

Influence of density and predators on metamorphic size in *Rhinella schneideri* tadpoles raised in mesocosm conditions

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Abstract: Amphibians exhibit extreme plasticity in the timing of metamorphosis, and several species respond to water availability, accelerating metamorphosis when their ponds dry. We analyzed the plasticity of the developmental response to water volume in *Rhinella schneideri* tadpoles. We raised tadpoles in mesocosm. Covariation between body size at metamorphosis and timing of development was positive. Nevertheless, the first approximately 53% of the metamorphoses finishing the cycle required between 34 and 56 days, and the covariation between body size at metamorphosis and timing of development was negative. For these tadpoles, the larval density and the presence of predators did not significantly affect their mass to metamorphosis. Nevertheless, predators affected time to metamorphosis. For the remainder of the tadpoles that reached metamorphosis at > 56 days, the relationship between body size at metamorphosis and timing of development was positive. For these tadpoles, larval density was important for mass at metamorphosis and presence of predators was also important for time to metamorphosis. Two dominant features were observed: (i) approximately 53% of metamorphs had morphological features similar to individuals developing in desiccating ponds, and (ii) the other individuals had morphological characteristics comparable to metamorphs developing in an unchanging environment.

Key words: tadpoles; phenotypic plasticity; density; predators; trade-off; r-strategist

Introduction

The influence of biotic and abiotic factors on tadpole life cycles differs depending on whether the environment is predictable or unpredictable. A temporary pond habitat has two important characteristics for tadpoles: first, explosive primary productivity stimulates rapid growth (Wassersug 1975); second, fish and other large predators (insects) are typically absent (Wilbur 1980).

Many studies have dealt with the effect of density on the intensity of competition at the specific or community level, and on predation (Kehr 1989; Relyea 2004; Bolnick & Preisser 2005; Teplitsky & Laurila 2007). High density (crowding) has been shown to negatively affect survivorship, growth rate, and the length of larval period of amphibian larvae (Kehr 1994; Relyea 2002).

Nevertheless, biotic factors can be minimized by the influence of abiotic effects (Warner et al. 1991). Pond desiccation is one of the most important abiotic factors regulating larval amphibian populations (Kehr 1997; Maciel & Juncá 2009; Richter-Boix et al. 2011). Natural selection may lead to several strategies to deal with habitat unpredictability. Strong selection for fast development would reduce the risk of death by desiccation and reduce phenotype variation in the environment. Natural selection can alter the phenotypic range of populations in the current generation by serving as a

filter that favors the survival of particular phenotypes (Kingsolver 1995; Van Buskirk & Relyea 1998).

Tadpoles can respond to predators in different ways. They may minimize the risk of predation by changing their habitat (Werner & Peacor 2003), their life history (Benard 2004; Fusco & Minelli 2010), behavior and growth (Relyea & Werner 1999; Babbitt 2001; Peacor 2002; Relyea 2007), or by developing chemical or morphological defenses. Predator-induced changes in body morphology have, however, been described in tadpoles (Relyea 2003, 2004; Jara & Perotti 2009; Gómez & Kehr 2011a, b, 2012). Another possibility is that the phenotype may be modified by developmental responses to environmental variations. Concurrently, environmental conditions can have substantial impacts on phenotypes by inducing changes in behavior, morphology, physiology, or life history of most organisms, and these changes are often in adaptive directions (West-Eberhard 1989; Tollrian & Harvell 1999). Although tadpoles may display phenotypic plasticity, several mechanisms may reduce variability in metamorphic size producing a dilution of this trade-off (Tejedo & Reques 1994).

Covariation between the length of the larval period and size at metamorphosis may be influenced both by environmental factors (Travis 1984; Griffiths 1991; Pfennig et al. 1991). A positive correlation between size at metamorphosis and length of the larval period sug-

gests a trade-off between growing large and metamorphosing quickly to avoid pond desiccation or predation. Nevertheless, negative correlations between size at metamorphosis and length of the larval cycle, implying no trade-off have been described occasionally in experimental and natural ponds (Pfenning et al. 1991; Tejado & Reques 1992, 1994).

This study was carried out with *Rhinella schneideri* (Werner, 1894) tadpoles. This common species in northeastern Argentina can reproduce continuously from August until March, depending on rainfall. This species breeds in either temporary or semipermanent ponds. The main goals of this work were: (i) to describe the effects of density; (ii) to observe the effect of predator presence, and (iii) to analyze covariation between time at metamorphosis and metamorph size in each treatment. Results were obtained by analyzing: a) mass at metamorphosis, b) survival, c) time to metamorphosis, and d) growth rate under various conditions.

Material and methods

Experimental design

The experiment was performed in 12 artificial ponds located in the field of the Centro de Ecología Aplicada del Litoral (CECOAL-CONICET) 12 km from Corrientes City, Argentina (27°30' S, 58°45' W). The artificial ponds (0.90 m in diameter and 0.80 m deep) were filled with 400 L of water one month before beginning the experiment. To each tank we added 0.5 L every 10 d of a standard inoculum of plankton and periphyton filtered from two natural ponds. In addition, every 10 days we added 2 g (dry weight) of lettuce previously boiled for 10 min and 3 g of fish food (commercial brand *Shulet*). During the experience the dissolved oxygen ranged between 3.3–3.8 mg L⁻¹, pH between 6.6–6.8, and the air mean temperature (°C) ranged between 22–26 (minimum 18°C and maximum 32°C).

We used a randomized block design with a factorial combination of two densities of tadpoles (150 and 300) and two predator presence (presence – absence). The four treatments were arranged in three randomly replicated spatial blocks with a total of 12 tanks. The predators used were *Belostoma micantulum* Stal, 1858 (Hemiptera: Belostomatidae). We used three individuals of *B. micantulum* in each tank.

The *R. schneideri* eggs were obtained from three different clutches within a large ephemeral pond located near Corrientes city. The tadpoles were mixed and added to the tanks when they were five days old (Stages 25–26, Gosner 1960). The experiment began on 18th September 1999.

We measured the effect of larval density and the presence of insect predators on larval survival, mass at metamorphosis, time to metamorphosis, and growth rate on *R. schneideri*.

Statistical analyses

The statistical test used was a Multivariate Analysis of Variance of two factors (MANOVA). This test allowed us to separate the effect of predators and density (both independent factors) on the following dependent variables: (1) mass at metamorphosis measured at tail absorption (stage 46; Gosner 1960) to the nearest 0.1 mg; (2) time to metamorphosis, or the interval from the start of the experiment until tail

absorption (Gosner stage 46), and (3) survival to metamorphosis, measured as the percent of metamorphosis in each tank. In those cases where significant MANOVA results were observed, this analysis was supplemented with a univariate variance analysis (ANOVA) to detect which factor exercised the most influence.

After the first metamorphs (Gosner 42–43) were discovered, tanks were checked daily and all metamorphs found were collected and maintained in the laboratory in plastic containers (15 cm diameter) arranged in an inclined plane of 15 degree and 2 cm of water until tail absorption was completed (Gosner stage 46).

Mass and time to metamorphosis were log_e transformed due to heterogeneity of variances among treatments. Survival data were angularly transformed by the arcsine square root transformation. With these transformations, the distribution of the data approached normality. A previous analysis showed that the main effect of blocks did not exhibit a significant effect on the response variables. Therefore, blocks were pooled across treatments. All statistical analyses were performed using the software SYSTAT 7.0 for Windows. All values are expressed as mean ± 1 SD.

The growth rates of tadpoles (mg day⁻¹) under different experimental conditions were compared using the “General Linear Model” (GLM) which allowed us to compare the homogeneity (“parallelism”) in the slope (b) of the regression between mass and time to metamorphosis. This was calculated by an ANCOVA using the variable “time” as the covariable of the pattern. This method permits comparison of the different values of the constant b (regression coefficient) of the regression equation.

In order to analyze the relationship between size at metamorphosis and time to metamorphosis, at different moments of development time, that span of time was divided into three periods of approximately 22 days (first period: between 34 and 56 days; second period: between 57 and 78 days, and third period: between 79 and 102 days). We considered these 68 first days (time between 34 and 102 days) as adequate for analyzing the relationship between the dependent variables (mass at metamorphosis and development time) and the two factors for each of the three periods predator presence and density (150 vs. 300 tadpoles). Survival for each period was not considered because we took the data from throughout the time of development and not for a particular time.

Results

A MANOVA test suggested that the combined responses of mass and time to metamorphosis, and survival to metamorphosis were significantly affected by presence of predators and larval density (Wilks' Lambda < 0.001) (Table 1). The MANOVA test also revealed that the interaction between predator presence and larval density affected significantly the three dependent variables. The following univariate results showed the variables responsible for these complete treatment effects.

Predators effects

Mass to metamorphosis differed significantly between treatments (Table 1). The mean mass of tadpoles in tanks without predators ($\bar{x} = 120.48 \pm 30.66$ mg, $n = 293$) was lower than tadpoles that developed in tanks

Table 1. Results of the analysis of MANOVA and ANOVA carried out with *Rhinella schneideri* tadpoles. Probabilities for the univariate test (ANOVA) were compared with the level of adjusted significance for the three dependent variables according to that proposed by Bonferroni ($\alpha = 0.016$).

Variable PREDATOR MANOVA (Wilk's Lambda = 0.700; df = 3, 342; $P < 0.001$)			
	df	F	P
Univariate F test			
Mass at metamorphosis	1, 344	10.499	0.001
Time to metamorphosis	1, 344	21.741	< 0.001
Survival to metamorphosis	1, 344	136.411	< 0.001
Variable DENSITY MANOVA (Wilk's Lambda = 0.656; df = 3, 342; $P < 0.001$)			
Univariate F test			
Mass at metamorphosis	1, 344	0.035	0.852
Time to metamorphosis	1, 344	27.039	< 0.001
Survival to metamorphosis	1, 344	167.754	< 0.001
Variable PREDATOR \times DENSITY MANOVA (Wilk's Lambda = 0.742; df = 3, 342; $P < 0.001$)			
Univariate F test			
Mass at metamorphosis	1, 344	8.448	0.004
Time to metamorphosis	1, 344	13.186	< 0.001
Survival to metamorphosis	1, 344	93.729	< 0.001

with predators ($\bar{x} = 134.00 \pm 39.88$ mg, $n = 55$). Furthermore, in tanks with predators the standard deviation was greater, reflecting major variability in the mass to metamorphosis of these tadpoles.

Time to metamorphosis was significantly different between treatments (Table 1). The mean time of tadpoles in the tanks without predators was shorter ($\bar{x} = 58.47 \pm 23.47$ days, $n = 293$) than the mean time to metamorphosis of tadpoles in tanks with predators ($\bar{x} = 81.30 \pm 19.04$ days, $n = 55$).

Survival to metamorphosis was significantly different between treatments (Table 1). Survival proportion of tadpoles in the tanks without predators was greater ($\bar{x} = 0.598 \pm 0.29$, $n = 293$) than tanks with predators ($\bar{x} = 0.06 \pm 0.03$, $n = 55$).

Larval density effects

Mass at metamorphosis was not significantly different between treatments (Table 1). The mean mass of tadpoles in tanks with low density (0.375 larvae per liter) ($\bar{x} = 121.99 \pm 29.45$ mg, $n = 277$) was slightly lower than tadpoles that developed in tanks with high density (0.75 larvae per liter) ($\bar{x} = 125.07 \pm 42.92$ mg, $n = 71$).

Time to metamorphosis was significantly different between treatments (Table 1). The mean time of tadpoles in tanks with low density was less ($\bar{x} = 55.88 \pm 16.50$ days, $n = 277$) than the mean time of tadpoles in tanks with high density ($\bar{x} = 86.25 \pm 33.24$ days, $n = 71$).

Survival to metamorphosis was significantly different between treatments (Table 1). The proportion of tadpoles that survived in tanks with low density was greater ($\bar{x} = 0.631 \pm 0.264$, $n = 277$) than the proportion of tadpoles in tanks with high density ($\bar{x} = 0.058 \pm 0.018$, $n = 71$).

General relationship between the mass to metamorphosis and development time

Size at metamorphosis for all metamorphs ($n = 348$), was positively correlated with development time in tadpoles raised under the two factors, predators and density.

Significant differences were observed (GLM ANOVA, $F_{1,344} = 5.32$, $P = 0.021$) in the growth rates of tadpoles in the presence of predators ($b = 0.97$) versus without predators ($b = 0.45$). Growth rates of tadpoles developing in different larval densities ($b = 0.68$ and $b = 0.64$; 150 and 300 tadpoles respectively) showed no significant differences (GLM ANOVA, $F_{1,344} = 0.087$, $P = 0.76$).

Relationship between mass and time to metamorphosis (34 and 56 days of development)

Predators effect

The mass of metamorphs that reached the metamorphosis between 34 and 56 days of development was not significantly different between tadpoles reared in tanks with presence and absence of predators (Table 2). The mean mass of tadpoles in tanks without predators ($\bar{x} = 110.38 \pm 21.79$ mg, $n = 181$) was higher than of those developed in tanks with predators ($\bar{x} = 102.00 \pm 21.68$ mg, $n = 5$).

Time to metamorphosis was significantly different between treatments (Table 2). The mean time of tadpoles in tanks without predators was lower ($\bar{x} = 45.19 \pm 5.37$ days, $n = 181$) than the mean time of tadpoles in tanks with predators ($\bar{x} = 50.80 \pm 3.76$ days, $n = 5$).

Density effects

Mass of metamorphs that reached metamorphosis between 34 and 56 days of development was not signifi-

Table 2. Summary of analysis of mass to metamorphosis and time to metamorphosis of *Rhinella schneideri* tadpoles with the factors predators (presence – absence) and densities (150 vs. 300 tadpoles) using ANOVA tests (dependent variables were previously transformed to natural logarithms). The three time periods correspond to time tadpoles spent until metamorphosis. For these analyses the first metamorphs needed 34 days and the latter 102 days. During this time 316 tadpoles reached to metamorphosis.

	Between 34 and 56 days ($n = 186$)			Between 57 and 78 days ($n = 93$)			Between 79 and 102 days ($n = 37$)		
	Predators	Density	Predators × density	Predators	Density	Predators × density	Predators	Density	Predators × density
Mass to meta-morphosis	$F_{(1,182)} < 0.001$; $P = 0.99$	$F_{(1,182)} = 0.015$; $P = 0.90$	$F_{(1,182)} = 8.36$; $P < 0.01^*$	$F_{(1,89)} = 1.97$; $P = 0.16$	$F_{(1,89)} = 11.47$; $P < 0.01^*$	$F_{(1,89)} = 5.37$; $P = 0.02^*$	$F_{(1,33)} = 0.32$; $P = 0.57$	$F_{(1,33)} = 0.101$; $P = 0.75$	$F_{(1,33)} = 5.397$; $P = 0.02^*$
Time to meta-morphosis	$F_{(1,182)} = 4.62$; $P = 0.03^*$	$F_{(1,182)} = 0.906$; $P = 0.34$	$F_{(1,182)} = 1.82$; $P = 0.18$	$F_{(1,89)} = 9.19$; $P < 0.01^*$	$F_{(1,89)} = 0.22$; $P = 0.63$	$F_{(1,89)} = 1.07$; $P = 0.30$	$F_{(1,33)} = 1.408$; $P = 0.24$	$F_{(1,33)} = 0.047$; $P = 0.83$	$F_{(1,33)} = 0.017$; $P = 0.89$

Table 3. Summary of *Rhinella schneideri* metamorphs between 34 and 102 days (first and last days) for each treatment. The regression equation corresponds to the relationship between weight on metamorphic climax and time of development. In those cases where adjustment of the data to regression was not significant, the Spearman Rank Correlations test was used. In the column corresponding to mass and time, the data correspond to the arithmetic mean and 1 standard deviation (in parenthesis). * significant.

	Treatment	Regression	Mass (mg)	Time (days)
Between 34 and 56 days	Predators	Spearman Rank; $R_s = 0.81$, $n = 5$, $P > 0.05$	102.00 (21.67)	50.80 (3.76)
	No predators	$y = 137.7 - 0.604 x$; $r = 0.15$; $F_{(1,179)} = 4.07$, $n = 181$, $P = 0.04^*$	110.38 (21.79)	45.19 (5.37)
	Density 150	$y = 141.20 - 0.639 x$; $r = 0.17$; $F_{(1,164)} = 5.02$, $n = 166$, $P = 0.02^*$	112.23 (20.22)	45.32 (5.45)
	Density 300	Spearman Rank; $R_s = -0.096$, $n = 20$, $P > 0.05$	93.00 (26.77)	45.55 (5.13)
Between 57 and 78 days	Predators	Spearman Rank; $R_s = 0.48$, $n = 22$, $P < 0.05^*$	119.54 (29.19)	68.54 (6.19)
	No predators	$y = -23.50 + 2.45 x$; $r = 0.35$; $F_{(1,69)} = 9.58$, $n = 71$, $P = 0.003^*$	132.71 (33.04)	63.64 (4.70)
	Density 150	$y = 38.59 + 1.45 x$; $r = 0.24$; $F_{(1,82)} = 5.12$, $n = 84$, $P = 0.026^*$	132.29 (31.34)	64.54 (5.23)
	Density 300	Spearman Rank; $R_s = 0.64$, $n = 9$, $P < 0.05^*$	104.44 (34.31)	67.44 (7.19)
Between 79 and 102 days	Predators	Spearman Rank; $R_s = 0.11$, $n = 28$, $P > 0.05$	151.07 (42.28)	96.78 (11.82)
	No predators	Spearman Rank; $R_s = -0.067$, $n = 41$, $P > 0.05$	143.90 (39.10)	108.14 (21.92)
	Density 300	$y = -196.28 + 3.93 x$; $r = 0.54$; $F_{(1,16)} = 6.62$, $n = 18$, $P = 0.02^*$	144.76 (39.89)	109.66 (20.42)

cantly different between the tadpoles reared at the two densities (Table 2). The mean mass of tadpoles in tanks with low density ($\bar{x} = 112.23 \pm 20.22$ mg, $n = 166$) was higher than of tadpoles that developed in tanks with predators ($\bar{x} = 93.00 \pm 26.77$ mg, $n = 20$).

Differences in time to metamorphosis were not significant between larval densities (Table 2). The mean time of tadpoles in tanks with low density ($\bar{x} = 45.32 \pm 5.45$ days, $n = 166$) was similar to the mean time of tadpoles in tanks with high density ($\bar{x} = 45.55 \pm 5.13$ days, $n = 20$).

The interaction between predators × density was significant for mass to metamorphosis but not for time of development.

During this period, the relation between size to metamorphosis and time of development, was linear, negative and significant for tadpoles reared in tanks without predators and in tanks with lower density (Fig. 1A; Table 3). Metamorphs that developed in tanks with higher density displayed a negative but not significant relationship. The few metamorphs that developed in tanks with predators ($n = 5$) showed a positive but not significant relationship between both variables.

Relationship between mass and time to metamorphosis (57 and 78 days of development)

Predators effect

Mass of metamorphs that reached metamorphosis between 57 and 78 days of development was not significantly different between tadpoles reared in the two conditions (Table 2). The mean mass of tadpoles in tanks without predators ($\bar{x} = 132.72 \pm 33.04$ mg, $n = 71$) was higher than of tadpoles that developed in tanks with predators ($\bar{x} = 119.54 \pm 29.19$ mg, $n = 22$).

Differences in time to metamorphosis were significant between the two conditions (Table 2). The mean time of tadpoles in tanks without predators ($\bar{x} = 63.65 \pm 4.70$ days, $n = 71$) was lower than the mean time of tadpoles in tanks with predators ($\bar{x} = 68.54 \pm 6.19$ days, $n = 22$).

Density effect

Mass of metamorphs that reached metamorphosis between 57 and 78 days of development was significantly different between tadpoles reared in the two densities (Table 2). The mean mass of tadpoles in tanks with low density ($\bar{x} = 132.30 \pm 31.34$ mg, $n = 84$) was higher than of those tadpoles that developed in tanks with

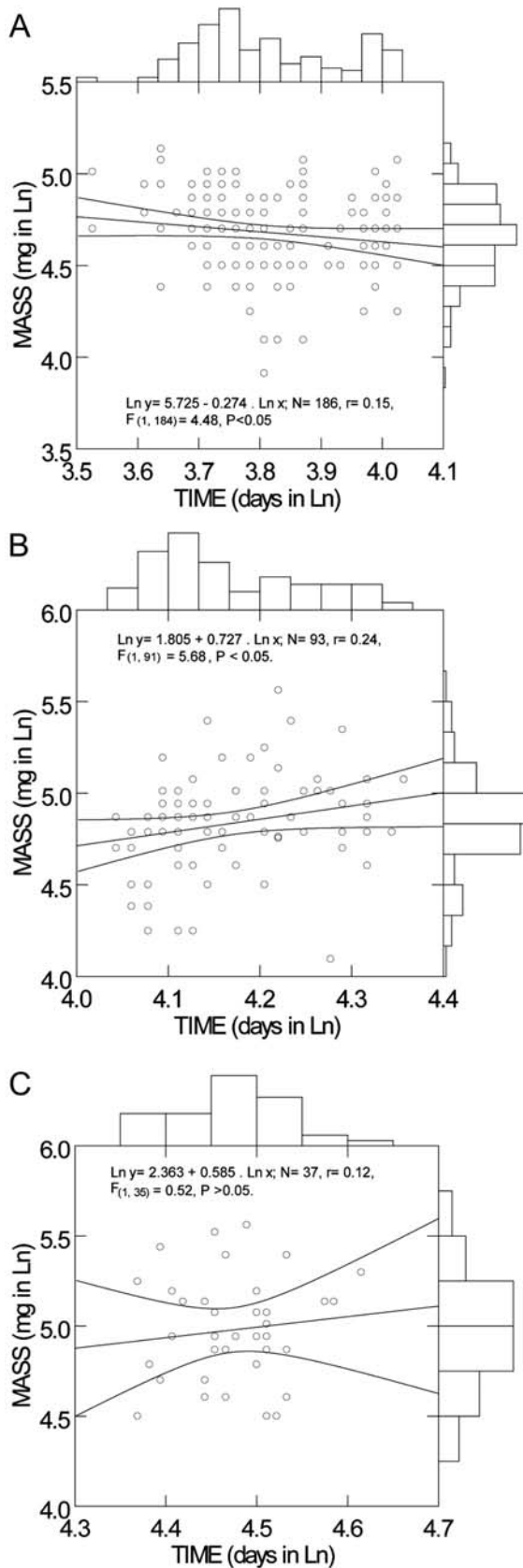


Fig. 1. Linear regression between mass to metamorphosis and time to metamorphosis during three periods of development of *Rhinella schneideri* tadpoles. A: metamorphosed during 34 to 56 days; B: metamorphosed during 57 to 78 days, and C: metamorphosed during 79 to 102 days. The histogram represents frequency of individuals in relation to mass and time to metamorphosis. In total, 316 individuals metamorphosed during 34 to 102 days.

high density ($\bar{x} = 104.44 \pm 34.32$ mg, $n = 9$).

Differences in time to metamorphosis were not significant between the densities (Table 2). The mean time to metamorphosis for tadpoles in tanks with low density ($\bar{x} = 64.52 \pm 5.23$ days, $n = 84$) was slightly lower than the mean time of tadpoles in tanks with high density ($\bar{x} = 67.44 \pm 7.19$ days, $n = 9$).

The interactions between predators \times density was significant for mass at metamorphosis but not significant for time of development.

During this period, the relationship between mass at metamorphosis and time of development was linear, positive and significant for all treatments (Fig. 1B; Table 3).

Relationship between mass and time to metamorphosis (79 and 102 days of development)

Predators effects

Mass of metamorphs that reached metamorphosis between 79 and 102 days of development was not significantly different between tadpoles reared in the two conditions (Table 2). The mean mass of tadpoles in tanks without predators ($\bar{x} = 148.95 \pm 48.75$ mg, $n = 19$) was slightly lower than of tadpoles that developed in tanks with predators ($\bar{x} = 153.90 \pm 42.03$ mg, $n = 18$).

Differences in time to metamorphosis were not significantly different between both conditions (Table 2). The mean time of tadpoles in tanks without predators ($\bar{x} = 86.32 \pm 4.24$ days, $n = 19$) was slightly lower than the mean time of tadpoles in tanks with predators ($\bar{x} = 89.27 \pm 5.99$ days, $n = 18$).

Density effect

Mass of metamorphs that reached metamorphosis between 79 and 102 days of development was not significantly different between tadpoles reared at the two densities (Table 2). The mean mass of tadpoles in tanks with low density ($\bar{x} = 150.52 \pm 47.54$ mg, $n = 19$) was similar to tadpoles that developed in tanks with high density ($\bar{x} = 152.22 \pm 43.59$ mg, $n = 18$).

Differences in time to metamorphosis were not significantly different between densities (Table 2). The mean time of tadpoles in tanks with low density ($\bar{x} = 86.74 \pm 4.49$ days, $n = 19$) was lower than the mean time of tadpoles in tanks with high density ($\bar{x} = 88.83 \pm 6.00$ days, $n = 18$).

The interactions between predators \times density was significant for mass to metamorphosis but not significant for time of development (Table 2).

During this period, there was no linear and significant relation between mass at metamorphosis and time of development (Fig. 1C; Table 3).

Discussion

General ecological implication

In general, the mass at metamorphosis of *Rhinella schneideri* tadpoles was dependent only on predator presence and independent of initial density. The time to metamorphosis and survival were dependent on two

factors: predators and density. The controversial results obtained concerning the lack of density dependence of mass to metamorphosis can be explained by the habitat stability, which influences the time to metamorphosis. More time in a stable habitat (e.g., water level without fluctuations), the mass of tadpoles reaches the maximum at the metamorphic climax. Simultaneously, the density changed as the tadpoles metamorphosed. This situation produced a general positive covariation between mass at metamorphosis and time to metamorphosis.

The responses on performance from a general standpoint are similar to other species in field experiments, where the magnitude of the effect caused by the density of tadpoles on metamorphic size was not large (Loman 2004). Density regulation produces a clear effect on tadpole performance in laboratory studies. In field conditions, the results are less clear. While Berven (1990) and Skelly (1995) have found evidence of density regulation in anuran larvae, other studies, (Loman 2004) have provided little evidence for such regulation. In other words, the real dimension of density regulation in anuran larvae can change according to different development time.

Ecological implications in particular time of reaching metamorphosis. 34 to 56 days of time to metamorphosis
This first period (between 34 and 56 days to reach metamorphosis) had the largest number of metamorphs reaching the climax ($n = 186$; 53%). The predators and density factors were not significant for mass at metamorphosis. Presence of predators in the tanks significantly affected time to metamorphosis and number of metamorphs reaching metamorphosis. For the individuals of *R. schneideri* reaching metamorphosis between 34 and 56 days of development (53% of all juveniles), under stable conditions, the density was less important than the stress produced by predator presence. Predators can have significant lethal and non-lethal effects on prey through chemically mediated interactions. Some amphibian larvae often respond to predators by altering behavior (i.e., decreasing mobility) or morphology (developing longer tails, deeper tail fins and pronounced tail musculature that increases swimming ability) (Petranka & Hayes 1998; Anderson & Petranka 2003; Gómez 2012).

During this period the covariation between mass at metamorphosis and time to metamorphosis was negative, implying no trade-off. This feature has been reported only rarely in experimental and natural ponds and is generally associated with seasonally decreasing food resources (Pfenning et al. 1991). Kehr & Marangoni (1999) reported a negative relationship between mass at metamorphosis and time to metamorphosis for tadpoles of *R. schneideri* in natural conditions by the gradual drying of the pond. Newman (1988) and Pfenning et al. (1991) also observed negative correlations between the size and the time of metamorphosis in tadpoles of the genus *Scaphiopus* in habitats whose duration was modified in an experimental array. Reques

& Tejedo (1995) also reported a negative relationship between these variables in larvae of *Bufo calamita* (Laurenti, 1768) in natural conditions, also due to increasing water temperature.

In our experiment, the water level was constant, without much fluctuation in water temperature throughout the larval development. We suggest that the absence of trade-off in the *R. schneideri* tadpoles in the first third of the time to reach metamorphosis was not influenced by abiotic or biotic interactions. In contrast, the stability of the environment lead to the observed trade-off changes in relation to the prolonged time to metamorphosis for the tadpoles that remained in the tanks. Absence of a trade-off for 53% of tadpoles arriving to metamorphic climax may be related to the life strategy. *Rhinella schneideri* is a typical r-selected species characterized by reproduction usually in unpredictable and temporary habitat immediately after rainfall. The adaptation of this species for rapid development and early metamorphosis for most tadpoles may be caused by genetic factors favoring tadpoles that grow fast with better characteristics, such as more mobility, greater growth rate and development rate.

Ecological implication for tadpoles arriving to metamorphosis between 57–78 days

In these individuals the mass to metamorphosis was significantly affected by the density and the interactions between predators and density, and not by the predators alone. The presence of predators directly influenced time to metamorphosis. Under stable conditions and when the time to metamorphosis is longer, density was important for mass to metamorphosis and the predators, similar to the first period, was also important for time to metamorphosis.

The covariation between mass and time at metamorphosis was a positive relationship. The mean mass to metamorphosis was greater than the mass obtained in the tadpoles that reached metamorphosis between 34–56 days. During this time, 93 (27% of the total) individuals reached metamorphic climax.

Ecological implication for tadpoles arriving to metamorphosis between 79–102 days

Between 79 and 102 days of development only the interactions between predators x density was significant on mass to metamorphosis. In this third period the two factors were not important on the metamorphs that finished their larval cycles.

Similar to the second period, a positive relationship in the covariation of mass at metamorphosis and development time was observed in the metamorphs of this period. Furthermore, the mean mass of metamorphs was greater than the mean mass of metamorphs from the two prior periods.

For the tadpoles that reached metamorphosis between 57–102 days of development, the presence of a trade-off between mass at metamorphosis and time to metamorphosis implies the reversibility of this feature. When there is a substantial cost of possessing a par-

ticular phenotype in the “wrong” environment, selection should favor the evolution of reversibility (Gabriel 1999). Variable environments should favor phenotypic reversibility in a species. Even if the water level were constant throughout the larval cycle, biotic factors would still change with development time (density, intraspecific competition, predator-prey relation, etc.).

In summary, for the *R. schneideri* tadpoles that usually breed in ephemeral ponds, the influences of biotic factors changed in response to habitat stability. As a typical newly born r-strategist, the main objective is to grow rapidly and reach a minimum size threshold to begin metamorphosis. For these individuals that have a negative covariation between mass at metamorphosis and time to metamorphosis, the priority is to reach the metamorphic climax, independent of habitat conditions. Furthermore, the coefficient of variation of the mass metamorphosis in these juveniles is lower than that recorded for individuals that metamorphosed between 57–78 and 79–102 days.

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