

## Why Should Reacquisition of Larval Stages by Desmognathine Salamanders Surprise Us?

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It is understandable that Bruce (2005) questions interpretation of our independent studies concerning plethodontid life-history evolution. Larval development is found in all ten families of salamanders, and only members of Plethodontidae exhibit direct development from deposited eggs (Salthe 1969). Nevertheless, all analyses of mitochondrial DNA (Macey 2005; Mueller et al. 2004; Titus and Larson 1994), nuclear DNA (Chippindale et al. 2004), and even of the combined morphological and molecular data (Chippindale et al. 2004), indicate that desmognathine salamanders with larval development are deeply nested within direct-developing clades. Given the new and robust phylogenetic hypotheses, the most parsimonious interpretation of these results is that the free-living larval phase has re-evolved. We think that explorations such as those presented by Bruce (2005) are important contributions toward explaining these findings. However, invocation of generalizations such as Dollo's "Law" (which states that complex states cannot re-evolve once lost; Hall 1998) is not the most appropriate approach to the analysis of real data sets. Empirical analyses of Dollo's Law show that genes controlling developmental processes can indeed remain dormant for long time periods, up to 10 million years, and then be reactivated to express traits previously "lost" within clades (e.g., Marshall et al. 1994). Within salamanders there have been demonstrable reversals of other life-history traits, notably a return of metamorphosis in taxa deeply nested within otherwise permanently larval forms (Bruce 1976; Chippindale et al. 2000; Shaffer 1984; Sweet 1982). Also, there is surprising plasticity in life-history traits associated with metamorphosis and even egg retention and oviductal development (Lanza and Piero 2001; Wake 1989; Wake and Hanken 1996).

Perhaps, as Bruce (2005) argues, life history evolution has been highly non-parsimonious (i.e., direct development has independently evolved many times within Plethodontidae), and what has been called "direct development" is, instead, a heterogeneous set of non-homologous states. Labeling his argument as a "point of view" is appropriate. We think that his arguments contain elements of speculation, whereas ours are based on direct interpretations of the phylogenetic data. Refuting the possibility of a reversal from direct development to a free-living aquatic larval stage in desmognathines would require the acceptance of at least four or five independent origins of direct development. This would also imply that "direct development" is not one but many states (e.g., Wake and Hanken 1996); if so, there has been far more diversity of life-history modes in plethodontids than has been recognized previously. Our new phylogenetic hypotheses are based largely

on new molecular datasets; because they are hypotheses, we are actively engaged in gathering additional data to test them. We need to develop other datasets and integrate our studies in such a manner as to effectively test and reject alternative hypotheses of plethodontid relationships until such hypotheses can generally be viewed as robust, even if they conflict with more classical hypotheses for the evolution of morphology and life-history evolution in this group. The newly discovered Asian plethodontid salamander, *Karsenia koreana*, also appears to be a direct developer. Given its placement in the phylogeny (Min et al. 2005), the case for a reversal in *Desmognathus* is even stronger; argument against reversal now requires the independent evolution of direct development in at least six clades (*Plethodon*, *Hydromantes* + *Ensatina*, *Karsenia*, *Aneides*, *Phaeognathus*, and *Desmognathus wrighti*).

What has become clear in this developing debate is the absence of relevant comparative ontogenetic and developmental morphological data for salamanders. Relatively few species have been studied, and even such common species as members of *Amphiuma* are imperfectly known. Direct development is best known from studies of a species that is only a marginal direct developer, *Desmognathus aeneus* (Marks 2000), and very little is known about the early ontogeny of the 328+ species of direct developing plethodontids, many of which may prove to be relevant to this controversy.

#### LITERATURE CITED

- BRUCE, R. C. 1976. Population structure, life history, and evolution of paedogenesis in the salamander *Eurycea neotenes*. *Copeia* 1976:242–249.
- . 2005. Did desmognathine salamanders reinvent the larval stage? *Herpetol. Rev.* 36:107–112.
- CHIPPINDALE, P. T., R. M. BONETT, A. S. BALDWIN, AND J. J. WIENS. 2004. Phylogenetic evidence for a major reversal of life-history evolution in plethodontid salamanders. *Evolution* 58:2809–2822.
- , A. H. PRICE, D. M. HILLIS, AND J. J. WIENS. 2000. Phylogenetic relationships and systematic revision of central Texas hemidactyliine plethodontid salamanders. *Herpetol. Monogr.* 14:1–80.
- HALL, B. K. 1998. *Evolutionary Developmental Biology*. Chapman and Hall, London. 489 pp.
- LANZA, B., AND L. PIERO. 2001. Prima osservazione sicura di riproduzione vivipara nel genere *Speleomantes* (Amphibia: Caudata: Plethodontidae). *Pianura* 13:317–319.
- MACEY, J. R. 2005. Plethodontid salamander mitochondrial genomics: a parsimony evaluation of character conflict and implications for historical biogeography. *Cladistics* 21:194–202.
- MARKS, S. B. 2000. Skull development in two plethodontid salamanders (Genus *Desmognathus*) with different life histories. In R. C. Bruce, R. G. Jaeger, and L. D. Houck (eds.), *The Biology of Plethodontid Salamanders*, pp. 261–276. Kluwer Academic/Plenum Publishers, New York.
- MARSHALL, C. R., E. C. RAFF, AND R. A. RAFF. 1994. Dollo's law and the death and resurrection of genes. *Proc. Natl. Acad. Sci.* 91:12283–12287.
- MIN, M. S., S. Y. YANG, R. M. BONETT, D. R. VIEITES, R. A. BRANDON, AND D. B. WAKE. 2005. Discovery of the first Asian plethodontid salamander. *Nature* 435:87–90.
- MUELLER, R. L., J. R. MACEY, M. JAEKEL, D. B. WAKE, AND J. L. BOORE. 2004. Morphological homoplasy, life history evolution, and historical biogeography of plethodontid salamanders inferred from complete mitochondrial genomes. *Proc. Natl. Acad. Sci.* 101:13820–13825.
- SALTHER, S. N. 1969. Reproductive modes and the number and sizes of ova in the urodeles. *Amer. Midl. Natur.* 81:467–490.

- SHAFFER, H. B. 1984. Evolution in a paedomorphic lineage. I. An electrophoretic analysis of the Mexican ambystomatid salamanders. *Evolution* 38:1194–1206.
- SWEET, S. S. 1982. A distributional analysis of epigeal populations of *Eurycea neotenes* in central Texas, with comments on the origin of troglomorphic populations. *Herpetologica* 38:430–444.
- TITUS, T. A., AND A. LARSON. 1996. Molecular phylogenetics of desmognathine salamanders (Caudata: Plethodontidae): a reevaluation of evolution in ecology, life history, and morphology. *Syst. Biol.* 45:451–472.
- WAKE, M. H. 1989. Phylogenesis of direct development and viviparity in vertebrates. Complex organismal functions: integration and evolution in vertebrates, In D. B. Wake and G. Roth (eds.), pp. 235–250, John Wiley & Sons Ltd., London.
- WAKE, D. B., AND J. HANKEN. 1996. Direct development in the lungless salamanders: what are the consequences for developmental biology, evolution and phylogenesis? *Int. J. Dev. Biol.* 40:859–869.