdom Butterfly Monitoring Scheme report for 2017* (p. 24). Retrieved from Centre for Ecology and Hydrology, Butterfly Conservation, British Trust for Ornithology and Joint Nature Conservation Committee website: http://www.ukbms.org/reportsandpublications

Briscoe, N. J., Elith, J., Salguero-Gómez, R., Lahoz-Monfort, J. J., Camac, J. S., Giljohann, K. M., … Guillera-Arroita, G. (2019). Forecasting species range dynamics with process-explicit models: matching methods to applications. *Ecology Letters*, *0*(0). doi: 10.1111/ele.13348

Bryant, S. R., Thomas, C. D., & Bale, J. S. (2000). Thermal ecology of gregarious and solitary nettle-feeding nymphalid butterfly larvae. *Oecologia*, *122*(1), 1–10. doi: 10.1007/PL00008825

Calosi, P., Bilton, D. T., & Spicer, J. I. (2008). Thermal tolerance, acclimatory capacity and vulnerability to global climate change. *Biology Letters*, *4*(1), 99–102. doi: 10.1098/rsbl.2007.0408

Casey, T. M. (1976). Activity Patterns, Body Temperature and Thermal Ecology in Two Desert Caterpillars (Lepidoptera: Sphingidae). *Ecology*, *57*(3), 485–497. doi: 10.2307/1936433

Curtis, R. J., & Isaac, N. J. B. (2015). The effect of temperature and habitat quality on abundance of the Glanville fritillary on the Isle of Wight: implications for conservation management in a warming climate. *Journal of Insect Conservation*, *19*(2), 217–225. doi: 10.1007/s10841-014-9738-1

Dennis, R. L. H., & Shreeve, T. G. (1991). Climatic change and the British butterfly fauna: Opportunities and constraints. *Biological Conservation*, *55*(1), 1–16. doi: 10.1016/0006-3207(91)90002-Q

Devictor, V., Julliard, R., Couvet, D., & Jiguet, F. (2008). Birds are tracking climate warming, but not fast enough. *Proceedings of the Royal Society B: Biological Sciences*, *275*(1652), 2743–2748. doi: 10.1098/rspb.2008.0878

Diamond, S. E., Chick, L., Penick, C. A., Nichols, L. M., Cahan, S. H., Dunn, R. R., … Gotelli, N. J. (2017). Heat tolerance predicts the importance of species interaction effects as the climate changes. *Integrative and Comparative Biology*, *57*(1), 112–120. doi: 10.1093/icb/icx008

Dormann, C. F., Elith, J., Bacher, S., Buchmann, C., Carl, G., Carré, G., … Lautenbach, S. (2013). Collinearity: a review of methods to deal with it and a simulation study evaluating their performance. *Ecography*, *36*(1), 27–46. doi: 10.1111/j.1600-0587.2012.07348.x

Dreisig, H. (1995). Thermoregulation and flight activity in territorial male graylings, Hipparchia semele (Satyridae), and large skippers, Ochlodes venata (Hesperiidae). *Oecologia*, *101*(2), 169–176. doi: 10.1007/BF00317280

Eeles, P. (2020). UK Butterflies. Retrieved from UK Butterflies website: https://ukbutterflies.co.uk/index.php

Fox, R., Brereton, T. M., Asher, J., August, T. A., Botham, M. S., Bourn, N. A. D., … Roy, D. B. (2015). *The State of the UK's Butterflies*. Retrieved from Butterfly Conservation and the Centre for Ecology & Hydrology website: https://butterfly-conservation.org/sites/default/files/soukb-2015.pdf

Franco, A. M. A., Hill, J. K., Kitschke, C., Collingham, Y. C., Roy, D. B., Fox, R., … Thomas, C. D. (2006). Impacts of climate warming and habitat loss on extinctions at species' low-latitude range boundaries. *Global Change Biology*, *12*(8), 1545–1553. doi: 10.1111/j.1365-2486.2006.01180.x

Gilchrist, G. W. (1990). The Consequences of Sexual Dimorphism in Body Size for Butterfly Flight and Thermoregulation. *Functional Ecology*, *4*(4), 475–487. doi: 10.2307/2389315

González-Megías, A., Menéndez, R., Roy, D., Brereton, T., & Thomas, C. D. (2008). Changes in the composition of British butterfly assemblages over two decades. *Global Change Biology*, *14*(7), 1464–1474. doi: 10.1111/j.1365-2486.2008.01592.x

Greenwood, O., Mossman, H. L., Suggitt, A. J., Curtis, R. J., & Maclean, I. M. D. (2016). Using in situ management to conserve biodiversity under climate change. *Journal of Applied Ecology*, *53*(3), 885–894. doi: 10.1111/1365-2664.12602

Hayes, M. P., Hitchcock, G. E., Knock, R. I., Lucas, C. B. H., & Turner, E. C. (2019). Temperature and territoriality in the Duke of Burgundy butterfly, Hamearis lucina. *Journal of Insect Conservation*, *23*(4), 739–750. doi: 10.1007/s10841-019-00166-6

Hayes, M. P., Rhodes, M. W., Turner, E. C., Hitchcock, G. E., Knock, R. I., Lucas, C. B. H., & Chaney, P. K. (2018). Determining the long-term habitat preferences of the Duke of Burgundy butterfly, Hamearis lucina, on a chalk grassland reserve in the UK. *Journal of Insect Conservation*, *22*(2), 329–343. doi: 10.1007/s10841-018-0065-9

Hill, J. K., Thomas, C. D., & Huntley, B. (1999). Climate and habitat availability determine 20th century changes in a butterfly's range margin. *Proceedings of the Royal Society of London. Series B: Biological Sciences*, *266*(1425), 1197–1206. doi: 10.1098/rspb.1999.0763

Ide, J. (2010). Weather factors affecting the male mate-locating tactics of the small copper butterfly (Lepidoptera: Lycaenidae). *European Journal of Entomology*, *107*, 369–376. doi: 10.14411/eje.2010.046

Ide, J.-Y. (2002). Seasonal changes in the territorial behaviour of the satyrine butterfly *Lethe diana* are mediated by temperature. *Journal of Ethology*, *20*(1), 71–78. doi: 10.1007/s10164-002-0056-9

Kearney, M. R., Shine, R., & Porter, W. P. (2009). The potential for behavioral thermoregulation to buffer '"cold-blooded"' animals against climate warming. *Proceedings of the National Academy of Sciences*, *106*(10), 3835–3840. doi: https://doi.org/10.1073/pnas.0808913106

Kelly, A., Godley, B. J., & Furness, R. W. (2004). Magpies, *Pica pica*, at the southern limit of their range actively select their thermal environment at high ambient temperatures. *Zoology in the Middle East*, *32*(1), 13–26. doi: 10.1080/09397140.2004.10638039

Kemp, D. J., & Krockenberger, A. K. (2002). A novel method of behavioural thermoregulation in butterflies. *Journal of Evolutionary Biology*, *15*(6), 922–929. doi: 10.1046/j.1420-9101.2002.00470.x

Kleckova, I., & Klecka, J. (2016). Facing the heat: thermoregulation and behaviour of lowland species of a cold-dwelling butterfly genus: Erebia. *PLoS ONE*, *11*(3), e0150393. doi: 10.1371/journal.pone.0150393

Kleckova, I., Konvicka, M., & Klecka, J. (2014). Thermoregulation and microhabitat use in mountain butterflies of the genus Erebia: Importance of fine-scale habitat heterogeneity. *Journal of Thermal Biology*, *41*, 50–58. doi: 10.1016/j.jtherbio.2014.02.002

Knapp, R., & Casey, T. M. (1986). Thermal Ecology, Behavior, and Growth of Gypsy Moth and Eastern Tent Caterpillars. *Ecology*, *67*(3), 598–608. doi: 10.2307/1937683

Lehikoinen, A., Jaatinen, K., Vähätalo, A. V., Clausen, P., Crowe, O., Deceuninck, B., … Fox, A. D. (2013). Rapid climate driven shifts in wintering distributions of three common waterbird species. *Global Change Biology*, *19*(7), 2071–2081. doi: 10.1111/gcb.12200

Lembrechts, J. J., Lenoir, J., Roth, N., Hattab, T., Milbau, A., Haider, S., … Nijs, I. (2019). Comparing temperature data sources for use in species distribution models: From in-situ logging to remote sensing. *Global Ecology and Biogeography*, *28*(11), 1578–1596. doi: 10.1111/geb.12974

Maclean, I. M. D., Suggitt, A. J., Wilson, R. J., Duffy, J. P., & Bennie, J. J. (2017). Fine-scale climate change: modelling spatial variation in biologically meaningful rates of warming. *Global Change Biology*, *23*(1), 256–268. doi: 10.1111/gcb.13343

Mason, S. C., Palmer, G., Fox, R., Gillings, S., Hill, J. K., Thomas, C. D., & Oliver, T. H. (2015). Geographical range margins of many taxonomic groups continue to shift polewards. *Biological Journal of the Linnean Society*, *115*(3), 586–597. doi: 10.1111/bij.12574

Menéndez, R., González-Megías, A., Collingham, Y., Fox, R., Roy, D. B., Ohlemüller, R., & Thomas, C. D. (2007). Direct and Indirect Effects of Climate and Habitat Factors on Butterfly Diversity. *Ecology*, *88*(3), 605–611. doi: 10.1890/06-0539

Menéndez, R., González-Megías, A., Hill, J. K., Braschler, B., Willis, S. G., Collingham, Y., … Thomas, C. D. (2006). Species richness changes lag behind climate change. *Proceedings of the Royal Society B: Biological Sciences*, *273*(1593), 1465–1470. doi: 10.1098/rspb.2006.3484

Mills, S. C., Oliver, T. H., Bradbury, R. B., Gregory, R. D., Brereton, T., Kühn, E., … Evans, K. L. (2017). European butterfly populations vary in sensitivity to weather across their geographical ranges. *Global Ecology and Biogeography*, *26*(12), 1374–1385. doi: 10.1111/geb.12659

Oliver, T. H., Thomas, C. D., Hill, J. K., Brereton, T., & Roy, D. B. (2012). Habitat associations of thermophilous butterflies are reduced despite climatic warming. *Global Change Biology*, *18*(9), 2720–2729. doi: 10.1111/j.1365-2486.2012.02737.x

Oliver, T., Hill, J. K., Thomas, C. D., Brereton, T., & Roy, D. B. (2009). Changes in habitat specificity of species at their climatic range boundaries. *Ecology Letters*, *12*(10), 1091–1102. doi: 10.1111/j.1461-0248.2009.01367.x

Parmesan, C., Ryrholm, N., Stefanescu, C., Hill, J. K., Thomas, C. D., Descimon, H., … Warren, M. (1999). Poleward shifts in geographical ranges of butterfly species associated with regional warming. *Nature*, *399*(6736), 579. doi: 10.1038/21181

Pivnick, K. A., & McNeil, J. N. (1986). Sexual Differences in the Thermoregulation of Thymelicus Lineola Adults (Lepidoptera: Hesperiidae). *Ecology*, *67*(4), 1024–1035. doi: 10.2307/1939825

Pörtner, H. O., & Knust, R. (2007). Climate Change Affects Marine Fishes Through the Oxygen Limitation of Thermal Tolerance. *Science*, *315*(5808), 95–97. doi: 10.1126/science.1135471

R Core Team. (2019). R: A language and environment for statistical computing. (Version 3.5.3). Retrieved from http://www.Rproject. org/

Rebaudo, F., & Rabhi, V.-B. (2018). Modeling temperature-dependent development rate and phenology in insects: review of major developments, challenges, and future directions. *Entomologia Experimentalis et Applicata*, *166*(8), 607–617. doi: 10.1111/eea.12693

Rutowski, R. L., Demlong, M. J., & Leffingwell, T. (1994). Behavioural thermoregulation at mate encounter sites by male butterflies (Asterocampa leilia, Nymphalidae). *Animal Behaviour*, *48*(4), 833–841. doi: 10.1006/anbe.1994.1307

Shanks, K., Senthilarasu, S., ffrench-Constant, R. H., & Mallick, T. K. (2015). White butterflies as solar photovoltaic concentrators. *Scientific Reports*, *5*(1), 12267. doi: 10.1038/srep12267

Smit, B., & McKechnie, A. E. (2015). Water and energy fluxes during summer in an arid-zone passerine bird. *Ibis*, *157*(4), 774–786. doi: 10.1111/ibi.12284

Sparks, T. H., & Yates, T. J. (1997). The effect of spring temperature on the appearance dates of British butterflies 1883–1993. *Ecography*, *20*(4), 368–374. doi: 10.1111/j.1600-0587.1997.tb00381.x

Suggitt, A. J., Gillingham, P. K., Hill, J. K., Huntley, B., Kunin, W. E., Roy, D. B., & Thomas, C. D. (2011). Habitat microclimates drive fine-scale variation in extreme temperatures. *Oikos*, *120*(1), 1–8. doi: 10.1111/j.1600-0706.2010.18270.x

Suggitt, A. J., Stefanescu, C., Páramo, F., Oliver, T., Anderson, B. J., Hill, J. K., … Thomas, C. D. (2012). Habitat associations of species show consistent but weak responses to climate. *Biology Letters*, *8*(4), 590–593. doi: 10.1098/rsbl.2012.0112

Suggitt, A. J., Wilson, R. J., August, T. A., Fox, R., Isaac, N. J. B., Macgregor, N. A., … Maclean, I. M. D. (2015). Microclimate affects landscape level persistence in the British Lepidoptera. *Journal of Insect Conservation*, *19*(2), 237–253. doi: 10.1007/s10841-014-9749-y

Suggitt, A. J., Wilson, R. J., Isaac, N. J. B., Beale, C. M., Auffret, A. G., August, T., … Maclean, I. M. D. (2018). Extinction risk from climate change is reduced by microclimatic buffering. *Nature Climate Change*, *8*(8), 713–717. doi: 10.1038/s41558-018-0231-9

Thackeray, S. J., Sparks, T. H., Frederiksen, M., Burthe, S., Bacon, P. J., Bell, J. R., … Wanless, S. (2010). Trophic level asynchrony in rates of phenological change for marine, freshwater and terrestrial environments. *Global Change Biology*, *16*(12), 3304–3313. doi: 10.1111/j.1365-2486.2010.02165.x

Thomas, J. A., & Simcox, D. J. (2005). Contrasting management requirements of *Maculinea arion* across latitudinal and altitudinal climatic gradients in west Europe. In J. Settele, E. Kuehn, & J. A. Thomas (Eds.), *Studies on the ecology and conservation of butterflies in Europe* (pp. 240–244). Sofia: Pensoft Publishers.

Thomas, Jeremy A., & Lewington, R. (2016). *The Butterflies of Britain and Ireland*. London: Bloomsbury.

Turlure, C., Radchuk, V., Baguette, M., Van Dyck, H., & Schtickzelle, N. (2011). On the significance of structural vegetation elements for caterpillar thermoregulation in two peat bog butterflies: Boloria eunomia and B. aquilonaris. *Journal of Thermal Biology*, *36*(3), 173–180. doi: 10.1016/j.jtherbio.2011.02.001

Turner, E. C., Granroth, H. M. V., Johnson, H. R., Lucas, C. B. H., Thompson, A. M., Froy, H., … Holdgate, R. (2009). Habitat preference and dispersal of the Duke of Burgundy butterfly (Hamearis lucina) on an abandoned chalk quarry in Bedfordshire, UK. *Journal of Insect Conservation*, *13*(5), 475–486. doi: 10.1007/s10841-008-9194-x

Warren, M. S., Hill, J. K., Thomas, J. A., Asher, J., Fox, R., Huntley, B., … Thomas, C. D. (2001). Rapid responses of British butterflies to opposing forces of climate and habitat change. *Nature*, *414*(6859), 65–69. doi: 10.1038/35102054

Wasserthal, L. T. (1975). The role of butterfly wings in regulation of body temperature. *Journal of Insect Physiology*, *21*(12), 1921–1930. doi: 10.1016/0022-1910(75)90224-3

Wilson, R. J., & Maclean, I. M. D. (2011). Recent evidence for the climate change threat to Lepidoptera and other insects. *Journal of Insect Conservation*, *15*(1), 259–268. doi: 10.1007/s10841-010-9342-y

Figures

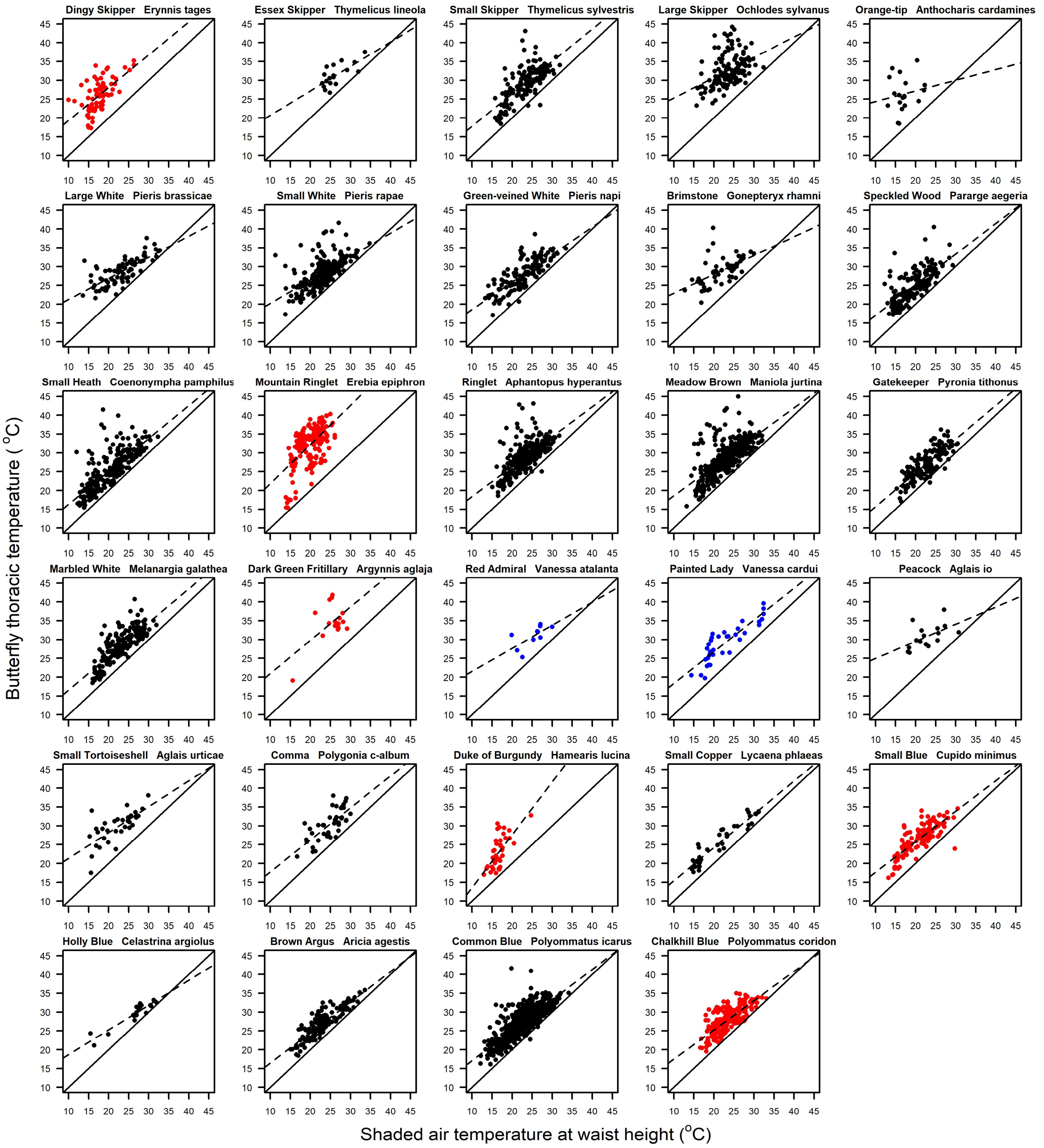


Fig. 1: The response of thoracic temperature to changes in air temperature in 29 butterfly species. Points show data from individual butterflies, coloured according to their habitat associations as listed by Butterfly Conservation (black = habitat generalist; dark grey/red = habitat specialist; pale grey/blue = migrant species). The dashed line on each figure represents the linear relationship of thoracic temperature against air temperature. The solid lines represent a simple 1:1 relationship between air

and butterfly temperature (the same for all panels), and are provided to aid interpretation of the relative gradients of the fitted relationship.

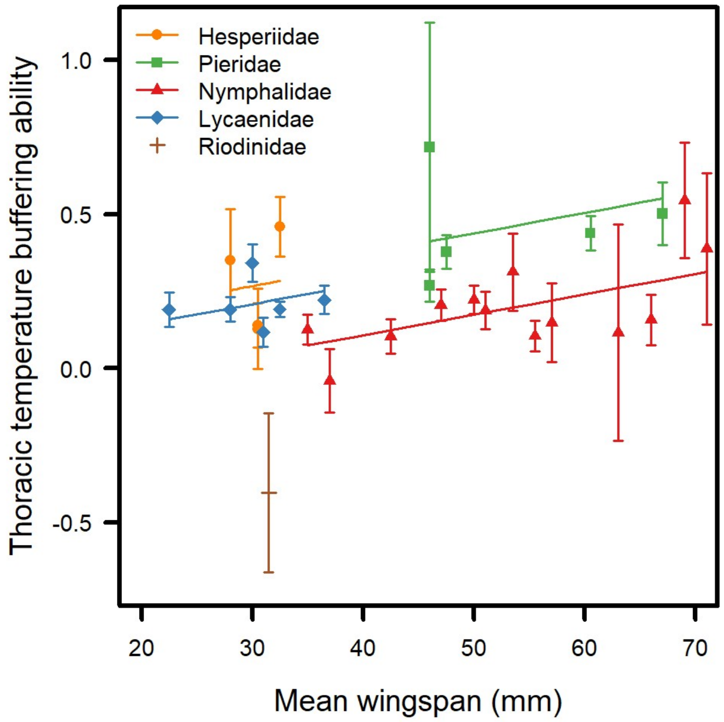
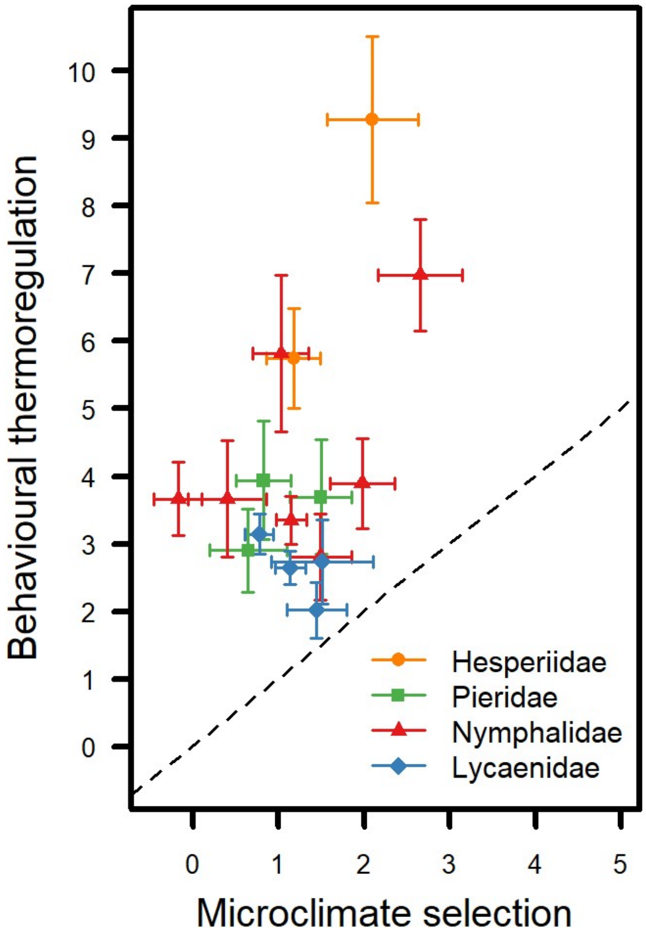


Fig. 2: Traits which influence species-specific thoracic temperature buffering ability (gradient of the regression line from Fig. 1, subtracted from one) for each of 29 butterfly species. Buffering ability differed between taxonomic families, and larger species were better at controlling their thoracic temperature than their smaller relatives. Points represent individual species ± one standard error for the estimate of the slope from Fig. 1; lines represent the predicted relationship for each Family. No line is presented for Riodinidae, as this is a monospecific Family in the UK, containing only the Duke of Burgundy (*Hamearis lucina*).



<u>Fig. 3:</u> The ability of 16 butterfly species to alter their thoracic temperature by using either microclimate selection or behavioural thermoregulation (such as altering the angle of their wings relative to the sun). "Microclimate selection" was calculated by subtracting the waist-height, shaded air temperature from the temperature one centimetre above the butterfly's chosen perch. "Behavioural thermoregulation" was calculated by subtracting temperature one centimetre above the butterfly's chosen perch from the butterfly's thoracic temperature. Points represent means for individual species ± one standard error. Data for individuals of each species are shown in Fig. S2.

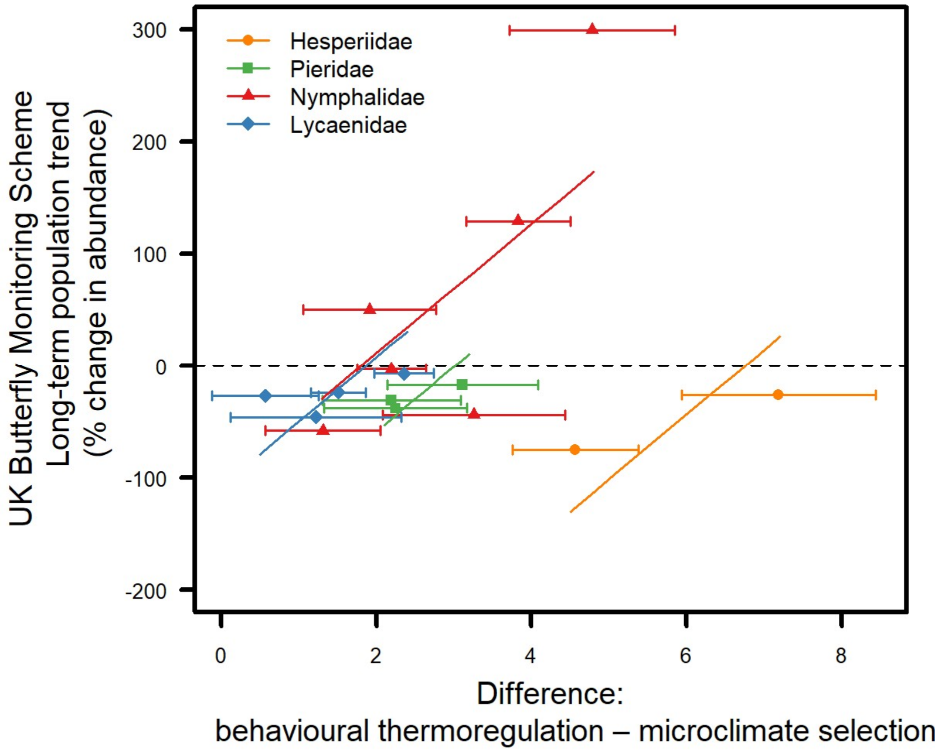


Fig. 4: Correlation between species' published long-term UK population trend (taken from the UK Butterfly Monitoring Scheme, (Brereton et al., 2018)) and the difference between each species' mean behavioural thermoregulation and mean microsite selection (higher values represent a greater ability to use behavioural mechanisms to buffer thoracic temperature) (Fig. 3, Fig. S2). Points show data for individual species ± one standard error for the mean difference between thermoregulatory strategies (standard errors for species' change in abundance are not published). Symbols and colours used represent species' taxonomic family. Lines represent fitted relationships for individual taxonomic families based upon the selected model in which no interaction term was retained.