



Morph-specific differences in escape behavior in a color polymorphic lizard

Kinsey M. Brock¹ · Indiana E. Madden²

Received: 30 March 2022 / Revised: 2 July 2022 / Accepted: 4 July 2022 / Published online: 14 July 2022
© The Author(s) 2022

Abstract

Variation in color morph behavior is an important factor in the maintenance of color polymorphism. Alternative anti-predator behaviors are often associated with morphological traits such as coloration, possibly because predator-mediated viability selection favors certain combinations of anti-predator behavior and color. The Aegean wall lizard, *Podarcis erhardii*, is color polymorphic and populations can have up to three monochromatic morphs: orange, yellow, and white. We investigated whether escape behaviors differ among coexisting color morphs, and if morph behaviors are repeatable across different populations with the same predator species. Specifically, we assessed color morph flight initiation distance (FID), distance to the nearest refuge (DNR), and distance to chosen refuge (DR) in two populations of Aegean wall lizards from Naxos island. We also analyzed the type of refugia color morphs selected and their re-emergence behavior following a standardized approach. We found that orange morphs have different escape behaviors from white and yellow morphs, and these differences are consistent in both populations we sampled. Orange morphs have shorter FIDs, DNRs, and DRs; select different refuge types; and re-emerge less often after being approached compared to white and yellow morphs. Observed differences in color morph escape behaviors support the idea that morphs have evolved alternative behavioral strategies that may play a role in population-level morph maintenance and loss.

Significance statement

Color polymorphic species often differ in behaviors related to reproduction, but differences in other behaviors are relatively underexplored. In this study, we use an experimental approach in two natural populations of color polymorphic lizards to determine that color morphs have diverged in their escape behaviors. By conducting our experiments in two different populations with similar predator regimes, we show for the first time that behavioral differences among intra-specific color morphs are repeatable across populations, suggesting that alternative behavioral strategies have evolved in this species. Using this experimental approach, we demonstrate that the brightest orange morph stays closer to refuge than other morphs, uses a different refuge type (vegetation) more often than other morphs (wall crevices), and take much longer to emerge from refuge after a simulated predation event than other morphs. Thus, selective pressures from visual predators may differ between morphs and play a role in the evolution and maintenance of color polymorphisms in these types of systems. Our study species, *Podarcis erhardii*, belongs to a highly color polymorphic genus (19/23 spp. are color polymorphic) that contains the same three color morphs, thus we believe our results may be relevant to more than just *P. erhardii*.

Keywords Color polymorphism · Alternative behavioral strategies · Escape behavior · Anti-predator behavior · Flight initiation distance · *Podarcis erhardii*

Communicated by: T. Madsen

✉ Kinsey M. Brock
kbrock@berkeley.edu

Extended author information available on the last page of the article

Introduction

Understanding how phenotypic variation is generated and maintained is a central goal in evolutionary biology. Color polymorphic species are good systems for studying mechanisms that regulate phenotypic variation because sympatric color morphs represent distinct, genetically determined

variation maintained by selection (Gray and McKinnon 2007; Svensson 2017). Color morphs usually, if not always, differ in additional traits (McKinnon and Pierotti 2010; Stuart-Fox et al. 2021) such as morphology (Brock et al. 2020), physiology (Sinervo et al. 2000; Border et al. 2019), diet (Lattanzio and Miles 2016), and behavior (Vercken and Clobert 2008). These alternative morph phenotypes are likely the result of correlational selection that produces genetic correlations between phenotypic traits under selection, such as color and behavior (Brodie 1989, 1992; Miles et al. 2007).

Predation can be a major selective force in the evolution of color and behavior (Brodie 1992; Lima and Dill 1990; Hurtado-Gonzales et al. 2010; Pérez i de Lanuza and Font 2015), and could play a role in maintenance of color polymorphism. Because color is a visual trait, certain color morphs could be more easily detected by predators and may use different tactics to avoid predation (Forsman and Appelqvist 1998). In insects (Forsman and Appelqvist 1998), fish (Maan et al. 2008) and reptiles (Forsman 1995), sympatric color morphs suffer different rates of predation in different environments due to differential visual detection by predators. While predators refine their hunting techniques to find and obtain prey, prey evolve adaptive strategies to avoid being detected and captured (Dawkins and Krebs 1979). For color morphs, avoiding detection by predators may require different strategies given differences in conspicuousness against different backgrounds and surroundings (Marshall et al. 2016). Prey that differ in color or pattern may evolve different anti-predator behaviors that involve occupying distinct microhabitat to enhance crypsis and avoid detection (Reid 1987; Sandoval 1994), different levels of boldness (Yewers et al. 2016), and different escape behaviors (Marcellini and Janssen 1991; Labra et al. 2007).

All prey species must balance the risk of predation with activities that allow them to meet their basic needs (Pérez-Cembranos et al. 2013), and color morphs may evolve different strategies for negotiating safety based on their risk of being visually detected by predators. The flight initiation distance (FID) is the distance between a prey animal and a perceived threat when the prey animal begins its escape (Ydenberg and Dill 1986; Runyan and Blumstein 2004). Generally, FIDs have been used as a metric of risk assessment and habituation (Blumstein 2003), where longer FIDs are interpreted as risk aversion, and shorter FIDs are associated with boldness or habituation (Blumstein 2006). FIDs have also been useful for quantifying variation in escape behavior and capture the benefits and costs to fleeing that prey animals must negotiate (Lima and Dill 1990). If prey flee at greater distances upon perceiving a predation threat, they may lose out on beneficial feeding, mating, social, or thermoregulatory opportunities that can increase prey fitness (Lima and Dill 1990). On the other hand, prey must weigh

the benefits of staying against the costs of staying too long when faced with a predation threat. Costs of staying too long can include an energy-extensive chase, injury, or death by predation (Snell et al. 1988). Ultimately, escape is predicted to begin when a predator approaches to a distance where the cost of staying and the cost of fleeing are equal (Ydenberg and Dill 1986; Cooper and Frederick 2007). Since prey usually flee to a refuge (a space a predator cannot access), deciding when to flee may be influenced by the distance a prey is from the nearest refuge (DNR). Prey must also quickly select what type of refuge to take and decide how long to wait to safely re-emerge at the expense of other activities. If predation risk is unequal among morphs that differ in color, we expect morphs to display consistent, distinct escape behaviors when confronted with a simulated predation event.

The present study focuses on color morph escape behavior in the color polymorphic Aegean wall lizard (*Podarcis erhardii*). This species has three monochromatic color morphs that are orange, yellow, or white and three mosaic morphs that are orange-white, yellow-orange, or white-yellow (Brock et al. 2022b). All sexes are color polymorphic in this species and can be any of the six color morphs (Brock et al. 2020). Male monochromatic color morphs differ in head and body size and female monochromatic color morphs tend to be more similar in size (Brock et al. 2020). Male monochromatic color morphs exhibit differences in several social behaviors like aggression, boldness, chemical signaling, and visual signaling frequency when in contest with each other over limited thermally-suitable space (Brock et al. 2022a), suggesting that color morphs in this species may have different behavioral strategies. We focused our study on monochromatic color morph risk assessment and escape behavior, including flight initiation distance (FID), distance to the nearest refuge (DNR), refuge choice (distance to chosen refuge and refuge type), and re-emergence behavior. In this study, we address the following questions:

- 1 Do color morphs differ in their escape behavior (flight initiation distance, distance to nearest refuge, and distance to chosen refuge) and are these morph differences in behavior the same across two populations with the same predator guilds?
- 2 Do color morphs differ in the type of refuge they select when fleeing from an approaching threat?
- 3 After taking refuge, do color morphs differ in the amount of time they wait to re-emerge from hiding?

To answer these questions, we measured the escape behavior of color morphs observed on dry stone walls from two populations on Naxos island, Greece. We hypothesized that color morphs within a site would exhibit significantly different escape behaviors (FID, DNR, DR, refuge selection, and re-emergence), and that these behavioral differences

would be the same among populations with the same predator guilds. Specifically, we expected white morphs to have shorter FIDs and longer DNRs and DRs based on their relatively less conspicuous coloration (Pérez i de Lanuza and Font 2015; Brock et al. 2020). We also expected orange morphs to have longer FIDs and shorter DNRs because it was determined that orange morphs in a closely related polymorphic *Podarcis* species with the same three color morphs were more conspicuous to conspecifics and predators than yellow or white morphs (Pérez i de Lanuza and Font 2015). We expected yellow morphs to exhibit intermediate escape behaviors since their color is intermediate to white and orange morphs (Brock et al. 2020) and yellow morphs tend to have intermediate behavioral (Brock et al. 2022a) and phenotypic traits compared to white and orange morphs in this species. We hypothesized that color morphs would differ in the types of refuge they select when fleeing from similar starting positions on dry stone walls. Here, we expected that orange morphs would seek refuge in vegetation more often than other color morphs since they are most often found in microhabitat that is shaded by leafy vegetation in this species (KMB, unpublished data), and other *Podarcis* species (Pérez i de Lanuza and Carretero 2018; Brock et al. 2022c). Finally, we hypothesized that color morphs would differ in their re-emergence behavior. Specifically, we expected white and yellow morphs to re-emerge from hiding more often than orange morphs, and take less time to do so, given their tendency to be more active, aggressive, and bold than orange morphs (Brock et al. 2022a).

Methods

Study sites

We conducted our study in June 2021, in the middle of the breeding season when lizards are most active (Valakos et al. 2008). We measured the escape behavior of color morphs in two populations from the island Naxos: Moni and Damarionas. Moni and Damarionas are separated by the Halki valley and are 7 km apart. Both sites have all three monochromatic color morphs and are similar environmentally and ecologically. Moni and Damarionas are small agricultural villages with olive groves, other fruit-bearing trees, and small fields primarily used for goat grazing. Both sites contain many dry stone walls that wall lizards occupy to shelter from predators, safely thermoregulate, and forage for invertebrates. The natural vegetation is characteristic of Mediterranean scrub habitat with sclerophyllous evergreen maquis and phrygana, a diverse species community of summer-deciduous shrubs (Fielding and Turland 2008). The predator guild is the same at both sites where snakes (*Elaphe quatuorlineata*, *Viper ammodytes*, *Natrix natrix*, and *Eryx jaculus*), small mammals (*Rattus rattus*, *Martes*

foina), introduced feral house cats, (*Felis catus*), and birds (*Falco tinnunculus* and *Buteo buteo*) prey upon *P. erhardii* (Brock et al. 2015).

Field sampling of escape behavior

Field sampling took place on warm, sunny days with low wind speeds when lizards were fully active. We recorded flight initiation distance (FID) using a method developed by Blumstein (2003) and utilized in many previous studies on various lizard species including *P. erhardii* (Li et al. 2014; Brock et al. 2015). To eliminate experimenter variation, the same person performed all FID approaches in the field while wearing the same clothes. We limited our sampling to lizards that were initially spotted on dry stone walls so that comparisons of morph escape behavior and refuge choice come from the same starting conditions. Upon sighting a lizard, the experimenter would confirm a lizard's color morph identity and sex with binoculars. Color morphs in this species can be reliably distinguished by eye (Brock et al. 2020), and females and males are easily distinguishable by their head and tail shapes. Once the color morph and sex of the individual was recorded, the experimenter walked directly toward the focal lizard from a head-on starting position perpendicular to the wall and starting distance of approximately 500 cm away to eliminate angle of approach and starting distance as possible sources of bias (Blumstein et al. 2015). Approaches were performed at a practiced pace (0.5 m/s) while maintaining eye contact and the experimenter stopped once the focal lizard moved away (i.e., initiated an escape response). The experimenter then recorded the distance between their stopping point and the point of lizard flight initiation (FID). The experimenter also recorded the distance from the point of flight initiation and the nearest available refuge (distance to nearest refuge, DNR). We defined the nearest available refuge as a space a lizard could enter and hide itself completely from view. The experimenter then recorded the distance from the point of flight initiation and where the lizard chose to seek refuge (distance to refuge, DR). Every lizard sampled fled to a refuge. We also recorded the type of refuge the lizard selected, which ultimately fell into one of two categories: either a crevice in the stone wall or vegetation. Finally, after each lizard entered a refuge, the experimenter waited 30 s and recorded whether or not the lizard re-emerged in that time frame and recorded the time in seconds.

To ensure that we did not measure the same individual twice, we recorded our transect tracks with a Garmin GPS unit using the waypoint finder feature, so we did not revisit the same area while collecting FIDs. The home range size of this species is small, approximately 1.5 m² on Naxos (BeVier et al. 2021), and we did not record individuals of the same sex and morph within this distance of each other. Due to the nature of our measurements, it was not possible

to record data blind because our study involved focal animals in the field.

Statistical analysis

Flight initiation distance can be influenced by the starting distance of the experimenter (Blumstein 2003). Therefore, we first tested for a statistical relationship between FID and starting distance to determine whether we needed to include starting distance as a covariate in our comparisons of FID. We did not find a significant linear relationship between FID and experimenter starting distance in our dataset ($r=0.025$, $df=418$, $P=0.61$), and proceeded to analyze morph differences in FID without starting distance as a covariate.

To test if color morph differed in mean flight initiation distance, distance to nearest refuge, and distance to refuge, we used a two-way ANOVA with an unbalanced design (Type III Sums of Squares) with morph, site, and their interaction as factors. To determine which groups differed from each other, we followed up two-way ANOVAs with post-hoc Tukey multiple pairwise comparison tests, which accounts for multiple pairwise-comparisons between groups. We log-transformed FID, DNR, and DR to meet the assumptions of ANOVA. To prevent the inflation of type I error rates, or rejecting the null when the null is true, adjustments to the p values were made using a Benjamini, Hochberg, and Yekutieli correction with the *p.adjust.method* = "BH" specification in R.

To test for color morph associations with refuge selection (e.g., stone wall crevice or vegetation), we used chi-squared tests of independence.

We tested for color morph differences in re-emergence behavior using Kruskal–Wallis tests. To determine which groups differed significantly from each other, we used a Mann–Whitney U post hoc test and used the Benjamini, Hochberg, and Yekutieli p value correction for multiple comparisons.

We performed all statistical analyses in R (R Core Team 2021).

Results

In total, we measured the escape behavior of 420 lizards. At Moni, we measured 50 of each monochromatic morph per sex, for a total of 300 lizards. At Damarionas, which is similar to Moni in habitat but smaller in area, we measured 20 of each morph per sex for a total of 120 lizards. Flight initiation distance ranged from 5 to 206 cm at Moni, and from 30 to 187 cm at Damarionas. Distance to nearest refuge ranged from 0 to 50 cm at Moni, and from 0 to 29 cm at Damarionas. Distance to chosen refuge ranged from 0 to 109 cm at Moni, and from 0 to 126 cm at Damarionas.

Morph differences in escape behavior

Color morphs exhibited differences in escape behaviors, and site had no effect on these behavioral differences (two-way ANOVA FID site $F=9.13$, $df=1$, $p=0.11$; DNR site $F=1.97$, $df=1$, $p=0.16$; DR site $F=3.07$, $df=1$, $p=0.08$). The interaction between morph and site was not significant for FID, DNR, or DR (two-way ANOVA FID morph:site $F=2.62$, $df=2$, $p=0.07$; DNR morph:site $F=0.72$, $df=2$, $p=0.49$; DR morph:site $F=1.54$, $df=2$, $p=0.22$). At both Moni and Damarionas, we found that orange morphs had significantly shorter FIDs than yellow and white morphs, yellow had a significantly different but intermediate FID, and white morphs had significantly longer FIDs than orange and yellow morphs (post-hoc Tukey HSD FID orange-white $p<0.001$, white-yellow $p<0.001$, yellow-orange $p<0.001$; Fig. 1). At both Moni and Damarionas, we found all morphs differed from each other in their DNR (post-hoc Tukey HSD DNR orange-white $p<0.001$, white-yellow $p<0.01$, yellow-orange $p<0.001$). At both sites, orange morphs had the shortest DNR, yellow morphs had an intermediate DNR, and white morphs had the longest DNR (Fig. 1). Finally, at both sites, we found that morphs differed significantly in the distance to their chosen refuge (post-hoc Tukey HSD DR orange-white $p<0.001$, white-yellow $p<0.001$, yellow-orange $p<0.001$). At both sites, orange morphs had the shortest DR and white morphs had the longest DR (Fig. 1). Morphs of the same color from the different sites did not differ in any behaviors (Fig. 1).

Morph refuge selection

At Moni and Damarionas, orange morphs chose refuge in vegetation significantly more often than the other morphs (Moni: $\chi^2=19.12$, $df=2$, $p<0.001$; Damarionas: $\chi^2=13.5$, $df=2$, $p=0.001$; Fig. 2). At Moni, 23 orange morphs, 3 white morphs, and 8 yellow morphs chose refuge in vegetation, and 77 orange morphs, 97 white morphs, and 92 yellow morphs chose refuge in dry stone wall crevices. At Damarionas, 10 orange morphs, 1 white morph, and 1 yellow morph chose refuge in vegetation, and 30 orange morphs, 39 white morphs, and 39 yellow morphs chose refuge in wall crevices. In general, all morphs chose refuge in the crevices of dry stone walls more often than vegetation (Fig. 2).

Morph re-emergence behavior

We found consistent, across-site differences in morph re-emergence behavior. At both Moni and Damarionas, color morphs differed significantly in their re-emergence behavior (Moni Kruskal–Wallis chi-squared = 150.1, $df=2$, $p<0.001$; Damarionas Kruskal–Wallis chi-squared = 61.027, $df=2$, $p<0.001$). A post hoc Mann–Whitney U test revealed that

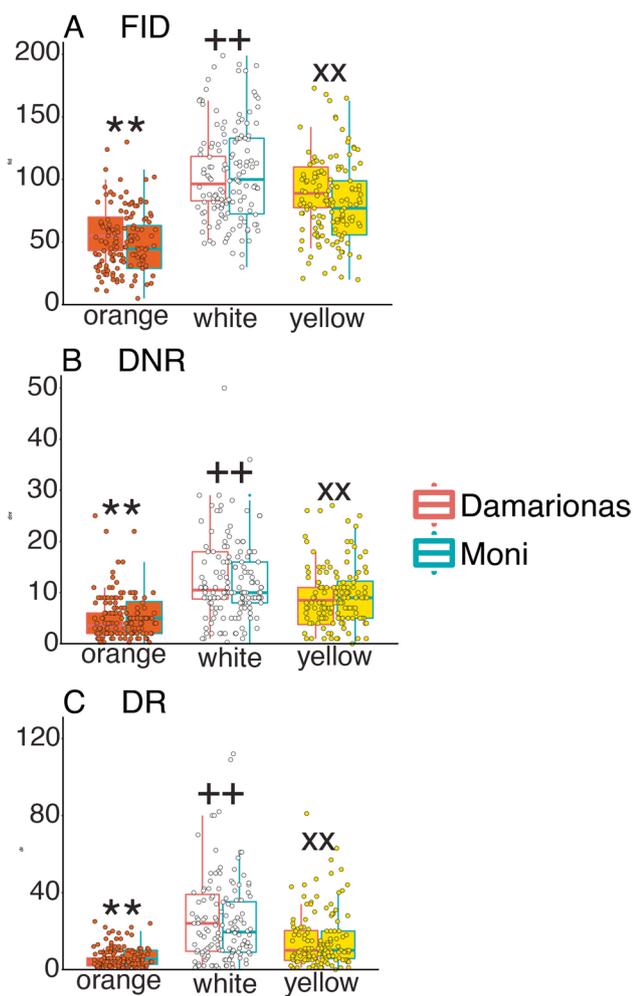


Fig. 1 Color morph differences in flight initiation distance FID (A), distance to nearest refuge DNR (B), and distance to chosen refuge DR (C). Boxes indicate the inter quartile range (IQR), with the central line depicting the median and the whiskers extending to 1.5*IQR. Different symbols indicate significant mean differences in log-transformed FID, DNR, and DR behaviors between groups ($\alpha < 0.05$). Note that datapoints presented in the Fig. are the raw data and not log-transformed

all color morphs differed significantly from each other at both sites (Moni Mann–Whitney U with Bonferroni correction, orange–white $p < 0.001$; white–yellow $p < 0.001$; yellow–orange $p < 0.001$; Damarionas Mann–Whitney U with Bonferroni correction, orange–white $p < 0.001$, white–yellow $p < 0.001$, yellow–orange $p < 0.001$). White morphs re-emerged most often, followed by yellow, and orange re-emerged the least often (Table 1).

At both Moni and Damarionas, very few orange morphs re-emerged from refuge within a 30-s period compared to other morphs (Table 1). At Moni, only 6% of orange morphs re-emerged from refuge and just 10% re-emerged at Damarionas. When orange morphs did re-emerge from hiding, they took about twice as long to do so (on-average)

compared to other morphs at their site (Table 1). Yellow morphs re-emerged from hiding 36% of the time at Moni, and 53% of the time at Damarionas. Yellow morphs were quicker to re-emerge than orange morphs and took a similar amount of time to re-emerge as white morphs at both sites. Finally, more white morphs re-emerged from hiding compared to other morphs at both sites, with a re-emergence rate of over 90% at both sites (Table 1).

Discussion

Distinct color morph-correlated behavior may play a role in the evolution and maintenance of polymorphisms (Sinervo and Lively 1996; Punzalan et al. 2005; Gray and McKinnon 2007). In this study, we examined color morph differences in escape behavior to determine if color morphs have distinct strategies for avoiding predation. We found that co-occurring color morphs exhibit differences in their flight initiation distance, distance from nearest refuge, distance to chosen refuge, type of refuge they select, and re-emergence behaviors. Specifically, orange morphs had significantly shorter FIDs, white morphs had significantly longer FIDs, and yellow morphs had intermediate FIDs. Orange morphs were found very close to refuge and used closer refugia than the other morphs. White morphs were found furthest away from refuge and used refugia that were significantly further away than other morphs. Orange morphs used vegetation as refuge more often than the yellow and white morphs. Orange morphs took longer to re-emerge and re-emerged less often after we approached them compared to other morphs. We also found that color morph-correlated anti-predator behaviors were the same in two different populations. Our results add to a growing body of evidence that color morphs in this species have evolved alternative life history traits and behavioral strategies (Brock et al. 2020, 2022a), which may play a role in color morph evolution and coexistence.

Visual predators may impose correlational selection on prey color and behavior (Forsman and Appelqvist 1998), and influence color morph evolution, frequencies, and persistence in populations (Shigemiyama 2004; Punzalan et al. 2005). Common wall lizards (*Podarcis muralis*) are a close relative to *P. erhardii* and have the same three monochromatic color morphs and three mosaic morphs (Andrade et al. 2019). In *P. muralis*, orange morphs are more conspicuous to conspecifics and predators than yellow or white morphs (Pérez i de Lanuza and Font 2015). In *P. erhardii*, orange, yellow, and white morphs have distinct and discernible spectra (Brock et al. 2020) and may also be discernible by predators with advanced visual systems, such as birds. Birds are known to impose apostatic selection, or disproportionately consume common color morphs in a

Fig. 2 Proportion of chosen refugia by color morph at each field site. At both Moni and Damarionas, orange morphs used vegetation as a refuge more often than white and yellow morphs. Asterisks indicate significant differences between morphs in their use of vegetation as a refuge ($\alpha < 0.05$)

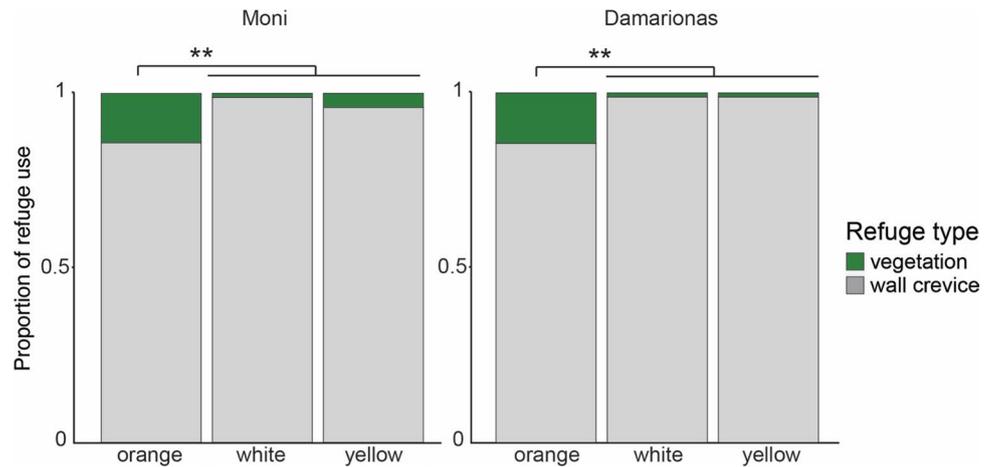


Table 1 Morph re-emergence frequencies and time to re-emergence from both field sites. At both Moni and Damarionas, significantly fewer orange morphs re-emerged in a 30-s period after taking refuge than white or yellow morphs. Of individuals that did re-emerge, orange morphs took significantly longer to do so

Site	Morph	N re-emerged (%)		Avg. seconds to re-emergence	
		Female	Male	Female	Male
Moni	orange	2/50 (4%)	4/50 (8%)	24.5 s	23 s
	yellow	10/50 (20%)	26/50 (52%)	13.5 s	13.8 s
	white	45/50 (90%)	46/50 (92%)	12.6 s	7.8 s
Damarionas	orange	0/20 (0%)	4/20 (20%)	–	14.8 s
	yellow	6/20 (30%)	15/20 (75%)	15 s	7.2 s
	white	19/20 (95%)	20/20 (100%)	7.8 s	5.3 s

population (Allen 1972; Allen et al. 1998). This apostatic selection is a form of negative frequency-dependent selection that could promote polymorphism by favoring a rare morph, allowing it to persist in the population. Morph frequencies in these populations in 2021 were similar to each other, and the white morph was by far the most common in both populations (Moni monochromatic morph frequencies: white = 69%, yellow = 11%, orange = 5%; Damarionas morph frequencies: white = 71%, yellow = 9%, orange = 6%). Long-term monitoring of polymorphic populations is needed to discern whether color morph frequencies change following a frequency-dependent pattern in this species. It is also worth noting that *P. erhardii* male color morphs have somewhat distinct chemical signal profiles in their femoral pore exudate (Brock et al. 2020). Although not evaluated in this study, chemosensory cues may make certain color morphs more susceptible to detection by certain types of predators, particularly snakes and mammals. It is known that different types of predators have different effects on FID and tail-shedding behavior in *P. erhardii* (Brock et al. 2015), but how those differences pertain to color morph evolution and

persistence is unknown. Further research on the visual and chemical ecology of Aegean wall lizards and their predators is needed to determine if certain color morphs are more conspicuous to predators, and under what circumstances.

Morph-correlated behaviors can be indicators of broader life-history differences and alternative strategies in color polymorphic species (Yewers et al. 2016). In color polymorphic lizards in the genus *Podarcis*, orange morphs tend to be associated with vegetated habitats close to water (Pérez i de Lanuza and Carretero 2018; Brock et al. 2022b). In our study, we found that orange morphs used vegetation as refuge more often, stayed the closest to refuge, and had shorter FIDs compared to other morphs. Orange morphs in *Podarcis* lizards may stay close to vegetation and refuge due to greater detectability by predators (Pérez i de Lanuza and Font 2015), and may have shorter FIDs due to maintaining close proximity to refuge. Other studies have found strong correlations between lizards with orange or red coloration on their throats (Pérez i de Lanuza and Font 2015) or dewlaps (Ng et al. 2013) and vegetated environments that are darker, possibly due to signal efficacy in more mesic environments compared to xeric environments. Alternatively, orange morphs may develop associations with vegetated habitat and shorter FIDs in response to more frequent predator harassment. If orange morphs are more easily detectable by predators, then they may experience being chased more often and develop higher risk thresholds for fleeing, which would translate to shorter FIDs. Time spent fleeing cannot be spent on other necessary and fitness-enhancing activities such as feeding, thermoregulating, socializing, and mating. Orange morphs may remain close to vegetation and refuge to avoid detection and reduce the amount of time being chased, resulting in shorter FIDs.

In color polymorphic *Podarcis* species, orange morphs tend to be brighter (Pérez i de Lanuza and Font 2015; Brock et al. 2020), which may make them more easily detectable by visual predators such as birds. Our results support the idea that orange morphs modify their behavior to avoid

detection by predators by remaining closer to refuge and waiting much longer to re-emerge from hiding compared to white and yellow morphs. Orange morphs were observed closer to vegetation and chose vegetation as a refuge more often than other morphs. Vegetation can provide shade that orange morphs may use to avoid detection by predators (Ng et al. 2013). Further research into color morph microhabitat selection could provide insights into how background matching and lighting environments interact with morph colors to increase signal efficacy to intended receivers versus decreasing detectability by different predator types.

Our results add support to the idea that wall lizard color morphs exhibit alternative behavioral strategies (Brock et al. 2022a). In this study, white morphs were observed significantly further away from refuge and chose more distant refugia than yellow and orange morphs, which are facets of boldness. Boldness is often correlated with high levels of aggression toward conspecifics (Coleman and Wilson 1998; Reaney and Backwell 2007). White and yellow male morphs, which were consistently found further from refuge than orange morphs in this study, exhibited higher levels of bold and aggressive behaviors than orange male morphs in one-on-one contest experiments (Brock et al. 2022a). Higher levels of boldness and aggression and larger body size are usually associated with dominance (Francis 1988; Sinervo and Lively 1996), and dominance can determine relative resource access and fitness among morphs. In this species, white and yellow morphs have higher levels of boldness and aggression (Brock et al. 2022a), and the orange morph has larger head and body sizes (Brock et al. 2020). Morph-correlated behaviors and morphological traits likely offer context-specific advantages, which could result in somewhat equal population-level morph fitness over time and balancing the polymorphism. Our study is an important step in characterizing the color polymorphism in this species, and provides another piece to the puzzle of polymorphism evolution and maintenance. More research that focuses on morph relative fitness and trait values is needed to provide insight into the mechanisms regulating polymorphisms.

In the well-studied color polymorphic *Podarcis* species, the orange morph tends to be more behaviorally and phenotypically distinct from white morphs (Huyghe et al. 2007, 2009; Abalos et al. 2016; Brock et al. 2020, 2022a), and the yellow morphs tend to be an intermediate phenotype. Morph-assortative mating could minimize the break-up of coadapted gene complexes and generate linkage disequilibrium built up by correlational selection (McKinnon and Pierotti 2010), resulting in phenotypic divergence between morphs. Homomorphic pairs tend to be more common than heteromorphic pairs in several *Podarcis* species (Huyghe et al. 2010; Pérez i de Lanuza et al. 2013; KMB unpublished data). Further, white and yellow morphs

tend to be found in more similar habitat and share more similar behavioral repertoires compared to the orange morph (Huyghe et al. 2007; Brock et al. 2022a, b). Thus, it is possible that white-yellow is a more common heteromorphic mate pairing than orange-white or yellow-orange pairings. If there is non-random mating in populations of color morphs, it could lead to deeper genetic divergences between certain morphs (Huyghe et al. 2010), with potential effects on phenotypic divergence. More work on color morph genomes is needed to understand the genetic basis of behavioral and phenotypic differences among color morphs and their implications in morph evolution and maintenance.

We demonstrated that color morph-correlated behaviors are consistent across different populations, a detail that is often assumed in other behavioral studies concerning color polymorphic species. Although color morph differences in behaviors such as boldness (Yewers et al. 2016), aggression (Abalos et al. 2016; Yewers et al. 2016), and mating strategies (Sinervo and Lively 1996) are often observed in color polymorphic species, these differences are rarely demonstrated to be the same for more than one population. Confirming consistency in alternative behavioral phenotypes is essential when trying to understand drivers of phenotypic variation. Future work should also explore color morph behavioral differences in populations with different ecological contexts. Whether or not different predation regimes have different effects on color morph behaviors, life histories, or persistence in populations remains an open question in this and many other color polymorphic species.

Supplementary Information The online version contains supplementary material available at <https://doi.org/10.1007/s00265-022-03211-8>.

Acknowledgements We thank the reviewers for their thoughtful and constructive feedback on our manuscript. This research was supported by a University of California President's Dissertation Fellowship and National Science Foundation Postdoctoral Research Fellowship in Biology (Award no. 2109710) awarded to KMB.

Data availability All data and code for this manuscript are available on Dryad (Reviewer URL: https://datadryad.org/stash/share/z_-tWrlZejsLyKKdxVJSmQpYAjLEwTp3IExEFqNS3k).

Declarations

Ethics approval Animals were not handled for this study and our experimental procedures were minimally invasive. All applicable international, national, and institution guidelines for the use of animals were followed. All procedures in this study were approved by the Greek Ministry for Environment and Energy (permit YΠEN/ΔΔΔ/43912/1357) and the University of California, Berkeley Institutional Animal Care and Use Committee (AUP-2021-08-14567).

Conflict of interest The authors declare no conflict of interest.

Open Access This article is licensed under a Creative Commons Attribution 4.0 International License, which permits use, sharing, adaptation, distribution and reproduction in any medium or format, as long as you give appropriate credit to the original author(s) and the source, provide a link to the Creative Commons licence, and indicate if changes were made. The images or other third party material in this article are included in the article's Creative Commons licence, unless indicated otherwise in a credit line to the material. If material is not included in the article's Creative Commons licence and your intended use is not permitted by statutory regulation or exceeds the permitted use, you will need to obtain permission directly from the copyright holder. To view a copy of this licence, visit <http://creativecommons.org/licenses/by/4.0/>.

References

- Abalos J, Pérez i de Lanuza G, Carazo P, Font E (2016) The role of male coloration in the outcome of staged contests in the European common wall lizard (*Podarcis muralis*). *Behaviour* 153:607–631
- Allen JA (1972) Evidence for stabilizing and apostatic selection by wild blackbirds. *Nature* 237:348–349
- Allen JA, Raison HE, Weale ME (1998) The influence of density on frequency-dependent selection by wild birds feeding on artificial prey. *Proc R Soc Lond B* 265:1031–1035
- Andrade P, PinhoPérez i de Lanuza CG et al (2019) Regulatory changes in pterin and carotenoid genes underlie balanced color polymorphisms in the wall lizard. *P Natl Acad Sci USA* 116:5633–5642
- BeVier GT, Brock KM, Foufopoulos J (2021) Life on an island: the effects of insularity on the ecology and home range size of the Aegean wall lizard (*Podarcis erhardii*). *Herpetol Conserv Bio* 16:394–404
- Blumstein DT (2003) Flight-initiation distance in birds is dependent on intruder starting distance. *J Wildlife Manage* 2003:852–857
- Blumstein DT (2006) Developing an evolutionary ecology of fear: how life history and natural history traits affect disturbance tolerance in birds. *Anim Behav* 71:389–399
- Blumstein DT, Samia DSM, Stankowich T, Cooper WE (2015) Best practices for the study of escape behavior. In: Cooper WE, Blumstein DT (eds) *Escaping from predators: an integrative view of escape decisions*. Cambridge University Press, Cambridge, pp 407–419
- Border SE, DeOliveira GM, Janeski HM, Piefke TJ, Brown TJ, Dijkstra PD (2019) Social rank, color morph, and social network metrics predict oxidative stress in a cichlid fish. *Behav Ecol* 30:490–499
- Brock KM, Bednekoff PA, Pafilis P, Foufopoulos J (2015) Evolution of antipredator behavior in an island lizard species, *Podarcis erhardii* (Reptilia: Lacertidae): The sum of all fears? *Evolution* 69:216–231
- Brock KM, Baeckens S, Donihue CM, Martín J, Pafilis P, Edwards DL (2020) Trait differences among discrete morphs of a color polymorphic lizard, *Podarcis Erhardii*. *Peerj* 8:e10284
- Brock KM, Madden IE, Rosso AA, Ramos C, Degen R, Stadler SR, Ayton C, Fernandez MEL, Reyes Servin J (2022b) Patterns of colour morph diversity across populations of the Aegean Wall Lizard, *Podarcis erhardii* (Bedriaga, 1882). *Herpetol Notes* 15:361–364
- Brock KM, McTavish EJ, Edwards DL (2022c) Color polymorphism is a driver of diversification in the lizard family Lacertidae. *Syst Biol* 71:24–39
- Brock KM, Chelini MC, Madden IE, Ramos C, Ayton C, Blois JL, Pafilis P, Edwards DL (2022a) Colour morph predicts social behaviour and contest outcomes in a polymorphic lizard (*Podarcis erhardii*). *Anim Behav* (published online)
- Brodie ED III (1989) Genetic correlations between morphology and antipredator behaviour in natural populations of the garter snake *Thamnophis ordinoides*. *Nature* 342:542–543
- Brodie ED III (1992) Correlational selection for color pattern and antipredator behavior in the garter snake *Thamnophis ordinoides*. *Evolution* 46:1284–1298
- Coleman K, Wilson DS (1998) Shyness and boldness in pumpkinseed sunfish: individual differences are context-specific. *Anim Behav* 56:927–936
- Cooper WE Jr, Frederick WG (2007) Optimal flight initiation distance. *J Theor Biol* 244:59–67
- Dawkins R, Krebs JR (1979) Arms races between and within species. *Proc R Soc Lond B* 205:489–511
- Fielding J, Turland N (2008) *Flowers of Crete*, 2nd edn. Kew Publishing, London, UK
- Forsman A (1995) Opposing fitness consequences of colour pattern in male and female snakes. *J Evol Biol* 8:53–70
- Forsman A, Appelqvist S (1998) Visual predators impose correlational selection on prey color pattern and behavior. *Behav Ecol* 9:409–413
- Francis RC (1988) On the relationship between aggression and social dominance. *J Ethol* 78:223–237
- Gray SM, McKinnon JS (2007) Linking color polymorphism maintenance and speciation. *Trends Ecol Evol* 22:71–79
- Hurtado-Gonzales JL, Baldassarre DT, Uy JAC (2010) Interaction between female mating preferences and predation may explain the maintenance of rare males in the pentamorphic fish *Poecilia parae*. *J Evol Biol* 23:1293–1301
- Huyghe K, Vanhooydonck B, Herrel A, Tadić Z, Van Damme R (2007) Morphology, performance, behavior and ecology of three color morphs in males of the lizard *Podarcis melisellensis*. *Integr Comp Biol* 47:211–220
- Huyghe K, Husak JF, Herrel A, Tadić Z, Moore IT, Van Damme R, Vanhooydonck B (2009) Relationships between hormones, physiological performance and immunocompetence in a color-polymorphic lizard species, *Podarcis melisellensis*. *Horm Behav* 55:488–494
- Huyghe K, Small M, Vanhooydonck B, Herrel A, Tadić Z, Van Damme R, Backeljau T (2010) Genetic divergence among sympatric colour morphs of the Dalmatian wall lizard (*Podarcis melisellensis*). *Genetica* 138:387–393
- Labra A, Sufán-Catalán J, Solís R, Penna M (2007) Hissing sounds by the lizard *Pristidactylus volcanensis*. *Copeia* 2007:1019–1023
- Lattanzio MS, Miles DB (2016) Trophic niche divergence among colour morphs that exhibit alternative mating tactics. *R Soc Open Sci* 3:150531
- Li B, Belasen A, Pafilis P, Bednekoff P, Foufopoulos J (2014) Effects of feral cats on the evolution of anti-predator behaviours in island reptiles: insights from an ancient introduction. *Proc R Soc B* 281:20140339
- Lima SL, Dill LM (1990) Behavioral decisions made under the risk of predation: a review and prospectus. *Can J Zool* 68:619–640
- Maan ME, Eshuis B, Haesler MP, Schneider MV, van Alphen JJ, Seehausen O (2008) Color polymorphism and predation in a Lake Victoria cichlid fish. *Copeia* 2008:621–629
- Marcellini DL, Janssen TA (1991) Avoidance learning by the curly-tailed lizard, *Leiocephalus schreibersi*: Implications for anti-predator behavior. *J Herpetol* 25:238–241
- Marshall KL, Philpot KE, Stevens M (2016) Microhabitat choice in island lizards enhances camouflage against avian predators. *Sci Rep* 6:1815
- McKinnon JS, Pierotti ME (2010) Colour polymorphism and correlated characters: genetic mechanisms and evolution. *Mol Ecol* 19:5101–5125

- Miles DB, Calsbeek R, Sinervo B (2007) Corticosterone, locomotor performance, and metabolism in side-blotched lizards (*Uta stansburiana*). *Horm Behav* 51:548–554
- Ng J, Landeen EL, Logsdon RM, Glor RE (2013) Correlation between *Anolis* dewlap phenotype and environmental variation indicates adaptive divergence of a signal important to sexual selection and species recognition. *Evolution* 67:573–582
- Pérez i de Lanuza G, Carretero MA (2018) Partial divergence in microhabitat use suggests environmental-dependent selection on a colour polymorphic lizard. *Behav Ecol Sociobiol* 72:138
- Pérez i de Lanuza G, Font E (2015) Differences in conspicuousness between alternative color morphs in a polychromatic lizard. *Behav Ecol* 26:1432–1446
- Pérez i de Lanuza G, Font E, Carazo P (2013) Color-assortative mating in a color-polymorphic lacertid lizard. *Behav Ecol* 24:273–279
- Pérez-Cembranos A, Pérez-Mellado AV, Cooper WE (2013) Predation risk and opportunity cost of fleeing while foraging on plants influence escape decisions of an insular lizard. *J Ethol* 119:522–530
- Punzalan D, Rodd FH, Hughes KA (2005) Perceptual processes and the maintenance of polymorphism through frequency-dependent predation. *Evol Ecol* 19:303–320
- R Core Team (2021) R: A language and environment for statistical computing. R Foundation for Statistical Computing, Vienna, Austria, <https://www.R-project.org/>. Accessed 1 Aug 2021
- Reaney LT, Backwell PR (2007) Risk-taking behavior predicts aggression and mating success in a fiddler crab. *Behav Ecol* 18:521–525
- Reid DG (1987) Natural selection for apostasy and crypsis acting on the shell colour polymorphism of a mangrove snail, *Littoraria filosa* (Sowerby) (Gastropoda: Littorinidae). *Biol J Linn Soc* 30:1–24
- Runyan AM, Blumstein DT (2004) Do individual differences influence flight initiation distance? *J Wildlife Manage* 68:1124–1129
- Sandoval CP (1994) Differential visual predation on morphs of *Timema cristinae* (Phasmatodeae: Timemidae) and its consequences for host range. *Biol J Linn Soc* 52:341–356
- Shigemiyama Y (2004) Reversible frequency-dependent predation of a puffer, *Takifugu niphobles* (Pisces: Tetraodontidae), related to spatial distribution of colour-polymorphic prey. *Biol J Linn Soc* 81:197–202
- Sinervo B, Lively CM (1996) The rock–paper–scissors game and the evolution of alternative male strategies. *Nature* 380:240–243
- Sinervo B, Miles DB, Frankino WA, Klukowski M, DeNardo DF (2000) Testosterone, endurance, and Darwinian fitness: natural and sexual selection on the physiological bases of alternative male behaviours in Side-blotched lizards. *Horm Behav* 38:222–233
- Snell HL, Jennings RD, Snell HM, Harcourt S (1988) Intrapopulation variation in predator-avoidance performance of Galapagos lava lizards: the interaction of sexual and natural selection. *Evol Ecol* 2:353–369
- Stuart-Fox D, Aulsebrook A, Rankin KJ, Dong CM, McLean CA (2021) Convergence and divergence in lizard colour polymorphisms. *Biol Rev* 96:289–309
- Svensson EI (2017) Back to basics: using colour polymorphisms to study evolutionary processes. *Mol Ecol* 26:2204–2211
- Valakos ED, Pafilis P, Sotiropoulos K, Lymberakis P, Maragou P, Foutopoulos J (2008) The amphibians and reptiles of Greece. Edition Chimaira, Frankfurt
- Vercken E, Clobert J (2008) Ventral colour polymorphism correlates with alternative behavioural patterns in female common lizards (*Lacerta vivipara*). *Ecoscience* 15:320–326
- Ydenberg RC, Dill LM (1986) The economics of fleeing from predators. *Adv Stud Behav* 16:229–249
- Yewers M, Pryke S, Stuart-Fox D (2016) Behavioural differences across contexts may indicate morph-specific strategies in the lizard *Ctenophorus decresii*. *Anim Behav* 111:329–339

Publisher's note Springer Nature remains neutral with regard to jurisdictional claims in published maps and institutional affiliations.

Authors and Affiliations

Kinsey M. Brock¹  · Indiana E. Madden²

¹ Department of Environmental Science, Policy, and Management, College of Natural Resources, University of California, Berkeley, Berkeley, CA, USA

² School of Veterinary Medicine, University of California, Davis, Davis, CA, USA