

RESEARCH ARTICLE

Physiological and morphological traits affect contemporary range expansion and implications for species distribution modelling in an amphibian species

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Funding information

NSF Grant, Grant/Award Number: DMS-2325196

Handling Editor: Lucie Kuczynski

Abstract

1. Species range shifts due to climate alterations have been increasingly well-documented. Although amphibians are one of the most sensitive groups of animals to environmental perturbations due to climate change, almost no studies have offered evidence of poleward distribution shifts in this taxon in response to climate warming. Range shifts would be facilitated by variation in traits associated with the ability of species to persist and/or shift their range in the face of climate change, but the extent and consequences of intraspecific variation in these traits is unclear.
2. We studied the role of intraspecific variation in the ongoing range shift of green treefrogs (*Hyla cinerea*) in response to climate change. We explored factors that are often associated with range shifts to test the hypothesis that there are differences in these traits between recently range-expanded and nearby historical populations. We then tested the consequences of intraspecific variation for modelling climate-induced range shifts by comparing species distribution models (SDMs) that used as input either data from the entire species range or separate inputs from 'subpopulations' corresponding to the historical range or the recently expanded range. We expected that building a separate SDM for each population would more accurately characterize the species range if historical and expanded populations differed in traits related to their response to climate.
3. We found that critical thermal minimum decreased and thermal breadth increased with latitude, but the effect of latitude was significantly stronger for expanded populations compared to historical populations. Additionally, we found that individuals from expanded populations had longer leg lengths when compared to their historical counterparts. Finally, we found higher model accuracy for one of the population-level SDMs than the species-level SDM.
4. Our results suggest that thermal tolerance and dispersal morphologies are associated with amphibian distributional shifts as these characteristics appear to facilitate rapid range expansion of a native anuran. Additionally, our modelling results emphasize that SDM accuracy could be improved by dividing a species range to

consider potential differences in traits associated with climate responses. Future research should identify the mechanisms underlying intraspecific variation along climate gradients to continue improving SDM prediction of range shifts under climate change.

KEYWORDS

climate change, dispersal, green treefrogs, habitat suitability, *Hyla cinerea*, Hylidae, range shift, thermal tolerance

1 | INTRODUCTION

Species' geographical distributions can shift in response to various global factors, including tectonic (Trewick, 2017), oceanographic (Pearcy & Schoener, 1987) and climatic (Cronin & Schneider, 1990) events. Anthropogenic-induced warming temperatures have resulted in especially rapid changes in species' distributions (Chen et al., 2011; Parmesan & Yohe, 2003), and the uncertainty surrounding the patterns of species and population-level distributional responses to global rises in temperature have become a prominent challenge and major topic in the field of ecology (Brodie et al., 2022). One major response to rising temperatures is that many species shift their distributions toward higher latitudes or elevations (Osland & Feher, 2020; Root et al., 2003; Thomas, 2010) to maintain stability in the temperatures they are exposed to. These distributional shifts have been detected in a multitude of taxonomic groups, including plants (Chen et al., 2020; Lenoir et al., 2008), invertebrates (Richardson, 2008; Yamano et al., 2011), and vertebrates (Virkkala & Lehikoinen, 2014; Zuckerberg et al., 2009). However, although amphibians are highly sensitive to climate change (Collins & Storfer, 2003; Stuart et al., 2004), previous studies have failed to detect evidence of climate-driven poleward shifts in amphibians (Enriquez-Urzelai et al., 2019; Hickling et al., 2006).

Distributional responses to climate change depend on multiple factors that often vary intraspecifically, including dispersal ability (Gaston & Blackburn, 2002), habitat availability (Mair et al., 2014), population abundance (Newton, 1997), species interactions (Svenning et al., 2014) and thermal tolerance (Sunday et al., 2012). As a species expands its range, individuals on the leading edge of the expansion are prone to experience novel environmental conditions, unique spatial processes, and different selective pressures compared to conspecifics that are not range expanding (Chuang & Peterson, 2016). Consequently, edge populations tend to have distinctive morphological (Phillips et al., 2006), behavioural (Liebl & Martin, 2014), and physiological (Kolbe et al., 2014) characteristics. Although it is possible that observed phenotypic differences between expanded and core populations can be caused by phenotypic plasticity (Baxter-Gilbert et al., 2021; Ghalambor et al., 2007), in many instances there has been an evolutionary change in phenotypes, as suggested by theoretical models (Dytham, 2009; Travis & Dytham, 2002) and common garden experiments (Bridle et al., 2014;

Simmons & Thomas, 2004). Regardless of the cause, trait variation in expanding populations likely affects the speed and extent of range shifts under climate change.

Thermal tolerance, commonly quantified as the critical thermal maximum (CT_{max}) and minimum (CT_{min}) (Cowles & Bogert, 1944; Huey & Stevenson, 1979), is an especially important factor in range expansions for ectothermic taxa because their behavioural and physiological capabilities are directly affected by environmental temperature (Huey et al., 2012). Hence, thermal tolerance plays a critical role in establishing a species' fundamental niche (Hutchinson, 1957), which ultimately dictates geographic distributions (Angilletta, 2009; Somero, 2010). The majority of climate impact research on ectothermic thermal tolerance has focused on CT_{max} (Baudier et al., 2015; Herrando-Pérez et al., 2019; Nowakowski et al., 2017), rather than CT_{min} (but see Kolbe et al., 2010). This emphasis on CT_{max} has been useful for studies that focus on tropical ectotherms, which are often more restricted by their upper thermal limit (Brusch et al., 2016; Huey et al., 2012; Madeira et al., 2012). However, for temperate species with broad geographic distributions in which seasonal variations in temperature are expected to be relatively extreme, especially toward the poles (Sunday et al., 2011), cold tolerance may be more important. Moreover, the climatic variability hypothesis (CVH) states that thermal tolerance breadth ($CT_{max} - CT_{min}$) should be higher in environments that experience more fluctuating temperatures (Stevens, 1989). Accordingly, ectotherms undergoing natural poleward range expansions are hypothesized to exhibit changes in thermal tolerance between populations at the range margin and those from core areas (Carbonell et al., 2021; Lancaster et al., 2015). In fact, patterns of variation in ectotherm thermal tolerance depend on where they are expanding (Lancaster, 2016), and CT_{min} has been found to change more than CT_{max} for ectothermic populations expanding to higher latitudes (Lancaster, 2016; Lancaster et al., 2015). Thus, it is important to characterize both CT_{min} and CT_{max} when addressing how thermal tolerance limits may facilitate or limit the range expansion of ectothermic species, especially those that are broadly distributed. However, besides the relatively few studies on invertebrate models (Carbonell et al., 2021; Lancaster, 2016; Lancaster et al., 2015), this hypothesis has not been rigorously tested.

In addition to thermal tolerance, climate-induced range shifts appear to be strongly correlated with dispersal because dispersal phenotypes often accumulate at the range edge (Phillips et al., 2010; Williams & Blois, 2018). For example, rapid changes in

dispersal-related morphological characteristics (e.g. wing size and shape) have been documented during the range expansions of insects (Simmons & Thomas, 2004; Taylor-Cox et al., 2020) and birds (Berthouly-Salazar et al., 2012; Gunnarsson et al., 2012). Within the context of the westward expansion of invasive cane toads (*Rhinella marina*) in Australia, toads at the invasion front exhibit distinctive morphologies (e.g. longer legs) when compared to conspecifics from the range core and these differences have been linked to locomotor performance, thus facilitating rapid dispersal (Hudson et al., 2020; Phillips et al., 2006). However, studies on amphibian dispersal syndromes and range expansions have primarily been conducted on invasive species (Gruber et al., 2017; Perkins et al., 2013; Phillips et al., 2006). Thus, there is still a lack of information on the relationships between morphological traits and range shifts in response to climate change for native amphibians. These knowledge gaps were partially addressed by a recent study showing that green treefrogs (*Hyla cinerea*) from expanded populations had significantly longer femur lengths than individuals from their historical range (Edwards et al., 2023). This study only focused on a single morphological trait, but locomotion is complex and measuring additional traits may give new insights.

The fact that range expanding populations often differ from core populations in several classes of phenotypic traits, including those related to climate tolerance, raises the important point that the effects of climate on distribution may vary across the species' range. However, large-scale analyses of climate effects on organisms rarely explicitly incorporate intraspecific variation in these effects, leading to uncertainty in understanding and projecting distributional shifts under climate change (Bestion et al., 2015; Valladares et al., 2014). For example, although there are global datasets on climate-related traits such as thermal tolerance that cover a broad range of taxa (Bennett et al., 2018), for most species there are data from only a single population. Furthermore, traditional species distribution models (SDM), which are widely used to inform conservation and management activities (Mainali et al., 2015; Zurell et al., 2016), assume all populations of a species respond identically to climate change (Booth et al., 2013; Wiens et al., 2009) because they are built as a single model that includes all data from the species regardless of any variation among populations. Assuming that the climatic niche does not vary across populations (i.e. niche conservatism) of a species is likely to be inappropriate for most species given local adaptation to climate conditions (DeMarche et al., 2019; Hällfors et al., 2016). To address this issue, recent studies have clustered occurrence data based on variation in habitat conditions and/or genetic features, and then built a separate SDM for each cluster (Biaou et al., 2023; Chardon et al., 2020). Thus, if intraspecific variation is detected in traits that are likely to affect the response to climate change, then SDMs built at the species level are likely to be inaccurate and subdividing the species range into groups that are differently affected by climate, and building separate models from these subdivisions, may be more effective.

We studied variation in thermal tolerance and dispersal ability in the green treefrog (*H. cinerea*) along a climate gradient, including

populations in both their historical and recently expanded range. We then compared the model accuracy of a species-level SDM to SDMs that split the range into smaller population clusters (historical and expanded range) to test if accounting for potential differences among populations in the climatic factors that affect distributions improves models of *H. cinerea* distribution. We tested three hypotheses. (1) There is a latitudinal gradient in thermal tolerance among populations of *H. cinerea* sampled from historical and expanded range locations, and thermal breadth will be greater in the expanded groups than the historical groups. (2) Expanded range populations of *H. cinerea* will have larger dispersal-related morphological characters compared to historical populations. (3) Separate SDMs built from subdividing the species range into historical and expanded populations will outperform the species-level model in terms of model accuracy.

2 | MATERIALS AND METHODS

2.1 | Study species

Green treefrogs (*H. cinerea*) are moderately large, elongate anurans (up to 57 mm body length) that are commonly found in wooded areas surrounding swamps, ponds, lakes, and streams across the central and southeastern United States (Powell et al., 2016). *H. cinerea* is a broad-ranging hylid, with its historical distribution occurring along the Atlantic and Gulf Coastal Plains, as well as a portion of the Interior Low Plateaus (Figure 1b; Dodd, 2023). *H. cinerea* was used as the model species in our study for several reasons. First, *H. cinerea* is a broadly distributed ectotherm that relies on the external environment to maintain body temperatures for stability of physiological functions, making it an ideal species to quantify intraspecific variation in thermal tolerance capacities (CT_{max} and CT_{min}) across its range. Second, populations of *H. cinerea* diverge in dispersal phenotypes, with populations from their expansion range having longer femur lengths compared to counterparts in the historical range (Edwards et al., 2023). Third, this species has undergone a rapid, long distance range expansion, moving northward and eastward into the midwestern United States (Lodato et al., 2014; Redmer et al., 1999; Tucker et al., 2008). According to Redmer et al. (1999), *H. cinerea* started range expanding northward and eastward within the state of Illinois as early as the mid-1980s, with initial explanations of their expansion based around the introduction of tadpoles within fisheries, the increased construction of novel aquatic habitats associated with impoundment reservoirs and farm ponds, and the ability of *H. cinerea* to colonize novel areas due to dispersal. In Illinois, *H. cinerea* has now expanded and travelled northward up to 110km (Tucker et al., 2008) from its historical, northernmost geographic record. In Indiana, *H. cinerea* first appeared as a state record in 2003, and since then has colonized eight counties adjacent to the Ohio river (Engbrecht et al., 2018; Lodato et al., 2014; Mirtl et al., 2020). In Kentucky, *H. cinerea* has also rapidly expanded northward and eastward with at least 22 county-level records since 1985 (J. MacGregor,

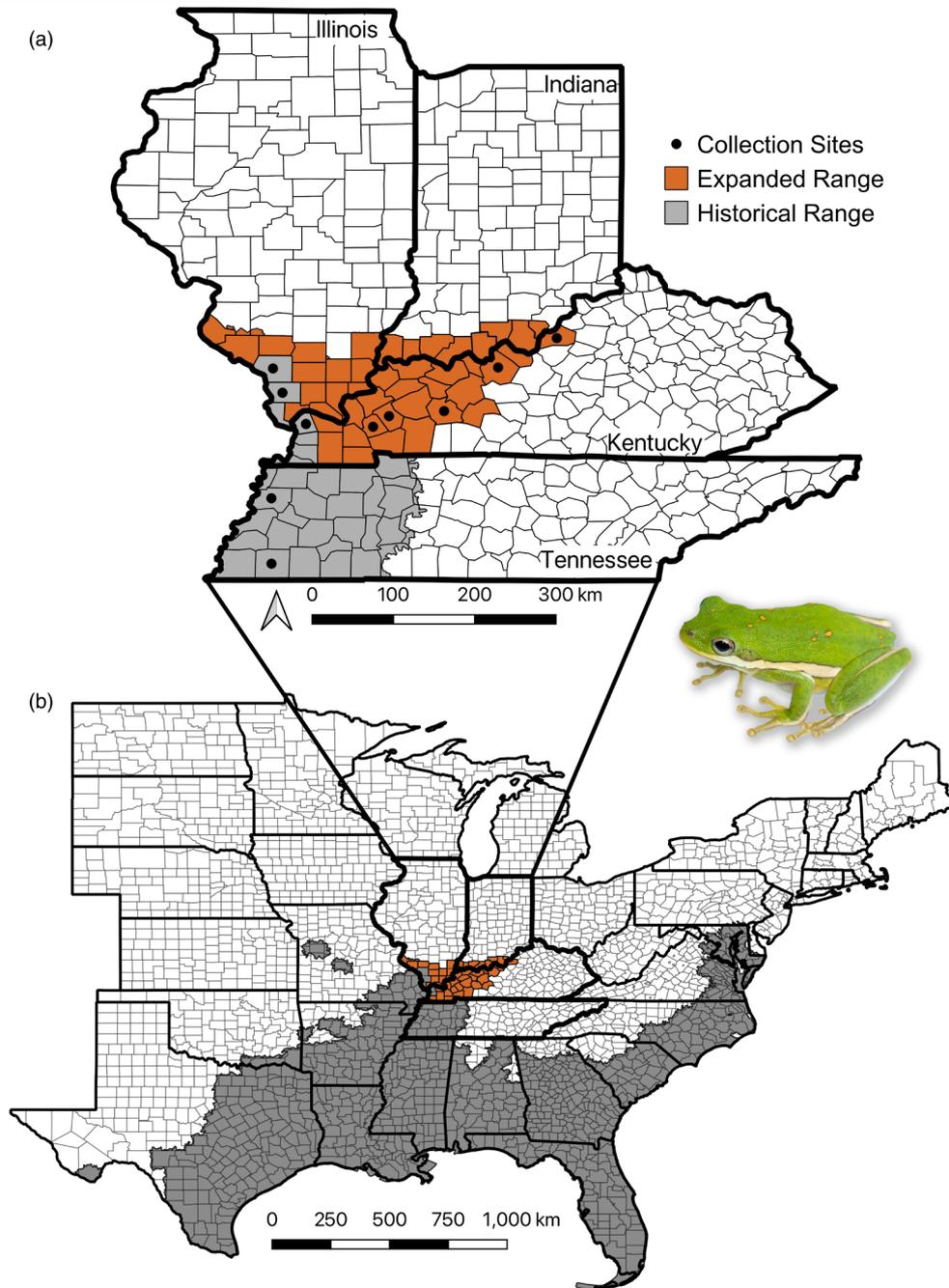


FIGURE 1 Partial map of the central and eastern United States displaying the green treefrog (*Hyla cinerea*) range expansion. (a) Black circles indicate the 10 frog populations sampled from the states of Tennessee ($N=2$), Kentucky ($N=6$), and Illinois ($N=2$). (b) Geographic distribution of historical-range frogs based on United States Geological Survey (grey area; USGS, 2018), and range expanded frogs based on historical and geographical distribution records (orange area; Table 1). Photo of *H. cinerea* taken by Owen M. Edwards.

personal communication; Kentucky Department of Fish & Wildlife Resources, 2024).

2.2 | Field sites

Frogs were collected from 10 sites (five expanded range and five historical range) in the states of Tennessee, Illinois and Kentucky (Table 1; Figure 1a). Expanded and historical localities for *H. cinerea*

were determined based on geographical and historical distribution records of these frogs (Garman, 1895; Kentucky Department of Fish & Wildlife Resources, 2024; Lodato et al., 2014; Redmer et al., 1999). Expanded-range localities in this study represent populations that have colonized new counties after 1985, while historical-range localities represent populations from either their original range or any populations that had colonized counties prior to 1985. A total of 100 individual *H. cinerea* from 10 counties and three states were sampled for this study.

TABLE 1 Localities of the collection sites for expansion and historical range populations of green treefrogs (*Hyla cinerea*), data for GPS coordinates is WGS84.

Colonization stage	Colonization year	Reference	Collection site	Latitude	Longitude	n	Collection date
Expansion	2014	KFWIS	Hopson Lake, Caldwell Co., Kentucky	36.988°N	87.845°W	10	June 2022
Expansion	2006	KFWIS	Cave Pond, Lyon Co., Kentucky	37.076°N	88.099°W	10	June 2022
Expansion	2014	KFWIS	Mill Creek, Jefferson Co., Kentucky	38.152°N	85.878°W	10	June 2022
Expansion	2003	KFWIS	Yellow Bank WMA, Breckinridge Co., Kentucky	38.031°N	86.511°W	10	July 2022
Expansion	2007	KFWIS	Peabody WMA, Muhlenberg Co., Kentucky	37.252°N	87.030°W	10	July 2022
Historical	1890	Garman (1895)	Upper Bluff Lake, Union Co., Illinois	37.392°N	89.348°W	10	June 2022
Historical	1980	Redmer et al. (1999)	Oakwood Bottoms, Jackson Co., Illinois	37.672°N	89.457°W	10	June 2022
Historical	1963	KFWIS	Ballard WMA, Ballard Co., Kentucky	37.172°N	89.026°W	10	June 2022
Historical	Original range	Powell et al. (2016)	Tigrett WMA, Dyer Co., Tennessee	35.994°N	89.265°W	10	July 2022
Historical	Original range	Powell et al. (2016)	Wolf River WMA, Fayette Co., Tennessee	35.028°N	89.271°W	10	July 2022

Abbreviations: KFWIS, Kentucky Fish and Wildlife Information System; WMA, Wildlife Management Area.

2.3 | Data collection

For all localities, 10 individual *H. cinerea* were collected by hand capture from active breeding choruses during the months of June and July 2022. Only adult males were collected because they are easier to detect, and their removal presumably is less impactful on the population relative to the removal of females because of the extremely skewed operational sex ratio (Morris, 1989). Males were identified based on either active calling upon collection, and/or the presence of an enlarged subgular vocal pouch. Prior to hand-capture, individual frog body temperatures (T_b) were measured with an ETEKCITY hand-held infrared thermometer (model Lasergrasp 774) held ~5 cm away aiming at the lower dorsal area near the thigh (Rowley & Alford, 2007). Collected frogs were transported to the University of Evansville within 6 h of capture and were housed in plastic perforated containers (10.5 cm width × 10.5 cm length × 6.5 cm height) for 24 h of acclimation at ~22–24°C before and between thermal assays (CT_{min} and CT_{max}). Frogs were weighed to the nearest 0.1 g on a digital scale and measured (snout-vent length [SVL]) to the nearest 0.01 mm using Vernier callipers prior to thermal assays. All protocols in this study were approved by the Oklahoma State University Institutional Animal Care and Use Committee, IACUC #22-32.

2.4 | Thermal tolerance measurements

Individual *H. cinerea* were placed in a 473 mL perforated plastic vessel (9.5 cm width × 9.5 cm length × 11.5 cm height) and were then

moved to a plastic bath (35.0 cm width × 42.0 cm length × 17 cm height) filled with water at a depth of ~1.5 cm (Figure S1b). A plastic top was placed onto the vessel containing the frog to create a water chamber with an air pocket to ensure the submersion of each frog and to prevent frogs from escaping their individual vessels (Figure S1a). CT_{max} trials were implemented first, in which the water bath temperature was gradually increased in steps of 0.5°C every 3 min by adding ~20–40 mL of dechlorinated water that was heated to 100°C. After 24 h, CT_{min} trials were then conducted in which the water bath temperature was gradually decreased by adding water that was chilled to ~1.0–1.5°C with ice. Because water additions caused gradual increases in the water bath level during CT_{max} and CT_{min} trials, small amounts of water were subsequently and gradually removed to prevent frogs from drowning. Temperature was measured using both a glass 76 mm Sargent-Welch thermometer (model 80005-B) as well as a handheld infrared thermometer. Both CT_{max} and CT_{min} were recorded at the exact temperature when individual frogs lost their righting reflex, defined as an individual's ability to right itself after being placed on its back (Navas et al., 2007). After each 0.5°C increase or decrease, we quickly removed frogs from their water chamber to flip them on their backs three times. Under normal conditions, anurans right themselves immediately after being forced to an upside-down position, thus we recorded CT_{max} and CT_{min} at the water temperature at which frogs failed to right themselves within a 15 s time period (Brusch et al., 2016; Lutterschmidt & Hutchison, 1997). Upon completion of recordings, all individuals were immediately placed in a recovery container of room temperature water. Prior to CT trials, control trials were implemented to confirm that the loss of righting reflex was specifically

a result of temperature changes rather than stress or habituation from the frogs being handled (Brusch et al., 2016). Control trials were performed by placing five randomly selected individuals on their backs repeatedly at room temperature (22°C) for 30 min. None of the individuals exhibited loss of righting reflex from the control trials.

2.5 | Morphometric measurements

Upon completion of thermal tolerance assays, frogs were immediately euthanized following standard protocols accepted by the American Society of Ichthyologists and Herpetologists Guidelines for Live Amphibians and Reptiles in Field and Laboratory Research. Individuals were then formalin fixed and preserved in 70% ethanol for morphological data collection. We obtained 10 readable radiographs of 100 frogs ($n=10$ per plate) using a DRJEM radiography system at Oklahoma State University with the settings of 84 μ A, 60 kV, and 1.35 mAs and with a ruler for scale on the detector plate. Radiographs were then converted to digital images using Asteris Omni version 108.0.0, and five morphological characters were measured on digitally stored images to the nearest 0.01 mm using a distance measuring tool on a Picture Archiving and Communications System (PACS) from the frog specimen radiographs (Figure S2): SVL, skull width (SW; centre of the quadrate 'U-curve' on the left to the right side), radioulna length (RL; maximum distance from olecranon process to the styloid process of the ulna), humerus length (HL; maximum distance from condylar ridge to head of humerus), femur length (FL; maximum distance from the medial condyle to the femur head), and tibiofibula length (TL; maximum distance from proximal articular facet to distal articular facet) of the right and left side of each individual. The first author measured each morphological character three times and was blind with respect to frog identification numbers, county names and colonization categories (expansion vs. historical). Measurements were obtained from the left and right of the sagittal plane of each individual for RL, HL, FL and TL and both sides were averaged. Voucher specimens of *H. cinerea* for this study have been deposited in the Sam Noble Oklahoma Museum of Natural History at the University of Oklahoma (Catalogue numbers OMNH 49434-49533).

2.6 | Statistical analysis

We used linear mixed models (LMMs) to examine the main and interaction effects of colonization stage (expanded and historical) and latitude on the thermal tolerance parameters: CT_{min} , CT_{max} , and thermal breadth, with population (county) entered as a random effect. The interaction effects were used to determine if the effects of latitude varied by colonization stage. Assumptions of linearity, equal variance, and normality were examined by the residual plots along the predictors, fitted values and the normal quantile plots, respectively. To scale skeletal morphological traits relative to body size, we

divided each trait by SVL to obtain a ratio. After checking for assumptions of normality and homogeneity of variance, we conducted LMMs to compare morphological trait ratios of *H. cinerea* between historical and expanded ranges using colonization stage as a fixed factor and population as a random effect. All the above statistical analyses were conducted in R version 4.3.2 (R Core Team, 2023) and the LMMs were built using the *lme4* package version 1.1-35.2 (Bates et al., 2015). Main effects in our fitted LMMs were all evaluated using Type III Wald *F*-tests with Kenward-Rogers degrees of freedom using the ANOVA function in the *car* package version 3.1-2 (Fox & Weisberg, 2019).

2.7 | Species distribution models

We built SDMs for *H. cinerea* by either building separate 'population-level' models for the historical and expanded range, respectively, or with a single model built at the species-level covering both the historical and expanded range. The population-level model for the historical range was defined as *H. cinerea* occurrence data within the depicted historical range map by the United States Geological Survey (Figure 2; USGS, 2018), while the expanded range population-level model was defined by the overlap between Level IV ecoregions and species occurrence data beyond the historical range of *H. cinerea* (Figure 2). We extracted the spatial presence data of *H. cinerea* from the Global Biodiversity Information Facility (<http://www.gbif.org>). We considered the effects of climate and elevation variables on the species distribution. The climate effects were quantified by 19 bioclimatic variables from the WorldClim database (Fick & Hijmans, 2017), covering different aspects of climate (e.g. temperature, precipitation) at different temporal scales (e.g. season, year). To minimize correlation between the bioclimatic variables, we conducted a principal component analysis to select the variables driving the climate differences between areas with and without the species present (Guisan et al., 2017). Our final model included four bioclimatic variables: minimum temperature of the coldest month, temperature seasonality, annual precipitation, and precipitation seasonality. The elevation data were compiled from the Shuttle Radar Topography Mission (SRTM) elevation data. We compiled the predictor variables at 2.5-minute resolution. We built the SDMs using four algorithms: Maximum entropy (maxent), generalized linear models (GLM), generalized boosting model (GBM) and random forest (RF), to ensure the model results were not biased by the choice of algorithm. We compared accuracy of the two population-level models and the species-level model with True Skills Statistics (TSS), which are widely used in SDM studies to measure model accuracy (Allouche et al., 2006). We also calculated the variable importance of each predictor, which quantifies the relative effect of a predictor on species distribution. We randomly split the original data into two samples: 70% of the species presence data used for training and 30% for validation sets, and the validation sets were used to evaluate the model accuracy. Finally, the suitability of habitats for *H. cinerea* in our study area was projected based on the

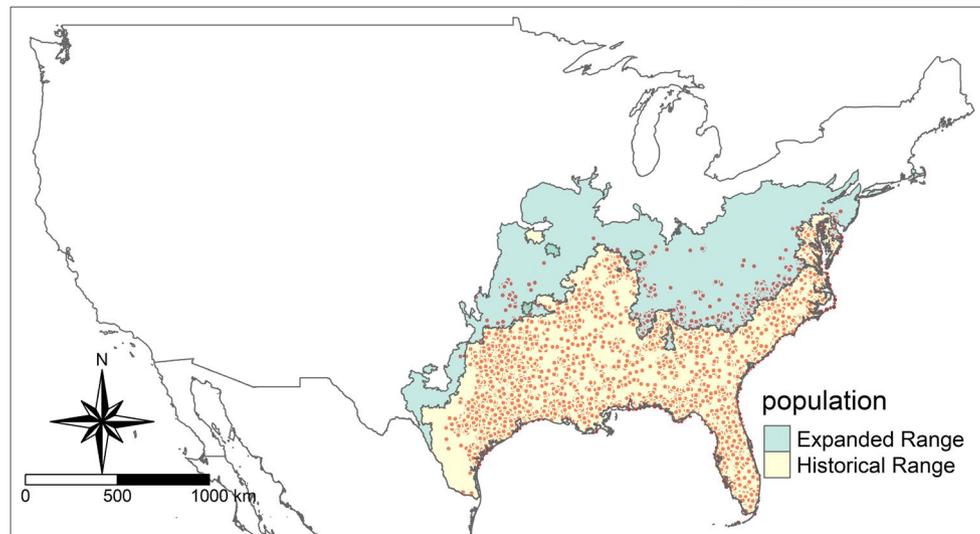


FIGURE 2 Map of the United States depicting the expanded and historical regions defined in this study. Red dots indicate green treefrog (*Hyla cinerea*) occurrences.

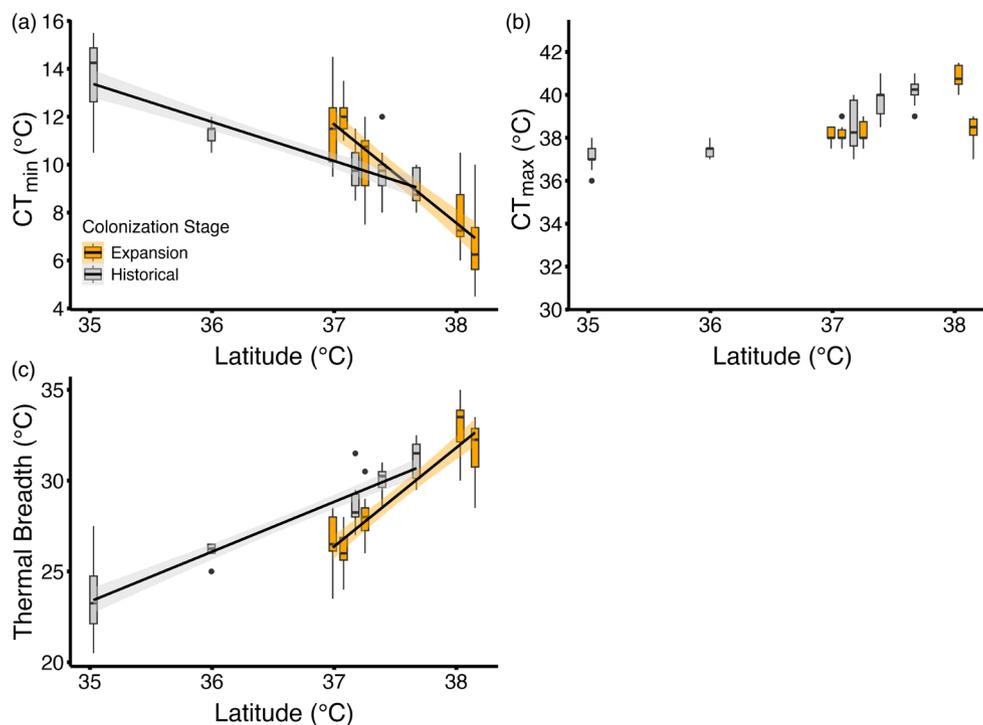


FIGURE 3 Critical thermal minimum (a), critical thermal maximum (b), and thermal breadth (c) with increasing latitude. Orange lines of best fit represent expanded-range populations of male green treefrogs (*Hyla cinerea*), while grey lines represent historical-range populations.

current climate conditions used to train the models. We conducted linear mixed effect models to analyse the effects of model type (two population vs. one species models) on model accuracy and variable importance of minimum temperature of the coldest month. The models included the random effects of the four algorithms and data splitting on model accuracy and variable importance. Assumptions of equal variance and normality were examined by the residual plots along the fitted values and the normal quantile plot, respectively. We conducted the above statistical analyses using the R package *nlme* version 3.1-164 (Pinheiro et al., 2023).

3 | RESULTS

3.1 | Differences in thermal tolerance across latitudes between historical and expanded populations

Latitude had a significantly negative effect on CT_{min} (Figure 3a; Table 2; $F_{1,6}=105.15$, $p<0.001$), and the decrease in CT_{min} with latitude exhibited a significant interaction effect with colonization stage, being stronger for expanded-range populations than historical

TABLE 2 Results of linear mixed models (LMMs) testing for the effects of latitude and colonization stage on the response variables.

Thermal measure	Fixed effects	Estimate (SE)	F-value	p-Value
CT _{min}	Latitude	-4.12 (0.40)	105.15	<0.001
	Colonization stage (historical)	-93.45 (16.73)	31.21	0.001
	Latitude: Colonization stage (historical)	2.48 (0.45)	30.76	0.001
CT _{max}	Latitude	1.32 (0.80)	2.73	0.15
	Colonization stage	8.55 (33.31)	0.07	0.806
	Latitude: Colonization stage	-0.21 (0.89)	0.05	0.825
Breadth	Latitude	5.44 (0.68)	64.39	<0.001
	Colonization stage (historical)	102.00 (28.23)	13.05	0.011
	Latitude: Colonization stage (historical)	-2.69 (0.76)	12.66	0.012

Note: The reference category for colonization stage was the expanded group. Boldface text indicates significance at the level of $p < 0.05$.

ones (Figure 3a; Table 2; $F_{1,6} = 30.76$, $p = 0.001$). Latitude had a significantly positive effect on thermal breadth (Figure 3c; Table 2; $F_{1,6} = 64.39$, $p < 0.001$), and the increase in breadth with latitude also exhibited a significant interaction effect with colonization stage, being stronger for expanded-range populations compared to historical ones (Figure 3c; Table 2; $F_{1,6} = 12.66$, $p = 0.012$). However, latitude had no effect on CT_{max} in either expanded or historical populations (Figure 3b; Table 2; $F_{1,6} = 2.73$, $p = 0.15$).

3.2 | Differences in morphological traits between historical and expanded populations

Historical and expanded populations of *H. cinerea* did not differ significantly in mean SVL (Historical = 51.6 mm; Expanded = 50.1 mm; Table 3; $F_{1,8} = 2.60$, $p = 0.145$). However, there were strong and significant differences between expanded and historical range populations for nearly every dispersal related morphology character (Figure 4; Table 3). Compared to historical range populations, expanded-range populations had relatively longer tibiafibula (all morphological traits were expressed as a ratio of SVL) (Figure 4a; Table 3; $F_{1,8} = 17.02$, $p = 0.003$), femur (Figure 4b; Table 3; $F_{1,8} = 15.88$, $p = 0.004$), humerus (Figure 4c; Table 3; $F = 32.09$, $p < 0.001$), and radioulna (Figure 4d; Table 3; $F_{1,8} = 41.08$, $p < 0.001$). However, there was no significant difference in skull width ratios for expanded and historical-range populations (Figure 4e; Table 3; $F_{1,8} = 0.51$, $p = 0.495$).

3.3 | Accuracy comparison in population-level and species-level SDMs

The SDM based on the expanded population showed significantly higher TSS values than the other two SDMs, and the TSS value of the historical population-based SDM was similar to the species-level model (Figure 5a). Between the two population-level models, minimum temperature had the lowest variable importance in the model based on the historical population (Figure 5b). For most study areas, the difference in projected suitability was small between

TABLE 3 Results of linear mixed models (LMMs) testing for the effects of colonization stage on the response variables.

Morphological trait	Estimate (SE)	F-value	p-Value
Snout-vent length	1.57 (0.97)	2.60	0.145
Tibiafibula length	-0.01 (0.004)	17.02	0.003
Femur length	-0.01 (0.004)	15.88	0.004
Humerus length	-0.02 (0.003)	32.09	<0.001
Radioulna length	-0.01 (0.002)	41.08	<0.001
Skull width	0.002 (0.003)	0.51	0.495

Note: The reference category for colonization stage was the expanded group. Boldface text indicates significance at the level of $p < 0.05$.

the population-level SDMs and species-level SDM, within 0.2. However, in some parts of the expanded range (e.g. the blue areas near the boundary between the historical and expanded ranges), the species-level SDM underestimated the suitability compared with the population-based SDMs (Figure 5c).

4 | DISCUSSION

Contemporary climate change has caused geographic distribution shifts of thousands of species (Chen et al., 2011; Parmesan & Yohe, 2003; Root et al., 2003), hence, accurately predicting where shifts in species' ranges will occur and examining traits associated with range expansions are crucial for understanding the long-term impact of climate change on organismal range shifts. Our work highlighted that there are significant differences in both thermal tolerance responses and morphological traits between populations located in historical and nearby expanded ranges. Thus, there can be large differences in trait values even between populations that are in close geographic proximity. As a result, building separate SDMs for subdivisions in a species range that account for these differences between populations in the response to climate in some cases outperformed a model that was built at the species-level.

We found that the SDMs built from the expanded range exhibited greater TSS (a measure of model accuracy) than the species-level

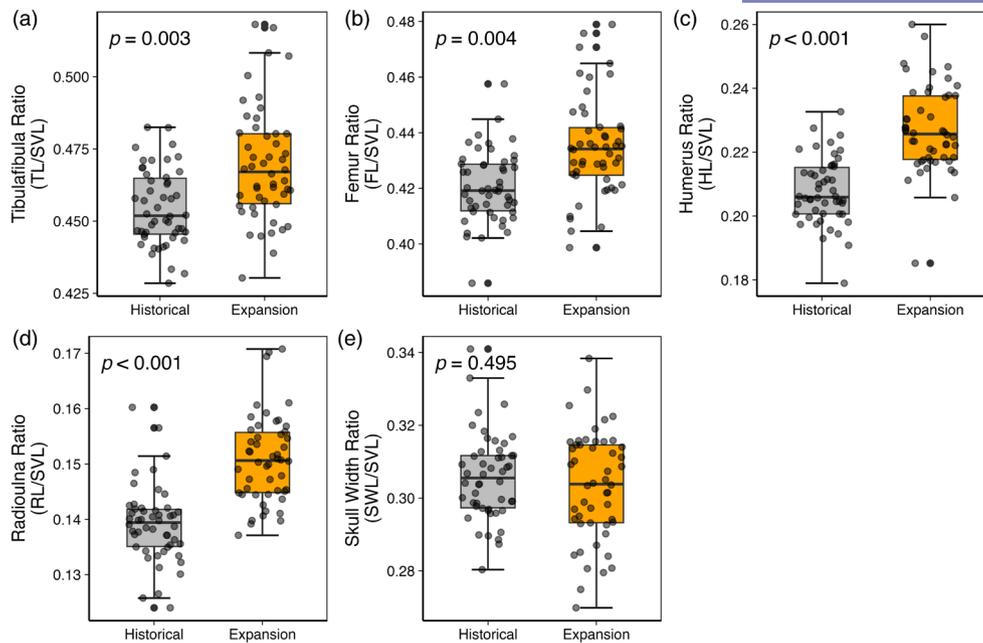


FIGURE 4 A comparison of relative character measurements (a) tibulafibula length, (b) femur length, (c) humerus length, (d) radioulna length, and (e) skull width (all quantified as ratios) of male green treefrogs (*Hyla cinerea*) from historical and expanded-range populations. The boxplots represent the median (line), interquartile range (box), range excluding outliers (whiskers), outliers (black dots), and jitter points of raw data (grey dots).

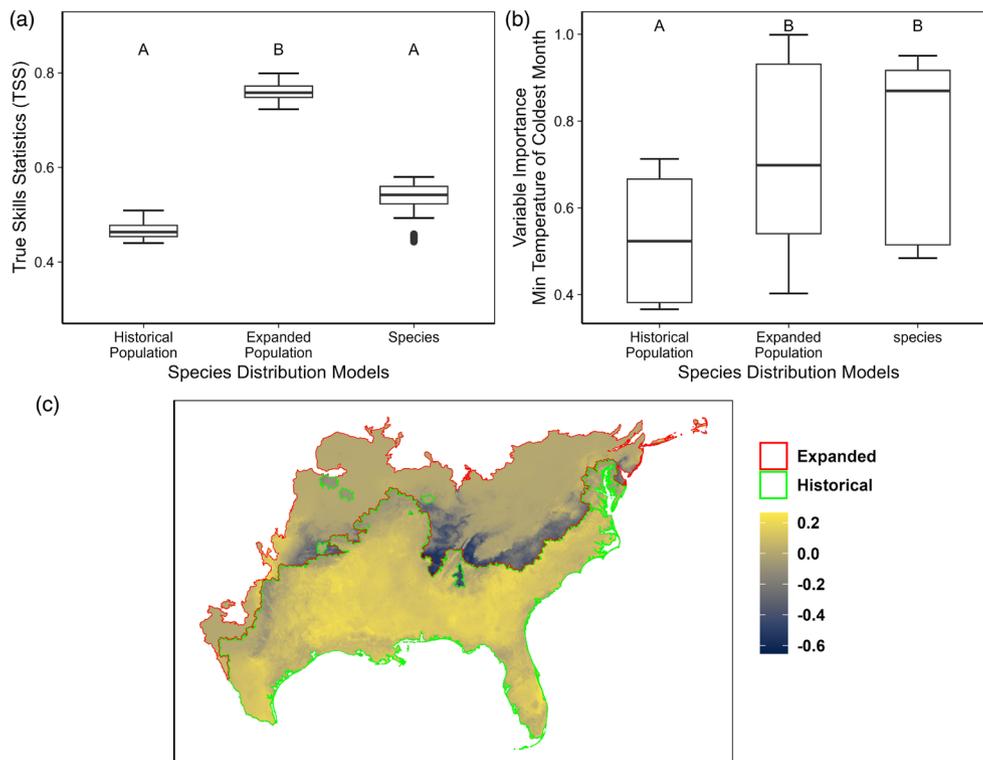


FIGURE 5 (a) The modelling accuracy defined by True Skill Statistic (TSS), (b) variable importance of the minimum temperature for the three types of species distribution model and (c) difference in projected suitability between the species- and population-level models. The positive values of the suitability difference denote that projected suitability is higher in the species- model than the population-level model, and the negative values denote the opposite. Green and red lines represent the boundary of the historical and expanded region, respectively. Note the data points (A, B) included the outputs from the four algorithms with repeated data splitting used to build the species distribution models.

model, suggesting that model accuracy could be generally improved by dividing a species range to consider potential differences in climate responses. Additionally, in some parts of the expanded range for *H. cinerea*, the species-level SDM underestimated the habitat suitability compared with the population-based SDM. Previous studies also found that accounting for intraspecific variation in genetic groups (Ikeda et al., 2017) and climatic conditions (Hällfors et al., 2016) improves the accuracy of SDMs. The extent to which SDMs are improved when subdividing the species range depends on the specific variables used to characterize intraspecific variation. For example, building SDMs based on genetic structure-based variation led to more model improvement in an arctic-alpine cushion plant (*Silene acaulis*) than did building SDMs based on variation in regions or habitats (Chardon et al., 2020). In this study, we built an SDM by subdividing the species range into historical and expanded areas based on our findings of differences between populations in these areas in thermal tolerance and dispersal morphology. Perhaps a more informative approach to modelling the species distribution would be to use a mechanistic or process-based model, which explicitly incorporates essential processes involving eco-evolutionary responses to environmental changes (Bocedi et al., 2021). Because traits like thermal tolerance and dispersal ability are critical variables that affect how organisms shift their range (Chuang & Peterson, 2016), it is possible that these variables would have better captured relevant intraspecific differences that could have been used to identify subdivisions of the species range to be modelled separately. However, we did not use the trait values themselves to parameterize our models due to our sampling only covering a limited part of the range of *H. cinerea*. Therefore, future studies modelling species ranges under climate change should aim to cluster populations by traits associated with range shifts to test if these variables can improve model predictions.

The accuracy of SDMs tends to increase when the background localities extend beyond the area where there are presence records (Anderson, 2012), and this may partially contribute to the relatively high accuracy of the expanded-range SDM because there were no presence records in a large portion of this region. One caveat is that the improvement in model accuracy we found in the expanded-range SDM may be a statistical artefact, if reducing the area covered by the model automatically increases performance. However, there was no improvement in accuracy for the historical range SDM when compared to the species-level SDM, indicating that making the area smaller when using population-level SDMs does not automatically improve model accuracy. These potential confounding factors could be further investigated in future studies to improve the mechanistic understanding of greater SDM accuracy by considering intraspecific variation. Lastly, future modelling studies should consider incorporating intraspecific trait variations to improve prediction of range shifts under climate change. For instance, mechanistic models, such as agent-based models that specify rules for the behaviour of individuals and allow these individuals to interact with each other and their environment (Bocedi et al., 2021; DeAngelis & Diaz, 2019), could be parameterized with the results from this study to better

forecast how changes in thermal tolerance and dispersal during range expansion may influence range expansion rate.

In our thermal tolerance results, we found that CT_{min} decreased while thermal breadth increased with increasing latitude among populations of *H. cinerea* from both historical and expansion range locations. This effect of latitude on thermal tolerance is consistent with other studies conducted on ectotherms (Addo-Bediako et al., 2000; Sunday et al., 2011, 2019), including anurans (Kolbe et al., 2010; Snyder & Weathers, 1975). Unlike CT_{min} , we did not detect any relationship between CT_{max} and latitude, suggesting that CT_{max} is more conserved in the *H. cinerea* populations sampled in this study. This result corresponds with previous studies that showed that over relatively narrow latitude ranges, CT_{min} is more strongly related to latitude than is CT_{max} (Deutsch et al., 2008; Sunday et al., 2011). Together, the increasing thermal breadth with increasing latitude may be indicative of the relative invariance of CT_{max} (Brett, 1956) compared to CT_{min} (Araújo et al., 2013; Bennett et al., 2019). Note that our sampling took place over a relatively narrow range of latitudes (~35–38°; Table 1), which may not have been large enough to detect an effect of latitude on CT_{max} , which typically changes by only ~0.3°C for every 10° of latitude in terrestrial ectotherms (Sunday et al., 2011).

Our SDM analyses showed greater effects of minimum temperature of coldest month on the distribution of *H. cinerea* in the expanded region than the historical, suggesting that *H. cinerea* populations from the cooler region are more sensitive to warming climate than those from the warmer region. The more sensitive response of *H. cinerea* from the cooler region is supported by our thermal tolerance results, which showed greater changes in CT_{min} and thermal breadth of northern populations along latitude relative to the more southern populations. The differences in these thermal measures between expanded and historical-range populations appear to be a consequence of the process of range expansion, as relaxed selection on CT_{max} and increased selection on CT_{min} can facilitate species range shifts (Lancaster et al., 2015; Parmesan, 2006). Populations on the poleward leading range edge encounter greater climatic variation (e.g. lower minimum temperature, higher annual temperature variation, extreme weather, etc.) as they colonize novel areas of their range (Janzen, 1967; Lancaster et al., 2015; Parmesan, 2006). Therefore, it is likely that expanded populations of *H. cinerea* have undergone changes in CT_{min} and thermal breadth, but not CT_{max} , due to the selective pressures of cooler and more variable thermal conditions at higher latitudes. Among insect thermal tolerances across the globe, thermal breadth was only found to increase in species recently or currently undergoing range expansion (Lancaster, 2016). This not only suggests that range expansion is an important driver of population-level shifts in thermal tolerance, but also suggests that frequently described macrophysiological patterns for ectotherms (e.g. CVH, invariance in CT_{max}) may only hold true for range expanding species (Addo-Bediako et al., 2000; Lancaster, 2016), just as observed in *H. cinerea* during its poleward expansion.

Under species range expansions, theoretical models project enhanced dispersal capacity at expanding range margins as a

consequence of spatial sorting in dispersal ability, in which highly dispersive phenotypes accumulate at the expansion front (Ochocki & Miller, 2017; Shine et al., 2011). As predicted, expanded-range populations of *H. cinerea* had significantly longer limb lengths when compared to historical populations (Figure 5). Because anuran limb length is strongly correlated with locomotion ability (Ficetola & De Bernardi, 2006; Nauwelaerts et al., 2007), our morphological results suggest that frogs found in the expanded range have phenotypic characteristics that likely enhance dispersal during the course of their range expansion. These results are consistent with several other studies examining dispersal-related morphological variation among species undergoing rapid range expansions (Berthouly-Salazar et al., 2012; Hudson et al., 2020; Therry et al., 2015). It is important to note, however, that without a direct understanding of how these range differences in limb lengths influence locomotion in *H. cinerea*, it is difficult to infer the functional significance of the changes in morphology observed when populations from historical and expanded ranges are compared. Regardless, these results are consistent with the notion that populations found at the expanding range edge consist of highly dispersive phenotypes (Burton et al., 2010; Phillips et al., 2010), which can have major consequences on the strength of range expansion.

Due to their general classification as poor dispersers (Smith & Green, 2005) and their high sensitivity and vulnerability to both terrestrial and aquatic fluctuations in temperature and precipitation (Duellman & Trueb, 1986), amphibians, unlike most other taxonomic groups, are not expected to track their climatic niche polewards during periods of global warming (Enriquez-Urzelai et al., 2019; Wells, 2007). However, over the last 40 years, since the initial start of their natural range expansion (Redmer et al., 1999), *H. cinerea* has remarkably expanded its range up to ~300 km in a northeast direction based on county boundaries (Figure 1a). Our results suggest that morphological and physiological traits are at some level facilitating a natural range expansion of a native amphibian species, and that these changes in phenotypes have likely occurred in less than half a century. The lower CT_{min} and higher thermal breadth exhibited by frogs in the expanded range may reflect adaptive/plastic change to both their environmental tolerance and spatial opportunities presented by their geographical expansion. Furthermore, the significant differences in dispersal morphology between expanded and historical range frogs are also consistent with rapid phenotypic evolution found in populations at the leading edge of an expansion front (Angert et al., 2020; Simmons & Thomas, 2004). Although the observed shifts in phenotypes in this study do reflect adaptive and/or spatially sorted changes involved in range expansions, the specific mechanisms causing variation in phenotypic expression were not within the scope of this study. Future studies should empirically test whether the intraspecific variation in thermal tolerance and morphology are due to plastic or adaptive processes, which may require genetic investigations or a common garden experimental approach. Such studies will also help elucidate the physiological mechanisms underlying the intraspecific differences in thermal tolerance observed here.

AUTHOR CONTRIBUTIONS

Owen M. Edwards, Michael S. Reichert, Lu Zhai and Bo Zhang conceived the ideas and designed methodology; Owen M. Edwards collected the field data; Owen M. Edwards, Lu Zhai and Bo Zhang analysed the data; Logan Ozment and Lu Zhai performed the SDM simulation and analysis, Owen M. Edwards, Lu Zhai and Bo Zhang led the writing of the manuscript; Owen M. Edwards, Michael S. Reichert, Ciaran A. Shaughnessy, Lu Zhai and Bo Zhang interpreted the results. All authors contributed critically to the result discussion and revisions of the manuscript.

ACKNOWLEDGEMENTS

We thank Dale Edwards and Brauly Sinclair for their assistance in collecting frogs in the field, and George Brusck for his assistance on the design of the thermal tolerance methods. We also thank Scott Ballard and Mark Guetersloh of the Illinois Department of Natural Resources, John MacGregor of the Kentucky Department of Fish and Wildlife Resources, and Brandon Gilbert of the Tennessee Wildlife Resource Agency for their assistance in identifying collecting sites. BZ acknowledge financial support from startup funds (IB 1-152281) and NSF grant (DMS-2325196). *Hyla cinerea* were collected under permits obtained from the Kentucky Department of Fish and Wildlife Resources (educational wildlife collecting permit #SC2211235, issued to OME), Illinois Department of Natural Resources (permit #HSCP 19-48, issued to D. Edwards) and Tennessee Wildlife Resources Agency (permit #5510, issued to BZ). Permission to collect frogs on sites in Shawnee National Forest was granted by Tim Pohlman, District Ranger of Shawnee National Forest's Hidden Springs/Mississippi Bluffs Ranger Districts.

CONFLICT OF INTEREST STATEMENT

The authors declare no conflicts of interest.

DATA AVAILABILITY STATEMENT

Data available from the Dryad Digital Repository <https://datadryad.org/stash/dataset/doi:10.5061/dryad.rbnzs7hjg> (Edwards et al., 2024).

STATEMENT ON INCLUSION

Our study brings together a team of authors from diverse identities and backgrounds, an essential quality for addressing problems in today's world. Our research has also been shared and discussed to personnel working in local agencies where the data was collected (e.g. Kentucky Department of Fish and Wildlife, Illinois Department of Natural Resources, Tennessee Wildlife Resource Agency, etc.).

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REFERENCES

- Addo-Bediako, A., Chown, S. L., & Gaston, K. J. (2000). Thermal tolerance, climatic variability and latitude. *Proceedings of the Royal Society of London. Series B: Biological Sciences*, 267(1445), 739–745. <https://doi.org/10.1098/rspb.2000.1065>
- Allouche, O., Tsoar, A., & Kadmon, R. (2006). Assessing the accuracy of species distribution models: Prevalence, kappa and the true skill statistic (TSS). *Journal of Applied Ecology*, 43(6), 1223–1232. <https://doi.org/10.1111/j.1365-2664.2006.01214.x>
- Anderson, R. P. (2012). Harnessing the world's biodiversity data: Promise and peril in ecological niche modeling of species distributions. *Annals of the New York Academy of Sciences*, 1260(1), 66–80. <https://doi.org/10.1111/j.1749-6632.2011.06440.x>
- Angert, A. L., Bontrager, M. G., & Ågren, J. (2020). What do we really know about adaptation at range edges? *Annual Review of Ecology, Evolution, and Systematics*, 51, 341–361. <https://doi.org/10.1146/annurev-ecolsys-012120-091002>
- Angilletta, M. J. (2009). *Thermal adaptation: A theoretical and empirical synthesis*. Oxford University Press.
- Araújo, M. B., Ferri-Yáñez, F., Bozinovic, F., Marquet, P. A., Valladares, F., & Chown, S. L. (2013). Heat freezes niche evolution. *Ecology Letters*, 16(9), 1206–1219. <https://doi.org/10.1111/ele.12155>
- Bates, D., Mächler, M., Bolker, B., & Walker, S. (2015). Fitting linear mixed-effects models using lme4. *Journal of Statistical Software*, 67(1), 1–48. <https://doi.org/10.18637/jss.v067.i01>
- Baudier, K. M., Mudd, A. E., Erickson, S. C., & O'Donnell, S. (2015). Microhabitat and body size effects on heat tolerance: Implications for responses to climate change (army ants: Formicidae, Ecitoninae). *Journal of Animal Ecology*, 84(5), 1322–1330. <https://doi.org/10.1111/1365-2656.12388>
- Baxter-Gilbert, J., Riley, J. L., & Measey, J. (2021). Fortune favors the bold toad: Urban-derived behavioral traits may provide advantages for invasive amphibian populations. *Behavioral Ecology and Sociobiology*, 75(9), 130. <https://doi.org/10.1007/s00265-021-03061-w>
- Bennett, J. M., Calosi, P., Clusella-Trullas, S., Martínez, B., Sunday, J., Algar, A. C., Araújo, M. B., Hawkins, B. A., Keith, S., Kühn, I., Rahbek, C., Rodríguez, L., Singer, A., Villalobos, F., Ángel Olalla-Tárraga, M., & Morales-Castilla, I. (2018). GlobTherm, a global database on thermal tolerances for aquatic and terrestrial organisms. *Scientific Data*, 5(1), 180022. <https://doi.org/10.1038/sdata.2018.22>
- Bennett, S., Duarte, C. M., Marbà, N., & Wernberg, T. (2019). Integrating within-species variation in thermal physiology into climate change ecology. *Philosophical Transactions of the Royal Society B*, 374(1778), 20180550. <https://doi.org/10.1098/rstb.2018.0550>
- Berthouly-Salazar, C., van Rensburg, B. J., Le Roux, J. J., Van Vuuren, B. J., & Hui, C. (2012). Spatial sorting drives morphological variation in the invasive bird, *Acridotheris tristis*. *PLoS One*, 7(5), e38145. <https://doi.org/10.1371/journal.pone.0038145>
- Bestion, E., Clobert, J., & Cote, J. (2015). Dispersal response to climate change: Scaling down to intraspecific variation. *Ecology Letters*, 18(11), 1226–1233. <https://doi.org/10.1111/ele.12502>
- Biaou, S., Gouwakinnou, G. N., Noulékoun, F., Salako, K. V., Houndjo Kpoviwanou, J. M. R., Houehanou, T. D., & Biaou, H. S. S. (2023). Incorporating intraspecific variation into species distribution models improves climate change analyses of a widespread West African tree species (*Pterocarpus erinaceus* Poir, Fabaceae). *Global Ecology and Conservation*, 45, e02538. <https://doi.org/10.1016/j.gecco.2023.e02538>
- Bocedi, G., Palmer, S. C. F., Malchow, A.-K., Zurell, D., Watts, K., & Travis, J. M. J. (2021). RangeShifter 2.0: An extended and enhanced platform for modelling spatial eco-evolutionary dynamics and species' responses to environmental changes. *Ecography*, 44(10), 1453–1462. <https://doi.org/10.1111/ecog.05687>
- Booth, T. H., Nix, H. A., & Busby, J. R. (2013). Bioclim: The first species distribution modelling package, its early applications and relevance to most current MaxEnt studies. *Diversity and Distributions*, 20, 1–9. <https://doi.org/10.1111/ddi.12144>
- Brett, J. R. (1956). Some principles in the thermal requirements of fishes. *The Quarterly Review of Biology*, 31(2), 75–87. <https://doi.org/10.1086/401257>
- Bridle, J. R., Buckley, J., Bodsworth, E. J., & Thomas, C. D. (2014). Evolution on the move: Specialization on widespread resources associated with rapid range expansion in response to climate change. *Proceedings of the Royal Society B: Biological Sciences*, 281(1776), 20131800. <https://doi.org/10.1098/rspb.2013.1800>
- Brodie, S., Smith, J. A., Muhling, B. A., Barnett, L. A. K., Carroll, G., Fiedler, P., Bograd, S. J., Hazen, E. L., Jacox, M. G., Andrews, K. S., Barnes, C. L., Crozier, L. G., Fiechter, J., Fredston, A., Haltuch, M. A., Harvey, C. J., Holmes, E., Karp, M. A., Liu, O. R., ... Kaplan, I. C. (2022). Recommendations for quantifying and reducing uncertainty in climate projections of species distributions. *Global Change Biology*, 28(22), 6586–6601. <https://doi.org/10.1111/gcb.16371>
- Brusch, G. A., Taylor, E. N., & Whitfield, S. M. (2016). Turn up the heat: Thermal tolerances of lizards at La Selva, Costa Rica. *Oecologia*, 180(2), 325–334. <https://doi.org/10.1007/s00442-015-3467-3>
- Burton, O. J., Phillips, B. L., & Travis, J. M. J. (2010). Trade-offs and the evolution of life-histories during range expansion. *Ecology Letters*, 13(10), 1210–1220. <https://doi.org/10.1111/j.1461-0248.2010.01505.x>
- Carbonell, J. A., Wang, Y.-J., & Stoks, R. (2021). Evolution of cold tolerance and thermal plasticity in life history, behaviour and physiology during a poleward range expansion. *Journal of Animal Ecology*, 90(7), 1666–1677. <https://doi.org/10.1111/1365-2656.13482>
- Chardon, N. I., Pironon, S., Peterson, M. L., & Doak, D. F. (2020). Incorporating intraspecific variation into species distribution models improves distribution predictions, but cannot predict species traits for a wide-spread plant species. *Ecography*, 43(1), 60–74. <https://doi.org/10.1111/ecog.04630>
- Chen, E., Blaze, J. A., Smith, R. S., Peng, S., & Byers, J. E. (2020). Freeze tolerance of poleward-spreading mangrove species weakened by soil properties of resident salt marsh competitor. *Journal of Ecology*, 108(4), 1725–1737. <https://doi.org/10.1111/1365-2745.13350>
- Chen, I.-C., Hill, J. K., Ohlemüller, R., Roy, D. B., & Thomas, C. D. (2011). Rapid range shifts of species associated with high levels of climate warming. *Science*, 333(6045), 1024–1026. <https://doi.org/10.1126/science.1206432>
- Chuang, A., & Peterson, C. R. (2016). Expanding population edges: Theories, traits, and trade-offs. *Global Change Biology*, 22(2), 494–512. <https://doi.org/10.1111/gcb.13107>
- Collins, J. P., & Storfer, A. (2003). Global amphibian declines: Sorting the hypotheses. *Diversity and Distributions*, 9(2), 89–98. <https://doi.org/10.1046/j.1472-4642.2003.00012.x>
- Cowles, R. B., & Bogert, C. M. (1944). Preliminary study of the thermal requirements of desert reptiles. *Bulletin of the American Museum of Natural History*, 83, 261–296.
- Cronin, T. M., & Schneider, C. E. (1990). Climatic influences on species: Evidence from the fossil record. *Trends in Ecology & Evolution*, 5(9), 275–279. [https://doi.org/10.1016/0169-5347\(90\)90080-W](https://doi.org/10.1016/0169-5347(90)90080-W)
- DeAngelis, D. L., & Diaz, S. G. (2019). Decision-making in agent-based modeling: A current review and future prospectus. *Frontiers in Ecology and Evolution*, 6, 237. <https://doi.org/10.3389/fevo.2018.00237>
- DeMarche, M. L., Doak, D. F., & Morris, W. F. (2019). Incorporating local adaptation into forecasts of species' distribution and abundance under climate change. *Global Change Biology*, 25(3), 775–793. <https://doi.org/10.1111/gcb.14562>
- Deutsch, C. A., Tewksbury, J. J., Huey, R. B., Sheldon, K. S., Ghalambor, C. K., Haak, D. C., & Martin, P. R. (2008). Impacts of climate warming on terrestrial ectotherms across latitude. *Proceedings of the National Academy of Sciences of the United States of America*, 105(18), 6668–6672. <https://doi.org/10.1073/pnas.0709472105>

- Dodd, C. K. (2023). *Frogs of the United States and Canada* (2nd ed.). JHU Press.
- Duellman, W. E., & Trueb, L. (1986). *Biology of amphibians*. McGraw Hill.
- Dytham, C. (2009). Evolved dispersal strategies at range margins. *Proceedings of the Royal Society B: Biological Sciences*, 276(1661), 1407–1413. <https://doi.org/10.1098/rspb.2008.1535>
- Edwards, O. M., Edwards, D. D., Duncan, S. I., Laurencio, D., & Goessling, J. M. (2023). Range expansion and dispersal traits of Green Treefrogs (*Hyla cinerea*). *Journal of Herpetology*, 57(2), 151–158. <https://doi.org/10.1670/21-063>
- Edwards, O. M., Zhai, L., Reichert, M. S., Shaughnessy, C. A., Ozment, L., & Zhang, B. (2024). Physiological and morphological traits affect contemporary range expansion and implications for species distribution modeling in an amphibian species. *Dryad Digital Repository*. <https://doi.org/10.5061/dryad.rbnzs7hj>
- Engbrecht, N. J., Mirtl, J. J., & Johnson, E. M. (2018). *Hyla cinerea* (Green Treefrog): Geographic distribution, Indiana, Perry County. *Herpetological Review*, 49(4), 708.
- Enriquez-Urzelai, U., Bernardo, N., Moreno-Rueda, G., Montori, A., & Llorente, G. (2019). Are amphibians tracking their climatic niches in response to climate warming? A test with Iberian amphibians. *Climatic Change*, 154(1–2), 289–301. <https://doi.org/10.1007/s10584-019-02422-9>
- Ficetola, G. F., & De Bernardi, F. (2006). Trade-off between larval development rate and post-metamorphic traits in the frog *Rana latastei*. *Evolutionary Ecology*, 20(2), 143–158. <https://doi.org/10.1007/s10682-005-5508-6>
- Fick, S. E., & Hijmans, R. J. (2017). WorldClim 2: New 1-km spatial resolution climate surfaces for global land areas. *International Journal of Climatology*, 37(12), 4302–4315. <https://doi.org/10.1002/joc.5086>
- Fox, J., & Weisberg, S. (2019). *An R companion to applied regression* (3rd ed.). Sage publications.
- Garman, H. L. (1895). Notes on Illinois reptiles and amphibians, including several species not before recorded from the northern states. *Illinois Natural History Survey Bulletin*, 3(1–15), 185–190. <https://doi.org/10.21900/j.inhs.v3.191>
- Gaston, K. J., & Blackburn, T. M. (2002). Large-scale dynamics in colonization and extinction for breeding birds in Britain. *Journal of Animal Ecology*, 71(3), 390–399. <https://doi.org/10.1046/j.1365-2656.2002.00607.x>
- Ghalambor, C. K., McKay, J. K., Carroll, S. P., & Reznick, D. N. (2007). Adaptive versus non-adaptive phenotypic plasticity and the potential for contemporary adaptation in new environments. *Functional Ecology*, 21(3), 394–407. <https://doi.org/10.1111/j.1365-2435.2007.01283.x>
- Gruber, J., Brown, G., Whiting, M. J., & Shine, R. (2017). Geographic divergence in dispersal-related behaviour in cane toads from range-front versus range-core populations in Australia. *Behavioral Ecology and Sociobiology*, 71(2), 38. <https://doi.org/10.1007/s00265-017-2266-8>
- Guisan, A., Thuiller, W., & Zimmermann, N. E. (2017). *Habitat suitability and distribution models: With applications in R*. Cambridge University Press.
- Gunnarsson, T. G., Sutherland, W. J., Alves, J. A., Potts, P. M., & Gill, J. A. (2012). Rapid changes in phenotype distribution during range expansion in a migratory bird. *Proceedings of the Royal Society B: Biological Sciences*, 279(1727), 411–416. <https://doi.org/10.1098/rspb.2011.0939>
- Hällfors, M. H., Liao, J., Dzurisin, J., Grundel, R., Hyvärinen, M., Towle, K., Wu, G. C., & Hellmann, J. J. (2016). Addressing potential local adaptation in species distribution models: Implications for conservation under climate change. *Ecological Applications*, 26(4), 1154–1169. <https://doi.org/10.1890/15-0926>
- Herrando-Pérez, S., Ferri-Yáñez, F., Monasterio, C., Beukema, W., Gomes, V., Belliure, J., Chown, S. L., Vieites, D. R., & Araújo, M. B. (2019). Intraspecific variation in lizard heat tolerance alters estimates of climate impact. *Journal of Animal Ecology*, 88(2), 247–257. <https://doi.org/10.1111/1365-2656.12914>
- Hickling, R., Roy, D. B., Hill, J. K., Fox, R., & Thomas, C. D. (2006). The distributions of a wide range of taxonomic groups are expanding polewards. *Global Change Biology*, 12(3), 450–455. <https://doi.org/10.1111/j.1365-2486.2006.01116.x>
- Hudson, C. M., Vidal-García, M., Murray, T. G., & Shine, R. (2020). The accelerating anuran: Evolution of locomotor performance in cane toads (*Rhinella marina*, Bufonidae) at an invasion front. *Proceedings of the Royal Society B: Biological Sciences*, 287(1938), 20201964. <https://doi.org/10.1098/rspb.2020.1964>
- Huey, R. B., Kearney, M. R., Krockenberger, A., Holtum, J. A., Jess, M., & Williams, S. E. (2012). Predicting organismal vulnerability to climate warming: Roles of behaviour, physiology and adaptation. *Philosophical Transactions of the Royal Society, B: Biological Sciences*, 367(1596), 1665–1679. <https://doi.org/10.1098/rstb.2012.0005>
- Huey, R. B., & Stevenson, R. D. (1979). Integrating thermal physiology and ecology of ectotherms: A discussion of approaches. *American Zoologist*, 19(1), 357–366. <https://doi.org/10.1093/icb/19.1.357>
- Hutchinson, G. E. (1957). Concluding remarks. *Cold Spring Harbor Symposium on Quantitative Biology*, 22, 415–427.
- Ikeda, D. H., Max, T. L., Allan, G. J., Lau, M. K., Shuster, S. M., & Whitham, T. G. (2017). Genetically informed ecological niche models improve climate change predictions. *Global Change Biology*, 23(1), 164–176. <https://doi.org/10.1111/gcb.13470>
- Janzen, D. H. (1967). Why mountain passes are higher in the tropics. *The American Naturalist*, 101(919), 233–249. <https://doi.org/10.1086/282487>
- Kentucky Department of Fish & Wildlife Resources. (2024). *Species information. County observation(s) for Amphibia. Green Treefrog (Hyla cinerea)*. <https://app.fw.ky.gov/speciesinfo/reference.asp?strElCode=AAABC02060&strCountyFips=7&strGroup=5>
- Kolbe, J. J., Ehrenberger, J. C., Moniz, H. A., & Angilletta, M. J., Jr. (2014). Physiological variation among invasive populations of the brown anole (*Anolis sagrei*). *Physiological and Biochemical Zoology*, 87(1), 92–104. <https://doi.org/10.1086/672157>
- Kolbe, J. J., Kearney, M., & Shine, R. (2010). Modeling the consequences of thermal trait variation for the cane toad invasion of Australia. *Ecological Applications*, 20(8), 2273–2285. <https://doi.org/10.1890/09-1973.1>
- Lancaster, L. T. (2016). Widespread range expansions shape latitudinal variation in insect thermal limits. *Nature Climate Change*, 6(6), 618–621. <https://doi.org/10.1038/nclimate2945>
- Lancaster, L. T., Dudaniec, R. Y., Hansson, B., & Svensson, E. I. (2015). Latitudinal shift in thermal niche breadth results from thermal release during a climate-mediated range expansion. *Journal of Biogeography*, 42(10), 1953–1963. <https://doi.org/10.1111/jbi.12553>
- Lenoir, J., Gégout, J. C., Marquet, P. A., De Ruffray, P., & Brisse, H. (2008). A significant upward shift in plant species optimum elevation during the 20th century. *Science*, 320(5884), 1768–1771. <https://doi.org/10.1126/science.1156831>
- Liebl, A. L., & Martin, L. B. (2014). Living on the edge: Range edge birds consume novel foods sooner than established ones. *Behavioral Ecology*, 25(5), 1089–1096. <https://doi.org/10.1093/beheco/aru089>
- Lodato, M. J., Engbrecht, N. J., Klueh-Mundy, S., & Walker, Z. (2014). The green treefrog, *Hyla cinerea* (Schneider), in Indiana. *Proceedings of the Indiana Academy of Sciences*, 123(2), 179–195.
- Lutterschmidt, W. I., & Hutchison, V. H. (1997). The critical thermal maximum: Data to support the onset of spasms as the definitive end point. *Canadian Journal of Zoology*, 75(10), 1553–1560. <https://doi.org/10.1139/z97-782>
- Madeira, D., Narciso, L., Cabral, H. N., & Vinagre, C. (2012). Thermal tolerance and potential impacts of climate change on coastal and estuarine organisms. *Journal of Sea Research*, 70, 32–41. <https://doi.org/10.1016/j.seares.2012.03.002>

- Mainali, K. P., Warren, D. L., Dhileepan, K., McConnachie, A., Strathie, L., Hassan, G., Karki, D., Shrestha, B. B., & Parmesan, C. (2015). Projecting future expansion of invasive species: Comparing and improving methodologies for species distribution modeling. *Global Change Biology*, 21(12), 4464–4480. <https://doi.org/10.1111/gcb.13038>
- Mair, L., Hill, J. K., Fox, R., Botham, M., Brereton, T., & Thomas, C. D. (2014). Abundance changes and habitat availability drive species' responses to climate change. *Nature Climate Change*, 4(2), 127–131. <https://doi.org/10.1038/nclimate2086>
- Mirtl, J. J., Truelock, Z. T., & Engbrecht, N. J. (2020). *Hyla cinerea* (Green Treefrog): Geographic distribution, Indiana, Crawford, Floyd, and Harrison counties. *Herpetological Review*, 51(1), 70.
- Morris, M. R. (1989). Female choice of large males in the treefrog *Hyla chrysoscelis*: The importance of identifying the scale of choice. *Behavioral Ecology and Sociobiology*, 25(4), 275–281. <https://doi.org/10.1007/BF00300054>
- Nauwelaerts, S., Ramsay, J., & Aerts, P. (2007). Morphological correlates of aquatic and terrestrial locomotion in a semi-aquatic frog, *Rana esculenta*: No evidence for a design conflict. *Journal of Anatomy*, 210, 304–317. <https://doi.org/10.1111/j.1469-7580.2007.00691.x>
- Navas, C. A., Antoniazzi, M. M., Carvalho, J. E., Suzuki, H., & Jared, C. (2007). Physiological basis for diurnal activity in dispersing juvenile *Bufo granulosus* in the Caatinga, a Brazilian semi-arid environment. *Comparative Biochemistry and Physiology Part A: Molecular & Integrative Physiology*, 147(3), 647–657. <https://doi.org/10.1016/j.cbpa.2006.04.035>
- Newton, I. (1997). Links between the abundance and distribution of birds. *Ecography*, 20(2), 137–145. <https://doi.org/10.1111/j.1600-0587.1997.tb00356.x>
- Nowakowski, A. J., Watling, J. I., Whitfield, S. M., Todd, B. D., Kurz, D. J., & Donnelly, M. A. (2017). Tropical amphibians in shifting thermal landscapes under land-use and climate change. *Conservation Biology*, 31(1), 96–105. <https://doi.org/10.1111/cobi.12769>
- Ochocki, B. M., & Miller, T. E. (2017). Rapid evolution of dispersal ability makes biological invasions faster and more variable. *Nature Communications*, 8(1), 14315. <https://doi.org/10.1038/ncomm514315>
- Osland, M. J., & Feher, L. C. (2020). Winter climate change and the poleward range expansion of a tropical invasive tree (Brazilian pepper—*Schinus terebinthifolius*). *Global Change Biology*, 26(2), 607–615. <https://doi.org/10.1111/gcb.14842>
- Parmesan, C. (2006). Ecological and evolutionary responses to recent climate change. *Annual Review of Ecology, Evolution, and Systematics*, 37(1), 637–669. <https://doi.org/10.1146/annurev.ecolsys.37.091305.110100>
- Parmesan, C., & Yohe, G. (2003). A globally coherent fingerprint of climate change impacts across natural systems. *Nature*, 421(6918), 37–42. <https://doi.org/10.1038/nature01286>
- Pearcy, W. G., & Schoener, A. (1987). Changes in the marine biota coincident with the 1982–1983 El Niño in the northeastern subarctic Pacific Ocean. *Journal of Geophysical Research: Oceans*, 92(C13), 14417–14428. <https://doi.org/10.1029/JC092iC13p14417>
- Perkins, A. T., Phillips, B. L., Baskett, M. L., & Hastings, A. (2013). Evolution of dispersal and life history interact to drive accelerating spread of an invasive species. *Ecology Letters*, 16(8), 1079–1087. <https://doi.org/10.1111/ele.12136>
- Phillips, B. L., Brown, G. P., & Shine, R. (2010). Life-history evolution in range-shifting populations. *Ecology*, 91(6), 1617–1627. <https://doi.org/10.1890/09-0910.1>
- Phillips, B. L., Brown, G. P., Webb, J. K., & Shine, R. (2006). Invasion and the evolution of speed in toads. *Nature*, 439(7078), 803. <https://doi.org/10.1038/439803a>
- Pinheiro, J., Bates, D., & R Core Team. (2023). *nlme: Linear and nonlinear mixed effects models* (3.1-164) [Computer software]. <https://CRAN.R-project.org/package=nlme>
- Powell, R., Conant, R., & Collins, J. (2016). *Peterson field guide to reptiles and amphibians of eastern and central North America* (4th ed.). Houghton Mifflin Harcourt Publishing.
- R Core Team. (2023). *R: A language and environment for statistical computing*. R Foundation for Statistical Computing. <https://www.R-project.org/>
- Redmer, M., Brown, B. E., & Ronald, B. A. (1999). Natural history of the bird-voiced treefrog (*Hyla avivoca*) and green treefrog (*Hyla cinerea*) in southern Illinois. *Illinois Natural History Survey Bulletin*, 36(2), 37–66. <https://doi.org/10.21900/j.inhs.v36.124>
- Richardson, A. J. (2008). In hot water: Zooplankton and climate change. *ICES Journal of Marine Science*, 65(3), 279–295. <https://doi.org/10.1093/icesjms/fsn028>
- Root, T. L., Price, J. T., Hall, K. R., Schneider, S. H., Rosenzweig, C., & Pounds, J. A. (2003). Fingerprints of global warming on wild animals and plants. *Nature*, 421(6918), 57–60. <https://doi.org/10.1038/nature01333>
- Rowley, J. J., & Alford, R. A. (2007). Non-contact infrared thermometers can accurately measure amphibian body temperatures. *Herpetological Review*, 38, 308–316.
- Shine, R., Brown, G. P., & Phillips, B. L. (2011). An evolutionary process that assembles phenotypes through space rather than through time. *Proceedings of the National Academy of Sciences of the United States of America*, 108(14), 5708–5711. <https://doi.org/10.1073/pnas.1018989108>
- Simmons, A. D., & Thomas, C. D. (2004). Changes in dispersal during species' range expansions. *The American Naturalist*, 164(3), 378–395. <https://doi.org/10.1086/423430>
- Smith, M. A., & Green, D. M. (2005). Dispersal and the metapopulation paradigm in amphibian ecology and conservation: Are all amphibian populations metapopulations? *Ecography*, 28(1), 110–128. <https://doi.org/10.1111/j.0906-7590.2005.04042.x>
- Snyder, G. K., & Weathers, W. W. (1975). Temperature adaptations in amphibians. *The American Naturalist*, 109(965), 93–101. <https://doi.org/10.1086/282976>
- Somero, G. N. (2010). The physiology of climate change: How potentials for acclimatization and genetic adaptation will determine 'winners' and 'losers'. *Journal of Experimental Biology*, 213(6), 912–920. <https://doi.org/10.1242/jeb.037473>
- Stevens, G. C. (1989). The latitudinal gradient in geographical range: How so many species coexist in the tropics. *The American Naturalist*, 133(2), 240–256. <https://doi.org/10.1086/284913>
- Stuart, S. N., Chanson, J. S., Cox, N. A., Young, B. E., Rodrigues, A. S., Fischman, D. L., & Waller, R. W. (2004). Status and trends of amphibian declines and extinctions worldwide. *Science*, 306(5702), 1783–1786. <https://doi.org/10.1126/science.1103538>
- Sunday, J. M., Bennett, J. M., Calosi, P., Clusella-Trullas, S., Gravel, S., Hargreaves, A. L., Leiva, F. P., Verberk, W. C. E. P., Olalla-Tárraga, M. Á., & Morales-Castilla, I. (2019). Thermal tolerance patterns across latitude and elevation. *Philosophical Transactions of the Royal Society, B: Biological Sciences*, 374(1778), 20190036. <https://doi.org/10.1098/rstb.2019.0036>
- Sunday, J. M., Bates, A. E., & Dulvy, N. K. (2011). Global analysis of thermal tolerance and latitude in ectotherms. *Proceedings of the Royal Society B: Biological Sciences*, 278(1713), 1823–1830. <https://doi.org/10.1098/rspb.2010.1295>
- Sunday, J. M., Bates, A. E., & Dulvy, N. K. (2012). Thermal tolerance and the global redistribution of animals. *Nature Climate Change*, 2(9), 686–690. <https://doi.org/10.1038/nclimate1539>
- Svenning, J.-C., Gravel, D., Holt, R. D., Schurr, F. M., Thuiller, W., Münkemüller, T., Schiffers, K. H., Dullinger, S., Edwards, T. C., Jr., & Hickler, T. (2014). The influence of interspecific interactions on species range expansion rates. *Ecography*, 37(12), 1198–1209. <https://doi.org/10.1111/j.1600-0587.2013.00574.x>
- Taylor-Cox, E. D., Macgregor, C. J., Corthine, A., Hill, J. K., Hodgson, J. A., & Saccheri, I. J. (2020). Wing morphological responses to latitude

- and colonisation in a range expanding butterfly. *PeerJ*, 8, e10352. <https://doi.org/10.7717/peerj.10352>
- Therry, L., Bonte, D., & Stoks, R. (2015). Higher investment in flight morphology does not trade off with fecundity estimates in a poleward range-expanding damselfly. *Ecological Entomology*, 40(2), 133–142. <https://doi.org/10.1111/een.12170>
- Thomas, C. D. (2010). Climate, climate change and range boundaries. *Diversity and Distributions*, 16(3), 488–495. <https://doi.org/10.1111/j.1472-4642.2010.00642.x>
- Travis, J. M., & Dytham, C. (2002). Dispersal evolution during invasions. *Evolutionary Ecology Research*, 4(8), 1119–1129.
- Trewick, S. (2017). Plate tectonics in biogeography. In D. Richardson, N. Castree, M. F. Goodchild, A. Kobayashi, W. Liu, & R. A. Marston (Eds.), *International encyclopedia of geography* (1st ed., pp. 1–9). Wiley. <https://doi.org/10.1002/9781118786352.wbieg0638>
- Tucker, J. K., Lamer, J. T., Dolan, C. R., & Avenue, M. (2008). A northern range expansion for the Green tree frog (*Hyla cinerea*) and trends in distributions of Illinois reptiles and amphibians. *Transactions of the Illinois State Academy of Science*, 101, 125–131.
- U.S. Geological Survey (USGS). (2018). *Gap analysis project. Green treefrog (Hyla cinerea) aGRTRx_CONUS_2001v1 range map* [Dataset]. <https://doi.org/10.5066/F7QR4W45>
- Valladares, F., Matesanz, S., Guilhaumon, F., Araújo, M. B., Balaguer, L., Benito-Garzon, M., Cornwell, W., Gianoli, E., Kleunen, M., Naya, D. E., Nicotra, A. B., Poorter, H., & Zavala, M. A. (2014). The effects of phenotypic plasticity and local adaptation on forecasts of species range shifts under climate change. *Ecology Letters*, 17(11), 1351–1364. <https://doi.org/10.1111/ele.12348>
- Virkkala, R., & Lehtikoinen, A. (2014). Patterns of climate-induced density shifts of species: Poleward shifts faster in northern boreal birds than in southern birds. *Global Change Biology*, 20(10), 2995–3003. <https://doi.org/10.1111/gcb.12573>
- Wells, K. D. (2007). *The ecology & behavior of amphibians*. The University of Chicago Press.
- Wiens, J. A., Stralberg, D., Jongsomjit, D., Howell, C. A., & Snyder, M. A. (2009). Niches, models, and climate change: Assessing the assumptions and uncertainties. *Proceedings of the National Academy of Sciences of the United States of America*, 106, 19729–19736. <https://doi.org/10.1073/pnas.0901639106>
- Williams, J. E., & Blois, J. L. (2018). Range shifts in response to past and future climate change: Can climate velocities and species' dispersal capabilities explain variation in mammalian range shifts? *Journal of Biogeography*, 45(9), 2175–2189. <https://doi.org/10.1111/jbi.13395>
- Yamano, H., Sugihara, K., & Nomura, K. (2011). Rapid poleward range expansion of tropical reef corals in response to rising sea surface temperatures. *Geophysical Research Letters*, 38(4), L04601. <https://doi.org/10.1029/2010GL046474>
- Zuckerberg, B., Woods, A. M., & Porter, W. F. (2009). Poleward shifts in breeding bird distributions in New York state. *Global Change Biology*, 15(8), 1866–1883. <https://doi.org/10.1111/j.1365-2486.2009.01878.x>
- Zurell, D., Thuiller, W., Pagel, J., Cabral, J. S., Münkemüller, T., Gravel, D., Dullinger, S., Normand, S., Schiffers, K. H., Moore, K. A., & Zimmermann, N. E. (2016). Benchmarking novel approaches for modelling species range dynamics. *Global Change Biology*, 22(8), 2651–2664. <https://doi.org/10.1111/gcb.13251>

SUPPORTING INFORMATION

Additional supporting information can be found online in the Supporting Information section at the end of this article.

Figure S1. An individual green treefrog (*Hyla cinerea*) shown inside a custom water chamber.

Figure S2. Radiograph image of a male green treefrog (*Hyla cinerea*) depicting landmarks for snout-vent length, skull width, radioulna length, humerus length, femur length, and tibiofibula length.

How to cite this article: Edwards, O. M., Zhai, L., Reichert, M. S., Shaughnessy, C. A., Ozment, L., & Zhang, B. (2025). Physiological and morphological traits affect contemporary range expansion and implications for species distribution modelling in an amphibian species. *Journal of Animal Ecology*, 94, 195–209. <https://doi.org/10.1111/1365-2656.14212>