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# Supplementary Data Catenazzi and Kupferberg 2013

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## **SUPPLEMENTARY DATA**

**Kml file.** The kml file shows sites used in this study: oviposition sites of *Rana boylei* where we monitored water temperature throughout the tadpoles' development period.

### **Appendix. Rearing experimental design details.**

We raised tadpoles from unhatched embryos to metamorphosis in four unregulated streams in the watershed of the South Fork Eel River on the Angelo Reserve in Mendocino Co. CA. The factorial rearing experiment had four levels of location and two levels of food (Fig. A1). The streams naturally differ in thermal regime with Fox Creek and Elder Creek the coldest, South Fork Eel intermediate, and Tenmile Creek the warmest. The streams also differ in watershed size, canopy cover, mean annual discharge and primary productivity (Table 1 of main article text). Although all four streams support adult and juvenile *R. boylei*, only Tenmile and SF Eel are used for breeding (circles in Fig A1).

**Food supplementation.** We collected embryos at the gastrula stage (Gosner 10) from two egg masses laid on the night of May 11, 2008. Groups of 30 embryos were separated from each clutch, placed in lidded vials with stream water and carried in backpacks to each of four rearing locations. We equalized the time spent in transport by hiking the same amount of time as it took to reach the most distant rearing location. We placed the embryos in flow-through enclosures constructed from plastic laundry baskets (55 x 38 cm interior dimensions) with 1 mm fiberglass mesh glued over the openings. The same mesh covered the baskets and was held in place with clothespins. This prevented colonization by other organisms such as ovipositing dragonflies. Water depth was approximately 15-20 cm. We placed native cobbles with epilithic periphyton from each rearing location (low quality food) in the enclosures sufficient to cover the bottom of

each basket. Thirteen baskets were deployed in each stream. Clutch had no effect on tadpole mortality and was henceforth excluded as a factor in our analyses. We randomly chose six baskets in each stream to receive food supplementation (high quality food). We harvested the filamentous macroalga *Cladophora glomerata* covered with a heavy growth of epiphytic diatoms (including species of *Epithemia*, *Cocconeis*, and *Gomphonema*) in the SF Eel River, taking care to select patches with the most dense cover of epiphytes. We picked through the algae with forceps to remove macroinvertebrates and then spun the algae in a lettuce spinner for 50 revolutions to attain similar dampness among batches. Each food supplemented enclosure received 40g damp mass of algae each week. All enclosures also had rocks swapped with fresh rocks weekly when tadpoles survival and developmental stage were observed.

***Tadpole density reductions.*** As tadpoles developed, we manipulated the density in enclosures for two reasons. First, to mimic declining density patterns in the open river when tadpoles disperse from their oviposition / hatching sites and attrition occurs, we periodically removed tadpoles (Table A.1). Second, to maintain equivalent densities among enclosures in the four study streams, it was necessary to remove tadpoles from enclosures in the warmer sites to match numbers in the coldest streams (Elder and Fox Creeks), where weekly mortality rates were greater. Experimental densities were at the high end of the range of natural densities, but below the maximum density observed, during the course of quadrat surveys conducted along 5.2 km of the South Fork Eel River from 1991-1994, and in 2009. To calculate the 99% confidence interval of density ( $\#/m^2$ ) we calculated the average developmental stage in each quadrat with tadpoles present, and grouped quadrats according to the ranges of developmental stages used in the removal schedule for enclosed tadpoles (25–31, 32–35, 36–42). We also calculated the 99% confidence interval for tadpole body size (mm) at each Gosner stage from 27–45 from

individuals measured during the quadrat sampling (n=647 tadpoles). The enclosed tadpoles, which were measured weekly with dial calipers and assessed for developmental stage with a 5x hand lens, were either larger than, or within the 99% CI of body size of free living tadpoles until stage 40 (Fig. A2). This result indicates that crowding effects did not occur until just prior to the experiment's completion.

***Spatial heterogeneity of water temperature.*** Our watershed wide analysis assumes that the thermal conditions in the habitat utilized by tadpoles can be accurately described by fixed sensors within the vicinity of one km. To validate this assumption, we compared temperatures where tadpoles occurred at one transect (observed throughout the day, Fig. A3) to temperatures from near-shore benthic sensors as well as a mid-column thalweg sensor (Fig. A4). At intervals of 1.5 hr from 8 AM to 8 PM we measured the temperature of 20 tadpole locations, and found that fixed sensors placed at appropriate depths and velocities within one kilometer closely mirrored the operative tadpole temperatures. A thermister placed in the thalweg, however was consistently 1-2 C cooler than the tadpole temperatures.

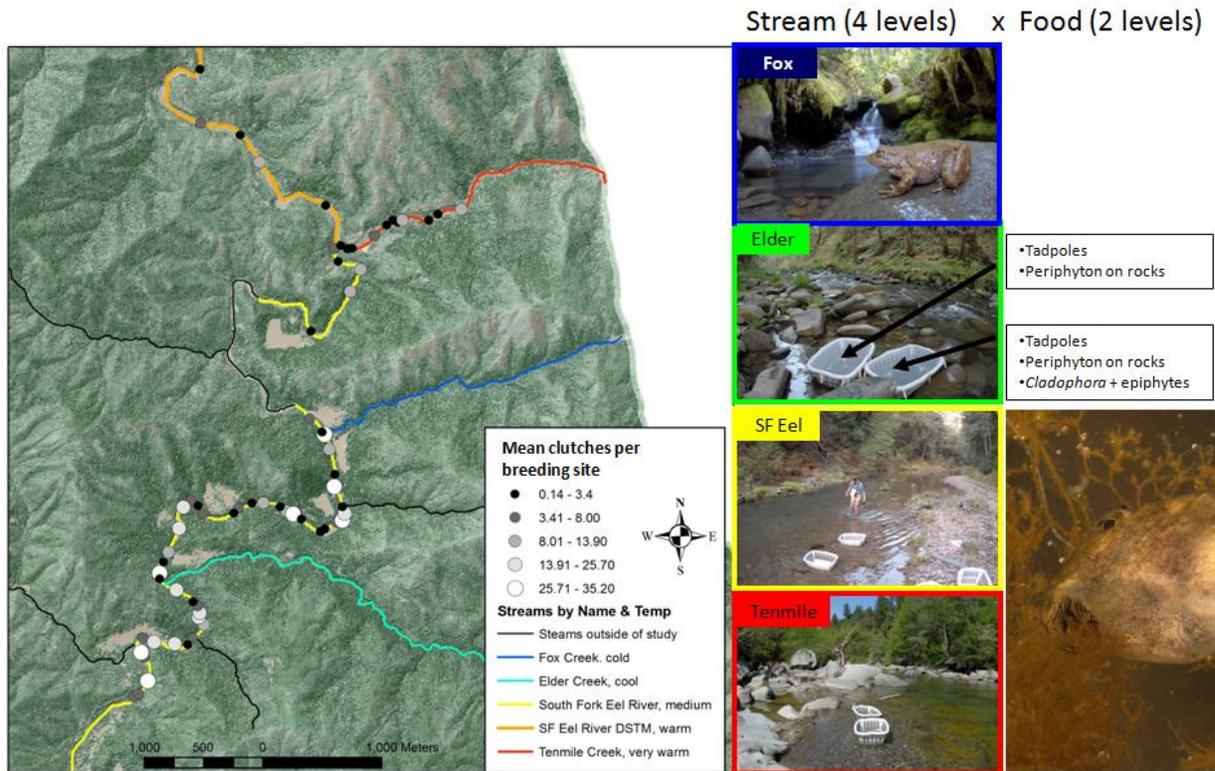
**Table A1.** Schedule of tadpole reduction in rearing experiment (initial density 30 embryos / enclosure) and comparison to open river densities.

Gosner stage	Density			Streams, warmest to coldest			
	Enclosed	Open river <sup>a</sup>		Tenmile	SF Eel	Elder	Fox
	#	#/m <sup>2</sup>	99% CI, max				
25	20	80		26 May	26 May	2 June	10 June
25 for ~1 wk	15	60	14.9–44.3, 428	1 June	2 June	11 June	17 June
32	10	40	5.4–9.1, 44	23 June	2 July	2 July	14 July
35 or 36	5	20	3.7–6.9, 52	1 July	20 July	3 Aug	3 Aug
42	1 <sup>b</sup>			≥20 July	≥20 July	≥24 Aug	≥31 Aug

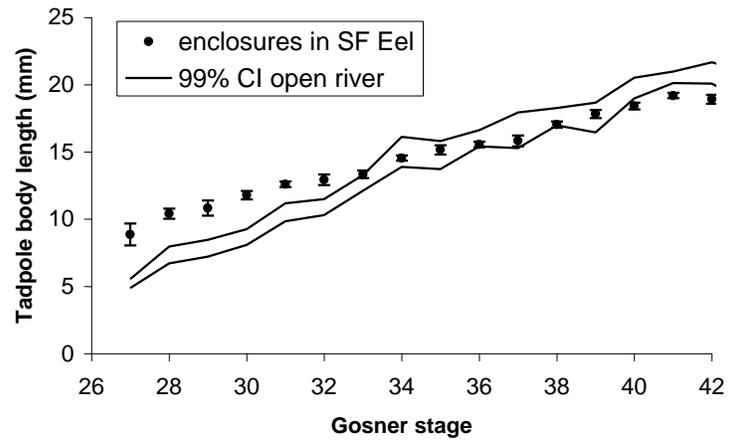
<sup>a</sup> Sample sizes = number of quadrats with tadpoles in the Gosner stage ranges:  $n_{25-31} = 94$ ,  $n_{32-35} = 104$ ,  $n_{36-42} = 119$ .

<sup>b</sup> Individuals removed from enclosures at front limb emergence, stage 42; remaining densities 1-4 / enclosure.

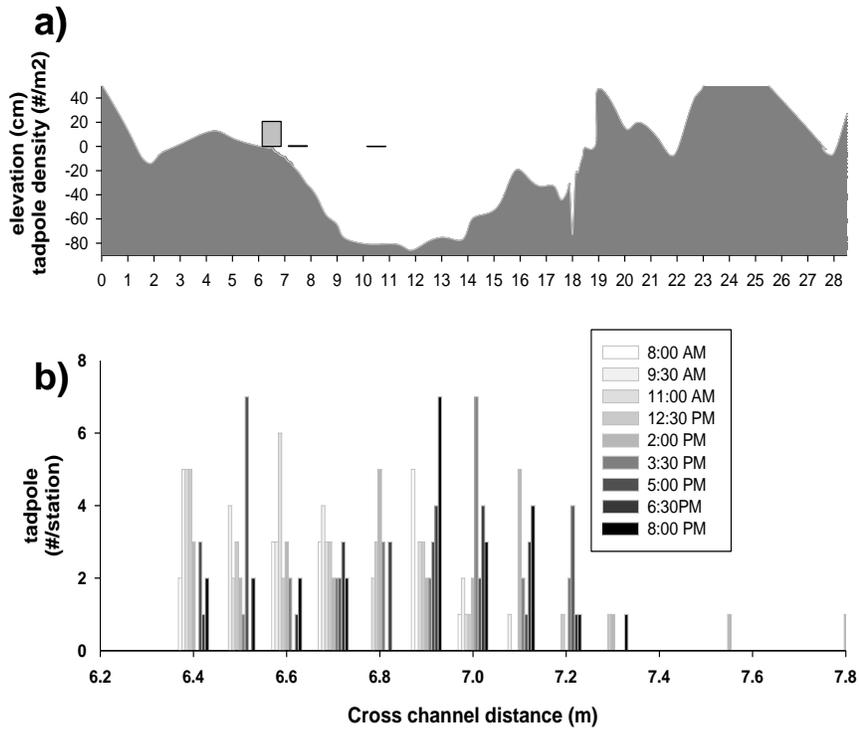
**Figure A1.** Experimental design of two factor rearing experiment.



**Figure A2. Mean ( $\pm 1$  SE) body size of *Rana boylei* tadpoles raised in enclosures compared to those in the open South Fork Eel River.**



**Figure A3.** Within a cross-section of the river channel, *Rana boylii* tadpoles utilize the shallow near shore environment (a). As tadpoles thermoregulate over the course of the day, they make small scale movements within the two meters closest to shore (b).



**Figure A4.** Thermal environment utilized by mobile *Rana boylii* tadpoles (box plots) over a 12 hour period (July 15, 2010) compared to temperatures (lines) measured at fixed data loggers placed on the benthos or mid-column at 3 different locations within approximately one km (either upstream, US, or downstream, DS) of the tadpole location observations

