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GONADAL COMPARISON BETWEEN MORPHS OF MALES *Sceloporus minor* LIZARD IN A POPULATION OF CENTRAL MEXICO

Comparación gonadal entre morfos de los machos de la lagartija *Sceloporus minor* en una población del centro de México

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ABSTRACT

To date, studies have found that polymorphism in a population of the lizard *Sceloporus minor* from central Mexico is not maintained by niche partitioning, thus leaving open the possibility that the polymorphism is being maintained by frequency-dependent selection or by variation in reproductive potential. Therefore, the objective of this study is to make a gross gonadal comparison between males of the yellow and red morphs of the lizard *S. minor* from a population (El Enzuelado) of the state of Hidalgo, Mexico. The results indicate that there are no significant differences in the volume and weight of the testicles in any of the years analyzed. Also, no significant differences were recorded in the weight of the liver or of the fat body. On the other hand, significant differences were recorded in the volume and weight of the testicles among different seasons of the year for both morphs. The lack of significant variation in testis size between morphs recorded in this study indirectly indicates that the two morphs have similar reproductive potential, and therefore the hypothesis that frequency-dependent selection is the force that maintains polymorphism in this population becomes more plausible.

Keywords: Fecundity, frequency-dependent selection, polymorphism, Squamata.

RESUMEN

A la fecha, algunos estudios han encontrado que el polimorfismo en una población de la lagartija *Sceloporus minor* del centro de México no se mantiene por partición de nicho, dejando abierta la posibilidad de que el polimorfismo se mantenga por selección dependiente de la frecuencia o por variación en el potencial reproductor. Por lo tanto, el objetivo de este estudio es hacer una comparación gonadal macroscópica entre machos del morfo amarillo y rojo de la lagartija *S. minor* de una población (El Enzuelado) del estado de Hidalgo, México. Los resultados indican que no hay diferencias significativas en el volumen y peso de los testículos en ninguno de los años analizados. Además, no se registraron diferencias significativas en el peso del hígado o del cuerpo graso. Por otra parte, se registraron diferencias significativas en el volumen y peso de los testículos entre las diferentes estaciones del año para ambos morfos. La falta de variación significativas en el tamaño de los testículos entre morfos registrada en este estudio, indicando indirectamente que los dos morfos tienen un potencial reproductor similar y, por lo tanto, la hipótesis de que la selección dependiente de la frecuencia es la fuerza que mantiene el polimorfismo en esta población, toma más relevancia.

Palabras clave: Fecundidad, polimorfismo, selección dependiente de la frecuencia, Squamata.

INTRODUCTION

In ecology, polymorphism is defined as the existence of two or more distinguishable forms (morphs) that coexist temporally and spatially in a population with interbreeding, in which each morph is associated with a specific genetic load (Gray and McKinnon, 2007; Andrade, 2021). In recent years, intrasexual color polymorphism in some species of Squamata reptiles has drawn the attention of evolutionary ecologists (Sinervo et al., 2000; Zamudio and Sinervo, 2000; Lattanzio and Miles, 2016: Scali et al., 2016; Sacchi et al., 2017; Yewers et al., 2017; Hernández-Gallegos et al., 2018; Paterson and Blouin-Demers, 2018; García-Rosales et al., 2019b). This interest has focused on species where colors vary with different ecological and/or reproductive strategies (Thompson and Moore, 1991, 1992; Sinervo and Lively, 1996; Galeotti et al., 2013). These strategies represent alternate pathways to reproductive success among members of a single sex, with the expression of multiple interrelated phenotypes that work together to maximize fitness (Svensson et al., 2001; Taborsky et al., 2008).

Alternate reproductive tactics (ARTs) are generally related to different morphological, behavioral, physiological, and life history traits (Sinervo and Lively, 1996; Svensson et al., 2001; Taborsky et al., 2008); for example, they may be determined by a conditioned pattern, and occur about physical size, as has been observed in some species of lizards, where the larger males are territorial, while the smaller ones generally do not defend territories, and wander around in the periphery of the area guarded by territorial males to try to steal some resource from them (satellite strategy; Sinervo and Lively, 1996; García-Rosales et al., 2021). Alternatively, these tactics can be related to population sizes, social or environmental variables, differences in effective fertilization, or offspring fitness, among others (Bounous, 2015). For example, in *Uta stansburiana* Bair and Girard, 1852, female reproductive strategies are dependent on population density; orange-throated females, who have many young that are small in size (r-strategy) are favored at low densities, because their young are more likely to survive as a consequence of little competition, while at high densities, yellow-throated females have high reproductive success (k-strategy), because their offspring are born larger and therefore are more competitive than the offspring of orange-throated females (Svensson et al., 2001).

The existence of multiple color morphotypes within the same population, is a characteristic presented by several species of lizards of the genus *Sceloporus* Wiegmann, 1828; for example, *S. horridus* Wiegmann, 1834 (Bustos-Zagal et al., 2014), *S. undulatus erythrocheilus* Maslin, 1956 (Rand, 1992), *S. grammicus* Wiegmann, 1828 (Bastiaans et al., 2013), *S. torquatus* Wiegmann, 1828 (Domínguez-Guerrero, 2015), and *S. minor* Cope, 1885 (Stephenson, 2010; García-Rosales et al., 2019b, 2021). In some of these studies, it has been observed that different morphs can show differences in their morphology, ecology, or behavior, among other traits (Stephenson, 2010; Bastiaans et al., 2013; Bustos-Zagal et al., 2014; Domínguez-Guerrero, 2015). For example, a morphological variation associated with color patterns has been recorded in the lizards *S. horridus* (Bustos-Zagal et al., 2014) and *S. torquatus* (Domínguez-Guerrero, 2015), while in *S. grammicus,* the existence of female choice has been recorded, where females prefer certain color morphs shown by males, more frequently discriminating against males that are not from their population (Bastiaans et al., 2014). Bastiaans et al. (2013) note that in this species, there is a variation in the aggressive behavior associated with the placement of patterns on individuals' throats. On the other hand, there are other species of the same genus where no differences between morphs have been recorded, such as *S. aeneus* Wiegmann, 1828, where no morphological or gonadal variation associated with color patterns is observed (Jiménez-Arcos, 2013; Hernández-Gallegos et al., 2018).

The species of the genus *Sceloporus* have been considered a good study model to learn about different aspects of ecological, physiological, and behavioral traits derived from polymorphism in color (Rand, 1992; Bastiaans et al., 2013; Bustos-Zagal et al., 2014; Hernández-Gallegos et al., 2018; García-Rosales et al., 2019b, 2021). This polymorphism associated with color (polychromatism) enables the potential morphological, behavioral, and reproductive variations to be evaluated (Stephenson, 2010; García-Rosales et al., 2019b; Andrade, 2021), characteristics that are essential for determining how polymorphism is being maintained in the different species and populations of the same species (Sinervo and Lively, 1996; Lattanzio and Miles, 2016; Scali et al., 2016; Paterson and Blouin-Demers, 2018). In this regard, it has been noted that to maintain polymorphism in a population, each morph must reach the same fitness over a long period (Gross, 1996; Taborsky et al., 2008), and this balance can be achieved through three processes; niche partitioning (Skúlason and Smith, 1995; Lattanzio and Miles, 2016; Scali et al., 2016), frequency-dependent selection (Sinervo and Lively, 1996; Pryke et al., 2007), or variation in reproductive potential (Galeotti et al., 2013).

The niche partitioning hypothesis proposes that individuals of different morphs exploit different environmental resources (e.g., space, shelter, or food) to avoid strong competition for the same resource (Skúlason and Smith, 1995; Lattanzio and Miles, 2016; Scali et al., 2016; Paterson and Blouin-Demers, 2018). Frequency-dependent selection can maintain polymorphism because this type of selection confers survival and/or reproductive advantages to rare morphs; thus, the fitness of a certain phenotype depends on the frequencies of the other phenotypes with which it competes (Sinervo and Lively, 1996; Pryke et al., 2007). The variation in reproductive potential hypothesis proposes that males of a particular morph (usually a satellite morph) have certain traits that enable them to father a greater number of offspring (e.g., larger testicles, higher sperm production) compared to those of other morphs with which it interacts. The lower number of females that this morph gets is compensated by its higher reproductive potential (greater postcopulatory reproductive success in a situation of sperm competition); therefore, all the morphs are evolutionarily stable because they reach the same reproductive success, although using different strategies (Olsson et al., 2007, 2009; McDiarmid et al., 2017).

Sceloporus minor is a good model of lizard species for which it is important to examine some of its life history traits due to its distribution range (Ramírez-Bautista et al., 2008, 2013; García-Rosales et al., 2019a). Various studies have been carried out with populations of the species, including studies focused on the biological importance of polymorphism (Stephenson, 2010), morphology and comparative genetics (García-Rosales et al., 2017), trophic ecology (García-Rosales et al., 2019b), spatial distribution, and aggressive and sexual behavior (García-Rosales et al., 2021), among others. The results of these studies indicate that the males of this species show a color polymorphism in the dorsal region of the body, where they can be blue, red, yellow, or brown; however, not all morphs are present in every population (Stephenson, 2010; García-Rosales et al., 2017). In this regard, García-Rosales et al. (2017, 2019b), note that in one population in Central Mexico (El Enzuelado), there are only two morphs (red and yellow), and as far as is known, these morphs do not show differences in size (SVL) or weight, consume the same types of prey, use the same microhabitat, and perch at the same height. Also, both are territorial, but the yellow morph is slightly more aggressive than the red morph (García-Rosales et al., 2019b, 2021).

Despite these findings, there are certain aspects of the polymorphism of this species that are still unknown; for example, the reproductive potential of the two morphs, which could help lead to an understanding of the forces that maintain polymorphism in this population. Therefore, the objective of this work is to carry out a gross gonadal comparison between the males of the yellow and red morphs of the lizard *S. minor* (Squamata: Phrynosomatidae) from a population in the state of Hidalgo (El Enzuelado), Mexico. The objectives are i) to compare the weight and volume of the gonads, as well as the weight of the fat body and liver between morphs, and ii) to analyze and compare the volume and weight of the gonads in three seasons of the year. According to what was reported by García-Rosales et al. (2019b, 2021), the polymorphism in the population of *S. minor* in the locality of El Enzuelado is not being maintained by niche partitioning, so this leaves open the possibility that the polymorphism is maintained by frequency-dependent selection or by variation in reproductive potential. Lozano et al. (2015), noted that the size of the testicles is related to the number of sperm produced. In this context, if the morphs show different gonadal sizes, it will indirectly indicate that the lizards have a different reproductive potential (Olsson et al., 2009). Usually, the male and/or morph that shows the greatest reproductive potential is the least competitive (generally the satellites). In this way they compensate for the lower number of females they can access (Olsson et al., 2007, 2009; McDiarmid et al., 2017). However, in this population of *S. minor*, both morphs are territorial, therefore we expect the size of the gonads (volume and weight) not to show differences between morphs in any of the years analyzed here. In addition, we expect that the volume and weight of the testicles will show differences among seasons in both morphs.

MATERIALS AND METHODS

Study area

The fieldwork was carried out in an area of approximately five hectares, near the community of El Enzuelado (20° 35′ N, and 98° 37′ W) in the municipality of San Agustín Metzquititlan, Hidalgo, Mexico. The study area is located at an elevation of 1,955 m, and the dominant vegetation is xeric scrub (Rzedowski, 1978). The average annual temperature is 17.5 °C and the average annual rainfall is 496.7 mm (Pavón and Meza-Sánchez, 2009).

Gonadal analysis

A total of 69 adult males of *Sceloporus minor* were used for gonadal analysis. Of these, 24 (eight yellow and 16 red) were collected in 2012, 32 (14 yellow and 18 red) were collected in 2017, and 13 in 2018 (six red and seven yellow). These individuals had previously been used for morphology and comparative genetics (García-Rosales et al., 2017) and trophic ecology studies (García-Rosales et al., 2019b); therefore, there was no need to sacrifice any further individuals for this study. In both samplings, the lizards were collected at different periods of their reproductive cycle. All individuals collected from December to March were considered to be within the post-breeding period. Those collected from April to August were considered to be within the pre-reproductive period, and the lizards collected from September to November were considered to be reproducers (Ramírez-Bautista et al., 2013).

Of the 24 males collected in 2012, six (two yellow and four red) were collected in the post-breeding period, nine (two yellow and seven red) were collected in the pre-breeding period, and nine (four yellow and five red) were collected in the reproductive period. Of the 45 males collected in 2017 and 2018, 20 were collected in June (pre-reproductive season; nine yellow and 11 red) 12 in November 2017 (reproductive period; five yellow and seven red) and 13 were collected in March 2018 (post-breeding period; six red and seven yellow). All lizards used in this analysis had been killed in the laboratory by intracoelomic injection of sodium pentobarbital. The samples were then fixed in 10 % formalin and preserved in 70 % ethanol (see García-Rosales et al., 2019b).

The specimens had been deposited in the Amphibian and Reptile Collection of the Center for Biological Research, Autonomous University of the State of Hidalgo. This study was carried out in accordance with the ethics and regulations for animal research of the Autonomous University of the State of Hidalgo, the AVMA Guidelines on Euthanasia (AVMA 2013) and the policies for handling animal specimens described in the Norma Oficial Mexicana NOM-033 -SAG/ZOO-2014 standard. We obtained approval for the use of animals under the permits SGPA/DGVS/11746/13 and SGPA/DGVS/06183/17 issued by the Ministry of the Environment and Natural Resources (SEMARNAT) of the government of Mexico.

Before fixing the individuals, we made a small cut in the middle part of the abdomen. Each of the gonads was removed, and then measured (length and width) with a Mitutoyo digital vernier caliper (± 0.01 mm) and weighed with an Adam brand analytical balance (± 0.0001 g). We calculated the volume of each gonad according to the recorded measurements (length and width), using the formula for an ellipsoid (Duré et al., 2009):

$$V = \frac{4}{3}\pi * \left[\left(\frac{length}{2} \right) * \left(\frac{width}{2} \right)^2 \right].$$

In addition, the mass of the liver and fat bodies were removed and weighed with the same Adam scale. Finally, both gonads, as well as the fatty body and the liver, were returned to the abdominal cavity of the respective individuals, and the lizards were tied with a thread in the middle part of the body to prevent any of the analyzed organs from coming out and getting lost.

Statistical analysis

To remove the effect of SVL on the other variables, multivariate regressions were performed, thereby obtaining the residuals for each variable, which were used as input to the other analyses. To evaluate the possible difference in the volume and weight of each of the testicles separately and the weight of the liver and fat body between morphs and between years, non-parametric Kruskal-Wallis tests were applied because the data do not show a normal distribution (Zar, 1999). In addition, we performed Kruskall-Wallis tests to evaluate possible differences in the volume and weight of the gonads among the three seasons in which the individuals were collected (pre-reproductive, reproductive, and post-breeding period). For this last analysis, we used the average value of both gonads, both for volume and weight. In addition, for the data from 2012, differences among seasons were only evaluated for the red morph, since the sample size for the yellow morph was too low. Also, we decided to pool the data from 2017 and 2018 because the time sampling was not too long; furthermore, by pooling the data we were able to cover three different periods in the reproductive cycle of this species. Tests were considered significant if $p \leq 0.05$. We used the free program PAST version 4.10 for all statistical analyses. Means are presented as $\bar{x}$ ± standard error.

RESULTS

The volume and weight of the testes between morphs of *Sceloporus minor* did not show significant differences in any of the years (Table 1). However, it is observed that the yellow males collected in 2012 have greater testicular volume and weight than the red males collected in the same year (Table 1), while for the lizards collected in 2017-2018, it was observed that the pattern is the other way around: the red males have greater testicular volume and weight than the yellow males collected in those years (Table 1). No significant differences were recorded in the weight of the liver or the fat body; however, in both traits, a greater weight was recorded in the red morph for the year 2012, while in 2017-2018 higher weight was registered in the yellow morph (Table 1).

The results of the Kruskall-Wallis test showed that there are significant differences in the volume and weight of the testicles at different times of the year for both morphs and periods (Fig. 1, Appendix 1). Significant differences were recorded in the volume ($H = 10.59$, $p = 0.005$) and weight ($H = 10.16$, $p = 0.006$) of the testicles for the red morph for the year 2012 (Fig. 1a-b, Appendix 1). Both traits reach their largest size in the reproductive season (volume: $\bar{x} = 214.66 \pm 52.53$ mm^3; weight: $\bar{x} = 0.20 \pm 0.05$ g), while the smallest gonadal volume ($\bar{x} = 34.35 \pm 3.26$ mm^3) was recorded in the pre-reproductive season and the smallest gonadal weight ($\bar{x} = 0.014 \pm 0.002$ g) was recorded in the post-breeding period. For 2017-2018, we recorded significant differences in the volume ($H = 10.95$, $p = 0.004$) and weight ($H = 10.92$, $p = 0.004$) of yellow morph testicles (Fig. 1c-d, Appendix 1). Both traits reach their largest size in the reproductive season (volume: $\bar{x} = 157.04 \pm 27.12$ mm^3; weight: $\bar{x} = 0.13 \pm 0.30$ g), while the smallest gonadal size was recorded in the post-breeding period (volume, $\bar{x} = 19.32 \pm 2.70$ mm^3; weight, $\bar{x} = 0.01 \pm 0.004$ g). A similar pattern was observed for the red morph for both traits in 2017-2018 (volume: $H = 15.1$, $p = 0.0005$; weight: $H = 14.8$, $p = 0.0006$; (Fig. 1c-d, Appendix 1). The largest testicular volume occurred in the reproductive season ($\bar{x} = 216.22 \pm 25.94$ mm^3), as well as the largest weight ($\bar{x} = 0.15 \pm 0.019$ g), while the smallest volume and weight were recorded in the post-reproductive period (volume, $\bar{x} = 21.23 \pm 4.60$ mm^3, weight; $\bar{x} = 0.008 \pm 0.001$ g; Fig. 1c-d, Appendix 1). The largest testicular volume for both morphs was reached in the reproductive period; but although the red morph showed a larger volume compared to the yellow morph in both years, the differences were not significant ($H = 5.90$, $p = 0.11$).

Table 1. Variables analyzed in the comparison of gonads, fatty body and liver between males of the yellow and red morphs of *Sceloporus minor*. Measurements are reported as mean ± standard error (range). The statistical test used was Kruskall-Wallis, where the value of H is observed followed by the probability (*p*).

	Males from El Enzuelado					
	2012		2017-2018			
	Yellow (n = 8)	Red (n = 16)	Yellow (n = 21)	Red (n = 24)	H	*p*
Right testis volumen (mm^3)	120.09 ± 36.92 (23.19-332.86)	97.95 ± 29.17 (17.74-379.98)	55.86 ± 13.71 (9.37-255.39)	75.89 ± 18.82 (5.20-343.28)	1.52	0.67
Right testis weight (g)	0.11 ± 0.04 (0.01-0.33)	0.08 ± 0.02 (0.007-0.35)	0.04 ± 0.01 (0.00-0.25)	0.05 ± 0.01 (0.00-0.23)	1.13	0.76
Left testicle volumen (mm^3)	101.51 ± 26.31 (17.86-222.93)	86.12 ± 23.81 (14.17-307.58)	57.54 ± 15.39 (6.33-232.17)	85.36 ± 20.50 (10.72- 361.63)	2.58	0.46
Left testicle weight (g)	0.08 ± 0.03 (0.01-0.19)	0.06 ± 0.02 (0.007-0.29)	0.04 ± 0.01 (0.00-0.22)	0.05 ± 0.01 (0.00-0.24)	1.24	0.74
Liver (g)	0.48 ± 0.04 (0.35-0.69)	0.49 ± 0.04 (0.240.93)	0.44 ± 0.02 (0.21-0.62)	0.39 ± 0.02 (0.25-0.62)	0.79	0.85
Fatty body (g)	0.18 ± 0.04 (0.02–0.39)	0.18 ± 0.03 (0.05-41)	0.15 ± 0.03 (0-0.75)	0.08 ± 0.01 (0.01-0.25)	1.85	0.60

DISCUSSION

Many studies have indicated that polymorphism can be maintained through three main forces: frequency-dependent selection, niche partitioning, or variation in reproductive potential (Sinervo and Lively, 1996; Galeotti et al., 2013; Lattanzio and Miles, 2016; Scali et al., 2016). The latter hypothesis proposes that the males of a particular morph (usually satellite morphs) have some traits that enable them to father more children than the other morphs with which they interact (Galeotti et al., 2013). The lower number of females that this morph gets is compensated by its higher reproductive potential (Olsson et al., 2007, 2009; McDiarmid et al., 2017). For example, Olsson et al. (2009) reported significant differences in the size of the testicles of the different morphs of *Ctenophorus pictus* Peters, 1866, with the yellow males having larger testicles than the red males. In turn, this greater testicular size is related to greater sperm production and a three times greater probability of paternity than red morph males; however, the red morph is better at defending its territory and has more females. Therefore, each morph is evolutionarily stable, because they reach the same reproductive success but using different strategies (Olsson et al., 2007, 2009; McDiarmid et al., 2017). However, this is not the case for *S. minor* from this population in any of the years analyzed, as no significant differences in testes volume or weight were recorded between the red morph and the yellow morph. Similarly, Hernández-Gallegos et al. (2018), did not record significant differences in the size of the testicles among the three morphs shown by the males (grey, yellow, and orange) of *S. aeneus*. Consequently, they state that the polymorphism in this species is being maintained by frequency-dependent selection (Jiménez-Arcos, 2013; Hernández-Gallegos et al., 2018), as has been suggested for *U. stansburiana* Baird and Girard, 1852 (Sinervo and Lively, 1996).

Since there are no differences in the size and weight of the testicles in the males of this population, one might postulate that there are also no differences in the amount of sperm produced by each morph (Lozano et al., 2015). Therefore, the reproductive potential of the two morphs could be similar, and consequently, the polymorphism in this population is not being maintained by differing reproductive potential. This information, together with that reported by García-Rosales et al. (2019b, 2021), where they note that the polymorphism in this population is not being maintained by niche divergence, leaves the possibility that its polymorphism is being maintained by frequency-dependent selection (García-Rosales et al., 2019b). However, these results should be taken with caution, since although we found no significant differences in gonadal size, the volume of the gonads was larger for the yellow morph in 2012 and larger for the red morph in 2017 and 2018. Nevertheless, these

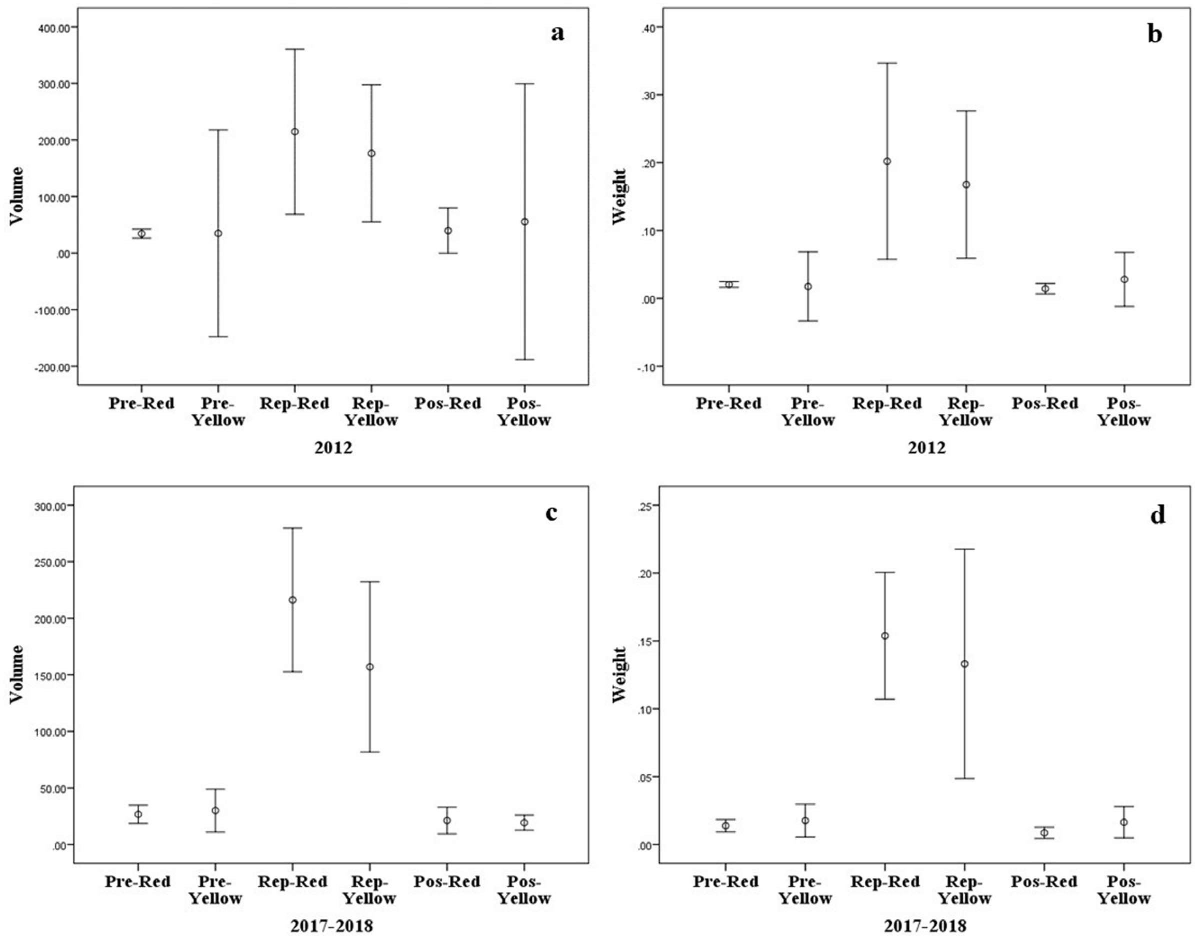

Figure 1. Variation in the volume (mm^3) and weight (g) of the gonads among seasons of the year for males of the two morphs of *S. minor*. a-b individuals of 2012 and c-b individuals of 2017–2018. Pre= pre-reproductive, Rep= reproductive and, Pos= post-reproductive.

differences in the gonad volume could be a direct consequence of the sample size. Therefore, future research should consider conducting this type of study with larger sample sizes, in addition to performing histological studies that include sperm counts, as well as paternity tests. In addition, it is necessary to carry out studies where the proportion of morphs is quantified over a long period, making it possible to corroborate or refute the hypothesis that frequency-dependent selection is the force maintaining polymorphism in this population of *S. minor*.

Color polymorphism is generally associated with a set of behavioral, physiological and life history traits (Sinervo and Lively, 1996; Zamudio and Sinervo, 2000; Svensson et al., 2001; Lattanzio and Miles, 2016: Scali et al., 2016; Sacchi et al., 2017; Yewers et al., 2017; Paterson and Blouin-Demers, 2018) and generally the yellow morphs tend to be the least dominant concerning the other morphs with which they interact (Stuart- Fox et al., 2020). Therefore, these morphs likely use alternative reproductive strategies (pre- or post-copulatory) to achieve reproductive success (Sinervo and Lively, 1996; Olsson et al. 2009). However, the lack of variation in test size recorded in this study and the aggressive behavior observed in both morphs (García-Rosales et al., 2021) suggest that the reproductive strategies in the two morphs may be similar. In this regard, Sacchi et al. (2017) note that aggressive behavior and average testosterone levels are similar among the morphs of *Podarcis muralis*; however, they did register variation in the seasonal levels of this hormone among morphs, which could increase aggressiveness, sexual behavior, and sperm production. Further study would be necessary to evaluate the seasonal levels of testosterone in the blood and its relationship with aggressive behavior and sperm production in the different morphs of *S. minor*.

On the other hand, it has been stated that in arid environments where resources are scarce and/or where population density is high, an aggressive strategy is more effective for defending resources (McLean et al., 2015). The individuals used for this study come from an arid environment, which could favor aggressive behavior in both morphs; however, there is no data on population density, proportion of morphs, or the quality and abundance of resources. Therefore, it is necessary to measure the population density and proportion of morphs over a long period, as well as to measure

Appendix 1. Variation in the volume and weight of the gonads among seasons of the year for males of the two morphs of *S. minor*. Measurements are reported as mean ± standard error (range). The statistical test used was Kruskall-Wallis, where the value of H followed by the probability (*P*) are observed.

	2012				2017-2018			
	Yellow		Red		Yellow		Red	
	Volume (mm^3)	Weight (gr)	Volume (mm^3)	Weight (gr)	Volume (mm^3)	Weight (gr)	Volume (mm^3)	Weight (gr)
Pre-reproductive	34.91±14.39 (20.52-49.30)	0.017±0.004 (0.013-0.021)	34.35±3.26 (25.98-46.73)	0.020±0.001 (0.013-0.028)	30.02±8.21 (11.42-79.19)	0.017±0.005 (0.006-0.05)	26.73±3.60 (8.24-46.82)	0.01±0.002 (0.006-0.02)
Reproductive	176.35±38.08 (109.09-264.50)	0.16±0.034 (0.11-0.25)	214.66±52.53 (96.28-339.55)	0.20±0.052 (0.073-0.32)	157.04±27.12 (93.94-243.78)	0.13±0.30 (0.05-0.23)	216.22±25.94 (128.80-352.45)	0.15±0.019 (0.086-0.24)
Pos-breeder	55.57±19.18 (36.38-74.76)	0.027±0.003 (0.024-0.031)	39.70±12.56 (15.96-74.76)	0.014±0.002 (0.007-0.018)	19.32±2.70 (8.94-27.49)	0.01±0.004 (0.0049-0.040)	21.23±4.60 (8.30-41.26)	0.008±0.0016 (0.003-0.14)
H	-	-	10.59	10.16	10.95	10.92	15.1	14.8
p	-	-	0.005	0.006	0.004	0.004	0.0005	0.0006

the availability, abundance, and quality of the resources that these lizards utilize, to generate more robust conclusions.

On the other hand, no significant differences were recorded in the weight of the liver nor of the fat body. In this regard, García-Rosales et al. (2019b, 2021), note that the two morphs show several similarities in specific characteristics that could alter the size of the fat body, such as body size (SVL), home range size (HR), and similar diet; furthermore, both morphs are territorial. Therefore, it is understandable that no differences were recorded in the weight of fat bodies. However, it is interesting to note that the yellow morph showed a greater fat body weight than the red morph in 2017–2018. This result could indicate that red morph individuals perform some activity that causes extra energy expenditure, and therefore their fat bodies are smaller in size (Derickson, 1976). García-Rosales et al. (2021) note that the red morph has a greater number of females within its HR compared to the yellow morph, which could translate into a greater number of copulations, and therefore a greater energy expenditure. In turn, it can translate into a smaller size of the fat bodies (due to energy expenditure); however, there is not enough evidence to confirm this idea. An interesting fact is that despite the greater aggressiveness and frequency of regenerated tails (a phenomenon that requires a lot of energy) shown by the yellow morph compared to the red morph (see García-Rosales et al., 2021), the yellow morph showed a higher amount of fat bodies, although the difference was not significant. Yet one would expect the relationship to be the reverse. Therefore, subsequent studies should consider a design that enables the behavior of the two morphs to be compared in different contexts, measuring the energy expenditure invested in each activity. It should be noted that during the study carried out by García-Rosales et al. (2021), the individuals were collected to analyze diet, as well as the size of the gonads, fat body, and liver (the latter is what is shown in this work). Therefore, our ideas regarding the size of fat bodies and their relationship with the activity shown by males in this population have previous support.

On the other hand, the Kruskal-Wallis test showed that there are significant differences in the volume and weight of the testicles among the different seasons of the year for both morphs and in both years (in 2012, the analysis was only done for the red morph). Both the weight and volume of the testicles were found to be greater in the reproductive season (September–November), similar to what was found by Ramírez-Bautista et al. (2013) for the same species and population, but in different years, where it is stated that September–November are when these lizards reach their maximum gonadal size; that is, testicular growth begins in July and August, peaking from September to November, then gradually decreasing from December to March. However, this period of maximum testicular growth recorded for September to November does not apply to all populations of *S. minor* (Ramírez-Bautista et al., 2008, 2013). Apparently, these differences are partially related to different environmental characteristics of the places where these populations live (Ramírez-Bautista et al., 2008, 2013).

CONCLUSION

In conclusion, no significant differences were recorded in the volume and weight of the testes between the yellow and red morphs of *S. minor*, which could indirectly suggest that the two morphs have a very similar reproductive potential. Therefore, the hypothesis that frequency-dependent selection is the force that maintains polymorphism in this population becomes more plausible. However, it would be necessary to carry out histological studies including sperm counts and paternity tests to generate more robust conclusions about the reproductive potential of each morph (Olsson et al., 2009; Lozano et al., 2015). Finally, it is necessary to measure the availability, abundance, and quality of the resources that these lizards utilize, and to carry out further field studies that can quantify the proportion of morphs over a long period, to see if this proportion changes over time. This would help an understanding of whether frequency-dependent selection is the force that maintains polymorphism in this population (Sinervo and Lively, 1996).

AUTHOR'S PARTICIPATION

A.V.H.L. formulated the idea for the manuscript, performed the statistical analyses, and drafted the first version of the manuscript. A.R.B. formulated the idea for the manuscript, provided the technical material, and revised the manuscript. A.G.R. performed the field wok, formulated the idea for the manuscript, performed the statistical analyses, and drafted and revised the manuscript. M.A.A.V. Drafted and revised the manuscript.

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CONFLICT OF INTEREST

The authors declare that they have no conflicts of interest.

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