

## Article

# Age structure of a lizard along an elevational gradient reveals nonlinear lifespan patterns with altitude

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## Abstract

Lifespan is one of the main components of life history. Shorter lifespans can be expected in marginal habitats. However, in the case of ectotherms, lifespan typically increases with altitude, even though temperature—one of the main factors to determine ectotherms' life history—declines with elevation. This pattern can be explained by the fact that a shorter activity time favors survival. In this study, we analyzed how lifespan and other life-history traits of the lizard *Psammotromus algirus* vary along a 2,200 m elevational gradient in Sierra Nevada (SE Spain). Populations at intermediate altitudes (1,200–1,700 m), corresponding to the optimal habitat for this species, had the shortest lifespans, whereas populations inhabiting marginal habitats (at both low and at high altitudes) lived longest. Therefore, this lizard did not follow the typical pattern of ectotherms, as it also lived longer at the lower limit of its distribution, nor did it show a longer lifespan in areas with optimal habitats. These results might be explained by a complex combination of different gradients along the mountain, namely that activity time decreases with altitude whereas food availability increases. This could explain why lifespan was maximum at both high (limited activity time) and low (limited food availability) altitudes, resulting in similar lifespans in areas with contrasting environmental conditions. Our findings also indicated that reproductive investment and body condition increase with elevation, suggesting that alpine populations are locally adapted.

**Key words:** age structure, elevation, life history, longevity, marginal habitats, relative clutch mass

The habitat of a particular species may be defined as a set of resources and conditions needed for survival and reproduction of individuals of that species (Chase and Leibold 2003). Accordingly, the central–marginal hypothesis states that zones with optimal or near-optimal conditions can be referred to as core habitats. Nevertheless, as one moves away from the core habitat areas, the environment usually becomes progressively less suitable for the species, implying lower survival probability and/or reproductive success, and hence decreased fitness

(Pironon et al. 2017). These habitats have border conditions that the species can tolerate for survival and reproduction and are, therefore, considered suboptimal or marginal (Kawecki 2008).

Core and marginal habitats for a given species can be found along altitudinal gradients. Mountain environments harbor a high level of ecological heterogeneity because several abiotic factors change with altitude; mainly, temperature and the partial pressure of oxygen decrease with altitude, whereas solar radiation increases

(Barry 2008). These abiotic factors exert selective pressures on animals and plants, causing communities to vary along the elevational gradient (e.g., Carothers et al. 2001; Navas 2002; Fu et al. 2007). Hence, a species inhabiting an elevational gradient may occupy core habitats as well as marginal ones in a relatively small geographical area. As such, elevational gradients provide researchers with a natural experimental setting to study how life-history varies according to habitat quality.

One of the main life-history traits is lifespan, which is both influenced by and influences other life-history traits, as it has a direct effect on several ecological and evolutionary outcomes (Metcalfe and Pavard 2007). Lifespan primarily depends on extrinsic mortality (Cichoń 1997), but life history theory suggests that it could also be shortened by selection for greater reproductive investment (Araya-Ajoy et al. 2018). Although lifespan shows a marked geographical variation (Valcu et al. 2014), we only have a limited understanding of how it varies with elevation in species distributed across a large range of altitudes and the causes of this variation. The available studies generally assume lifespan varies linearly with altitude and provide contradictory results where lifespan lengthens, shortens or remains relatively unchanged along the altitudinal gradient (e.g., review for birds in Boyle et al. 2016).

Furthermore, the elevational pattern in lifespan could differ between ectotherms and endotherms, because temperature, which is the main abiotic factor to vary with elevation (Körner 2007), has a more significant effect on the physiology of ectotherms than that of endotherms (Angilletta 2009). Environments become harsher at high elevations, where storms, strong winds, and snow are frequent, resulting in low thermal quality and long hibernation periods. This scenario may bring about early death and, therefore, reduce lifespan (Sears 2005). Most studies on ectotherms, however, report that lifespan increases with altitude (Zhang and Lu 2012). This pattern is attributed to several concomitant factors: 1) shorter activity seasons at high elevations, which reduces metabolic damage and time exposed to predators; 2) reduced predator pressure with altitude; and 3) changes in life history, as populations at high elevations often lead a slower pace of life and invest less in reproduction and more in self-preservation (review in Cabezas-Cartes et al. 2018).

In this study, we evaluate how lizard lifespan varies across a wide elevational gradient by studying the lizard *Psammotromus algirus* in the Sierra Nevada mountain (SE Spain). We assumed that habitat quality for this species in Sierra Nevada is maximum at mid-elevations (1,200–1,700 m) and diminishes as the species ascends or descends the elevational gradient. This assumption is based on 2 lines of evidence. First, thermal quality is one of the main factors affecting habitat quality in ectotherms (Angilletta 2009), and for this species in Sierra Nevada it becomes maximal at intermediate elevations (Zamora-Camacho et al. 2016). Second, population density is typically lower in marginal habitats than in core ones (Sagarin et al. 2006) and the density of this species in Sierra Nevada is highest at mid-elevations (Zamora-Camacho et al. 2013). Different environmental conditions can lead to a suboptimal habitat and hence, depending on the trait or traits affecting lizard fitness and how they vary with elevation, lifespan may present different elevational patterns. Accordingly, in this study, we tested competitive hypotheses developed to explain how lifespan varies with elevation in *P. algirus* (Table 1).

The central–marginal hypothesis: This hypothesis affirms that fitness is lower in low-quality habitats because adult survival decreases with habitat quality. As such, we expect the maximum lifespan in the core habitat (mid-elevations), with lifespans being shorter in marginal habitats situated at both high and low elevations. Adult survival may be shorter at high elevations due to harsh

and extreme climate conditions (Sears 2005). Meanwhile, at the lower limit, interspecific competition is often more intense (Comas et al. 2014), which can have negative consequences for adult survival (Dunham 1980). Moreover, at low elevations, predation risk may increase due to the presence of more and a greater diversity of predators (Fox et al. 1994), whereas at high elevations it can increase due to greater conspicuousness since more time is devoted to thermoregulation (Alford and Lutterschmidt 2012).

The resource allocation hypothesis: this hypothesis claims that fitness is higher in better quality habitats because they favor successful reproduction (this is also predicted by the central–marginal hypothesis). So, animals could be selected for greater investment in reproduction in core habitats, hence allocating fewer resources to self-preservation and thereby reducing lifespan (Kirkwood and Rose 1991). For example, *Crinia pseudinsignifera* frogs invest more in reproduction when living in more favorable areas, reducing their lifespan with respect to harsher areas (Reniers et al. 2015). In such cases, the lifespan would be longer in marginal habitats than in core habitats. The assumptions made in this hypothesis are supported by studies in other populations of *P. algirus*, which suggest that reproductive success is impaired at both high and low elevations. In highlands, low soil temperatures delay hatching and harm hatchlings' body condition (Monasterio et al. 2011), whereas at lower altitudes, hatchling survival is lower as a consequence of scarcer food availability (Iraeta et al. 2006). Moreover, predators are typically more abundant in lowland areas and tend to have a greater impact on juveniles than on adults (Ballinger 1979). In addition, juvenile mortality is very high during hibernation (Civantos and Forsman 2000), which is longer at high elevations.

The rate-of-living hypothesis and the activity-exposition hypothesis: the relevance of temperature for ectotherms may prevail, as it is the main determinant of lifespan across the gradient. Indeed, ectotherms frequently show an increased lifespan with altitude and when inhabiting cold environments in general (Morrison and Hero 2003; Munch and Salinas 2009; Zhang and Lu 2012; Scharf et al. 2015; Cabezas-Cartes et al. 2018; Stark et al. 2018). This generalized pattern can be explained by 2 different hypotheses. According to the rate-of-living hypothesis, in cold environments, ectotherms live through shorter activity seasons, which reduce metabolic damage and consequently increase survival and lifespan (Speakman 2005). Supporting this contention, Bestion et al. (2015) experimentally showed that increased temperatures in the lizard *Zootoca vivipara* enhances growth and reproductive investment, resulting in reduced longevity. Furthermore, according to the activity-exposition hypothesis, reduced activity would also reduce exposure to predators, and therefore increase survival at high altitudes (Adolph and Porter 1993). Several studies with ectotherms show that longevity increases for shorter activity seasons (Cvetković et al. 2009; Liao et al. 2016; Cabezas-Cartes et al. 2018), although the exact mechanism behind this pattern is still not completely known. In our study population, the length of the activity season decreases with elevation (Zamora-Camacho et al. 2013), and, as an evidence of reduced metabolic damage, oxidative stress also decreases with elevation (Reguera et al. 2014a, 2015). In other *P. algirus* populations, lizard survival during the activity period was lower at low altitudes, presumably due to longer activity time (Iraeta et al. 2015).

In general, different demographics and life-history traits are expected near the upper and lower distribution boundaries with respect to the core distribution. Accurate assessments of lifespan are, therefore, necessary to discern between competing hypotheses concerning the nature of selective forces driving life-history evolution.

**Table 1.** Summary of the hypotheses used to explain altitudinal lifespan patterns in lizards with an explanation of each hypothesis and its prediction for our study system

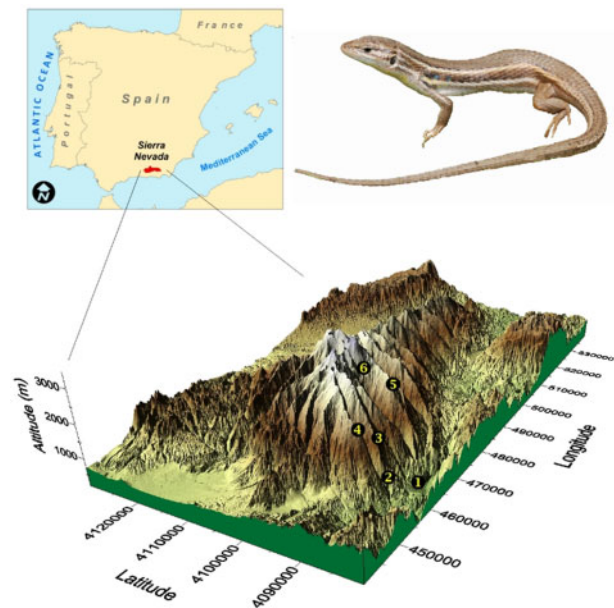
Hypotheses	Assumptions and predictions	Altitudinal pattern predicted
The central–marginal hypothesis	Adult survival decreases with decreased habitat quality	∩-shaped pattern
The resource allocation hypothesis	Animals are selected for greater investment in reproduction in core habitats, allocating fewer resources to self-preservation and thereby reducing lifespan	U-shaped pattern
The rate-of-living hypothesis	In cold environments, the length of activity seasons for ectotherms is shorter and reduced activity implies lower metabolic damage, thereby increasing survival and lifespan	Linear increase
The activity exposition hypothesis	In cold environments, the length of activity seasons for ectotherms is shorter and reduced activity implies lower exposure to predators, which increases survival	Linear increase

Indeed, the 3 hypotheses make different predictions on how lifespan should vary with elevation in our study system (Table 1): peak at mid-elevations (central–marginal hypothesis), U-shaped (resource allocation hypothesis), and linear increase (both the rate-of-living hypothesis and the activity-exposition hypothesis). In this study, we estimate the age structure (by means of skeletochronology) of a population of *P. algirus* across 2,200 m of an elevational gradient. Moreover, to disentangle the causes of elevational variation in lifespan, we present additional data to test the assumption that habitat quality is optimal at mid-elevations. Specifically, to differentiate between core and marginal populations, we test for different proxies of habitat quality (following Hoffmann and Blows 1994): the proportion of juveniles (which is expected to be higher in optimal habitats), population density, and a measure of a fitness-related trait such as body condition. Furthermore, given that the resource allocation hypothesis implies longer lifespans in marginal habitats as a consequence of life-history trade-offs, this hypothesis also predicts greater reproductive investment (estimated as relative clutch mass) in core habitats.

## Material and Methods

### General procedures

The lizard *P. algirus* is a medium-large lacertid (53–80 mm snout-vent length [SVL], in our study area) that inhabits shrubby habitats in the Mediterranean region of south-west Europe and north-west Africa (Salvador 2015). The fieldwork was performed in the Sierra Nevada mountain system (SE Spain), where *P. algirus* is found from 200 to 2,600 m above sea level (hereafter, m asl) (Fernández-Cardenete et al. 2000). We sampled from 6 sites, at 300, 700, 1,200, 1,700, 2,200, and 2,500 m asl (Figure 1). Lizards were captured by hand during their activity season in Sierra Nevada, which spans from March to September (Zamora-Camacho et al. 2013). We assessed a total of 125 individuals over 4 years (sample size per year; 2010: 9, 2011: 39, 2012: 72, and 2013: 5 individuals). We tried to assess equal numbers of each sex at each elevation (samples sizes of females/males for each altitude: 300, 12/11; 700, 11/8; 1,200, 10/10; 1,700, 11/10; 2,200, 9/10; 2,500, 11/12). Males were distinguished by their wider heads, larger and more numerous femoral pores in the hind limbs, and orange spots in the corners of their mouths (Carretero 2002; Iraeta et al. 2011). Because the lizards were part of a long-term study, they were marked by toe clipping. These toe samples were used to estimate lizards' age using phalanx skeletochronology (more details below). Toe clipping is a marking



**Figure 1.** Location of the Sierra Nevada mountain range in the Iberian Peninsula (top, left panel) and a 3-dimensional map of Sierra Nevada (lower panel), showing the location of the sampling sites (1–6 correspond to the sites at 300, 700, 1,200, 1,700, 2,200, and 2,500 m asl, respectively). An image of the lizard *P. algirus* appears in the top-right panel.

method frequently used in lizards with limited impact on their welfare (Perry et al. 2011).

We measured the lizards' SVL to the nearest 1 mm with a metal ruler and body mass to the nearest 0.01 g with a digital balance (Model Radwag WTB200). With these data, we estimated the body condition index (BCI) as the residuals of regressing log mass on log SVL. This is a widely used index that represents the relative energy reserves of an animal (Schulte-Hostedde et al. 2005). Lizards often detach their tails as a defensive mechanism. The lack of a complete tail could affect BCI calculation. However, a multiple regression with individuals possessing a full tail showed that most of the body mass was explained by SVL (partial correlation  $r = 0.85$ ,  $r^2 = 0.72$ ,  $P < 0.001$ ), tail length having a nonsignificant effect on body mass (partial correlation  $r = 0.11$ ,  $r^2 = 0.01$ ,  $P = 0.32$ ). Therefore, the presence of individuals with a partial tail should have a negligible effect on the BCI estimation.

In 2010, to quantify lizard relative abundance, we sampled 500-m transects every 2 weeks at each sampling site during the annual

activity season. Censuses were repeated every 2 h from sunrise until sunset. We recorded the number of active adults and juveniles seen in each transect. Juveniles were discriminated from adults based on body size and coloration, especially the tail (redder in juveniles). We assumed lizard detectability to be the same at all sampling stations and that the number of active individuals counted by this procedure correlated positively with the real population density (Blomberg and Shine 1996). From these transects, we estimated relative abundance at each site as the mean number of adults detected. We also estimated the percentage of juveniles and took it as a birth rate indicator for the population.

A subset of gravid females ( $n=102$ ) not included in previous analyses was used to estimate reproductive investment along the elevational gradient. We recognized gravid females by manual palpation of developing eggs. Gravid females were transported to a lab and placed in individual terrariums ( $100 \times 20 \times 40$  cm) with water (in form of aqueous nutritious gel) and food (*Tenebrio molitor* larvae) *ad libitum*, indirect access to sunlight, and a heat cable at one end of the cage, switched on 3 h/day (11–14 h) to allow thermoregulation. The substrate was bare soil from the study area. When females laid eggs, we recorded clutch mass and estimated relative clutch mass, an indicator of their reproductive investment (Shine 1980), as a percentage of female body mass. Females and their offspring were released at the point where the female had been caught. No lizard died or suffered permanent injury in this study.

### Skeletochronology age estimation

The age of the lizards was determined by phalanx skeletochronology (Comas et al. 2016), which is one of the most accurate age estimation techniques in animals (Zhao et al. 2019). Ectotherms with indeterminate growth may present a cyclic growth pattern in hard body structures, corresponding to alternate periods of growth and resting. Therefore, age can be estimated by examining cyclic growth patterns in bones (Figure 2). Phalanx skeletochronology provides age estimation by counting annual growth rings in the phalanges (Comas et al. 2016). One toe of each lizard was clipped and preserved in ethanol 70%, after which the wound was properly disinfected with chlorhexidine. The toes were decalcified in 3% nitric acid for 3.5 h. Cross-sections ( $10 \mu\text{m}$ ) were prepared using a freezing microtome (CM1850 Leica) at the Centre of Scientific Instrumentation, University of Granada. Cross-sections were stained with Harris hematoxylin for 20 min, dehydrated through an alcohol chain (70%, 96%, 100%; 5 min each), and washed in xylol for 15 min. They were then fixed with DPX (histology mounting medium), mounted on slides, and examined for the presence of lines of arrested growth (LAGs) using a light microscope (Leitz Dialux 20, Leica Microsystems, Wetzlar, Germany) at  $400\times$  magnification. We

took 10–20 photographs (with a ProgresC3 camera) of several representative cross-sections for each individual, discarding any cuts with unclear LAGs. We selected diaphyseal sections where the size of the medullary cavity was at its minimum and that of the periosteal bone at its maximum (Comas et al. 2016). The number of LAGs detected in the periosteal bone was counted on 3 separate occasions by the same person (MC) while blinded to the specimen identification. Each LAG may approach 1 year of life, so the number of LAGs indicates the lizard's approximate age with an accuracy of  $\pm 1$  year (Figure 2).

We used the skeletochronological data to estimate adult lifespan, that is, the expected average longevity of individuals that had reached maturity, by means of Seber's (1973) formula:  $\text{Lifespan} = 0.5 + 1/(1-S)$ , where  $S$  is the survival rate. Survival rate was calculated according to Robson and Chapman's (1961) formula:  $S = T/(R + T - 1)$ , where  $S$  is the finite annual survival rate estimate,  $T = N_1 + 2N_2 + 3N_3 + 4N_4$  and so on to complete age classes,  $R = \sum N_i$ , and  $N_i$  is the number of individuals in the age class  $i$ .

### Statistical analyses

A chi-square test was used to test for any differences in age structure between sexes or elevations. Since there were no 5-year-old males, we used 4 age categories to avoid creating cells with a value of 0: 1, 2, 3, and  $>3$  years (4 and 5 years together). To examine simultaneously the effect of sex and altitude on age, we used 2 approximations. On the one hand, we tested whether the lizards' average age varied with elevation and sex by using an ANOVA, taking altitude (6 levels, corresponding to the 6 sites sampled), sex (2 levels), and interaction as factors. We also employed a multinomial model with age (4 levels) as the dependent variable, and altitude (6 levels), sex (2 levels), and their interaction as predictors. To consider possible cohort effects, we repeated the previous analyses including the year of capture (2011 and 2012) as a factor (years 2010 and 2013 were not included in this analysis because of the small sample size). Analysis of variance (ANOVA) was also used to test for elevational variation in relative abundance, percentage of juveniles, BCI, and relative clutch mass. In these analyses, percentage of juveniles and relative clutch mass were arcsine-transformed (Quinn and Keough 2002). Data were checked for outliers, normality, and homoscedasticity following Zuur et al. (2010).

### Results

The age structure of the lizards did not differ between sexes ( $\chi^2_3 = 1.47$ ,  $P = 0.69$ ; Figure 3A; sample sizes given in the figure). Nevertheless, females had a maximum lifespan of 5 years and males

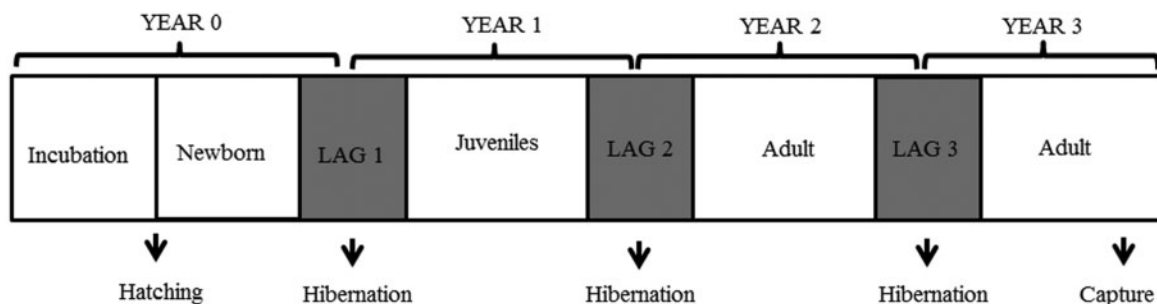
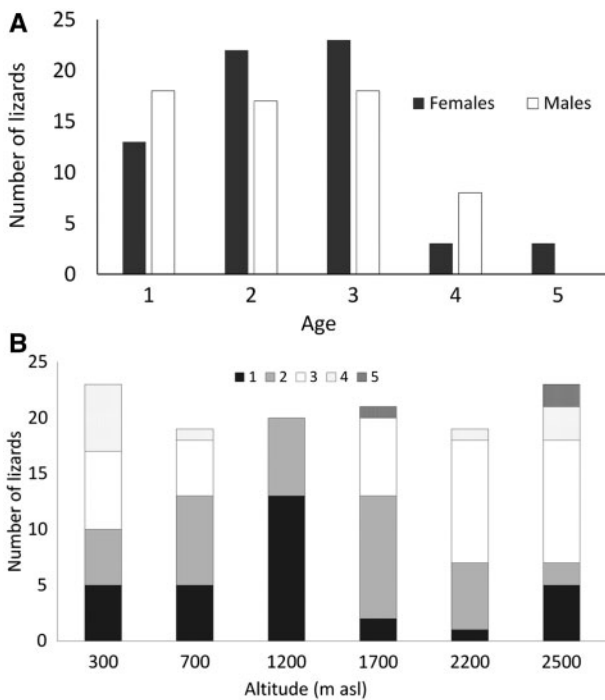


Figure 2. Life cycle of the lizard *P. algirus* with the example of a lizard estimated to be 3 years old.





**Figure 3.** (A) Frequency (number of lizards) of female (black bars) and male (white bars) lizards according to estimated age. Age structure is similar between sexes, but only females reached the age of 5. (B) Frequency of lizards at each age class (black: 1 year; gray: 2 years; white: 3 years; dotted: 4 years; hatched: 5 years) according to elevation.

of 4 years. The lizards showed a similar frequency of individuals aged 1, 2, and 3 years, but there was a decrease of almost 50% in the number of lizards reaching the age of 4 years. The age frequency distribution varied significantly in function of altitude ( $\chi^2_{15} = 36.58$ ,  $P = 0.001$ ; Figure 3B; sample sizes given in the figure). The frequency of individuals aged  $\geq 4$  years was lower at medium elevations than at low and high ones (only 1 of the 41 individuals at mid altitudes  $> 3$  years old, versus 7/42 at low and 6/42 at high elevations). The annual survival rate was  $\sim 0.70$  at each elevation, except at 1,200 m where the rate was 0.60. Similarly, the lifespan was  $\sim 4$  years at each elevation, except at 1,200 m where it was only 3 years (Figure 4).

The lizards' mean age varied with elevation ( $F_{5,113} = 5.89$ ,  $P < 0.001$ ;  $n = 125$ ), following a U-shaped pattern (Figure 5). Mean age did not differ with sex ( $F_{1,113} = 0.99$ ,  $P = 0.32$ ). However, the pattern with altitude differed slightly between sexes, males having a higher average age than females at 300 m, whereas females were older than males at all other elevations (interaction sex  $\times$  altitude,  $F_{5,113} = 3.55$ ,  $P = 0.005$ ; Figure 5). The multinomial model gave similar results, with a significant effect of elevation ( $\chi^2_{15} = 40.12$ ,  $P = 0.0004$ ) and the interaction sex  $\times$  altitude ( $\chi^2_9 = 21.03$ ,  $P = 0.01$ ) on lizard age, but no effect of sex ( $\chi^2_3 = 1.81$ ,  $P = 0.61$ ). When the analyses were repeated including year of capture as a factor, the results were qualitatively the same (data not shown for simplicity), with no significant effect of year or the interactions year  $\times$  altitude, year  $\times$  sex, and triple interaction. There were no differences in age structure with elevation between 2011 and 2012 ( $\chi^2_5 = 1.18$ ,  $P = 0.95$ ).

The relative abundance of adult lizards varied significantly with elevation ( $F_{5,49} = 5.09$ ,  $P < 0.001$ ,  $n = 55$  samplings), presenting a maximum at mid-elevations (1,200 and 1,700 m; Figure 6). The

percentage of juveniles detected in transects ranged between 43.4% at 2,500 m and 74.3% at 2,200 m, but did not differ significantly between elevations ( $F_{5,34} = 0.78$ ,  $P = 0.57$ ,  $n = 40$  samplings in which at least 1 juvenile was detected; Figure 6). Meanwhile, body condition was minimal at 700 m, and improved with elevation ( $F_{5,119} = 3.26$ ,  $P = 0.0085$ ,  $n = 125$ ; Figure 7). The reproductive investment registered minimal values at low elevations and followed a tendency to increase with elevation ( $F_{5,96} = 2.43$ ,  $P = 0.04$ ,  $n = 102$  gravid females; Figure 8).

## Discussion

Our findings (summarized in Table 2) show that the age structure of the lizard *P. algirus* in the Sierra Nevada mountain changes with altitude following a curvilinear pattern: populations at low and high elevations (presumed to be marginal habitats) harbor older individuals in comparison with populations at mid-elevations. We discarded a cohort effect because the elevational age structure did not vary with sampling year and the altitudinal effect remained after controlling for year of capture. The findings cannot be easily explained through either nonlinear clines in predators or parasites in our study system. Although we have no accurate data on predator pressure, anecdotal observations during fieldwork suggest that predator abundance is lower at high elevations (see also Fox et al. 1994; Camacho and Avilés 2019). Meanwhile, parasites showed a complex pattern: the prevalence of ectoparasites (mites) decreased linearly with ascending elevation, whereas hemoparasites increased linearly (Álvarez-Ruiz et al. 2018). Consequently, the altitudinal pattern observed in age structure initially appears to support the resource allocation hypothesis (see Table 1), which postulates that lizards should have a faster pace of life, invest less in self-preservation and, therefore, present shorter lifespans in core habitats where reproduction is favored. However, the resource allocation hypothesis relies on the assumption of higher reproductive investment at mid-elevations, but our data did not support this prediction, given that reproductive investment, measured as relative clutch mass, tended to increase with altitude. In fact, the resource allocation hypothesis is based on the life-history theory, which predicts an inverse relationship (i.e., a trade-off) between lifespan and reproductive investment (Stearns 1992; Roff 2002). Although such a trade-off has been evidenced in reptiles in general (Scharf et al. 2015), it is unclear whether it applies to lacertids (Bauwens and Díaz-Urriarte 1997). Therefore, the observed pattern seems more complex than predicted by the competitive hypotheses presented in the Introduction, and the longer lifespan in highland and lowland lizards could be due to different ecological processes.

An initial question is why *P. algirus* lizards live longer at high elevations than at intermediate elevations. We assumed that alpine zones constitute marginal habitats because lizard density and thermal quality were lower than those at the middle elevations (Zamora-Camacho et al. 2013, 2016). Moreover, *P. algirus* is a lacertid typical of North Africa and Mediterranean environments in the Iberian Peninsula (Carranza et al. 2006), and thus, a priori, it is presumed to be poorly adapted to alpine habitats (see Monasterio et al. 2011). However, while data presented in this study support that lizard densities are greater at middle rather than high elevations, other proxies of habitat quality show a more complex picture: the proportion of juvenile lizards, as a measurement of population growth, did not differ across elevations, whereas body condition increased with altitude. This calls into question the assumption that alpine habitats are suboptimal for *P. algirus*. In our study

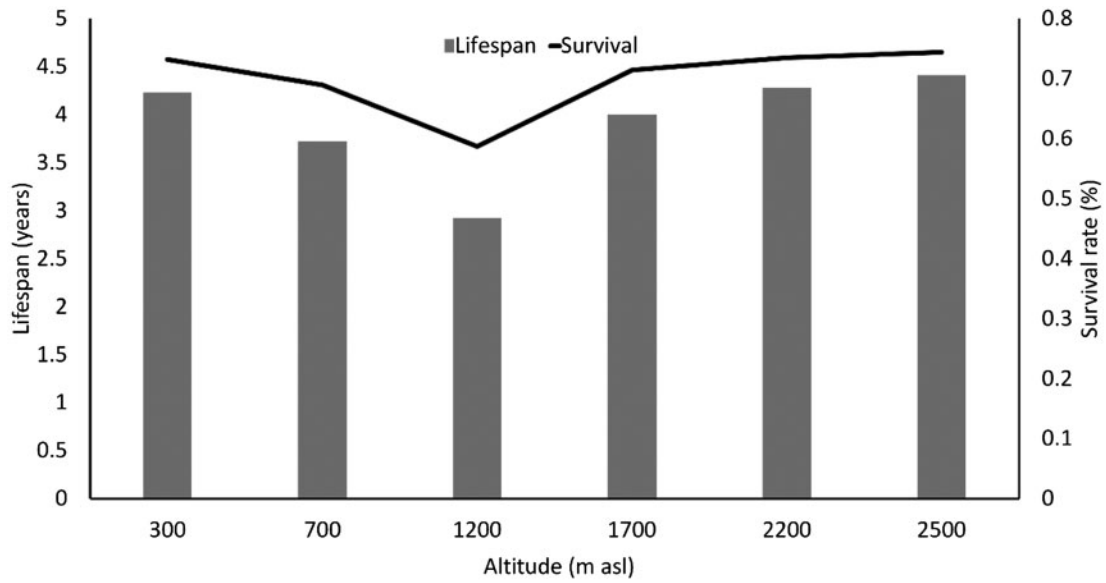


Figure 4. Estimated lifespan (gray bars) and survival rate (black line) for *P. algirus* lizards according to elevation (m asl).

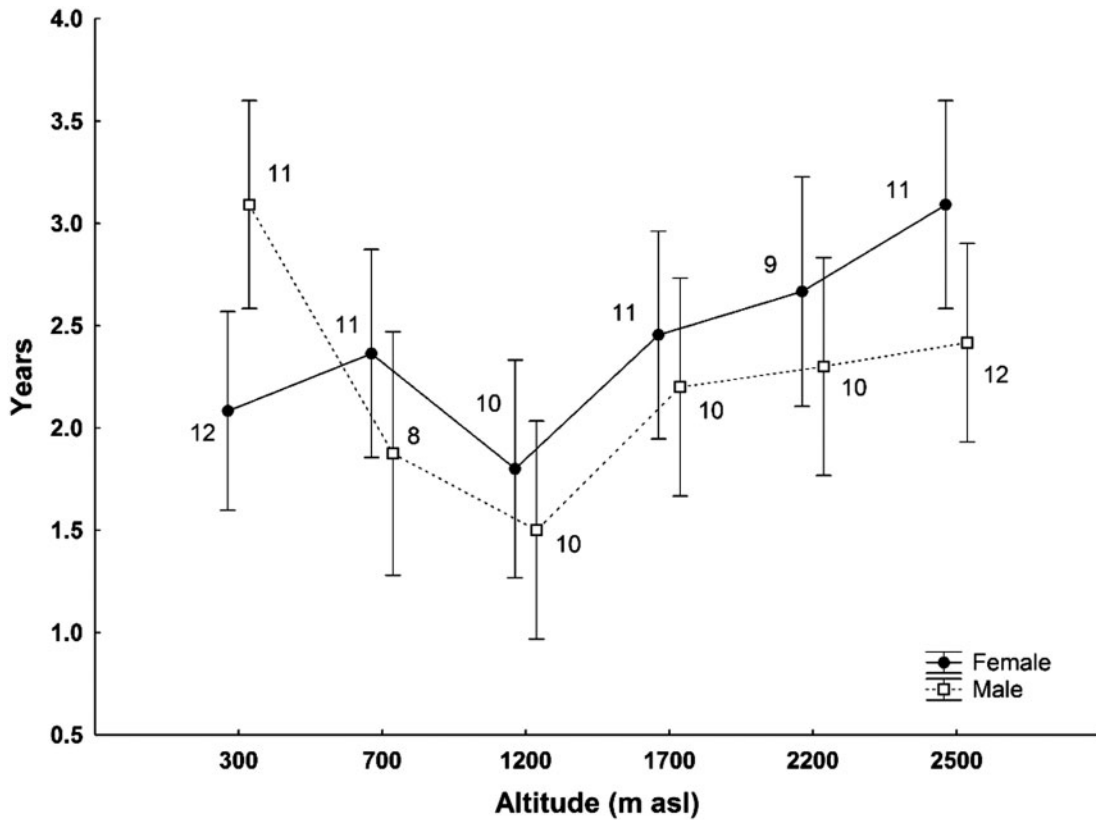
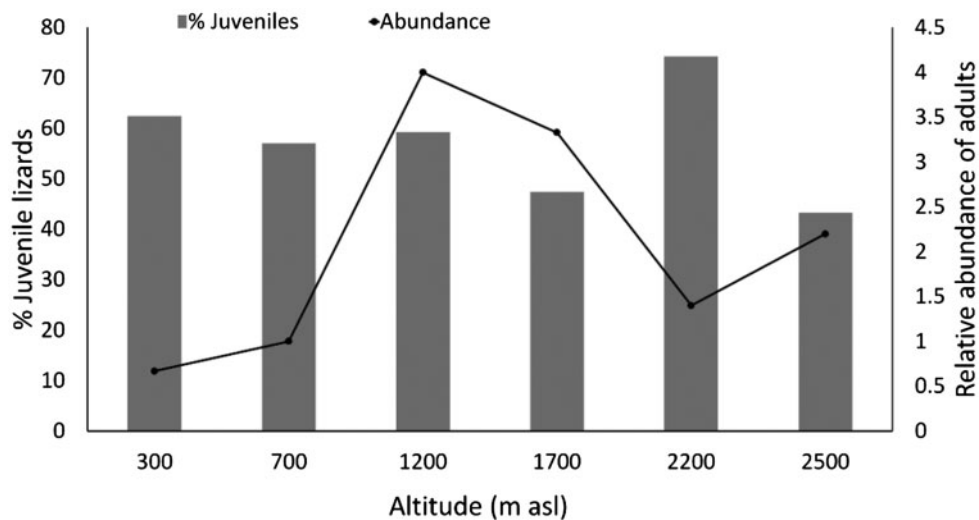


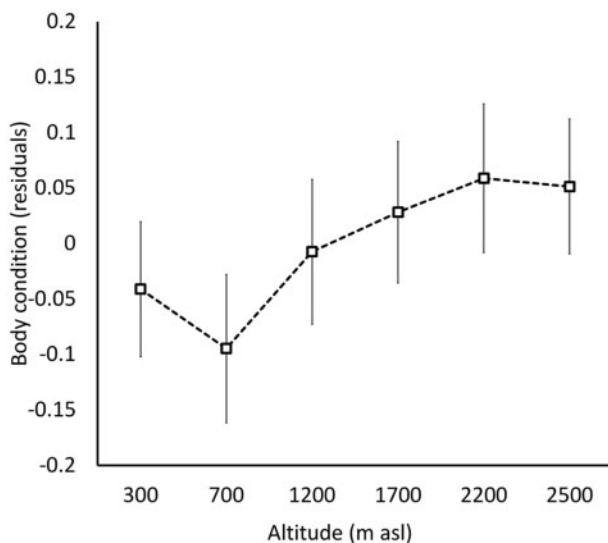
Figure 5. Average age (with 95% confidence interval (CI), vertical bars) of female (black dots, solid line) and male (white squares, dashed line) lizards depending on altitude. Sample size for each category is indicated on the graph, close to the corresponding data point.

population, alpine lizards show a number of phenotypic traits, well-differentiated from lizards at middle and low altitudes, such as a darker coloration and larger body size, which appear to be adaptations to cope with alpine habitats by improving thermoregulation in cold environments (Reguera et al. 2014b; Zamora-Camacho et al. 2014). These adaptations may mean this lizard is locally adapted to

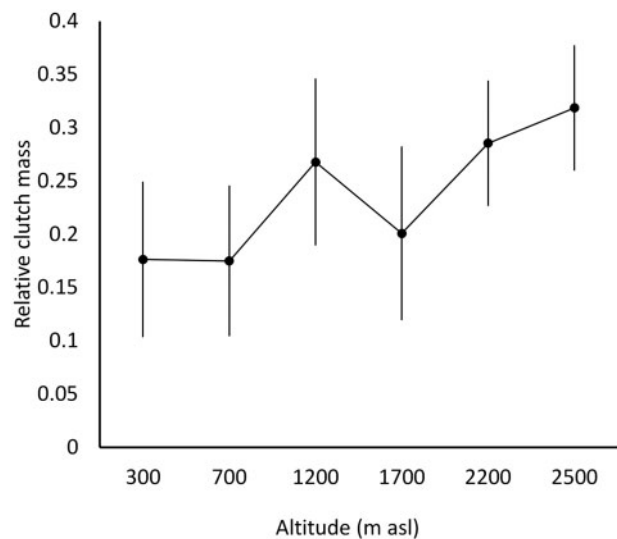
alpine zones in Sierra Nevada, thus the alpine zone may not be a suboptimal habitat. Furthermore, food availability is greater at higher elevations in our study system (Moreno-Rueda et al. 2018). So, the increased food availability in the alpine zone (Moreno-Rueda et al. 2018), in combination with low oxidative stress (Reguera et al. 2014a, 2015) and activity time (Zamora-Camacho



**Figure 6.** Maximal abundance values registered in transects for adult lizards (black line) and percentage of juvenile lizards (gray bars) depending on altitude.



**Figure 7.** Average body condition (residuals of the body mass regarding the SVL, both log-transformed) with 95% CI (bars) in function of altitude.



**Figure 8.** Average reproductive investment with 95% CI (estimated as relative investment in the clutch) depending on altitude.

et al. 2013), implies highland lizards could invest more in reproduction without a cost in the form of reduced lifespan.

By contrast, our findings support the idea that lowlands harbor suboptimal habitat for the lizard *P. algirus*. In lowlands, thermal quality and food availability were the lowest, whereas oxidative stress was maximal (Table 2). Consequently, lizard density, body condition, and reproductive investment were the lowest (Table 2). In fact, in Mediterranean environments, lowlands show low precipitation and high temperatures during summer, which can be very restrictive for lizards. Indeed, several studies in Mediterranean areas report a lower food availability and growth rate in lowland habitats than those at 1,200–1,800 m, which is the midland range in our study area (Iraeta et al. 2006; Ortega et al. 2015, 2017). However, in spite of the harmful environmental conditions and the long activity time (Zamora-Camacho et al. 2013), lifespan was not the shortest in the lowlands. A possible explanation is that the lower food availability would lead to poor body condition and so too low reproductive investment (see Bronikowski and Arnold 1999), thereby

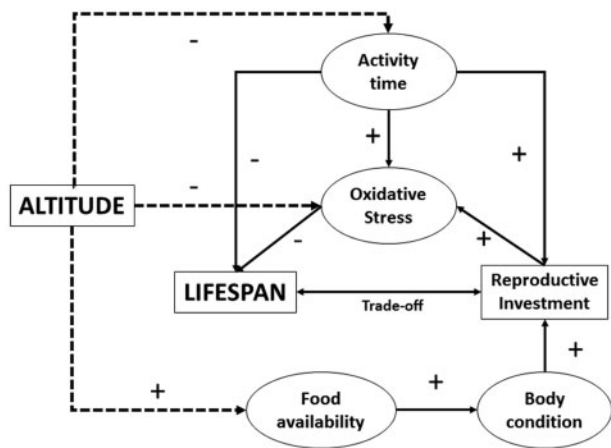
lengthening lifespan and balancing the negative impact derived from high activity time and oxidative damage (Figure 9).

Alternatively, the altitudinal pattern found for age structure could be a consequence of elevational variation in intraspecific competition. Intraspecific competition may be an important selective agent (Calsbeek and Cox 2010), and strong intraspecific competition may reduce survival (Balbontín and Møller 2015), at least under certain circumstances. In fact, *P. algirus* is a strongly territorial lizard in which aggressions are frequent (Civantos 2000). Effectively, when density is high, aggressions between lizards may affect their survival (Le Galliard et al. 2005). Consistent with this idea, *P. algirus* lifespan and survival were the lowest at mid-elevations, where abundance was the highest.

Our results also reveal that the lizards had similar survival rates until they were 3 years old and then survival declined sharply. That is, few lizards reached 4 years old and only 3 females attained the maximum lifespan of 5 years in our study area. The peak of mortality after 3 years may be a consequence of senescence. Senescence

**Table 2.** Summary of results describing altitudinal pattern in this study and in previous studies performed in the same study system

Variable	Altitudinal pattern	Reference
This study		
Mean age	U-shaped pattern	
Annual survival	U-shaped pattern	
Lifespan	U-shaped pattern	
Lizard abundance	∩-shaped pattern	
Percentage of juveniles	No altitudinal pattern	
Body condition	Increased with altitude	
Reproductive investment	Increased with altitude	
Previous studies		
Thermal quality	∩-shaped pattern	Zamora-Camacho et al. (2016)
Lizard abundance	∩-shaped pattern	Zamora-Camacho et al. (2013)
Activity season length	Decreases with altitude	Zamora-Camacho et al. (2013)
Oxidative stress	Decreases with altitude	Reguera et al. (2014a, 2015)
Ectoparasites (mites)	Decreases with altitude	Álvarez-Ruiz et al. (2018)
Hemoparasites	Increases with altitude	Álvarez-Ruiz et al. (2018)
Dorsal coloration	Darker with altitude	Reguera et al. (2014b)
Body size	Increases with altitude	Zamora-Camacho et al. (2014)
Food availability	Increases with altitude	Moreno-Rueda et al. (2018)

**Figure 9.** Flow chart of the interactions that could explain the elevational variation in lifespan of the lizard *P. algirus*. There is a trade-off between lifespan and reproductive investment. Activity time and oxidative stress decrease with altitude, whereas food availability increases. Both activity time and oxidative stress have negative effects on lifespan; activity time increases reproductive investment, which, in turn, increases oxidative stress. Meanwhile, food availability improves body condition, which at the same time boosts reproductive investment.

implies a deterioration of physiological conditions in older individuals, resulting in greater mortality (Massot et al. 2011). In fact, senescence often implies a deterioration of the immune system (Zamora-Camacho and Comas 2018), which also leads to higher mortality

due to pathogens and parasites. However, senescence is strongly determined by telomere length (Hausmann and Marchetto 2010) and, in our study population, telomeres lengthen up to 4 years old, and then shorten (Burraco et al. 2019). Therefore, it is still unclear why mortality sharply increases when lizards are 4 years old.

In conclusion, our findings contrast with most of those published to date on lizards (and ectotherms in general), which typically report greater longevity at higher altitudes. As summarized in Figure 9, several factors may affect lifespan in complex ways. Consequently, a pattern of enhanced longevity with altitude is not universal and our study highlights some causes that could be responsible for exceptions to the rule.

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