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Peaks of Otter Salamander (*Plethodon hubrichti*) Condition Declines along an Elevational Gradient

Olivia de Araujo, Jesse Hughes, Josh Twiddy, Liam Cusack, Cass Rupert, Timothy R. Brophy PhD, Paul Sattler PhD, and Norman Reichenbach PhD

Abstract

The Peaks of Otter Salamander, *Plethodon hubrichti*, is a montane species found at elevations above 442 m within a 117 km² area of the Blue Ridge Mountains in central Virginia, USA. In allopatric areas (areas without the Eastern Red-backed Salamander, *P. cinereus*, a potential competitor) salamander condition was hypothesized to decrease at lower elevations due to increased temperatures and lower humidities, which may adversely affect the ability of salamanders to forage effectively. On 28 October 2017, Peaks of Otter salamanders were collected by turning over rocks and logs at three sites ranging in elevation from 503 to 991 m. Mass, snout-vent-length (SVL), and gender were recorded in the field for 22 females and 21 males. From the SVL and mass data, salamander condition was calculated using the residual method. Condition index values were then regressed against elevation where a significant relationship was found with salamander condition declining with elevation. Average condition declined from a high of 0.10 at 991 m to a low of -0.19 at 503 m. Based on these preliminary results, the residual condition index has the potential to be a simple method for assessing the effects of elevation, or other stress factors such as competition, on this montane salamander species. Additionally, condition indices can provide researchers with a surrogate measure for density, survival, growth, and reproduction rates which also decrease with elevation. Furthermore, since this method requires significantly less time and effort than traditional methods for measuring survival and growth rates, it could be very useful in the conservation of this montane species with a very limited distribution. In the future, we plan to collect and assess the condition of salamanders from sites at several additional elevations.

Introduction

The Peaks of Otter Salamander has a very limited distribution, about 117 km², in the Blue Ridge Mountains of central Virginia. This species is only found in mature, deciduous forests at elevations greater than 442 m. Elevation may be the key factor limiting the distribution of the species, as the relative health of salamander populations appear to decline with elevation. Past studies have hypothesized that this relationship is due to the increase in temperature and decrease in relative humidity as elevation declines (Reichenbach and Brophy, 2017). *Plethodon hubrichti*'s intolerance to these conditions was first suggested by Thurow in 1957, who observed that the species possessed a lower tolerance for increased temperatures and evaporation rates than others, such as *P. cinereus*. Other salamander studies have suggested that their inability to inhabit low elevations may be the most critical constraint upon their distributions (Hairston, 1951, 1981; Kozak and Wiens, 2006). The densities of surface-active salamanders, survival rates, growth rates, eggs per female, and relative reproductive output all decrease with elevation, as the conditions become warmer and drier (Reichenbach and Brophy, 2017). Therefore low elevation sites represent fragile environments for the Peaks of Otter salamander and should be managed carefully so that it is not extirpated in these locations.

Assessing the impact of the warmer and drier conditions on the health of a montane salamander might be done by using a body condition index. Ecologists have long sought a reliable method for measuring an animal's body condition, which is an indicator of fitness, ability to cope with environmental pressures, and ultimately reproductive success. A good condition index should not vary with body size. Condition index is generally calculated as body mass divided by a linear measure of body size. The residual index, however, regresses body mass against body size, and estimates condition based on the distance of individual animals from the resulting regression line. The animals that fall above the regression line are considered healthier compared to those below the line (Jakob et al, 1996). The condition indices of surface-active *P. hubrichti* were hypothesized to decrease at lower elevations due to increased temperatures and lower relative humidities which may physiologically stress montane salamander species.

Methods & Materials

On 28 October 2017, a team of eight people collected *P. hubrichti* during the cool daytime hours, several days after rain. Salamanders were collected by carefully turning over rocks and logs at three different allopatric sites along an elevational gradient (503, 665, and 991 m). The goal was to collect ten male and ten female salamanders per site. Approximately one hour of collection and measurement was performed at each site. *Plethodon hubrichti* were temporarily placed in zip-lock bags along with moistened paper towels. Salamander masses (grams) were then determined using an electronic balance. Salamanders were also measured (SVL) using calipers and the "salamander-stick method" (Walston and Mullin, 2005; Figure 1), and finally examined for sex determination. A non-destructive, visual method using an LED light was used to determine the presence or absence of the vas deferens. Additionally, vent shape and size was used to determine gender, with males generally having more oblong vents than females (Figures 2-3). All salamanders were returned to their capture location immediately following processing.

Residual condition index values were calculated for salamanders, separately for each elevation, by regressing mass against SVL. Salamanders above the regression line were the fatter ones (positive condition index value) while those below the regression line were considered skinnier (negative condition index value). These condition index values were initially compared between the sexes, for elevations where adequate numbers of males and females were collected. This was done by comparing residuals using t-tests to determine whether data from both sexes could be combined when comparing condition index versus elevation. Residual condition index values were then calculated for all salamanders across all elevations by regressing mass against SVL. These condition index values were then regressed against elevation. Finally, comparisons were made graphically between average condition index values per elevation relative to data collected previously on survival rates, eggs per female, and salamander densities. These data were collected at the same or similar locations in 2008 and 2009 (Reichenbach and Brophy, 2017).



Photo by Olivia de Araujo.



Figure 1. Salamander stick and SVL measurement. Photo by Norman Reichenbach.



Figure 2. Male specimen with oblong, football-shaped vent. Photo by Andrew Kniowski. Used with permission.



Figure 3. Female specimen with slit-shaped vent. Photo by Andrew Kniowski. Used with permission.



Photo by Andrew Kniowski. Used with permission.

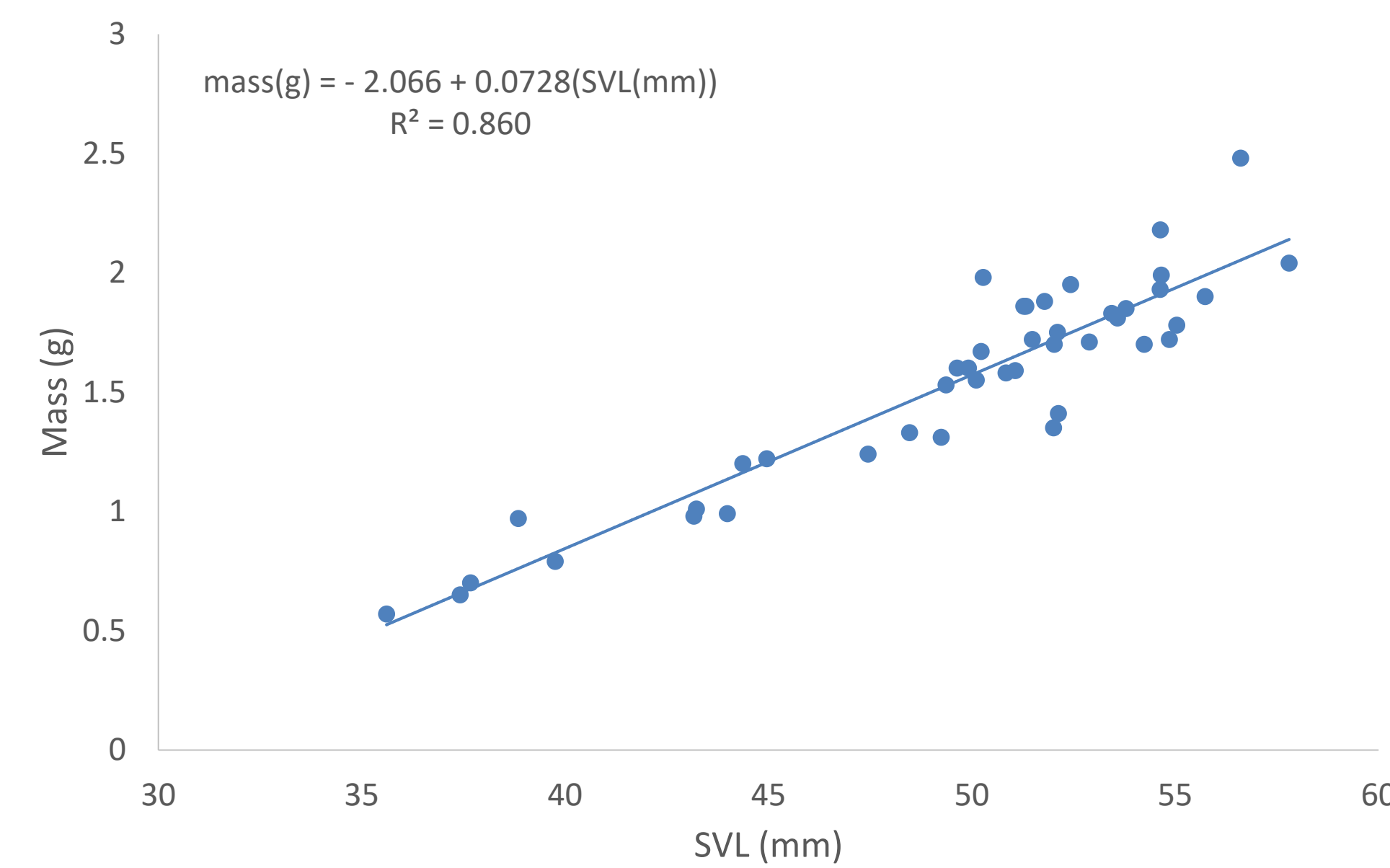


Figure 4. Relationship between mass and SVL.

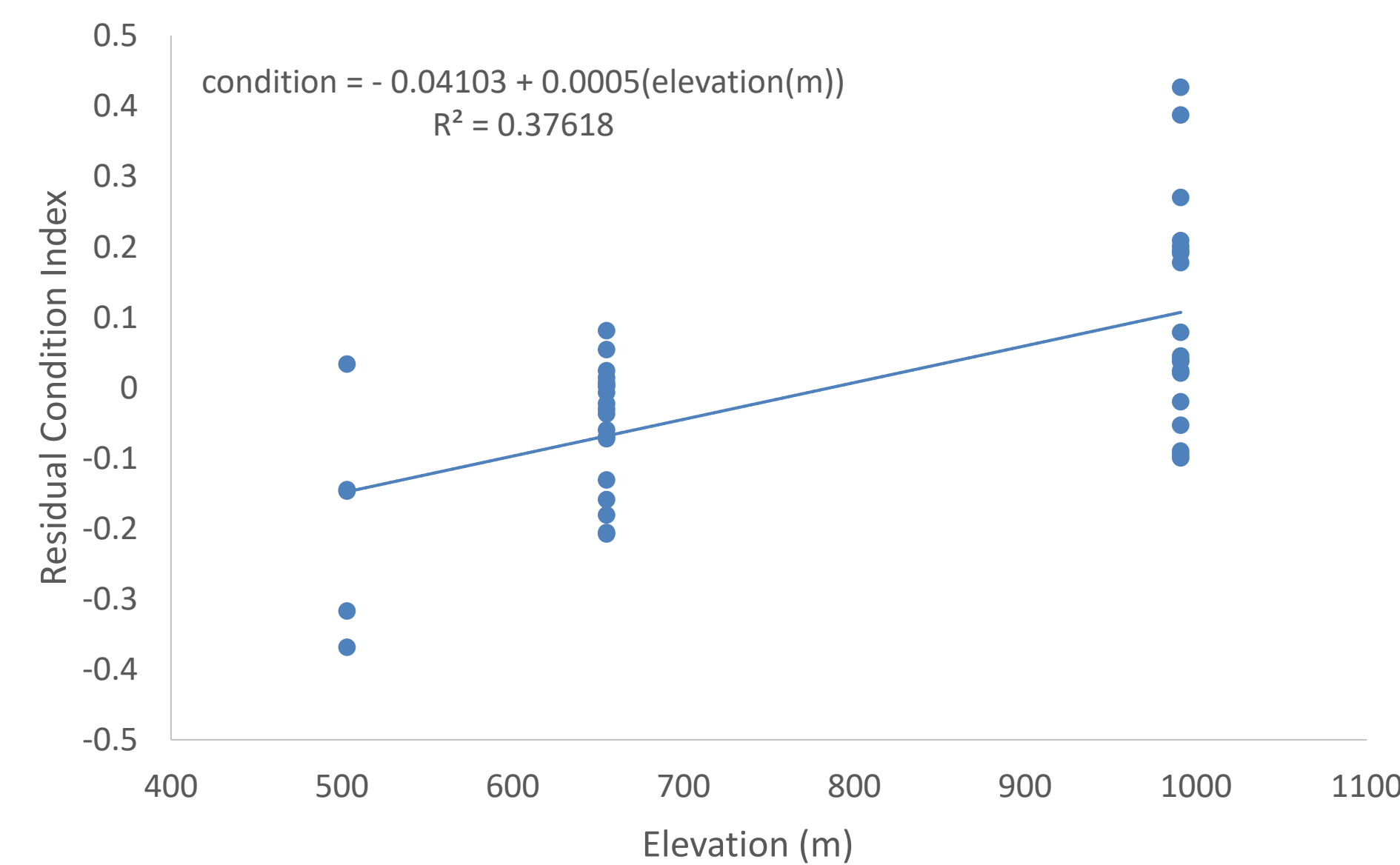


Figure 6. Relationship between elevation and condition index at all three sites.

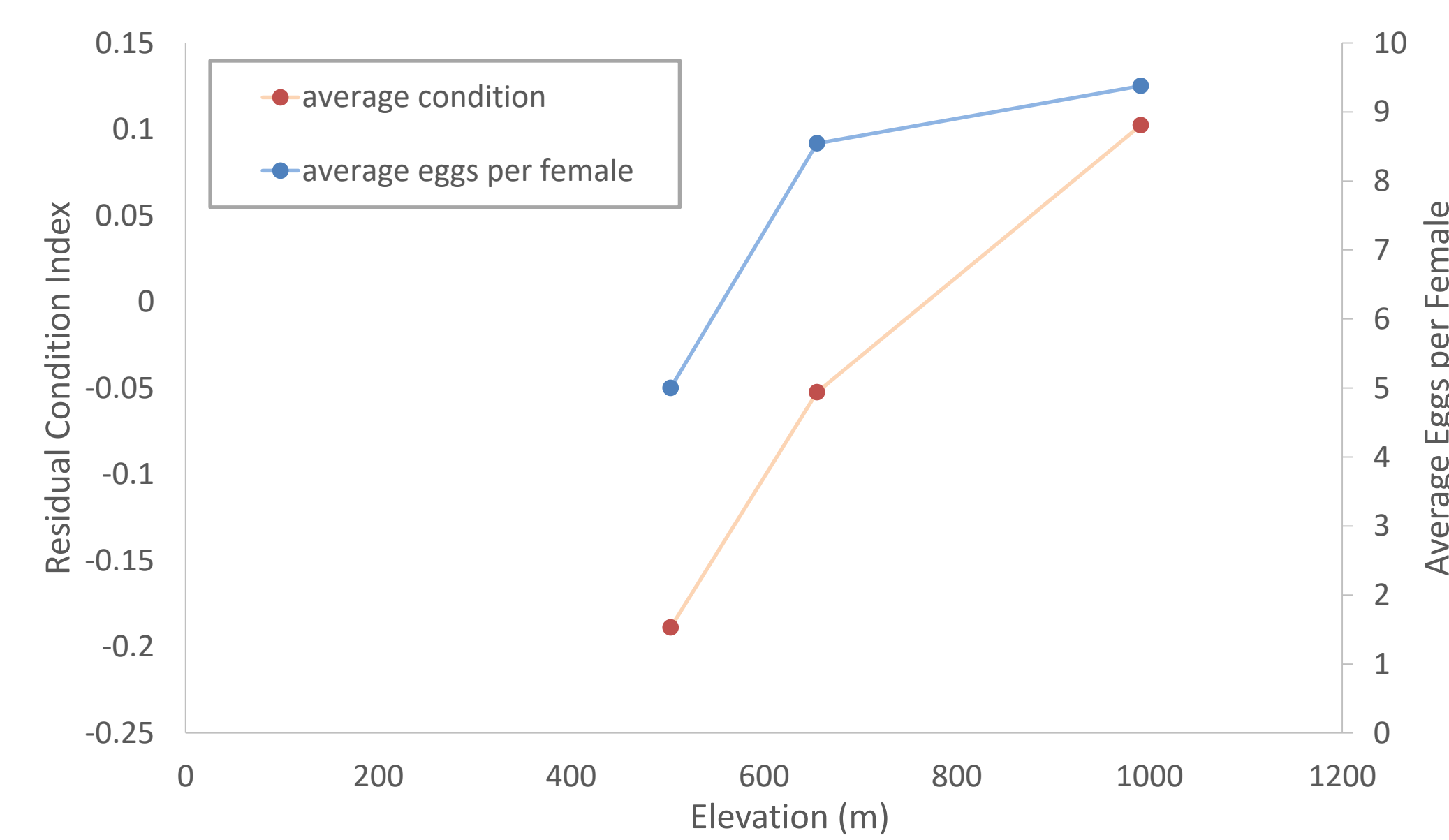


Figure 8. Relationship between condition index, elevation, and eggs per female.

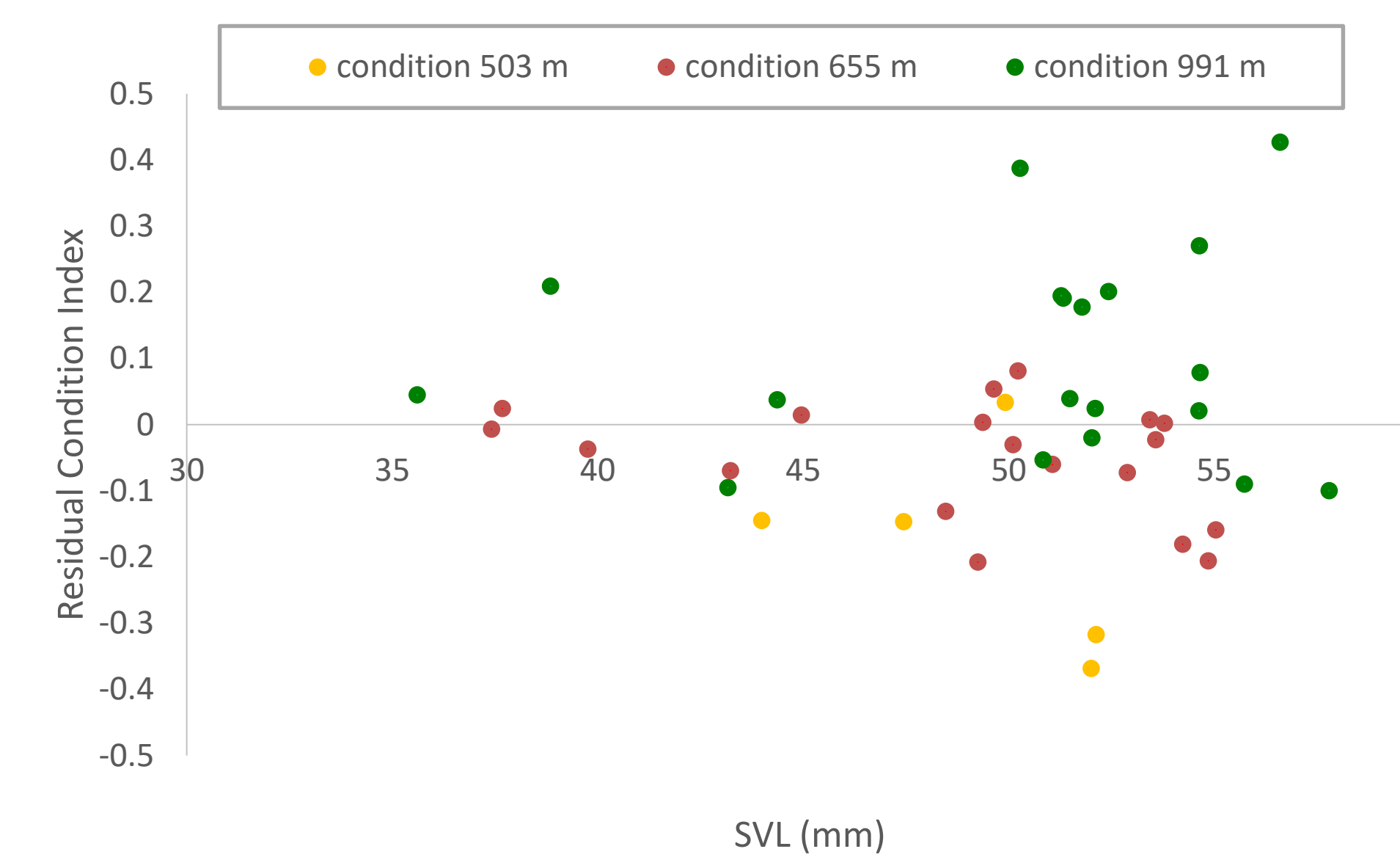


Figure 5. Relationship between condition index and SVL with respect to elevation.

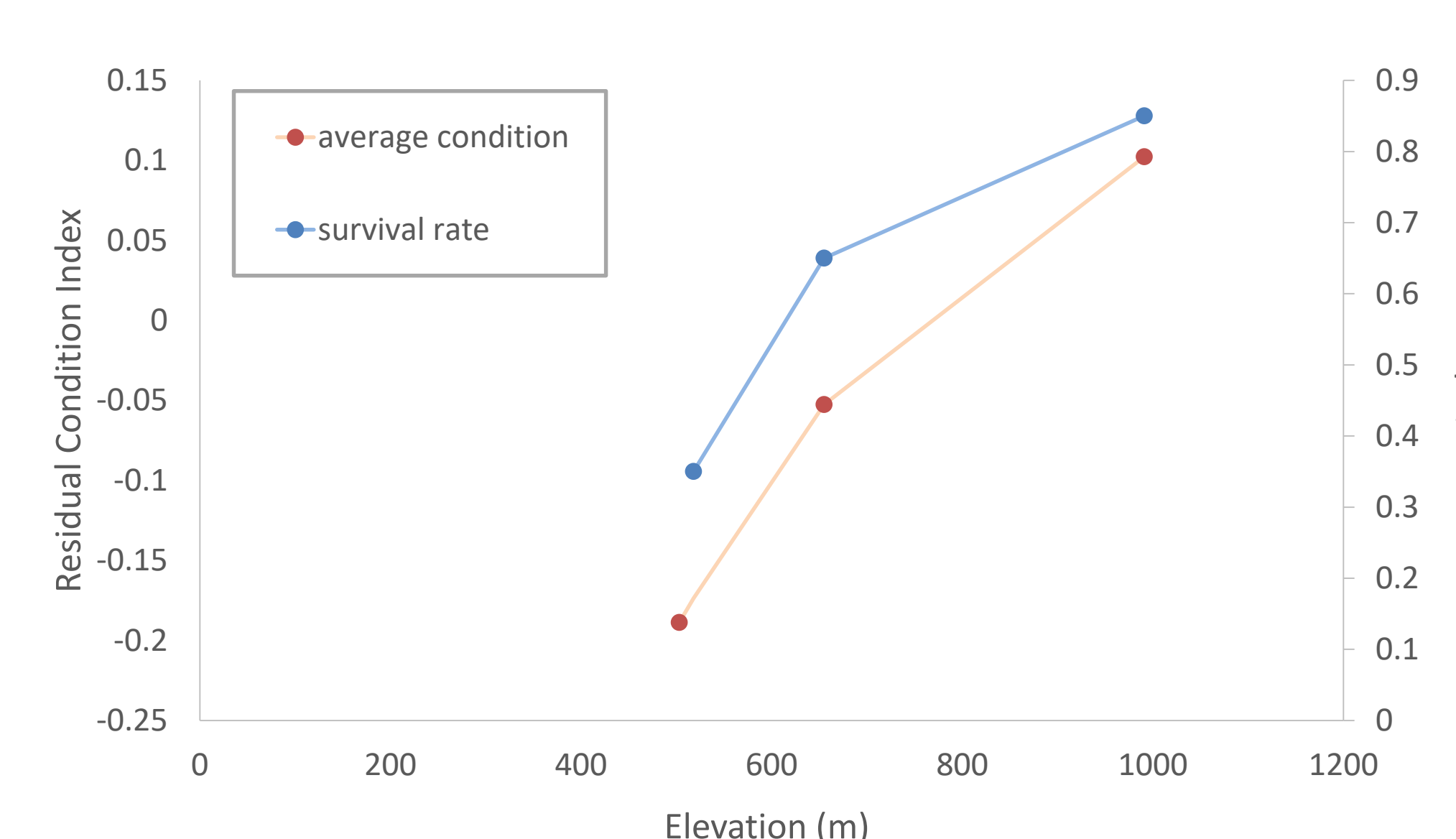


Figure 7. Relationship between condition index, elevation, and survival rate.

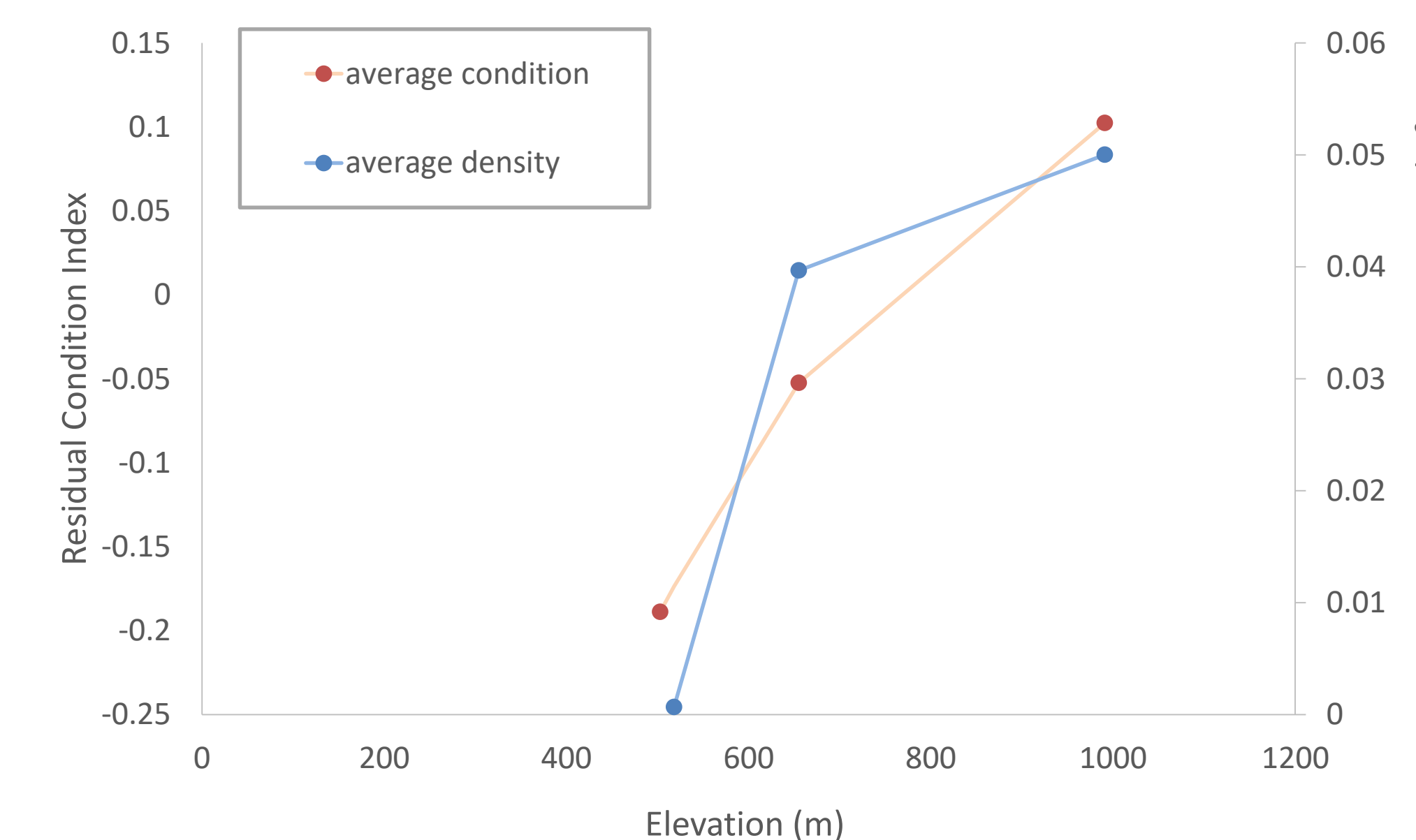


Figure 9. Relationship between condition index, elevation, and density.

Results and Conclusion

Approximately twenty animals were easily found from the sites at the two higher elevations (655 and 991 m), whereas collecting animals at the 503 m site was difficult due to the low salamander densities, and only five animals were found after extensive searching. Four males and one female, nine males and eleven females, and nine males and ten females were collected from the 503, 655, and 991 m sites, respectively.

Condition index values were not significantly different between sexes at the 655 m site ($t=0.62$, $d.f.=17$, $p=0.54$), but were significantly different at the 991 m site ($t=2.19$, $d.f.=17$, $p=0.04$). Since condition index values were significantly different at only one elevation, and since only one female was collected at the 503 m site, sexes were combined when regressing mass against SVL across all elevations to calculate condition index values. The mass of one male at the 655 m site was half that of salamanders of similar SVL. Data for this animal were dropped from analyses due to suspected measurement or transcription error.

As SVL increased, mass increased linearly ($\text{mass(g)} = -2.066 + 0.0728(\text{SVL(mm)})$) ($F=252.7$, $df=1, 41$, $P<0.001$, $r^2=0.86$; Figure 4). When condition index values were plotted against SVL, and data points were color-coded by elevation, salamanders from higher elevations had primarily positive condition index values while those from lower elevations had negative values (Figure 5). In addition, condition index values increased linearly with elevation ($\text{condition} = -0.04103 + 0.0005(\text{elevation(m)})$) ($F=24.7$, $df=1, 41$, $P<0.001$, $r^2=0.38$; Figure 6). As elevation increases from 503 to 991 m, salamanders become heavier per unit length, indicating better overall health. It is believed that this trend is primarily linked to foraging efficiency. When the forest is very moist, such as immediately following rainfall, humidity levels are high enough to permit salamanders to climb vegetation without desiccating. Salamanders foraging on vegetation do so with greater efficiency relative to those foraging on the forest floor (Jaeger, 1978). At high elevations there may be many days during the active season where conditions would allow the salamanders to forage on vegetation and thus develop into healthier animals overall. In contrast, at low elevations, there may be far fewer days available to forage on vegetation since these areas are relatively warmer and drier compared to high elevation sites (Reichenbach and Brophy, 2017). Average condition index values calculated from this study showed similar trends when compared to survival rates, eggs per female, and salamander densities across similar elevations (Figures 7-9). Survival rates declined 59%, average eggs per female 47%, and average salamander density 99% between the highest (991 m) and lowest (503 m) elevations. Similarly, average condition index declined 285% between these two elevations, albeit on a larger scale.

Overall, the residual condition index is a relatively simple method for assessing the state of Peaks of Otter salamander populations. It requires little time and effort, and the results parallel those of traditional, more complicated procedures. This method has the potential to assess the effects of elevation, or other stress factors such as competition, on this montane salamander species. Similarly, condition indices may provide researchers with a surrogate measure for density, survival, growth, and reproductive rates which also decrease with elevation. In summary, the residual condition index may prove to be useful in the conservation of this montane salamander species with a very limited distribution.

Future Work

1. Collect Peaks of Otter salamanders at additional elevations to assess their condition using the residual condition index.
2. Collect Peaks of Otter salamanders in allopatric and sympatric sites (sites with the Eastern Red-backed salamander) at comparable elevations to assess their condition and number of eggs per female. This will test the effects of interspecific competition on Peaks of Otter salamander condition and reproductive output.
3. Compare fraction of the population foraging on vegetation at low and high elevations sites.

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