

OPINION

Climate change and shrinking salamanders: alternative mechanisms for changes in plethodontid salamander body size

GRANT M. CONNETTE¹, JOHN A. CRAWFORD² and WILLIAM E. PETERMAN³¹Division of Biological Sciences, University of Missouri, Columbia, MO 65211, USA, ²National Great Rivers Research and Education Center, East Alton, IL 62024, USA, ³Prairie Research Institute, Illinois Natural History Survey, University of Illinois, Champaign, IL 61820, USA

Abstract

An increasing number of studies have demonstrated relationships between climate trends and body size change of organisms. In many cases, climate might be expected to influence body size by altering thermoregulation, energetics or food availability. However, observed body size change can result from a variety of ecological processes (e.g. growth, selection, population dynamics) or imperfect observation of biological systems. We used two extensive datasets to evaluate alternative mechanisms for recently reported changes in the observed body size of plethodontid salamanders. We found that mean adult body size of salamanders can be highly sensitive to survey conditions, particularly rainfall. This systematic bias in the detection of larger or smaller individuals could result in a signature of body size change in relation to reported climate trends when it is simply observation error. We also identify considerable variability in body size distributions among years and find that individual growth rates can be strongly influenced by weather. Finally, our study demonstrates that measures of mean adult body size can be highly variable among surveys and that large sample sizes may be required to make reliable inferences. Identifying the effects of climate change is a critical area of research in ecology and conservation. Researchers should be aware that observed changes in certain organisms can result from multiple ecological processes or systematic bias due to nonrandom sampling of populations.

Keywords: amphibian, declines, detection probability, growth, population dynamics, sampling bias, weather

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Introduction

Global climate change has been shown to impact the geographic distributions of species (Parmesan & Yohe, 2003; Perry *et al.*, 2005; Hickling *et al.*, 2006) as well as timing of life cycle events such as breeding and migration (Root *et al.*, 2003; Menzel *et al.*, 2006). Recent studies have also highlighted the potential for climate change to drive body size change in organisms (Daufresne *et al.*, 2009; Gardner *et al.*, 2011; Sheridan & Bickford, 2011; Baudron *et al.*, 2014). Altered thermoregulation, energetics or food availability may ultimately cause climate-related changes in body size (Gardner *et al.*, 2011), yet shifts in population size distributions may result from multiple mechanisms. For instance, climate may directly influence size at age (e.g. size at maturity) or, alternatively, may cause a shift in population age structure which also leads to a directional change in body size (Daufresne *et al.*, 2009). In

the latter case, changes in a number of demographic processes such as recruitment, growth or survival may be responsible for population body size trends.

Given the imperfect ability of researchers to observe many ecological systems, there is also a very real possibility that systematic sampling bias can confound inferences concerning underlying ecological processes such as climate-driven body size change. Sampling bias occurs when individuals are systematically unequal in their probabilities of being sampled from a population of interest. In demographic studies, this may occur due to differences in the availability of individuals for detection, such as when certain individuals are more likely to leave the sampling area (i.e. 'temporary emigration') or when present individuals differ in their respective probabilities of capture or detection due to survey methods (Kendall & Bjorkland, 2001). Such sampling biases may arise due to a number of factors such as trap efficiency (Smith *et al.*, 2004; Willson *et al.*, 2008; Driscoll *et al.*, 2012), observer skill (Cunningham *et al.*, 1999; Freckleton *et al.*, 2006; Kéry *et al.*, 2009), habitat

Correspondence: Grant M. Connette, tel. 573 882 1421, fax 573 882 0123, e-mail: grmcco@gmail.com

characteristics (Chen *et al.*, 2013; Peterman & Semlitsch, 2013), and weather conditions (Pellet & Schmidt, 2005; Chandler & King, 2011; O'Donnell *et al.*, 2014). In many of the above examples, sampling bias was related to body size and could lead to larger individuals or species being disproportionately represented in ecological datasets (Cunningham *et al.*, 1999; Smith *et al.*, 2004; Freckleton *et al.*, 2006; Willson *et al.*, 2008). Although relative demographic changes may be correctly identified with consistent bias in sampling, a relationship between detection and a factor of biological interest (e.g. body size, habitat, weather) may be erroneously interpreted as a true demographic shift.

We caution that observed changes in the body size of organisms may result from a number of true ecological processes or systematic bias due to nonrandom sampling of populations. Here, we consider possible hypotheses for recently reported changes in the observed body size of plethodontid salamanders (Caruso *et al.*, 2014). First, it is plausible that climate change has resulted in selection for smaller adult body size in recent decades. Second, individual growth may vary with weather conditions. Third, population age structure may be variable among years (potentially in relation to weather). Fourth, individuals may differ in their exposure to sampling due to survey conditions. In our study, we examine trends in the observed body size distributions of two species of *Plethodon* salamanders using data collected in southwestern North Carolina during separate studies examining the effects of forest management practices on salamander populations. We leverage these extensive datasets to examine the possibility that observed body size distributions are sensitive to proximate survey conditions, to quantify the observed variability in body size among years, and to highlight the need for adequate sample sizes when making inference concerning body size trends in *Plethodon* salamanders.

Materials and methods

Dataset I: Relative counts of *Plethodon metcalfi*

From 2004 to 2005, we conducted count surveys for salamanders in 32 (100 × 5 m) sampling plots distributed across 16 sites (two plots per site) located in the Nantahala National Forest, Macon County, NC, USA. All sites were located between 718 and 1248 m in elevation and were at least 1 km apart. Ten of these sites were sampled in both 2004 and 2005, while four sites were sampled only in 2004 and two sites were sampled only in 2005. Each plot was sampled three times during a season (i.e. plots that were sampled in both 2004 and 2005 had a total of six visits). We used a nighttime visual encounter search of each plot (survey order was randomized across sites to reduce bias related to seasonal activity) to capture surface-active salamanders. Surveys were performed between 22:00

and 03:00 EST from mid-May to mid-August and generally lasted 30 min to 2 h per plot (survey duration was dependent upon the number of salamanders captured). A researcher walked a straight line through the middle of the plot and exhaustively searched 2.5 m to the right and left. We identified all captured salamanders to species, weighed, and measured for snout-vent length (SVL) and subsequently released all salamanders at the point of capture. We determined age class (adult or juvenile) by comparing the SVL of each individual to published size classes (Bruce, 1967). For the purposes of analyses, we consider 50 mm to be the body size threshold between juveniles and adults, as males of this size often exhibit the secondary sex characteristic of mental gland development. A total of 1940 *P. metcalfi* were captured across the two field seasons ($N = 798$ adults).

Dataset II: Mark-recapture of *Plethodon shermani*

From 2009 to 2014, we conducted capture-mark-recapture surveys for salamanders in 16 (25 × 25 m) survey plots on the Nantahala National Forest, Clay County, NC, USA. These plots were located at similar elevation (~1200 m) in terrestrial habitat, and 8 of the 16 plots had timber removed between 2011 and 2013. In this study, we consider only data collected preharvest or in unharvested control plots. Within years, nighttime area-constrained surveys at each plot were staggered at approximately 1-month intervals from mid-May and mid-August and occurred between 21:30 and 05:45 EST. During surveys, the entire plot area was exhaustively searched by GMC and one trained assistant (occasionally 2+). All surface-active salamanders observed were hand-captured and surveys generally lasted 1–2 h per plot. We individually marked each salamander with visual implant elastomer (e.g. Heemeyer *et al.*, 2007) and recorded its sex, mass, and SVL prior to returning it to within 5 m of their unique capture location, almost always on the second night after capture.

In 6 years, we visited each plot 14–16 times and recorded 17 332 total captures of 10 salamander species. Red-legged salamanders (*P. shermani*) represented the majority of captures, with most individuals showing some morphological evidence of hybridization with *P. teyahalee* (e.g. Walls, 2009). We thinned our dataset to 12 289 *P. shermani* captures ($N = 5275$ adults) to exclude captures that occurred after timber harvest.

Mixed effects modeling

To evaluate the effects of weather and season on mean adult SVL and number of adult *P. metcalfi* counted per survey, we used linear mixed effects models with Gaussian and Poisson error distributions, respectively. For each analysis, we conducted model selection on six *a priori* models (Appendix S1), which included days since a soaking rain event and Julian day as fixed effects, each scaled and centered. A soaking rain event was when ≥5 mm of rain fell within a 24-h period, as this amount of rainfall is sufficient to reach the forest floor and moisten leaf litter (O'Connor *et al.*, 2006). We pooled observations from all sites surveyed in a night to determine the mean SVL and number of adult *P. metcalfi* collected for each survey

night ($N = 42$ nights) and used these measures as our response variables to examine the effects of rainfall and survey date on observed body size and salamander counts. Snout-vent length was used as our measure of body size in all analyses due to the potential for mass to change rapidly with salamander hydration state (Spight, 1968; Spotila, 1972). Year was considered as a random effect in all models, and the number of sites surveyed on a night was used as an offset in Poisson count models. All models were constructed using *lme4* (Bates *et al.*, 2013), and model selection was performed using AIC_c as calculated in *AICcmodavg* (Mazerolle, 2012).

Mixture analysis

We used our 6-year dataset for *P. shermani* to examine among-year variation in the observed distribution of individual body sizes (SVL). Each year, the overall size distribution of captured individuals showed three separate peaks (Fig. 1). Based on growth data from 2365 recaptures, these three distributions are known to correspond to distinct age classes. The smallest size distribution is composed of hatchling individuals, which are available to sample for the first time in the current season. The middle distribution consists of 2nd-year individuals and overlaps slightly with the largest body size distribution of individuals 3 years and older. Individuals in the largest size class are considered adults, as male salamanders on the smaller end of this size class will often show mental gland development. We performed a Bayesian analysis of a normal mixture model to identify shifts in the mean body size of the adult (3+ years) age class among years. A basic model represents the overall body size distribution of individuals as the weighted sum of three Gaussian densities.

$$S_i \sim \sum_{c=1}^3 \omega_c \text{Normal}(\mu_c, \sigma_c)$$

Here, body size (S) of each individual i is distributed according to a mixture of three normal probability density functions, where ω_c represents the probability that any individual belongs in a given body size class, c . Thus, our analysis treats the size class membership of each individual as an unobserved (latent) variable that is estimated directly from the data. The parameters μ_c and σ_c represent the mean and standard deviation describing the distribution for each body size class, c . In our analysis, we treat year as a fixed effect and separately estimate the parameters ω_c , μ_c and σ_c for each of our five survey years. We can then easily compute the estimated pairwise differences in mean adult body size among years, along with corresponding credible intervals (CRIs).

Mark–recapture analyses

To isolate potential mechanisms for observed changes in body size, we conducted two additional analyses of our capture–recapture data for *P. shermani*. First, we fit a von Bertalanffy growth curve to individual body size (SVL) data using a Bayesian hierarchical modeling approach that accounts for interindividual variation in growth rates as well as measure-

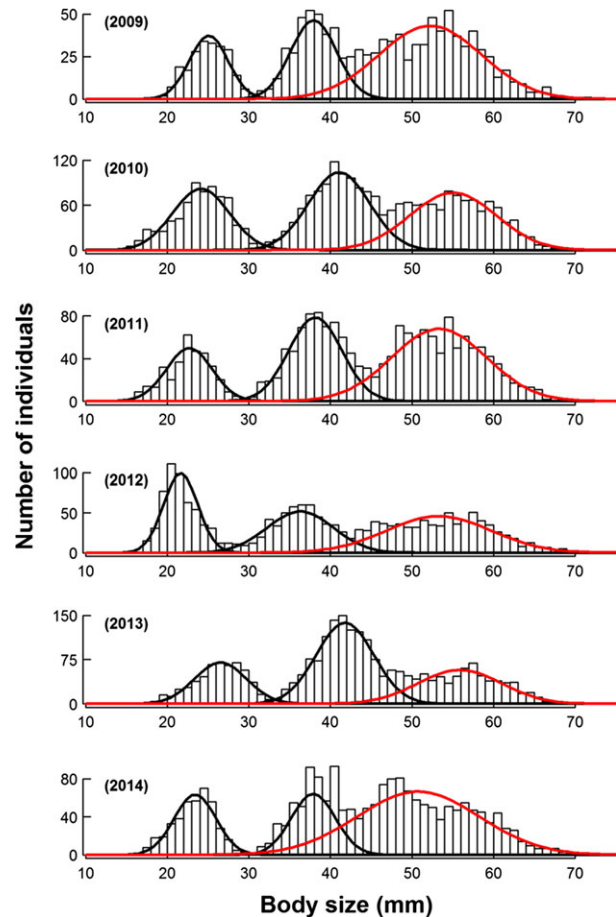


Fig. 1 Histograms depict observed body size distributions of *P. shermani* from 2009 (top panel) through 2014 (bottom panel). Lines indicate estimated densities by size class based on analysis of a normal mixture model.

ment error (Eaton & Link, 2011). We extended this model by incorporating a log-link function to estimate and account for the effects of weather covariates on the von Bertalanffy growth rate parameter, k . We considered both mean daily rainfall and mean daily temperature across the recapture interval as predictors of growth rate, as well as an indicator variable for season (summer vs. overwinter). To estimate survival and detection probabilities of adult (>45 mm SVL) *P. shermani*, we used a state-space representation of the Cormack–Jolly–Seber model (Royle, 2008; Kery & Schaub, 2012). In the current analysis, we estimate a constant survival probability and use a logit-link function to introduce covariates for body size at last capture (SVL) and days since a soaking rain event on individual detection probability. We also consider an interaction between body size and days since rain, which would indicate a difference between large and small individuals in their responses to rainfall. We considered just adults for this analysis, using a size threshold of 45 mm, which is the size at which males often exhibit the secondary sex characteristic of mental gland development. See Appendix S2 for additional model details, prior specification, and R code for these analyses.

Data resampling

To determine the sampling effort required to have confidence in mean adult body size estimates of *P. metcalfi* and *P. shermani*, we repeatedly pooled individual body size (SVL) measurements from randomly selected surveys and calculated mean adult body size for each new sample of individuals. We varied the number of surveys contributing to the new sample of body size measurements from 1 to 50 and performed 10 000 resamplings for each hypothetical number of surveys. We then calculated the mean percent difference of these samples from the observed mean adult body size based on all original surveys.

Results

Mean adult body size of *P. metcalfi* was highly sensitive to survey conditions and was best-predicted by a model including rainfall, Julian date, and a quadratic term for Julian date (Table 1). This result suggests that large adults were disproportionately represented in samples collected shortly after rainfall and toward the middle of the summer active season (Fig. 2). The effects of rainfall and Julian date were considerable in this study; mean body size of a sample was predicted to vary by up to 11.3% across the observed range of rainfall and by up to 8.3% in relation to Julian date. This equated to a predicted 11.8% difference in mean body size across the range of observed sampling conditions. The number of adult *P. metcalfi* observed was also best-explained by rainfall and Julian date, with counts predicted to decrease with time since rainfall and in correspondence with Julian date (Table 1, Fig. 2).

Using our *P. shermani* dataset, we found evidence for large annual variation in mean body size of our three age classes (Fig. 1). In particular, we note the high vari-

ability in both the relative frequencies and body size ranges of the hatchling and juvenile age classes, suggesting substantial annual variation in growth, recruitment, or age-dependent survival rates. Although the adult size class appeared to show the least variability among years, we still observed an overall 7.0% increase (95% CRI; 4.6 to 9.4) in mean adult body size between 2009 and 2013, followed by a 9.1% (95% CRI; 7.0 to 11.1) reduction in 2014 (Fig. 3). In addition, the difference in mean adult body size averaged 4.7% (95% CRI; 3.6 to 5.7) among sequential years. Mean adult size also tended to be higher in years with greater cumulative rainfall over the study period (Fig. 4).

Using recapture data, we considered variation in growth rates and sampling bias as potential mechanisms for observed shifts in population body size distributions. Growth rates of *P. shermani* between capture occasions showed a strong positive relationship with mean daily rainfall ($\beta_{\text{rain}} = 1.33$, 95% CRI; 1.10 to 1.58) and were considerably lower across the overwinter period ($\beta_{\text{season}} = -0.84$, 95% CRI; -1.12 to -0.52). Although evidence for a negative effect of mean daily temperature on growth rates was more equivocal ($\beta_{\text{temp}} = -0.02$, 95% CRI; -0.05 to 0.02), we recovered a posterior probability of 85.8% that temperature was negatively related to growth rates. Under average rainfall and temperature conditions, the expected SVL of a salamander entering its second or third summer was 36.3 mm (95% CRI; 35.3 to 37.5) and 46.7 mm (95% CRI; 45.6 to 48.0), respectively. From our analysis of a Cormack–Jolly–Seber model, we found that large and small adults differ in the degree to which their probabilities of detection decline with time since rainfall. Specifically, detection probability for large adults (60 mm) was predicted to decline by 57.9% (95% CRI; 44.8 to 68.2) from 0 to 9 days postrain, compared to 84.7% (95% CRI; 73.0 to 89.9) for small adults (45 mm). Regardless of conditions, large adults were more likely to be detected than small adults ($\beta_{\text{size}} = 0.07$, 95% CRI; 0.04 to 0.10).

Due to the variation in mean adult body size among surveys (Fig. 5a, b), our data resampling exercise indicates that repeated surveys were required to obtain consistently unbiased estimates of mean adult body size for each multiyear study (Fig. 5c, d). The mean number of adults captured per survey was 19.0 ± 19.8 (SD) for *P. metcalfi* and 28.2 ± 17.4 (SD) for *P. shermani*. Based on all captures, we observed a mean adult body size of 58.6 ± 6.3 mm (SD) for *P. metcalfi* and 54.0 ± 5.5 mm (SD) for *P. shermani*. With just one survey of a population, the survey mean would be expected to differ from the overall mean by an average of 3.96% in *P. metcalfi* and by 2.06% in *P. shermani*. In addition, there is a considerable probability of even

Table 1 Ranking of models for mean adult body size and number of adult *P. metcalfi* encountered

	K	ΔAICc	ω	Cum. Wt.
Adult body size				
Global*	6	0.00	0.89	0.89
Rain + Date	5	5.21	0.07	0.96
Rain	4	6.96	0.03	0.99
Date ²	5	9.61	0.01	0.99
Number of adults				
Rain + Date	4	0.00	0.61	0.69
Global*	5	0.92	0.39	1.00

ΔAICc represents the difference in AICc value between each model and the best model in the set. ω gives the Akaike weight for each model. Table includes only models with $\omega > 0$.

*Global models include Rain (days since ≥ 5 mm), Date (Julian day), and Date².

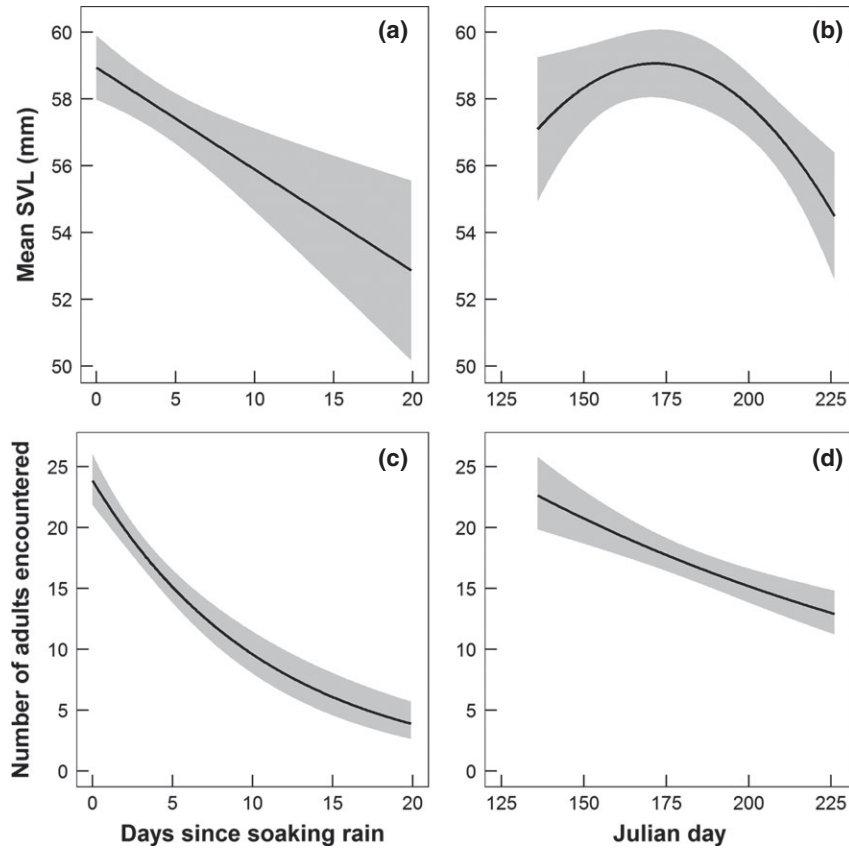


Fig. 2 Estimated effects of time since a soaking rain (a) and Julian day (b) on mean adult SVL of *P. metcalfi*. Estimated effects of time since soaking rain (c) and Julian day (d) on the number of adults encountered per site. Gray shaded regions represent 95% confidence intervals.

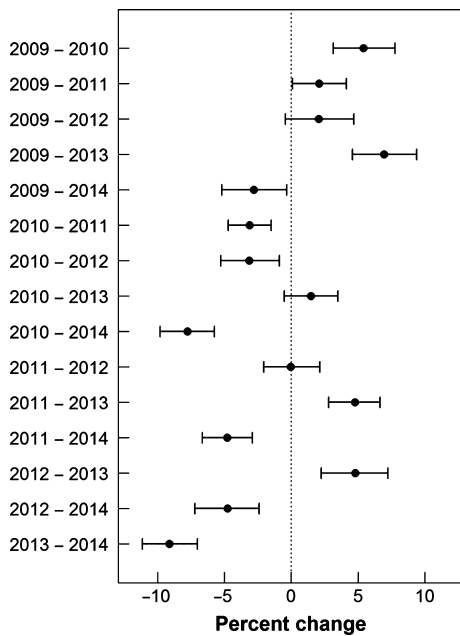


Fig. 3 Pairwise estimates of percent change among years in the mean adult body size of *P. shermani*. Error bars indicate 95% credible intervals for each pairwise difference.

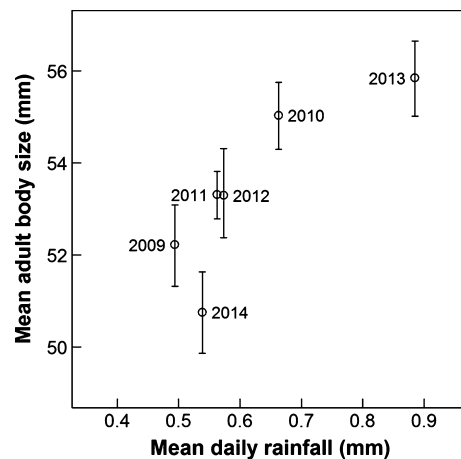


Fig. 4 Estimated mean adult body size of *P. shermani* based on a normal mixture model. Error bars indicate 95% credible intervals (CRI) for each annual estimate. Annual rainfall totals are calculated from May 15 to August 15, representing the typical survey window during our study.

greater bias in individual sample means as the 95% confidence region extends to 12.4% and 7.9% in *P. metcalfi* and *P. shermani*, respectively. Random selection of

seven or more surveys, averaging 19.0 individuals, was required to obtain an expected bias of <1% in *P. metcalfi* (~133 individuals). For *P. shermani*, four random surveys, averaging 28.2 individuals, resulted in an expected bias of <1% (~113 individuals). We also observe that, in both species, the 95% confidence region still extended to >3% bias at this threshold.

Discussion

We report considerable variation in the observed body size distributions of two plethodontid salamanders, both among years and in relation to survey-specific conditions. Significantly, we found evidence that individuals of different size varied in their relative capture

probabilities across a range of survey conditions. As a result of the high degree of variability in mean adult body size, both within and among years, large sample sizes and repeated surveys would be required to make reliable inferences concerning body size shifts for both of our salamander species. In addition, we found that individual growth rates of recaptured individuals corresponded strongly with rainfall, which indicates that context-dependent growth may be an important mechanism for rapid short-term fluctuations in salamander body size distributions. Not only do sampling variation and fluctuating population size distributions provide a complex background from which to isolate long-term trends, but the systematic effects of weather on sampling and demographic processes could potentially

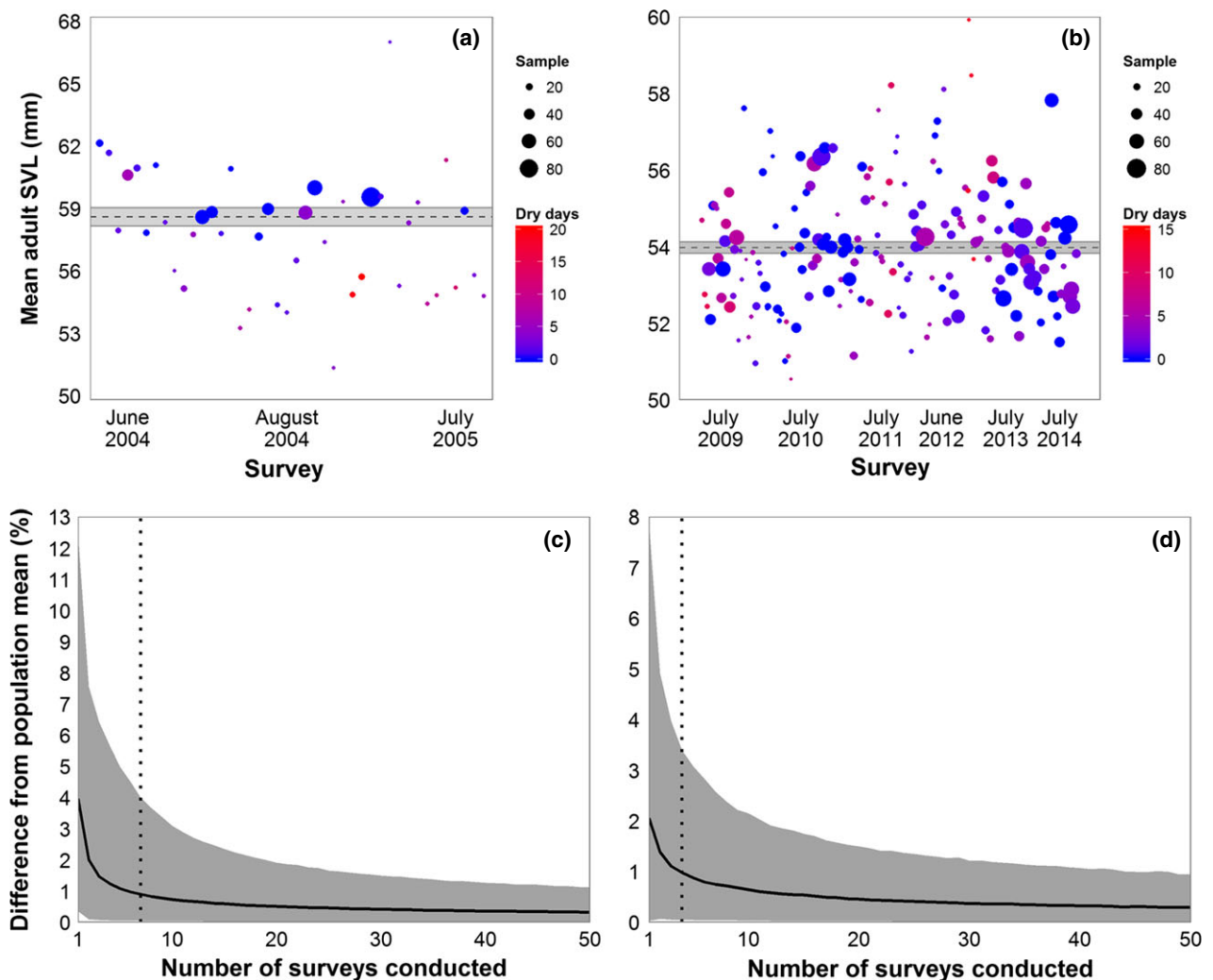


Fig. 5 Top panels represent survey means of adult body size for *P. metcalfi* (a) and *P. shermani* (b) with the gray shaded region indicating a 95% confidence band around the overall mean based on all sampled individuals. Bottom panels indicate the average bias (black line) of mean adult body size calculated from randomly selected surveys of *P. metcalfi* (c) and *P. shermani* (d). The gray shaded area indicates the 95% confidence region, and the vertical dashed line denotes where the difference between the sample mean and the overall population mean is <1%.

result in emergent patterns over decades, which might be misconstrued as directional selection.

Although selection of our study populations was nonrandom and influenced by logistical constraints and the spatial arrangement of forest management activities, we believe that the observed weather dependence of plethodontid salamander ecology and behavior are characteristics shared across populations and species. Previous research has established that evaporative water loss rates are high in plethodontid salamanders (Spight, 1968; Spotila, 1972), which limits the amount of time salamanders can spend away from moist refuges (Feder, 1983; Feder & Londos, 1984). These salamanders extensively use belowground retreats or cover objects (Taub, 1961; Petranka & Murray, 2001; Grover, 2006), and detectability of many species is related to rainfall (Petranka & Murray, 2001; Connette & Semlitsch, 2013; Peterman & Semlitsch, 2013). Thus, although imperfect observation is a reality of ecological studies across taxa, plethodontid salamanders may be particularly susceptible due to their highly fossorial nature and sensitivity to temperature and moisture conditions. This can be a serious concern when using raw counts of individuals to assess trends in population growth through time (Grant, 2014).

Our current study provides additional evidence that counts of salamanders are related to rainfall, but also that they vary temporally throughout the summer active season for *P. metcalfi*. Furthermore, we found that individuals within a population may not be equally available for sampling under certain survey conditions (a form of temporary emigration). Mean adult body size of *P. metcalfi* was strongly predicted by survey date and was negatively related to time since rainfall. Adult body size was also a significant predictor of the degree to which *P. shermani* capture probabilities declined with time since rainfall, with large adults being disproportionately active under drier conditions. Although our results suggest that differences may exist among species, the possibility for individuals of differing size to be systematically over- or underrepresented in samples due to rainfall suggests that long-term patterns in rainfall may also generate related trends in observed body size that are simply due to nonrandom sampling of individuals.

High variability in mean adult body size across surveys poses an additional problem for studies of body size change. For the species we considered, it is expected that mean body size of salamanders based on small numbers of surveys will be substantially biased relative to the mean from a much larger sample (Fig. 5). Specifically, our results indicate that >110–130 body size measurements from across 4–7 temporally spaced surveys may be required to generate reliable short-term

point estimates for mean body size of certain *Plethodon* salamanders. During our studies, such a sampling effort would have achieved an expected bias of <1% relative to the mean from more intensive sampling, with a 95% probability that such a sample will deviate by no more than approximately 4%. We note that large, pervasive body size declines are not apparent in Caruso *et al.* (2014) when we consider only species that had >130 total measurements ($N = 9$). For this calculation, we used the estimated annual change in SVL from Table 1 of Caruso *et al.* (2014; 'Slope' values) to recover the expected body size change over 55 years for each species and found an average change of just -0.57% relative to a baseline of the mean size reported for the first decade of sampling. In addition, there was a clear tendency for percent body size change to be greatest for species with fewer individual measurements and populations considered (Fig. 6). Such sample size issues have also been suggested as a likely cause for spurious identification of latitudinal clines in amphibian body size (Adams & Church, 2008) and for mischaracterization of species range shifts (Bates *et al.*, 2015).

Finally, our study demonstrates that individual growth and short-term demographic changes can be responsible for observed trends in body size. We observed complete shifts in the size ranges of the three *P. shermani* age classes among years (Fig. 1), whereas sampling bias would only be expected to influence the relative capture frequencies of individuals within a size distribution. For instance, the mean size of hatchlings, juveniles, and adults were all considerably larger in 2013, a particularly wet year, than in 2012 (+22.8%, +15.1% and +4.8%, respectively; Fig. 1). Although we have just 6 years to consider, there was a general correspondence between the amount of rainfall over the active season and the mean body size of adults (Fig. 4). The 7.0% increase in adult body size of *P. shermani* from 2009 to 2013 and subsequent 9.1% decrease from 2013 to 2014 suggest that rapid body size change in this species can be observed in less than a generation and that such changes are not likely driven by a genetic shift in size at maturity. This is evidenced by the fact that size did not change continuously over the 6-year period, but generally fluctuated up and down in correspondence with active season rainfall. We also found that the relative proportions of hatchling, juvenile, and adult individuals changed substantially through time (Fig. 1). The timing and influence of such population cycles on long-term patterns of body size are difficult to predict, but it is possible that years of high reproductive success and/or juvenile survival could subsequently result in smaller mean adult body size as large numbers of juveniles are recruited into the adult age class. Thus, it is important to recognize that a reduction

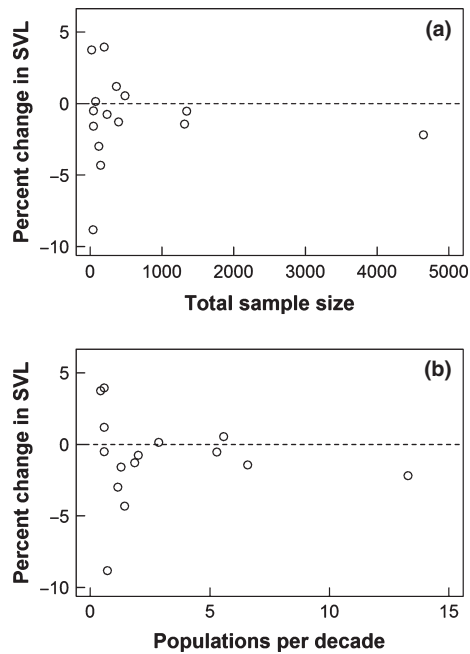


Fig. 6 Data from Table 1 of Caruso *et al.* (2014). Relationship between sampling effort and predicted percent change in snout-vent length (SVL) by species. The top panel represents sampling effort as the total number of individuals measured, while the bottom panel represents sampling effort as the mean number of populations surveyed per decade. Predicted percent change over 55 years was calculated using the estimated annual change in SVL (reported 'Slope' values from species-specific linear regression or linear mixed effects models) and the average SVL from the first decade of sampling as a reference point.

in mean body size may be due to an increased frequency of small individuals (a possible indicator of increasingly healthy, productive populations) or a change in the size and/or frequency of large individuals.

Temperature is of critical importance for ectothermic organisms, and climate warming is predicted to affect body size through numerous interacting pathways (Ohlberger, 2013). However, climate change is also a spatially and temporally complex phenomenon, and variation in local climate conditions due to landscape topography may be considerable (Dobrowski, 2011; Sears *et al.*, 2011). As a result, species may be able to behaviorally mitigate the effects of climate change, to a certain extent, by taking advantage of favorable local or microclimatic conditions (Sears *et al.*, 2011). Identification of mechanisms for body size shifts in populations may be further complicated by the fact that climate change more generally entails changes in the periodicity and intensity of precipitation events (Kundzewicz *et al.*, 2007). Our results, and those of Bendik & Gluesenkamp (2013), suggest that precipitation may be a

fundamental driver of ground surface activity, population dynamics, and individual growth rates in plethodontid salamanders. Milanovich *et al.* (2006) have also found a relationship between clutch size and annual precipitation, further highlighting the potential contribution of precipitation to population size structure. Thus, it is important to identify whether changes in body size are directly related to temperature as opposed to change in other environmental conditions (Gardner *et al.*, 2011).

Climate change has the potential to profoundly impact species, either directly by altering thermodynamics and the energetic cost of routine daily activities (Ohlberger, 2013) or indirectly by modifying food availability, predator-prey dynamics, and community composition (Durant *et al.*, 2007). Although such factors have been previously reported to influence body size distributions of populations, researchers should be aware that observed body size change can result from a variety of ecological processes (e.g. growth, selection, population dynamics), yet may also be due to imperfect observation. In particular, when individuals in a population vary in their availability for sampling under certain weather conditions, there is a high probability that observed population trends relative to climate will be substantially, if not entirely, driven by systematic bias in the sampling or observation process itself. Museum data can provide a valuable reference for historical comparison, yet failure to account for potential biases and basic ecological drivers of population change implicitly assumes that such factors contribute only to uncertainty in estimated effects (i.e. noise) rather than systematically influencing the observed dynamics of a system (e.g. Caruso *et al.*, 2014). Based on data we present in this study and the extensive literature demonstrating sampling issues in plethodontid salamanders (Hyde, 2001; Petranka & Murray, 2001; Bailey *et al.*, 2004a,b,c,d; Dodd & Dorazio, 2004; Buderman & Liebgold, 2012; Connette & Semlitsch, 2013; Peterman & Semlitsch, 2013), we believe that the correspondence of both natural ecological processes and sampling biases with rainfall has high potential to be misidentified as long-term trends of body size change and should be considered simultaneously. We would encourage researchers to objectively evaluate multiple hypotheses for observed changes in species (e.g. Daufresne *et al.*, 2009), which includes acknowledging and accounting for the fact that contributing mechanisms may be statistical in nature or artifacts of study design (Grant, 2014). Understanding the underlying causes of body size change is an important step toward appropriately directing species conservation and management efforts (Ohlberger, 2013). Because such management actions should ideally be rapid and decisive, a misguided sense

of certainty concerning existing threats could be detrimental to future management and conservation efforts due to a misdirection of resources toward problems that do not truly exist.

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Supporting Information

Additional Supporting Information may be found in the online version of this article:

Appendix S1. *A priori* models considered as predictors of mean adult body size (SVL) and counts in mixed models as well as parameter estimates for the best-supported models.
Appendix S2. Additional details on the analysis of the mixture, growth, and Cormack–Jolly–Seber models, prior specification, and R code.