
REPRODUCTIVE INTERFERENCE BY AN INVASIVE SPECIES: AN EVOLUTIONARY TRAP?

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Abstract.—We detected 43 cases of interspecific amplexus over three years between a threatened amphibian, *Rana draytonii*, and its invasive competitor and predator, *Rana catesbeiana*. In each case, smaller adult male *R. draytonii* clasped larger juvenile *R. catesbeiana*. In the same ponds over the same period, we observed only five *R. draytonii* in conspecific amplexus. *Rana draytonii* are the largest native frogs in western North America, females are larger than males on average, and female anuran size positively correlates with fecundity. The introduction of *R. catesbeiana* throughout the range of *R. draytonii* may disrupt the adaptive mechanisms of sexual selection, thus creating an evolutionary trap.

Key Words.—amphibian; conservation; evolutionary trap; interspecific amplexus; *Rana catesbeiana*; *Rana draytonii*

INTRODUCTION

Invasive species have widespread impacts on ecological communities throughout the world (Groombridge et al. 1992; Wilcove et al. 1998), but the mechanism behind these impacts and consequences for native species are not well characterized (Gurevitch and Padilla 2004). Invaders are often competitors or predators of native species, but they also may exact a broad range of powerful indirect and less-recognized direct impacts. For example, invaders may expand the range of certain diseases (Tompkins and Gleeson 2006) and otherwise expose native species to novel challenges, potentially altering evolutionary pathways (Mooney and Cleland 2001). Invaders may modify the habitat in ways that promote the successful establishment of other invasive species (Simberloff and Von Holle 1999) or make habitat unsuitable for native species (Levi and Francour 2004). The most complex effects occur when an invasive species simultaneously exacts multiple interacting direct and indirect effects on a native species.

Amphibians are experiencing worldwide declines and at least one-third of all amphibian species face extinction (Stuart et al. 2004; McCallum 2007; Wake 2008). Proposed reasons for the declines include the effects of invasive species (Kats and Ferrer 2003). Specifically, invasive predators are one of many causes of the extirpation of amphibian populations (Bradford 1991, 1994; Matthews et al. 2001). The most common aquatic invasive predators implicated in these declines include non-native fish, crayfish and American Bullfrogs (*Rana catesbeiana*; Kats and Ferrer 2003).

Rana catesbeiana are native to regions of North America east of the Rocky Mountains, but were introduced to western North America in the late 1800s (Hayes and Jennings 1986). The impact of *R. catesbeiana* is manifested upon their extirpation: when these large frogs go locally extinct within a pond system, the number of congeners increases four-fold (Hecnar and M'Closkey 1997). *Rana catesbeiana* presence affects the numbers of successful larvae produced by native frogs and is associated with negative impacts on measures of native amphibian body condition (Kiesecker and Blaustein 1997; Kupferberg 1997; Kiesecker et al. 2001).

A variety of sub-lethal effects may contribute to the population-level impact of an invader on a native species, including any impact that an invader has on reproductive success. For example, the presence of juvenile *R. catesbeiana* in pond systems in Oregon appears to cause reproductive interference for some native species when native adults attempt to mate with juvenile *R. catesbeiana* rather than conspecifics (Pearl et al. 2005). Pearl and colleagues hypothesized that for populations dealing with other stressors, this wasted reproductive effort may significantly impact population trends. Data were lacking to indicate that interspecific amplexus was widespread, or might occur more frequently than conspecific amplexus.

Here, we present novel observations of interspecific amplexus between males of a declining native species, *Rana draytonii* (California Red-legged Frogs), and juvenile *R. catesbeiana* (Fig. 1). The frequency of interspecific amplexus was much higher (in terms of intra/inter specific amplexus observed) than reported for



FIGURE 1. Adult male *Rana draytonii* in amplexus with a juvenile *Rana catesbeiana* in BYHP 1 Pond on the Elkhorn Ranch, Central California. (Photographed by Antonia D'Amore)

other western North American species. We suggest some drivers and potential consequences for this behavior, and detail the need for manipulative experiments of this phenomenon in a non-threatened species.

MATERIALS AND METHODS

We conducted three years of nighttime surveys (approximately 350 field-hours per year) in 25 ponds in central California as a part of a larger study on metapopulation dynamics of *Rana draytonii*. The behavioral observations presented here were not part of a formally designed research project on the subject, but rather evolved as the striking numbers of interspecific pairings became evident.

The total estimated *R. draytonii* population in the study area was approximately 1,000 animals throughout this study while the estimated *R. catesbeiana* population declined: 1,000 individuals in 2005, 500 in 2006, and 300 in 2007, with approximately 2/3 of those individuals being juveniles. This decline in *R. catesbeiana* is attributed to a removal effort performed at the study sites.

During the 2005 *R. draytonii* breeding season (January–March at our field site), we monitored all amplexus events visually to avoid disturbing the pairs, recording the relative size of each frog, whether they were the same species, and which partner was larger. In the 2006 and 2007 breeding seasons, we captured both individuals when possible and measured snout-vent length (SVL) of each *R. draytonii* individual of the *R. draytonii*-*R. catesbeiana* pair. We then visually estimated if the clasped *R. catesbeiana* was larger or smaller (in terms of SVL) than the *R. draytonii*. In 2005–2007, when a *R. draytonii* was captured during amplexus or otherwise, we marked the animal using a PIT tag, following standard procedures (Brown 1997). The total marked individuals ($n = 600$) allowed us to

gain accurate estimates of the sex ratio in these ponds, as well as relative male and female body size.

We tagged four males that we found engaged in interspecific amplexus over this period. Permitting requirements and difficulty in capturing amplexed pairs prevented us from using larger numbers in 2005. Regardless, we were able to analyze these data to determine whether the Bullfrog-clasping behavior was unique to a few individuals, or if many males were engaging in the behavior.

For one pond with sufficiently detailed data, we calculated the expected and observed levels of interspecific and conspecific amplexus. We defined expected levels by the numbers of pairs we would have seen in each category were mate choice entirely random (i.e., males simply clasp any frog that passes them). We based the calculations on the average number of *R. draytonii* and juvenile *R. catesbeiana* seen in the pond each year during the breeding season of *R. draytonii* (Table 1). We calculated expected interspecific amplexus by estimating the number of female *R. draytonii* in the pond (average number of *R. draytonii* seen, divided by two because the male/female sex ratio calculated by mark-recapture data was 1:1). We then added this number to the number of juvenile *R. catesbeiana* seen to obtain the total number of potential amplexus partners. We used the percentage of juvenile *R. catesbeiana* out of the total potential amplexus partners as the expected percentage of interspecific amplexus, if 'mate' choice were random. We used the percentage of female *R. draytonii* out of the total potential amplexus partners as the expected percentage of interspecific amplexus if 'mate' choice were random. We then used binomial distribution hypothesis testing to determine if interspecific amplexus occurred significantly more frequently than expected by chance alone.

RESULTS

We documented 43 different instances of interspecific amplexus between *R. draytonii* males and juvenile *R. catesbeiana* of indeterminate sex. All observations of interspecific amplexus occurred only in the three ponds that contained substantial numbers of juvenile *R. catesbeiana* during the *R. draytonii* breeding season of 2005–2007. The breeding season for *R. catesbeiana* at this site occurred between May and July and we observed no amplexus of *R. catesbeiana* males with *R. draytonii*. We found *R. draytonii* in the stomach contents of four adult *R. catesbeiana* we opportunistically caught and dissected.

The sex ratio of all marked frogs was 284 females: 275 males, close to a 1:1 sex ratio, which was important for calculating rates of expected conspecific amplexus. While we observed 43 interspecific pairs, we saw *R./*

TABLE 1. Averages shown for BHYP 1 Pond during the breeding season of *R. draytonii* in each year with an average of four visits in each year. Interspecific amplexus occurred more than expected by chance alone, and this is the only pond where conspecific amplexus occurred. (Binomial probability of the observed amplexus: 2005, $P = 0.0009$, 2006 $P = 0.109$, 2007, $P = 0.080$).

Year	Juvenile <i>Rana catesbeiana</i>	Adult <i>Rana draytonii</i>	Observed interspecific amplexus	Observed conspecific amplexus	Expected % interspecific amplexus	Expected % conspecific amplexus
2005	1.5	3	10 (100%)	0	50%	50%
2006	3	5.75	6 (75%)	2 (25%)	51%	49%
2007	7.5	10.25	8 (72%)	3 (28%)	59%	41%

draytonii conspecific amplexus five times during the course of this study: twice in 2006 and three times in 2007. Interspecific amplexus was more than eight times more common than *R. draytonii* conspecific amplexus, even though *R. draytonii* outnumber the invasive juvenile *R. catesbeiana*. We only noted *R. catesbeiana* conspecific amplexus in one pond. We calculated expected and observed rates of *R. catesbeiana* conspecific and interspecific amplexus for this pond in all three years (Table 1). Interspecific amplexus was observed more frequently than expected, with significant results in 2005 ($P = 0.0009$), no significance in 2006 ($P = 0.109$), and marginal significance in 2007 ($P = 0.080$).

In all cases, the clasping *R. draytonii* males (mean = 7.5 cm) were smaller than *R. draytonii* females (mean = 8.1 cm) or clasped *R. catesbeiana*. The male *R. draytonii* we measured ranged in size from SVL = 6.8–8.2 cm SVL. Female California Red-legged Frogs were much larger than males on average. Consistent with this pattern, all juvenile *R. catesbeiana* clasped were larger than the clasping male *R. draytonii*.

We caught two of the four marked *R. draytonii* males in amplexus with *R. catesbeiana* multiple times (two and three times respectively) during the breeding season of 2007. Interspecific amplexus is not likely due to a few errant individuals, as we observed the behavior in three

different ponds, over a three-year period, and up to six different interspecific pairs mating during any single visit to a pond (Fig. 2).

DISCUSSION

Frequency of interspecific amplexus.—While this behavior has been reported for ranids within the introduced range of the Bullfrog (Pearl et al. 2005), little work has been performed to estimate the frequency of this behavior or its potential impact on native congeners. The frequency with which we observed interspecific versus intraspecific amplexus was striking, especially as we estimated that *R. draytonii* were as, or more abundant than juvenile *R. catesbeiana* at these ponds. Even in the pond where conspecific amplexus occasionally occurred, the observed elevated frequency of interspecific amplexus was determined to be non-random in two of three years. Three non-exclusive explanations for our observations are possible: (1) Strong preference by male *R. draytonii* for juvenile *R. catesbeiana*; (2) longer duration of interspecific amplexus or; (3) because juvenile *R. catesbeiana* do not deposit eggs during these interspecific amplexus events, they do not swim towards emergent vegetation that would act as a suitable oviposition site and a cue for the clasping *R. draytonii* to release. We speculate that these last two possibilities would likely lead to more observations of interspecific pairs.

With the lack of an egg-laying response or proper release calls, interspecific amplexus may be of longer duration. Male ranids attempt amplexus with other males and non-receptive females, and each species has a specific release call presumably to avoid wasted reproductive effort (Aronson 1944; Blair 1968). Both individuals benefit from the conservation of energy, time, and reproductive effort. Research on the endocrine system of *R. catesbeiana* indicates that calling behaviors, including release calls, have hormonal control mechanisms (Boyd 1992). Juveniles may lack adult hormones specific to breeding vocalizations and thus may not be able to emit release calls (Boyd 1997). Thus, the absence of cues by juvenile *R. catesbeiana* (egg laying or release calls) that would act to terminate the clasping behavior of male *R. draytonii* may lead to longer pairings and explain the elevated frequency of interspecific amplexus observations. While these

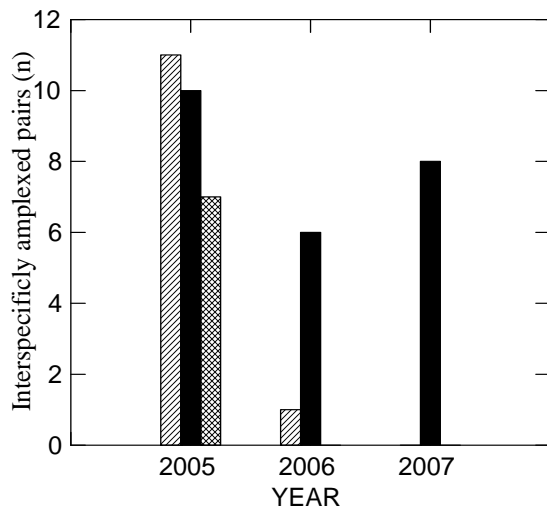


FIGURE 2. The number of interspecific pairs of *R. draytonii* and *R. catesbeiana* seen in three ponds (striped, solid, cross-hatched) in central California in 2005, 2006, and 2007.

explanations suggest that interspecific amplexus may well be of longer average duration, we also suspect that male mate preferences will make the initiation of interspecific pairings common, as we address in the next section.

Male preference as an evolutionary trap.—Male *R. draytonii* mate choice of juvenile *R. catesbeiana* is likely highly maladaptive. At a minimum, pursuing amplexus with a non-receptive individual wastes time and energy that could be better spent foraging or attracting conspecifics. Amplexus, even with conspecifics, may increase predation rates (Bernard 2007). If interspecific amplexus is of longer duration or is more conspicuous than intraspecific amplexus, then involved individuals may suffer particularly high predation levels. Furthermore, any *R. draytonii* that approach larger *R. catesbeiana* to initiate amplexus risk predation by *R. catesbeiana*, a documented predator of *R. draytonii* (Pers. Obs.). Why then, does this behavior occur?

We hypothesize that male preference for large body size of mates drives this behavior and that males do not employ other more specific criteria when selecting mates. *Rana draytonii* were historically the largest frogs in western North America (Wright and Wright 1949). The correlation between female anuran body size and female fecundity suggests that male preference for large females is generally adaptive (Castellano 2004; Gilbert et al. 1994; Berven 1981). Males that pursue amplexus with larger females leave more offspring and may pass on this tendency to mate with the largest possible female, if this preference is heritable. Adult female *R. draytonii* are typically larger by an average of 19% than males (Hayes and Miyamoto 1984). Hence, prior to the arrival of *R. catesbeiana*, mating with the largest receptive individual may have been a simple, yet adequate, mate choice strategy. The introduction of the larger *R. catesbeiana* challenged the efficacy of this strategy.

Other anurans show maladaptive male preference for larger heterospecifics (Engeler and Reyer 2001), and in systems where male ranid preference for larger females does not exist, this lack of preference may arise specifically to prevent interspecific mating with larger congeners (Hettyey et al. 2005). We suggest that male preference for large body size of mates by *R. draytonii* is an “evolutionary trap.” “Evolutionary trap” is a situation where sudden environmental change, through anthropogenic impact or species addition, renders a formerly adaptive behavior maladaptive (Schlaepfer et al. 2002). Schlaepfer and colleagues categorized certain impacts of invasive species on native species as examples of evolutionary traps (Schlaepfer et al. 2005).

Potential for population-level consequences.—Discovery of a seemingly maladaptive behavior does not

necessitate significant population-level consequences, but we hypothesize that in ponds with small *R. draytonii* populations and large numbers of juvenile *R. catesbeiana*, a reduction in population growth rates may result from this activity. Males engaged in amplexus do not call to attract further mates, may have higher risk of predation, and therefore prolonged amplexus may lead to reduced breeding choruses and subsequent reduction in female recruitment to a freshwater site. This affects both major demographic processes of survival and reproduction.

Another possibility is that presence of juvenile *R. catesbeiana* alters the social environment and criteria for *R. draytonii* mate selection. In the snake *Thamnophis sirtalis parietalis*, female size corresponds with fecundity and male mate choice is influenced by the size of the females that a male encounters (Shine et al. 2006). Males exposed to larger females are less likely to mate with the smaller females they encounter. This raises the possibility that a pond system with abundant juvenile *R. catesbeiana* may make smaller *R. draytonii* adult females unattractive to male conspecifics, leaving females with a potential shortage of mates. We would likely have seen interspecific amplexus at higher frequencies, particularly in 2006 and 2007, if it were not for Bullfrog control efforts, indicating that this behavior may occur more frequently in sites with a greater proportion of *R. catesbeiana*.

In conclusion, we find that potential for reproductive interference may exist between invasive *R. catesbeiana* and native ranids throughout their introduced range. We specifically hypothesize that these interspecific interactions may be particularly problematic for species that did not coevolve with larger congeners. As larger female ranids have higher fecundity, a larger invasive congener may then be attractive, and potentially more so than conspecific females. In these situations, the additional reproductive barriers may not be in place to prevent males from choosing mates based on size alone.

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