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## EFFECTS OF AGE AND GROUP SIZE ON HABITAT SELECTION AND ACTIVITY LEVEL IN *XENOPUS LAEVIS* TADPOLES

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### ABSTRACT

We experimentally examined the effects of age and group size on habitat selection and activity level in *Xenopus laevis* tadpoles. Tadpoles were introduced into an experimental arena at two ages (early and late) and at three group sizes (one, two, and six tadpoles). Group size did not affect habitat selection or activity level. Late tadpoles were more active and used the bottom of the arena more than early tadpoles. Our results suggest that an intrinsic cue (age) is important in determining the behavior of these tadpoles.

† † †

In amphibian larvae, habitat selection and activity level may be altered in the presence of particular cues or environments. For example, in some amphibian larvae, activity level is influenced by chemical cues emitted by both the predator and the prey item (Bridges and Gutzke 1997, Kiesecker et al. 1996, Petranka et al. 1987). A tadpole's activity level can also depend on food levels or the presence or absence of a predator (Anholt and Werner 1995). Such responses may result in the formation of aggregates of tadpoles (Watt et al. 1997). Habitat selection behavior in amphibian larvae is also affected by environmental cues. Previous studies have demonstrated that tadpoles select habitats in response to oxygen concentration, population density, water temperature, water depth, vegetation density, predators, time of day, and substrate type or pattern (Johnson 1991, Kiesecker and Blaustein 1998, Nie et al. 1999, Noland and Ultsch 1981, Peterson et al. 1992, Schley et al. 1998, Waringer-Löschenkohl 1988).

Age or developmental stage affects habitat selection in some amphibian larvae. In some cases habitat preference or use changes with ontogeny or size (Alford 1986, Alford and Crump 1982, Smith 1999; Werner

1992). In *Xenopus laevis*, tadpoles change substrate color preference, preferring white early and black late in development (Moriya et al. 1996). In another case, *Rana pipiens* tadpoles develop a strong tendency to favor a white background, regardless of the background on which they were reared (Dunlap and Satterfield 1982). Background preference is established by experience early in development of *Rana aurora* tadpoles and persists in isolation from the background (Wiens 1970). Less is known about the effect of age on activity level; however, swimming ability appears to be affected by age or size (Brown and Taylor 1995, Jung and Jago 1995, McCollum and Leimberger 1997), and so it would seem likely that activity levels may change with age as tadpole swimming ability changes.

Group size is another possible factor influencing habitat selection and activity levels in tadpoles. Previous studies have suggested that there is some variation between species in the response of tadpoles to the number of conspecifics. Some species increase activity or change habitat use, whereas other species show no change in behavior (Griffiths and Foster 1998; Lefcort 1998)

We describe the effects of group size and age on activity levels and habitat selection of tadpoles of *Xenopus laevis* in the laboratory. Since amphibian larvae often behave in such a way as to reduce the risk of predation (e.g., Anholt and Werner 1995, Kupferberg 1998, Lefcort 1998), we predicted that the *X. laevis* tadpoles would alter their behavior in response to indirect cues of predation risk, in particular, an intrinsic cue, age/ body size; and an extrinsic cue, group size. Predation risk of anuran larvae often decreases with size (e.g., Semlitsch 1990), thus we predicted that younger, smaller tadpoles would use habitats that could

be perceived as less risky (e.g., increased use of a vegetated habitat, decreased use of the open water column) and to lower activity, which tends to reduce predation risk (e.g., Kupferberg 1998, Skelly 1994). Group size may influence an individual's perception of predation risk, the larger the group the lower the individual's perceived risk (e.g., Watt et al. 1997). We therefore predicted that tadpoles alone or in small groups would use less risky habitats and have lower levels of activity compared to tadpoles in larger groups. We also expected that the effect of group size would decrease in older, larger tadpoles since the perception of risk would be lower, thus group size may not have any additional effect (i.e., we predicted a significant interaction term).

## MATERIALS AND METHODS

*Xenopus laevis* tadpoles are relatively unique among anuran larvae (see Wassersug 1996 for a review). Among their unique characteristics are their morphology (both in their feeding structures and in the anatomy of their spinal cord and associated muscles) (Wassersug 1996). *Xenopus laevis* tadpoles are positively buoyant and are obligate air-breathers (Wassersug 1996). In addition, they often aggregate and tend to school in a fashion similar to fish, using a lateral line system to help hold school shape (Wassersug 1996). Unfortunately, very little is known about the biology of *X. laevis* tadpoles in the wild (e.g., natural predators).

*Xenopus laevis* eggs were obtained from the Carolina Biological Supply Company. Eggs were incubated at room temperature (19°C), and hatched after 10 d of incubation. Tadpoles were maintained under similar conditions in opaque plastic containers. Two to three days after hatching, we began to feed tadpoles crushed adult *Xenopus* food. Tadpoles were fed and containers cleaned daily.

Test arenas were 19-L aquaria with a shallow bed of pebbles covering the bottom, with larger rocks and artificial vegetation placed on opposite ends of the tank, leaving an open area in the center. The test arenas were filled half-full with filtered stream water. After each trial test arenas were emptied and refilled to eliminate any cues that may have been left by the previous tadpole(s).

The general testing procedure consisted of carefully placing the appropriate number of tadpoles into the test arena and then allowing 15 min for the tadpoles to acclimate to the arena. Following the acclimation period, the position and activity level for each tadpole were noted every minute for 15 min. Specific information collected included: 1) location in the tank (bottom = resting on the substrate, open = floating or swimming in water column, against glass = resting

against the side of the aquarium, but not resting on the substrate), 2) habitat (rock, neutral, vegetated), and 3) activity level (low = no movement, medium = some tail movement, high = actively swimming). Four replicates were performed for each tadpole density (one, two, or six).

The first set of trials occurred two weeks after hatching. Tadpoles in the first set of trials were stage 25 (Gosner 1960). The second set of trials occurred four weeks after the completion of the first set of trials (seven weeks post-hatching). Tadpoles in the second set of trials were stages 36–38 (Gosner 1960). Tadpoles in the second set of trials were substantially larger than those used in the first set of trials (on the order of 3 to 4 times larger). Tadpoles were not used in more than one set of experiments.

To analyze the data, we generated a mean proportion of tadpoles observed in each location, habitat, or at each activity level for a given trial by averaging the proportions from each individual minute's observation. Thus, for each trial we had a mean proportion for each location (bottom, open, against glass), habitat (rock, neutral, vegetated), and activity level (low, medium, high).

For each type of variable (location, habitat, and activity level), we ran a separate two-way ANOVA with age and group size as the independent variables for each type of observation. For example, to analyze the location data we did separate ANOVAs on bottom use, glass use, and open water use. Because of the multiple tests we use a corrected  $\alpha$ -value of 0.017. Prior to analysis we transformed all proportion data with an arcsin square-root transformation. All means are given  $\pm 1$  standard error (SE), and represent the untransformed proportion data.

## RESULTS

Late tadpoles were more likely to be observed on the bottom than early tadpoles ( $0.450 \pm 0.074$  vs.  $0.173 \pm 0.061$ ,  $N = 12$  in both cases;  $F_{1,18} = 14.04$ ,  $P = 0.0015$ ). However, the size of the group interacted with the age of the tadpoles to modify the general trend (Fig. 1;  $F_{2,18} = 7.10$ ,  $P = 0.005$ ). For lone tadpoles, early tadpoles spent more time on the bottom; however, for pairs and groups, late tadpoles spent more time on the bottom. Group size by itself did not significantly affect the use of the bottom of the test arena ( $F_{2,18} = 0.40$ ,  $P = 0.68$ ). Age ( $F_{1,18} = 4.12$ ,  $P = 0.057$ ), group size ( $F_{2,18} = 0.094$ ,  $P = 0.91$ ), and the interaction between age and group size ( $F_{2,18} = 2.89$ ,  $P = 0.076$ ) were not significant influences on the use of glass by the tadpoles. Similarly, none of these factors significantly influenced tadpole use of the open water column (age:  $F_{1,18} = 2.35$ ,  $P = 0.14$ ; group

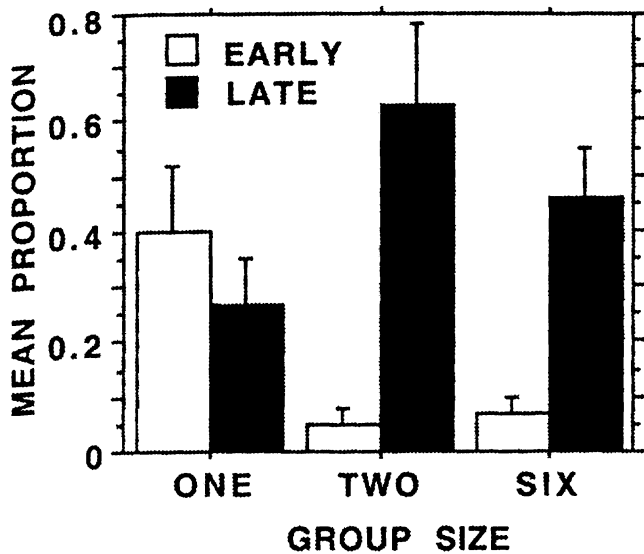


Figure 1. Mean ( $\pm 1$  S.E.) proportion of time spent on the bottom of the test arena as influenced by age and group size of *Xenopus laevis* tadpoles. Values given are untransformed raw means.

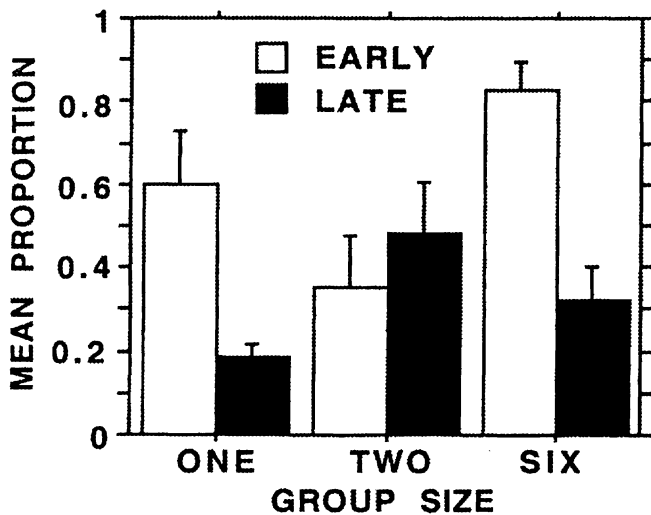


Figure 2. Mean ( $\pm 1$  S.E.) proportion of time spent at low activity levels as influenced by age and group size of *Xenopus laevis* tadpoles. Values given are untransformed raw means.

size:  $F_{2,18} = 3.29, P = 0.060$ ; age  $\times$  group size:  $F_{2,18} = 2.48, P = 0.11$ .

Age ( $F_{1,18} = 3.14, P = 0.093$ ), group size ( $F_{2,18} = 3.05, P = 0.072$ ), and the age by group size interaction ( $F_{2,18} = 1.50, P = 0.25$ ) did not significantly influence tadpole use of the rock habitat. Group size did not affect the proportion of time tadpoles spent using the vegetated side of the test arena ( $F_{2,18} = 0.99, P = 0.39$ ). Age also did not affect tadpole use of the vegetated habitat ( $F_{1,18} = 1.52, P = 0.23$ ). The age by group size interaction was also not significant ( $F_{2,18} = 0.64, P =$

0.54). Similar results were found for tadpole use of the neutral habitat (age:  $F_{1,18} = 0.028, P = 0.87$ ; group size:  $F_{2,18} = 0.60, P = 0.56$ ; age by group size:  $F_{2,18} = 0.027, P = 0.97$ ).

Overall, early tadpoles were more likely to be observed at low activity levels than late tadpoles ( $0.592 \pm 0.083$  vs.  $0.329 \pm 0.058, N = 12$  in both cases;  $F_{1,18} = 7.44, P = 0.014$ ). Lone early tadpoles and early tadpoles in groups spent more time at low activity levels than lone and group late tadpoles, respectively, whereas late tadpoles in pairs spent more time at low activity levels than early tadpoles in pairs (Fig. 2; interaction:  $F_{2,18} = 5.36, P = 0.015$ ). Group size had no significant influence on the proportion of tadpoles observed at low activity levels ( $F_{2,18} = 1.98, P = 0.17$ ). Age ( $F_{1,18} = 0.27, P = 0.61$ ), group size ( $F_{2,18} = 1.03, P = 0.38$ ), and the age by group size interaction ( $F_{2,18} = 1.14, P = 0.34$ ) did not significantly affect the proportion of time spent at medium activity level. Older tadpoles were more often seen at high activity levels than young tadpoles ( $0.444 \pm 0.070$  vs.  $0.126 \pm 0.034, N = 12$  in both cases;  $F_{1,18} = 19.2, P = 0.0004$ ). The proportion of time tadpoles spent at high activity level was not affected by group size ( $F_{2,18} = 1.22, P = 0.32$ ). The interaction between age and group size for high activity was not significant ( $F_{2,18} = 2.75, P = 0.091$ ).

### DISCUSSION

*Xenopus laevis* tadpoles showed no change in habitat selection or activity level with changes in group size. Other experiments have shown tadpoles alter behavioral patterns in the presence of conspecifics, often increasing activity levels or the use of open habitats (Griffiths and Foster 1998, Lefcort 1998, Rödel and Linsenmair 1997). However, still other species show no effect of group size on behavior of tadpoles (Griffiths and Foster 1998). Further investigations into why such differences exist between taxa in the response to the presence of conspecifics would be very informative.

Changes in behavior did occur as a function of age. Early *X. laevis* tadpoles were less active and used the glass more than the bottom, whereas older tadpoles were more active and used the bottom more than the glass. *Xenopus laevis* tadpoles are positively buoyant (Wassersug 1996) and thus may be able to use the water column even though they may not be strong swimmers. Also, large tadpoles may attain a size where they are no longer able to be eaten by a gape-limited predator (see Caldwell et al. 1980, Semlitsch 1990); thus larger tadpoles may perceive lower predation risk and use riskier, more rewarding habitats or behaviors. Alternatively, a tadpole's developmental stage or size may influence its swimming ability (e.g., Brown and Taylor 1995, Jung and Jago 1995, McCollum and

Leimberger 1997). Indeed, the early tadpoles had just become free swimming tadpoles and may avoid, or not be able to exploit as well, habitats requiring active swimming such as the open water column.

Amphibian larval behavior is determined, in part, by the interaction of intrinsic factors (e.g., age or developmental stage) and extrinsic factors (e.g., group size, predation pressure). Our study suggests that age, an intrinsic factor, may play an important role in determining the behavior of larval *X. laevis*. Age may also interact with the number of conspecifics, and extrinsic factor, to determine a tadpole's behavior. Our study also suggests that the evaluation of additional potential influences on amphibian larval behavior are warranted, as are studies assessing the effects of the interactions of multiple cues, both intrinsic and extrinsic, on tadpole behavior. Additional studies on the flexibility of tadpole responses to extrinsic and intrinsic cues would also be very enlightening.

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#### LITERATURE CITED

- Alford, R. A. 1986. Habitat use and positional behavior of anuran larvae in a northern Florida temporary pond. *Copeia* 1986: 408–423.
- , and M. L. Crump. 1982. Habitat partitioning among size classes of larval southern leopard frogs, *Rana utricularia*. *Copeia* 1982: 367–373.
- Anholt, B. R., and E. E. Werner. 1995. Interaction between food availability and predation mortality mediated by adaptive behavior. *Ecology* 76: 2230–2234.
- Bridges, C. M., and W. H. N. Gutzke. 1997. Effects of environmental history, sibship, and age on predator-avoidance responses of tadpoles. *Canadian Journal of Zoology* 75: 87–93.
- Brown, R. M., and D. H. Taylor. 1995. Compensatory escape mode trade-offs between swimming performance and maneuvering behavior through larval ontogeny of the wood frog, *Rana sylvatica*. *Copeia* 1995: 1–7.
- Caldwell, J. P., J. H. Thorp, and T. O. Jervey. 1980. Predator-prey relationships among larval dragonflies, salamanders, and frogs. *Oecologia* 46: 285–289.
- Dunlap, D. G., and C. K. Satterfield. 1982. Habitat selection in larval anurans: early experience and substrate pattern selection in *Rana pipiens*. *Developmental Psychobiology* 18: 37–58.
- Gosner, K. L. 1960. A simplified table for staging anuran embryos and larvae with notes on identification. *Herpetologica* 16: 183–190.
- Griffiths, R. A., and J. P. Foster. 1998. The effect of social interactions on tadpole activity and growth in the British anuran amphibians (*Bufo bufo*, *B. calamita*, and *Rana temporaria*). *Journal of Zoology* 245: 431–437.
- Johnson, L. M. 1991. Growth and development of larval northern cricket frogs (*Acris crepitans*) in relation to phytoplankton abundance. *Freshwater Biology* 25: 51–59.
- Jung, R. E., and C. H. Jagoe. 1995. Effects of low pH and aluminum on body size, swimming performance, and susceptibility to predation of green tree frog (*Hyla cinerea*) tadpoles. *Canadian Journal of Zoology* 73: 2171–2183.
- Kiesecker, J. M., and A. R. Blaustein. 1998. Effects of introduced bullfrogs and smallmouth bass on microhabitat use, growth, and survivorship of native red-legged frogs (*Rana aurora*). *Conservation Biology* 12: 776–787.
- , D. P. Chivers, A. R. Blaustein. 1996. The use of chemical cues in predator recognition by western toad tadpoles. *Animal Behaviour* 52: 1237–1245.
- Kupferberg, S. J. 1998. Predator mediated patch use by tadpoles (*Hyla regilla*): risk balancing or consequence of motionlessness? *Journal of Herpetology* 32: 84–92.
- Lefcort, H. 1998. Chemically mediated fright response in southern toad (*Bufo terrestris*) tadpoles. *Copeia* 1998: 445–450.
- McCollum, S. A., and J. D. Leimberger. 1997. Predator-induced morphological changes in an amphibian: predation by dragonflies affects tadpole color, shape, and growth rate. *Oecologia* 109: 615–621.
- Moriya, T., K. Kito, Y. Miyashita, and K. Asami. 1996. Preference for background color of the *Xenopus laevis* tadpole. *Journal of Experimental Zoology* 276: 335–344.
- Nie, M., J. D. Crim, and G. R. Ultsch. 1999. Dissolved oxygen, temperature, and habitat selection by bullfrog (*Rana catesbeiana*) tadpoles. *Copeia* 1999: 153–162.
- Noland, R., and G. R. Ultsch. 1981. The roles of temperature and dissolved oxygen in microhabitat selection by the tadpole of a frog (*Rana pipiens*) and a toad (*Bufo terrestris*). *Copeia* 1981: 645–652.
- Peterson, A. G., C. M. Bull, L. M. Wheeler. 1992. Habitat choice and predator avoidance in tadpoles. *Journal of Herpetology* 26: 142–146.
- Petranka, J. W., L. B. Kats, and A. Sih. 1987. Predator-prey interactions among fish and larval amphibians: use of chemical cues to detect predatory fish. *Animal Behaviour* 35: 420–425.
- Rödel, M.-O. and K. E. Linsenmair. 1997. Predator-

- induced swarms in the tadpoles of an African savanna frog, *Phrynomantis microps*. *Ethology* 103: 902–914.
- Schley, L., R. A. Griffiths, A. Román. 1998. Activity patterns and microhabitat selection of Mallorcan midwife toad (*Alytes muletensis*) tadpoles in natural torrent pools. *Amphibia-Reptilia* 19: 143–151.
- Semlitsch, R. D. 1990. Effects of body size, sibship, and tail injury on the susceptibility of tadpoles to dragonfly predation. *Canadian Journal of Zoology* 68: 1027–1030.
- Skelly, D. K. 1994. Activity level and the susceptibility of anuran larvae to predation. *Animal Behaviour* 47: 465–468.
- , and E. E. Werner. 1990. Behavioral and life-historical responses of larval American toads to an odonate predator. *Ecology* 71: 2313–2322.
- Smith, G. R. 1999. Microhabitat preferences of bullfrog tadpoles (*Rana catesbeiana*) of different ages. *Transactions of the Nebraska Academy of Sciences* 25: 73–76.
- Waringer-Löschenkohl, A. 1988. An experimental study of microhabitat selection and microhabitat shifts in European tadpoles. *Amphibia-Reptilia* 9: 219–236.
- Wassersug, R. 1996. The biology of *Xenopus* tadpoles. In: R. C. Tinsley and H. R. Kobel (eds.), *The Biology of Xenopus*. Oxford, Oxford University Press: 195–211.
- Watt, P. J., S. F. Nottingham, and S. Young. 1997. Toad tadpole aggregation behavior: evidence for a predator avoidance function. *Animal Behaviour* 54: 865–872.
- Werner, E. E. 1992. Individual behavior and higher-order species interactions. *American Naturalist* 140: S5–S32.
- Wiens, J. A. 1970. Effects of early experience on substrate pattern selection in *Rana aurora* tadpoles. *Copeia* 1970: 543–548.