# Larval Life History of *Lithobates sphenocephalus* (Southern Leopard Frog) in Southeast Louisiana

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**Abstract -** Most field-based life-history studies of amphibians only monitor the first occurrence of major life stages, missing important details of the intervening periods. To help fill this gap, we surveyed the larval growth of *Lithobates sphenocephalus* (Southern Leopard Frog) at a breeding site in Hammond, Tangipahoa Parish, LA. We repeatedly sampled an ephemeral pool for Southern Leopard Frog tadpoles shortly after oviposition through metamorphosis. We first observed eggs on 21 January 2016. From 29 January 2016 through the following 99–117 d, tadpoles grew an average of 0.163 mm/day in body length before completing metamorphosis between 6 and 23 May. These values are similar to previous literature estimates for the Southern Leopard Frog, but provide novel information on the timing and rates of changes in size and ontogenetic stage, and their interrelationship.

## Introduction

*Lithobates sphenocephalus* Cope (Southern Leopard Frog) is a medium-sized semi-aquatic true frog (Ranidae) ranging across a large portion of the eastern US. This species is abundant throughout its range and is found predominantly in shallow freshwater habitats. In Louisiana, the Southern Leopard Frog is found in ephemeral and permanent water bodies statewide (Dundee and Rossman 1989). Despite their ubiquity, Dundee and Rossman (1989) stated that these frogs were often displaced by *Lithobates clamitans* Latreille (Green Frog), and local decline from habitat loss has been noted (Hudson 1956, Minton 2001).

There is considerable geographic variation in the reproductive habits of Southern Leopard Frogs. Breeding can occur in every month of the year in southern states, but typically occurs in the early spring (summarized in Butterfield et al. 2005, Dodd 2013). In southeastern Louisiana and adjacent areas, breeding is most common from December through February (Doody and Young 1995, Dundee and

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Rossman 1989, Mount 1975). Eggs are laid in clumps of 300–6500 near the surface of the water and the clumps measure 7.5–16 cm wide; eggs hatch in 3 to 20 d (e.g., Altig and McDiarmid 2015, Dundee and Rossman 1989, Wright 1932). Our goal was to survey the larval life history of the Southern Leopard Frog in an ephemeral pool in southeast Louisiana to quantify larval growth rate, developmental staging series, and the larval period as both an empirical test of observational reports and to provide the first detailed account of this short but critical stage of a well-known southeastern native species.

## Field-site Description

The study site is located on the periphery of a wooded area at the north end of North Oak Park, a recreational area owned by Southeastern Louisiana University in Hammond, Tangipahoa Parish, LA (30°31'36"N, 90°28'40"W). The site is a well-drained artificial canal that infrequently flows into the adjacent Ponchatoula Creek, but otherwise forms a series of intermittently connected pools of varying depth. The ephemeral pool sampled in this study was approxiamately 20 m x 3 m and had a maximal depth of 20 cm. Over the course of the study, the pool varied in size from 10 m x 1.5 m to completely inundated and continuous with all nearby water bodies, and in maximal depth from ~1 cm to 160 cm. The bottom of the pool consisted of mud and leaf litter from the canopy trees, primarily *Quercus michauxii* Nuttall (Swamp Chestnut Oak), *Q. stellata* Wangenheim (Post Oak), *Q. nigra* L.(Water Oak), *Magnolia grandiflora* L. (Southern Magnolia), *M. virginiana* L.(Sweetbay Magnolia), and *Acer rubrum* L. (Red Maple). The pool contained significant woody debris in the form of overhanging damaged and detached tree branches and small twigs with little emergent vegetation. Submergent vegetation consisted of clumps of filamentous algae. We observed and captured several potential predatory species during sampling, including *Gambusia affinis* Baird and Girard (Mosquitofish), *Aphredoderus sayanus* Gilliams (Pirate Perch), Centrarchidae (sunfish), *Agkistrodon piscivorous* Lacépède (Cottonmouth), *Nerodia erythrogaster* Forster (Plain-bellied Watersnake), Cambaridae (crayfish), Belostomatidae (giant water bug), Notonectidae (backswimmer), Corixidae (water boatman), Odonata (dragonfly and damselfly nymph), and Hirudiniformes (leech). The pool is also used for breeding by Green Frogs, and tadpoles of this species were present throughout the sampling period.

## Methods

### Collection

We dip-netted specimens from the study pool twice weekly beginning on 29 January 2016, when we first observed larvae hatching from egg masses laid ~8 d prior. We limited sampling efforts to 6 person-hours per collection with a goal of measuring 100 tadpoles each trip. When we had either collected a sufficient sample or reached the time limit, we placed the tadpoles in a glass tray with a calibration ruler and photographed them. We used a thermometer to record the temperature at the edges and center of the study site mid-depth in the water column. On the second sampling trip of the week, we collected a random subset of 10 or fewer tadpoles and preserved them in 10% formalin for museum voucher specimens, identification, and developmental staging. We deposited the specimens in lots into the Herpetology Collection at the Southeastern Louisiana University Vertebrate Museum (catalog numbers SLU 6618–6630).

**Analysis**

We made all measurements in ImageJ® software (Schneider et al. 2012), measuring total length (tip of snout to tip of tail) and body length (tip of snout to central muscle attachment point; following McDiarmid and Altig 1999) of each clearly imaged specimen. We measured egg and ovum dimensions in ImageJ®; we also measured egg size from interpolation of egg volume (displacement). Egg and ovum definitions followed standard terminology (Altig and McDiarmid 2015). We estimated growth and development rates using linear regression. To reduce bias, the same person (J.A. Erdmann) staged all tadpoles (following Gosner 1960) and made all software measurements.

## Results

On 21 January 2016, we observed about 6 clutches of freshly oviposited eggs (0–2 d old) on small emergent branches near the center of the pool. On 29 January, we found hatchlings (0–1 d old) attached, and in close proximity, to their natal egg mass. By 5 February, the clutch from which the first sample came appeared to have finished hatching (no developing embryos remaining and the egg mass mainly dissolved), resulting in variation in hatching time from 7 to16 d (Fig. 1). We collected the last tadpoles (Gosner stages 36–37) on 6 May, and collected a single metamorph (25-mm snout–vent length) on 23 May, providing a 17-d window in which metamorphosis likely occurred in the remaining tadpoles (Fig. 1). Assuming all larvae hatched by 23 May (none were collected then), we estimate a larval period of 99–117 d, and a complete pre-metamorphic period of 108–127 d.

The body-length and total-length measurements taken over the 99-d sampling period of Southern Leopard Frog larvae are summarized in Figure 2. The tadpoles exhibited a linear growth pattern with a body-growth rate of 0.1632 mm /d ($R^2$ = 0.728) and varied in size from 2.7 mm at hatching to a recorded maximum of 22 mm (Figs. 2, 3). Total length also showed a linear growth pattern, with a growth rate of 0.4751 mm/d ($R^2$ = 0.683), varying from 6.3 mm at hatching to 60.5 mm maximum (Figs. 2, 3). The size distribution for each sampling period is summarized in Figure 3. Gosner stage appeared to proceed in punctuated, nonlinear fashion, with 3 distinct periods of relative stasis in development (Gosner stages 25–26, 27–29, and 36–37; Fig. 1; linear $R^2$ = 0.728).

On 31 January 2017, we found a recently laid clutch and used this for egg and egg-mass measurements. At the time of discovery, the larvae had begun hatching. The egg mass consisted of ~2800 eggs (570 mL volumetric displacement based on a known count of 123 eggs = 25.0 mL). The average egg thus had a diameter of 7.3 mm assuming a spherical volume/diameter ratio. This finding is consistent with photographic measurement of 2 individual eggs that were 6.5 mm and 7.9 mm maximum diameter (but see discussion). The ovum contained within one of the eggs, in arrested development (ca. Gosner stage 13), measured 3.0 mm x 3.2 mm diameter.

## Discussion

The wide range in sizes and life-history periods reported in Altig and McDiarmid (2015) appear to encompass most of the natural range one might encounter across the geographic range of a given species. As such, these ranges may be broader than is likely to occur at a given locality for a widely distributed species. Egg size appears to be the exception to this breadth. Altig and McDiarmid (2015:236) report a range of 1.0–2.2 mm ovum diameter and 3.4–7.0 mm egg diameter for the Southern Leopard Frog. Both measurements of the sample taken in January 2017 exceed these measurements by ~1 mm diameter. Although the age of an egg plays a role in its dimensions (Volpe et al. 1961), ovum volume remains relatively fixed and ranid egg masses often maintain structure at least until hatching (Altig and McDiarmid

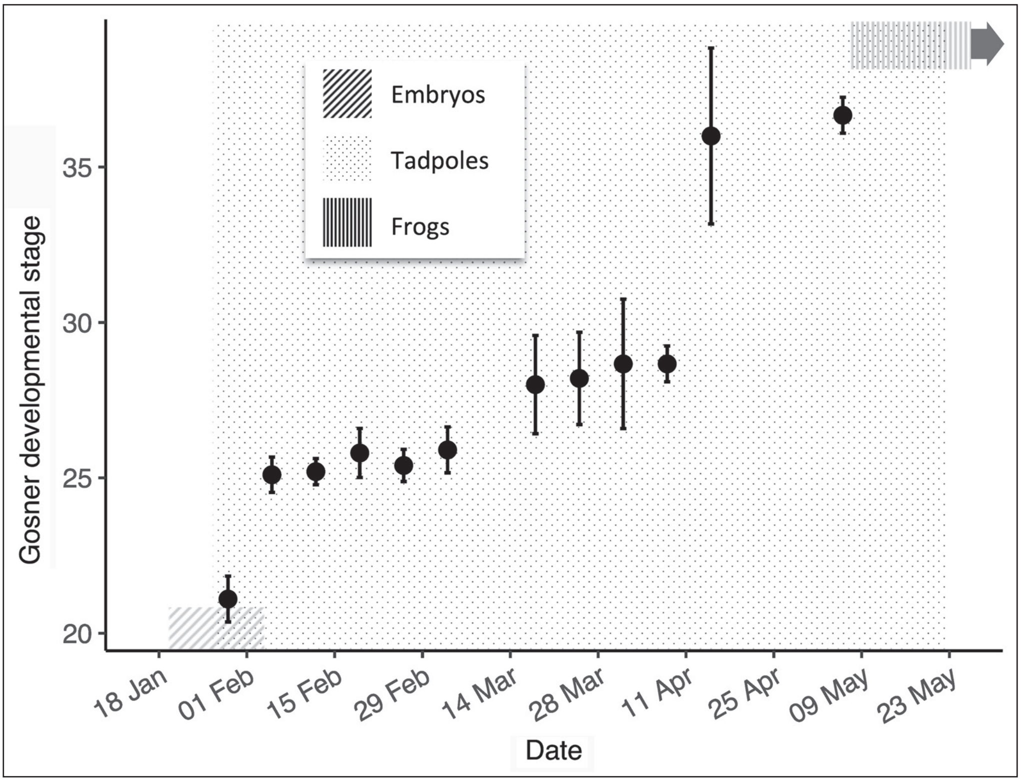


Figure 1. Ontogeny of Southern Leopard Frog tadpoles sampled at a breeding pool in Hammond, LA, in 2016. Shadings indicate when major developmental stages were detected ($x$-axis) as well as approximate developmental stages for defining classes ($y$-axis). Overlap between shadings represent the estimated maximum date ranges over which transitions (hatching and metamorphosis) occurred. Values represent mean ± 1 standard deviation.

2015). At our site, we saw remnants of egg masses a week after tadpoles were developed enough to disperse away from the clumps.

The 7–16-d range for hatching in Southern Leopard Frog eggs found in this study is a longer estimate than most previous values. Ashton and Ashton (1988) estimated a 4–5 d hatching time for frogs in Florida, which is similar to the estimate of 3–5 d in southern Georgia (Wright 1932). In northern reaches of the Southern Leopard Frog range, hatching takes upwards of 2 weeks (Johnson 1992, Martof et al. 1980). Altig and McDiarmid (2015) state that hatching time can happen between 3 d and 20 d. Crayfish were abundant in the pool, and their presence as predators is known to accelerate hatching in Southern Leopard Frog eggs (Saenz et al. 2003). Although this accelerated hatching would lead to a predicted reduction in hatching time relative to other estimates, lower temperatures (discussed below) and other variables may explain our unexpected extension in the estimated hatching period (Licht 1971, McLaren and Cooley 1972).

Our observed larval period of Southern Leopard Frogs is consistent with field guides that state a larval period of about 3 months (Ashton and Ashton 1988, Martof et al. 1980, Smith 1961). Interestingly, Wright (1932) hypothesized a "true" larval period of 50–75 d, and Wright and Wright (1942) proposed a period of 67–86 d. Our estimate of 99–117 days is longer than most, but is still encompassed by that

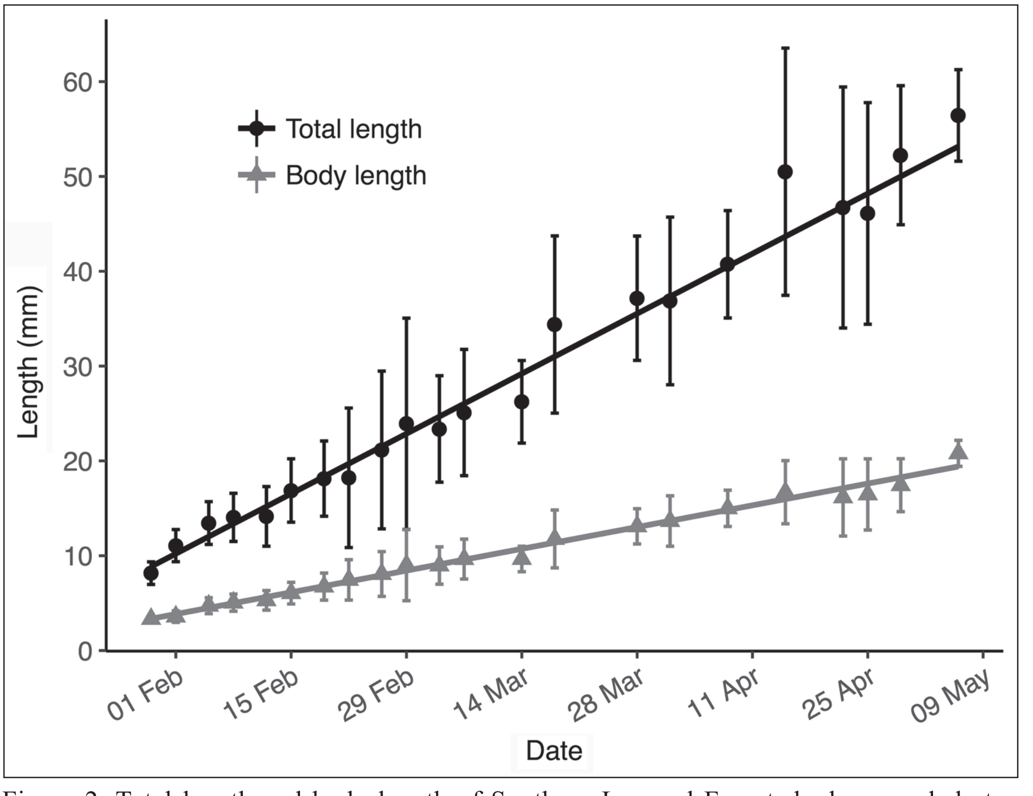


Figure 2. Total length and body length of Southern Leopard Frog tadpoles sampled at a breeding pool in Hammond, LA, in 2016. Values represent mean ± 1 standard deviation.

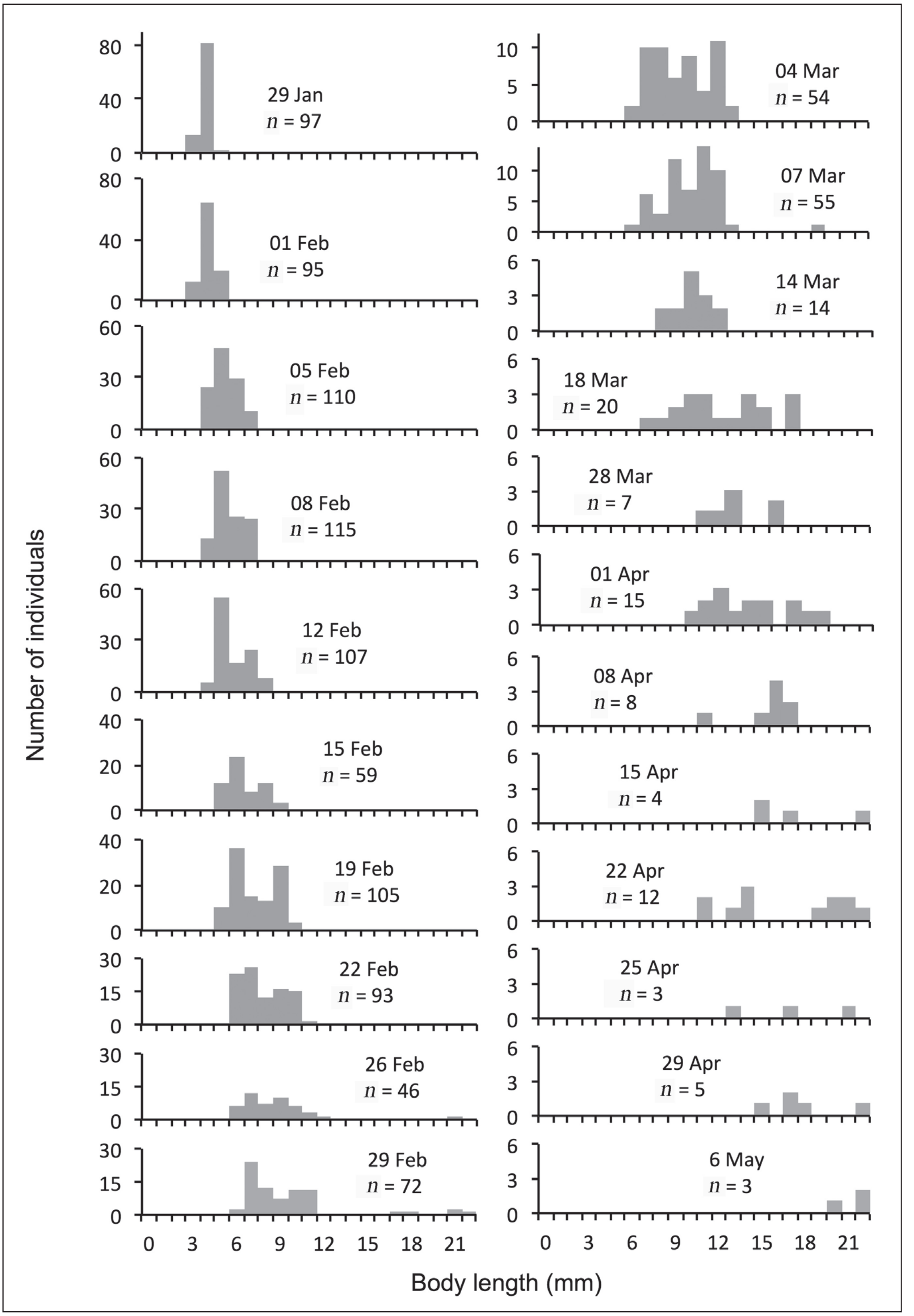


Figure 3. Histogram of size distributions of Southern Leopard Frog tadpoles sampled at a breeding pool in Hammond, LA, in 2016.

of Altig and McDiarmid (2015). It appears that the larval period is plastic, and depends on much more than just altitude and latitude across its range. Temperature differences can cause variation in metamorphic timing in amphibians, and lower temperatures cause delays in both oviposition and metamorphosis (Beachy 1995, Hayes et al. 1993, Uhlenhuth 1919, Voss 1993). Our sampling location is mostly shaded with few sunlit patches. Average pool temperature during sampling was 18 °C (min–max: 11–22 °C). It is not unusual for ponds that contain Southern Leopard Frog tadpoles to have temperatures that are much higher (Caldwell 1986). Abundant predators, including nymphal odonates, are also known to cause delays in Southern Leopard Frog metamorphosis (Babbitt 2001). A flood event between 7 and 14 March 2016 caused a reduction in tadpole abundance in the pool (Fig. 3), which may have resulted in delayed metamorphosis (Richter et al. 2009). Lastly, the multiple clutches observed may have been laid over multiple nights, adding to the reported breadth in larval period.

Altig and Crother (2016) recently discussed the value of staging in tadpoles, and demonstrate that, because of environmentally induced variability, staging is an unreliable method of tracking the growth and development of tadpoles both within and across taxa. Our data support the principle that tadpole ontogeny (as staged here and in most studies) does not precisely reflect the age of a tadpole. However, body-size data gathered in this study performed no better as a predictor of age than developmental stage ($r^2$-value of 0.728 for both), whereas total length was the worst for predicting age ($r^2$-value of 0.683). Differences are likely pronounced in species with extended larval periods, such as the sympatric *Lithobates catesbeianus* Shaw (American Bullfrog), where a single tadpole can grow in size over a span of months with little to no change in the outward developmental condition.

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