SHORTER COMMUNICATIONS

Predators and Mates: Conflicting Selection on the Size of Male Pacific Treefrogs (*Pseudacris regilla*)

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ABSTRACT.—Despite the large amount of work on frog mating systems, the potential role of predators as an agent of selection on breeding adults has received very little study. Here, I use data from multiple populations of Pacific Treefrogs (*Pseudacris regilla*) to demonstrate that sexual selection from mating success favors larger males, but natural selection from predation by giant water bugs favors smaller males. Additionally, I found no relationship between male body condition and mating success or predation risk. This result demonstrates that predation is a potentially important agent of selection that counteracts sexual selection in anurans.

Many traits that increase male mating success also have the potential to make those males more susceptible to capture by predators. Examples of this phenomenon, in which natural selection from predation risk counteracts sexual selection from mating success, are being discovered in a broad range of taxa (e.g., Ryan et al., 1982; Magnhagen, 1991: Kotiaho et al., 1998; Godin and McDonogh, 2003). When natural selection and sexual selection conflict, predation risk can limit or reverse the evolutionary response to sexual selection. For instance, in Green Swordtails, *Xiphophorus helleri*, males with long tails and large body size are favored by sexual selection, but males from populations that coexist with predators had relatively shorter tails than males from predator-free populations (Basolo and Wagner, 2004). This indicates that selection from predators has limited the evolutionary response to sexual selection. Thus, studies estimating selection from predators on traits influencing mating success are necessary to understand the evolution of those traits.

Anurans with lek mating systems are a group in which it seems likely that natural selection from predation should act in opposition to sexual selection from mating success. These anurans breed in large aggregations with conspicuous calling behavior, characteristics that make them an excellent model system for the study of sexual selection and mating systems (Ryan, 1991; Gerhardt, 1994). Yet because of their conspicuous breeding behavior, the male frogs are vulnerable to a diverse range of predators (Ryan et al., 1982; Hinshaw and Sullivan, 1990; Lodé et al., 2004; Toledo, 2005). If any of these predators exert selection on the same traits as does mating success, they could act to inhibit the evolutionary response to sexual selection. However, within anurans only the classic work on the Túngara Frog (*Physalaemus pustulosus*) has investigated this, demonstrating that natural selection from predation conflicts with sexual selection from female choice (e.g., Ryan et al., 1982). Despite this finding, and the importance of anurans as a model system for studying sexual selection and mating systems, the role of predators as an agent of

selection on the phenotypes of breeding male frogs has been overlooked by empirical studies for over two decades.

Here, I present field estimates of the relationship between male size and mating success and predation risk in Pacific Treefrogs (*Pseudacris regilla*) collected over multiple years and populations. This data set allowed me to test whether male size influenced predation risk and mating success in nature and whether these two types of selection pressure were in conflict with one another.

MATERIALS AND METHODS

Study system.—The Pacific Treefrog, *P. regilla*, is a geographically widespread anuran that has a prolonged breeding season during which males gather at bodies of water, call for mates, and engage in aggressive interactions with other males (Whitney and Krebs, 1975; Whitney, 1980; Perrill, 1984). Females exhibit a preference for those males that initiate bouts of calling (Whitney and Krebs, 1975), but a previous study with a limited sample size found no evidence that male size influenced male mating success (Perrill, 1984). While chorusing, male *P. regilla* are exposed to a variety of predators, such as the giant water bug (*Lethocerus americanus*, Fig. 1A). Giant water bugs are well-known predators of adult anurans around the world (Hinshaw and Sullivan, 1990; Hirai and Hidaka, 2002; Toledo, 2005).

Data collection and analysis.—Data on natural selection was collected as part of a five-year study of *P. regilla* population dynamics on the University of California Natural Reserve System's Quail Ridge Reserve (Napa County, California). As part of this study, I regularly visited ponds on the reserve and collected adult frogs that were later measured, individually marked, and released. The snout–vent length (SVL) of the frogs was measured with dial calipers (to the nearest 0.5 mm), and the mass of the frogs was measured on a digital balance (to the nearest 0.01g). With this data, I calculated the body condition of the males as each individual's cube-root transformed mass divided by its SVL. I haphazardly selected some of these nights to estimate the relationship between male size and body condition and mating success at three ponds (Decker Canyon Pond, Far Pond, and Fordyce Pond). To accomplish this, I

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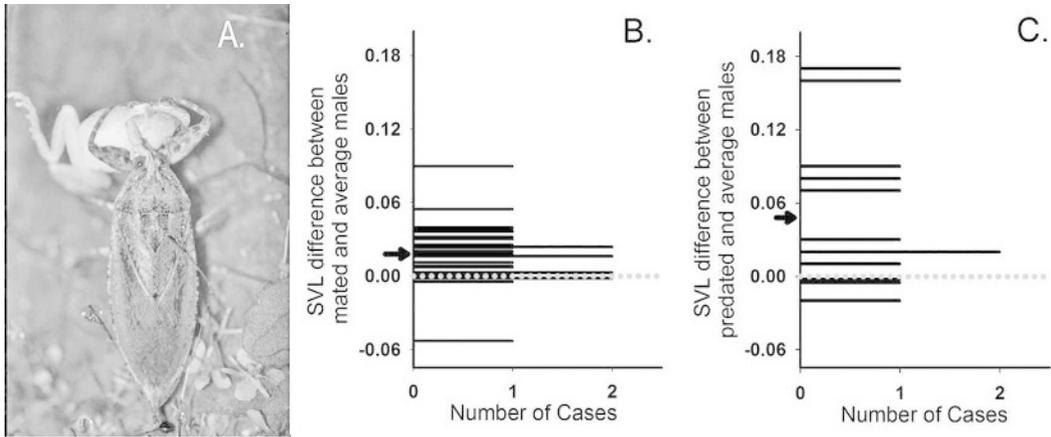
Fig. 1. Conflicting selection on the size of breeding male frogs. (A) Photograph of adult giant water bug (Hemiptera: *Lethocerus americanus*) eating a male Pacific Treefrog (*Pseudacris regilla*). Histograms demonstrating difference between (B) males collected in amplexus and the average male on that night and (C) average male and male captured by giant water bug on that night. The dashed gray line demarcates zero, and the arrows indicate the mean for each measurement.

collected pairs of frogs in amplexus and later measured them in the laboratory. This allowed me to compare the size of males captured in amplexus to the size of unmated males on each night. This type of comparison is a standard method to estimate sexual selection on body size in breeding populations of anurans (e.g., Gerhardt et al., 1987; Friedl and Klump, 2005). I estimated the effect of male SVL on mating success as the difference in SVL of males collected in amplexus minus the mean SVL of all males collected that night at that pond, on nights from 2003 through 2005. Each measure was standardized by the mean male SVL of that night. I made similar estimates for male mass and body conditions. For this study, I only included pond-nights in which three or more amplexing pairs were collected. I tested whether the means of the estimates of size-bias and condition-bias in mating success risk were significantly different from zero with a Wilcoxon Signed-Rank Test.

I collected similar data from 12 predation events by *L. americanus* at two ponds (Far Pond and Fordyce Pond) from 2002–2005. Giant water bugs capture prey by grasping them with large raptorial forelimbs, then pierce their prey with mandibles and inject digestive fluid, and finally consume the frog by sucking the digested innards of the frog. I collected frogs shortly after they were captured by a giant water bug, and before the injection of digestive fluid damaged the frogs, allowing me to accurately measure the captured frogs. These predation events were observed within 2 m of shore while I walked around the pond. Additional predation events were observed during the course of the study, but predation events were only included if they were observed upon my initial arrival at the pond (i.e., before my frog collecting activity could inadvertently cause a predation event) and if the predation event was relatively recent (i.e., before the injection of digestive fluids caused the captured frog to be digested and, thus, not reliably measured).

I calculated the effect of size on predation risk as the difference in the SVL of the male killed by the giant water bug minus the mean SVL of all of the males collected that night. Each measure was standardized by the mean male SVL of that night. I made similar estimates for male mass and body condition (cube-root transformed mass divided by SVL). However, I unfortunately did not record the mass of *Lethocerus*-killed frogs in 2004 and, hence, have only seven estimates of the effect of mass and body condition on predation risk. Thus, I have less statistical power to detect a significant effect of mass and body condition on predation risk. I tested whether the means of the estimates of size-bias and condition-bias in predation risk were significantly different from zero with a Wilcoxon Signed-Rank Test. Means are given $\pm$ 1 SE.

RESULTS

Larger male Pacific Treefrogs were more likely to mate than smaller Pacific Treefrogs, but larger males were also more likely to be captured by giant water bugs. Males in amplexus were on average 1.8 $\pm$ 0.5% larger in SVL than the average male (Fig. 1B, mean difference is significantly different from zero, $N = 26$, Wilcoxon Signed-Rank Test $T = 136$, $P < 0.001$). Additionally, males in amplexus were on average 5.2 $\pm$ 1.4% heavier than the average male ($N = 26$, Wilcoxon Signed-Rank Test $T = 136.5$, $P < 0.001$). However, there was no difference in the body condition of males in amplexus and lone males (mean difference = 0.2 $\pm$ 0.3%; $N = 26$, Wilcoxon Signed-Rank Test $T = 9.5$, $P = 0.82$). The results for mating success are not qualitatively changed if the data from Decker Pond, in which *L. americanus* were never observed, is dropped from the analysis.

Males captured by giant water bugs were on average 4.7 $\pm$ 1.8% larger in SVL than the average male (Fig. 1C, mean difference is significantly different from zero, $N = 12$, Wilcoxon Signed-Rank Test $T = 27$, $P < 0.03$). Similarly, males captured by giant

water bugs were on average 8.4 ± 7.4% heavier than the average male, although this difference was not statistically significant ($N = 7$, Wilcoxon Signed-Rank Test $T = 7.0$, $P = 0.11$). There was no substantial difference in body condition between males killed and the average male that night (0.8 ± 1.1%; $N = 7$, Wilcoxon Signed-Rank Test $T = -5.0$, $P = 0.47$).

<div align="center">DISCUSSION</div>

These results demonstrate a clear conflict in the direction of natural selection on male size from mating success and predation risk in two natural populations over multiple breeding seasons. In many organisms, including anurans, some of the variation in body size has an underlying genetic component (e.g., Berven, 1987; Blouin, 1992); thus, it is likely that either of these opposing selection pressures could cause an evolutionary response in *P. regilla* populations. One potential outcome is that, if populations were to differ in predation risk, they may evolutionarily diverge in adult body size (e.g., Basolo and Wagner, 2004). Additionally, there can be evolutionary consequences for traits correlated with body size. This includes sexually selected traits such as call characteristics (e.g., call frequency is correlated with male size in *P. regilla*, Snyder and Jameson, 1965), as well as traits in other life-history stages such as larval survival and growth rates (Mitchell, 1990; Semlitsch, 1994; Welch et al., 1998). Thus, the evolutionary implications for different selection pressures on adult body size are broad.

Giant water bugs (*Lethocerus* sp.) are a common predator on anurans around the world; some species are even dependent upon breeding anurans as their primary food source during parts of the year (Hinshaw and Sullivan, 1990; Hirai and Hidaka, 2002; Toldeo, 2005). Thus, the natural selection on male size observed here is likely to occur in many systems. I was not able to quantify the strength of natural selection (e.g., as a selection differential, Falconer and Mackay, 1996), because I do not have data on the total number of breeding frogs captured annually by the predators and, thus, cannot estimate how the mean size of the adult population was changed by mortality . However, I frequently observed frogs that had been killed by *L. americanus* (easily recognized from the unique manner in which *L. americanus* kill prey), suggesting that there may be substantial rates of predation and, thus, the potential for strong natural selection, on male frogs. The potentially high magnitude of the strength of selection on male size is also supported by observations of high frequencies (~8 % of the population) of predation by *L. americanus* on *Hyla versicolor* (Hinshaw and Sullivan, 1990) and because adult frogs are the primary prey item of *L. deyrollei* during the spring (Hirai and Hidaka, 2002).

The observed bias in mating success favoring larger male *P. regilla* stands in contrast to a recent summary of studies of treefrogs (family Hylidae), in which it was argued that there was rarely selection favoring large males in many lek-breeding treefrogs (Friedl and Klump, 2005). Why, then, is there a mating advantage for larger males in my study of *P. regilla* but not in the studies of many other hylids? One hypothesis is that size is a good correlate of a male's genetic quality in the populations of *P. regilla* that I studied but not in

the populations reviewed by Friedl and Klump. Alternatively, the populations of anurans in the studies reviewed by Friedl and Klump may have been close to their phenotypic optima under stabilizing selection for sexual selection on size; thus, studies of a size-effect on mating success would have failed to detect any selection. To evaluate these different hypotheses, experimental tests for the effect of interpopulation variation in ecological characteristics on sexual selection on male frogs would be valuable. The existence of intraspecific variation in the presence of sexual selection on male size is supported by comparison between Friedl and Klump's review and both my work and Gatz's (1981) description of a large-male advantage in mating success in both *H. versicolor* and *Psuedacris crucifer*.

A relationship between body condition and mating success has rarely been tested for in anurans, but it has been shown to occur in one other hylid, *Litoria xanthomera* (Morrison et al., 2001). In contrast, I found no evidence for an effect of *P. regilla* body condition on either male mating success or predation risk. Body condition in other species of anurans has been associated with mating behaviors, such as the frequency of chorus attendance and whether an individual acts as a calling male or a satellite male (Cherry, 1993; Eggert and Guyétant, 2003; Leary et al., 2004), but its relationship to mating behaviors or call characteristics has not been examined in *P. regilla*.

Although my results identify the direction of selection from mating success and predation, they do not identify the underlying mechanism responsible for the pattern. The positive relationship between male size and both mating success and predation risk may arise directly through similar attractions of both females and predators for the same trait (i.e., body size in *P. regilla*), a mechanism which has been documented in other taxa, such as the Túngara Frog, and guppies (Ryan et al., 1982; Godin and McDonogh, 2003; Bernal et al., in press). Alternatively, the conflict between selection from predation risk and mating success may arise indirectly through a correlation between male size and other traits, such as calling activity. For instance, if larger males call more frequently, they will be more likely to mate with females (Whitney and Krebs, 1975), but more frequent calls may attract *L. americanus*. Support for the presence of correlational selection on male size comes from the observation that in seven years of visits to this population, I never observed a lone female frog captured by a giant water bug, despite the fact that female *P. regilla* are on average larger than male *P. regilla*. Regardless of the mechanism, the natural selection on male traits found in this study demonstrates an important but overlooked factor that can potentially counteract and thus prevent evolutionary responses to sexual selection in anurans.

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