

Origins of the other metazoan body plans: the evolution of larval forms

Rudolf A. Raff^{1,2,*}

¹*Department of Biology, Indiana University, Bloomington, IN 47405, USA*

²*School of Biological Sciences, University of Sydney, Sydney, New South Wales 2006, Australia*

Bilaterian animal body plan origins are not solely about adult forms. Most animals have larvae with body plans, ontogenies and ecologies distinct from adults. There are two primary hypotheses for larval origins. The first hypothesis suggests that the first animals were small pelagic forms similar to modern larvae, with adult bilaterian body plans evolved subsequently. The second hypothesis suggests that adult bilaterian body plans evolved first and that larval body plans arose by interpolation of features into direct-developing ontogenies. The two hypotheses have different consequences for understanding parsimony in evolution of larvae and of developmental genetic mechanisms. If primitive metazoans were like modern larvae and distinct adult forms evolved independently, there should be little commonality of patterning genes among adult body plans. However, sharing of patterning genes is observed. If larvae arose by co-option of adult bilaterian-expressed genes into independently evolved larval forms, larvae may show morphological convergence, but with distinct patterning genes, and this is observed. Thus, comparative studies of gene expression support independent origins of larval features. Precambrian and Cambrian embryonic fossils are also consistent with direct development of the adult as being primitive, with planktonic larvae arising during the Cambrian. Larvae have continued to co-opt genes and evolve new features, allowing study of developmental evolution.

Keywords: metazoan origins; larval evolution; body plans; Cambrian radiation; regulatory gene evolution; evolutionary developmental biology

1. EVOLUTION, THE STUDY OF ADULTS

It is striking that studies of evolutionary histories are nearly all about the evolution of adults. Palaeontologists, having only a few fossil larval forms, perforce have to study adults, which make up most of the fossil record. Transitions that can be studied are nearly inevitably those of adult characters. In popular presentations, this translates into computer animations where fins transform into legs, dinosaurs into birds or apes into hominids—beguiling but misleading images. The bias extends to phylogeny. Most available characters are adult ones. Our definitions of the body plans of phyla are of adult body plans. This bias persists in evo–devo, which largely focuses on evolution of novel adult features (e.g. loss of legs in snakes and the origin of the turtle shell). These examples are now approached by studies that combine morphological, palaeontological and gene regulatory data (Cohn & Tickle 1999; Gilbert *et al.* 2001). Developmental biology also focuses primarily on the development of adults. This is largely dictated by interest in major body parts (e.g. insect wings or tetrapod legs). Another source of the bias arises from our genetic and developmental model systems, limited to a few chosen for ease of laboratory use (Bolker 1995; Jenner & Wills 2007). Essentially all genetic

model systems are direct developers, where the adult body plan of the phylum is generated progressively in development, even if some form of metamorphosis occurs. This is true of *Caenorhabditis elegans*, a nematode, *Drosophila melanogaster*, an arthropod, or the zebra fish, frog or mouse—all vertebrates. The evolution of adult bias does not mean that early development is ignored, but that it is largely the study of early development leading to adult characters.

2. MOST PHYLA HAVE A SECOND BODY PLAN

Not only has our focus been on origins of adult body plans, but also on vertebrates and arthropods that receive most attention in studies of the evolution of the Bilateria. Both have been highly successful in terrestrial as well as marine environments. They are primitively direct developers. The result of focus on these phyla is that the second largest episode of metazoan body plan evolution, that of larvae, has been less appreciated. The majority of the 35 or so bilaterian phyla are primarily or exclusively marine and exhibit indirect development in which a larval form with a body plan distinct from that of the adult is present (figure 1). A radical metamorphic event finally releases the adult form at the end of larval development. These phyla thus have a distinct second life-history stage—that of their larval forms. The larvae differ greatly from the adults in ecology, and generally are planktonic filter feeders, whereas their adults are benthic and often effectively sessile. Such larvae

* Address for correspondence: Department of Biology, Indiana University, Bloomington, IN 47405, USA (raffr@indiana.edu).

One contribution of 17 to a Discussion Meeting Issue 'Evolution of the animals: a Linnean tercentenary celebration'.

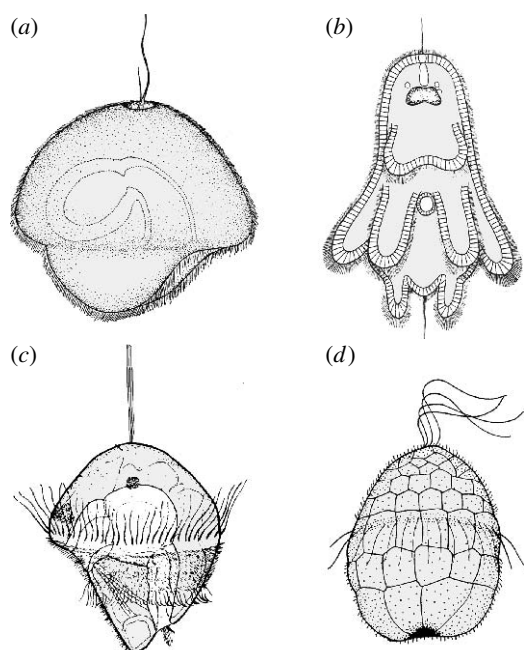


Figure 1. Examples of larval forms (four lophotrochozoans): (a) planktotrophic pilidium larva of a nemertine worm; (b) planktotrophic Müller's larva of a platyhelminth flatworm; (c) planktotrophic trochophore of a polychaete annelid; and (d) non-feeding trochophore-like larva of a basal mollusc. All oriented with the apical tuft up. Ciliary bands of various types are present on all. Guts are diagrammed for larvae as shown in (a,c). Adapted with permission from Rouse (2000). © Blackwell Science. Figures courtesy of G. Rouse.

historically are called 'primary larvae', on the basis of the historical idea that larval forms represent the primitive body plans of ancestral metazoans.

3. ADULT OR LARVAL BODY PLANS FIRST?

How did two distinct kinds of body plans evolve? The classical view, which derives from Haeckel's recapitulation theory, is that the first metazoans were similar to living larvae. Jägersten (1972) summarized it in this way: '... the two phases of the life cycle arose when the adult of the primeval ancestor of the metazoans, *viz.*, the holopelagic, radially symmetrical *Blastaea*, descended to life on the bottom (and became bilateral), while its juvenile stage remained in the pelagic zone'. Nielsen & Norrevang (1985) and Nielsen (1995) in the same vein suggested that a pelagic *Gastraea* animal evolved into a pelagic *Trochaea* animal (i.e. an adult holopelagic resembling a particular type of living feeding larva), which was ancestral to protostome and deuterostome phyla. This hypothesis was incorporated in the developmental genetic era to mesh with inferences about gene regulatory systems (Davidson *et al.* 1995). Gene regulatory systems of ancestral planktonic animals were hypothesized to resemble those found in living marine larvae (figure 2). Bilaterian adults were suggested to have evolved through the innovation of imaginal 'set aside' cells distinct from the majority of differentiated larval cells. The imaginal cells gave rise to tissues of a new adult stage and metamorphosis evolved to complete the transition. The new adults evolved a gene regulatory system similar to those of living adult bilaterians, including

novel use of Hox genes to pattern the anterior–posterior body axis. This scheme explains the lack of an early metazoan body or trace fossil record as all evolution took place in tiny planktonic adults. It ties larval forms into a phylogenetic scheme in which larval forms provide accessible proxies for the unfossilizable ancestors, and gives a developmental twist to the Cambrian radiation—the first fossil animals resulted from the appearance of new body plans. There are difficulties for this inter-linked suite of hypotheses (Sly *et al.* 2003; Peterson 2005; Peterson *et al.* 2005). Notably, the larva-first hypothesis requires a vast number of convergent events, accounting for the massive molecular convergences in the use of Hox and other regulatory genes in independently evolved descendent clades with benthic body plans. Furthermore, somehow a selective role for set-aside cells has to be accounted for before a new bilateral and benthic adult stage has evolved, which requires selection for novel developmental elements prior to need.

The planktonic metazoan ancestor has little evidence supporting it beyond analogies between the ontogeny of living larval forms and evolution of hypothetical ancestors. There is a second evolutionary possibility, that the first bilaterians were just that, small benthic bilaterally symmetric triploblastic animals similar to living acoel flatworms (figure 2). Molecular phylogenetic studies indicate that acoels are the most basal living bilaterians (Ruiz-Trillo *et al.* 2004; Sempere *et al.* 2007). Acoels are direct developers and possess anterior, middle and posterior group Hox genes (Ramachandra *et al.* 2002; Baganà & Riutort 2004). The last common ancestor of protostomes + deuterostomes (PD ancestor) probably was somewhat more complex than acoels, and possessed the genetic machinery basic to eye development, nephridia, heart and other mesodermal tissues (Erwin & Davidson 2002; Erwin 2006). This does not mean that these structures were present in as derived states as in living protostomes or deuterostomes. It means that acquisition of bilaterian features was stepwise, with some features attained between the split from cnidarians to the acoelomorph grade, and further acquisitions from there to the PD ancestor. Further evolution of features characterizing the stem groups of phyla would have represented a third stage in evolution of features (Baganà & Riutort 2004).

Proposal of an ancestral benthic bilaterian ancestor requires a hypothesis for the secondary evolution of the indirect-developing planktonic larvae, in place of the ancestral larval hypothesis. This inference has led to the intercalation model of larval origins (Valentine & Collins 2000; Sly *et al.* 2003). In this hypothesis, ancestral bilaterians are hypothesized to be small worm-like creatures, perhaps part of an acoelomorph radiation. These ancestral bilaterians were direct developers and had evolved the basic developmental gene regulatory systems of bilaterian development. With the opening of the Cambrian radiation, the evolution of more divergent bilaterians accelerated and produced the basal clades that gave rise to modern phyla and their precursors (Budd & Jensen 2000), but planktonic larvae and their body plans evolved secondarily.

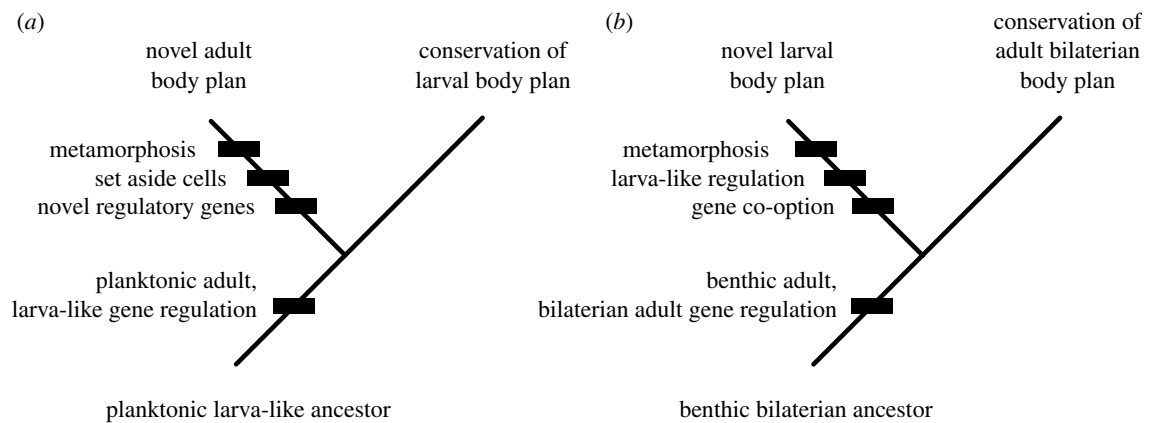


Figure 2. Conflicting larva-first and adult-first hypotheses of bilaterian origins. The hypotheses posit amounts of evolutionary change along branches leading to more derived developmental changes. (a) In the larva-first hypothesis, most evolution of developmental characters lies on the branch leading to the benthic adult, with the larva retaining ancestral features. (b) In the benthic adult-first hypothesis, most evolution lies in the line to the planktonic larva, with the adult retaining ancestral features. Both hypotheses illustrate single lineages, but in the metazoan radiation, numerous lineages evolved in parallel. A large degree of homoplasy resulted in either case. The amount of convergence required to evolve planktonic larvae with their relatively simple organization is substantially less than that to evolve the entire basic suite of adult bilaterian features in 35 or so lineages.

The requirements for a planktonic larva are simpler than for the larger benthic reproductive adult. Table 1 separates the characters of the benthic PD ancestor from those selected for in the evolution of a planktonic larva. Larvae require ciliary bands for swimming and capture of microscopic prey. A mouth and gut are needed to process prey. Simple neural systems allow some control of muscle cell contraction (e.g. in the pharynx). Other sensory information allows avoidance responses and ultimately detection of signals from the substrate biofilm to induce metamorphosis. For development of a coherent larval symmetry, systems for the determination of the larval axes (animal–vegetal, dorsoventral and left–right) are needed. In order to switch from larval to adult development, a developmental switch system that controls cellular fates has to be assembled from existing signalling systems in more primitive metazoans (Matus *et al.* 2006). Finally, a system for metamorphosis evolves, which probably initially involves transformation of most larval cells and tissue into adult tissues. However, slow metamorphosis increases vulnerability, and selection should favour evolution of a more rapid and efficient system using imaginal cells set aside as adult precursors within the larva to assure rapid metamorphosis.

Sly *et al.* (2003) predicted that some portion of genes required for adult development and life history would have been co-opted to direct the acquisition of a set of features involved in the simpler larval ontogeny required to produce a new life-history stage of an indirect-developing feeding larva. The acquisition of features would have involved stepwise intercalation of genes already used in the adult to generate features of the larva. The most basic requirement for feeding structures was probably met by the use of some of the adult gut programme. We have found evidence to support this idea in the common expression of genes in the sea urchin pluteus larval gut and in the adult gut (Love *et al.* 2008). Other features (e.g. the apical plate with its ciliary tuft) have co-opted unrelated sets of regulatory genes in sea urchin versus mollusc larvae (Dunn *et al.* 2007). Larval evolution would have been a build-up of features that would have diverted the

Table 1. Characters required to evolve a planktonic feeding larva from a benthic bilaterian.

characters required in larvae	adult characters not required in larvae
ciliary bands	locomotory appendages
gut	respiratory system
mouth	reproductive organs
simple neural/sensory system	brain
axial determination	strongly expressed A–P axes
developmental switch to adult	nephridia
feature ontogeny	eyes
metamorphosis	circulatory system
	skeleton

ancestral course of development into two temporally distinct streams, one that first produced a feeding larva and a second stream that, from larval tissue, developed the juvenile adult. Imaginal cells and a discrete metamorphosis would have more sharply separated the two ontogenetic trajectories.

The second consequence of the intercalation hypothesis is that different metazoan lineages would simultaneously have evolved planktonic larvae. Convergence would have been highly prevalent as the rise of feeding larvae followed in time the splitting of metazoan phyla or their precursor lineages. These evolving lineages would have evolved planktonic larvae with features noted in table 1. These features were gained by co-option of different suites of regulatory genes to accomplish the control of development of broadly similar larval morphological structures. Nonetheless, the convergence required would have been far less profound than that needed to evolve independently many lineages of bilaterians with the more complex PD ancestor features (table 1).

4. METAZOAN PHYLOGENY AND LARVAL ORIGINS

The two hypotheses have distinct phylogenetic consequences with respect to mapping of developmental features onto evolutionary history. The larva-like first

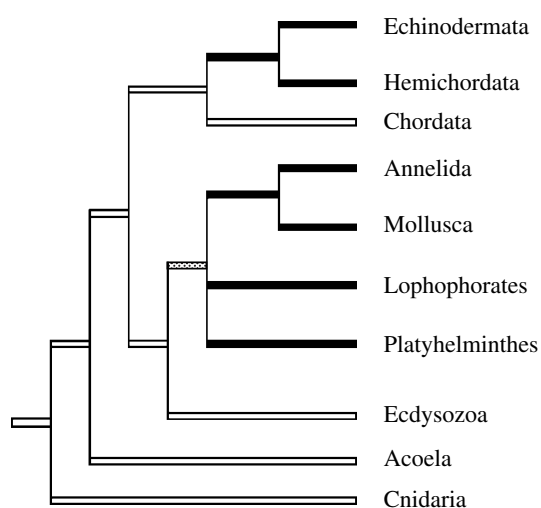


Figure 3. Developmental modes plotted on a bilaterian phylogenetic tree (open bars, direct development; stippled bar, ambiguous developmental mode; filled bars, planktotrophic indirect development; after Jenner (2000) and Peterson *et al.* (2005)).

scheme is difficult to reconcile with recent phylogenies of bilaterian metazoan clades. First, molecular phylogenetic analyses do not support a metazoan phylogeny in which basal clades are indirect developers. This was specifically pointed out by Jenner (2000) who noted that the strongest data allowing a decision on primitive developmental mode would come from phylogenetic studies, in which a wide range of 'minor' non-coelomate phyla were included. He tested the occurrence of indirect versus direct modes of development using a phylogenetic tree on which minor as well as major phyla were mapped. Figure 3 shows an analogous tree. Direct development is primitive in bilaterians and indirect-developing planktonic larvae have arisen independently in lophotrochozoans among the protostomes, and in the echinoderm plus hemichordate clade of deuterostomes. The other deuterostome clade, the chordates, is direct developing. The echinoderms and hemichordates share a planktonic larval form, but the highly diverse lophotrochozoan clades (molluscs, annelids, brachiopods, bryozoans, nemertines and platyhelminthes) have diverse larvae indicating a more complex history of multiple planktonic larval origins in the lophotrochozoan clade (Rouse 2000; Peterson 2005). Other protostome clades, notably the ecdysozoans (which includes arthropods and nematodes and others), are direct developing. Finally, the basal acoels and other minor clades (not shown) are direct developers. The mapping of the presence of planktonic larvae supports direct development as primitive in bilaterians, with separate origins of planktonic larvae in the echinoderm plus hemichordate clade and in the lophotrochozoans.

5. EVIDENCE FROM GENE EXPRESSION PATTERNS

One potentially strong discriminator for homologous features is patterns of expression of developmental regulatory genes. This approach has had mixed success, because there has been extensive co-option of genes in evolution. There have been a small number of

comparisons of gene expression patterns comparing protostome trochophore larvae (annelids and molluscs) with deuterostome dipleurula larvae (echinoderms plus hemichordates) to test for possible homologies (table 2). Some genes show similar expression patterns whereas others do not. The collection of genes is small and the sampling incomplete. The case of *nodal* illustrates the uncertainties. *Nodal* is involved in left-right determination in echinoderms and vertebrates. However, it operates in a different domain (right side in echinoderms, left side in chordates), and interpretations of axial homologies are not yet possible (Duboc & Lepage 2006). *Nodal* appears to have no role in *Drosophila*, an ecdysozoan, and it has not been reported from lophotrochozoans. The tabulation of gene patterns from the small sample shown in table 2 indicates some similarities in larval gene expression. That could arise from a case of homology between the trochophore and the dipleurula but, taken with the phylogenetic considerations, it appears more likely to represent a convergence in evolution of larval features accompanied by some convergence in gene regulation. Convergence is probable because the structure of larvae is simpler than the structure of adult bilaterians, and because co-option of genes may have been related to shared adult and larval functions. Thus, the patterns of expression of *Brachyury*, *Gsc* and *Otx* might represent co-option of adult oral developmental gene expression into development of similar larval oral structures: a sort of serial homology. Fully defining phylogenies and comparative gene expression will be advanced by genomic data. Most genome sequencing has concentrated on model or medically significant vertebrates, arthropods and nematodes. A sea urchin genome has now been sequenced (Sodergren *et al.* 2006), but the genomes of lophotrochozoans, especially marine annelids and molluscs with planktotrophic larvae, are still needed.

6. HUNTING THE LARVAL REVOLUTION IN THE FOSSIL RECORD

We have good fossil time markers for the visible appearance of diverse complex bilaterians in the fossil record—Early to Mid-Cambrian, 544–505 Myr ago. The origin of bilaterians lies in the Late Precambrian. Recent estimates suggest somewhere between 580 and 600 Myr ago (Peterson *et al.* 2005). An estimate of the timing for evolution of planktonic larvae of approximately 500 Myr ago is emerging, which if correct puts the origin of these second body plans 100 Myr later than the divergence of the basal bilaterian benthic adult. Signor & Vermeij (1994) noted that the Cambrian fossil record showed relatively few benthic suspension feeders or planktonic forms. They suggested that the evolution of planktonic feeding larvae took place in the Late Cambrian to Early Ordovician, driven by an expansion of plankton and sanctuary from predation—a point reinforced by Peterson (2005). Direct fossil evidence for larval evolution comes from exquisite phosphoritic preservation of Late Proterozoic and Cambrian cleavage-stage embryos of unknown taxa, larval forms of cnidarians and small ecdysozoans (Donoghue *et al.* 2006). An understanding of how embryos can be

Table 2. Homology and homoplasy in protostome (trochophore) and deuterostome (dipleurula) indirect-developing larval gene expression.

gene	trochophore	dipleurula	reference
<i>Brachyury</i>	foregut	foregut	Arendt <i>et al.</i> (2001)
<i>Gsc</i>	foregut	foregut	Arendt <i>et al.</i> (2001)
<i>Otx</i>	oral ciliary bands	oral ciliary bands	Arendt <i>et al.</i> (2001)
<i>NK2.1</i>	not apical plate	apical plate	Dunn <i>et al.</i> (2007)
<i>HNF6</i>	not apical plate	apical plate	Dunn <i>et al.</i> (2007)
<i>NK2.1</i>	foregut	foregut	Dunn <i>et al.</i> (2007)
<i>FoxA</i>	foregut	foregut	Dunn <i>et al.</i> (2007)
<i>Nodal</i>	?	right ecto+coelom	Duboc & Lepage (2006)
<i>Hox 2–5</i>	early larval	adult rudiment	Arenas-Mena <i>et al.</i> (1998), Kulakova <i>et al.</i> (2007)
<i>Hox 1</i>	left–right	aboral (dorsal)	Ishii <i>et al.</i> (1999), Kulakova <i>et al.</i> (2007)
<i>Hox 7</i>	post-gut	oral (ventral)	Ishii <i>et al.</i> (1999), Kulakova <i>et al.</i> (2007)

preserved for mineralization is emerging (Briggs 2003; Raff *et al.* 2006). The early fossil embryos so far described are large, ranging from 350 to 1100 μm (Xiao & Knoll 2000) for Late Precambrian embryos to 350–750 μm for Early–Mid-Cambrian embryos (Steiner *et al.* 2004; Donoghue *et al.* 2006). There are biases in the record, notably low taxonomic diversity (Donoghue *et al.* 2006). The possibility that small embryos typical of indirect-developing marine animals exist in the 50–200 μm size range has been checked by Donoghue *et al.* (2006), but not found. This observation of course does not prove that small embryos were not in existence. Fossil embryo evidence for the appearance of indirect-developing forms is still scarce. Nützel *et al.* (2006) have observed that Cambrian larval mollusc shells are larger than those of the Ordovician and Silurian, consistent with a shift from direct to indirect development.

7. FOSSILS, LARVAE AND LINNAEUS

Linnaeus propounded a systematic approach that created a static hierarchical system of classification, which has lent itself to evolutionary interpretation. Larval and adult characters have produced homoplasies that yield some contradictory phylogenetic inferences among some of the deepest Linnean taxa. Thus, the trochophore larvae of annelids and molluscs carry a different phylogenetic signal than their adult body plan features. Rather than seeing these characters as conflicting, a better knowledge of the Cambrian fossil record of clades basal to living phyla allows us to dissect more finely the timing of both adult and larval body plan evolution. Halwaxiids and their kin are sclerite-bearing middle Cambrian animals that lie somewhere basal in a clade that includes molluscs, annelids and brachiopods (Conway Morris & Caron 2007).

The characters of larval forms show some linkages between phyla obscured by changes in adult morphology, and in fact agree with phylogenetic inferences based on gene sequence data. Thus, the trochophore shared by annelids and molluscs belies segmentation and paired appendages shared by annelids and arthropods, the so-called articulata. The existence of these forms suggests that the primitive trochophore larva may have its origin in a Cambrian clade living before the split of the lophotrochozoan phyla. This would move the time of larval origin to lie earlier in the Cambrian. This might suggest that the earliest

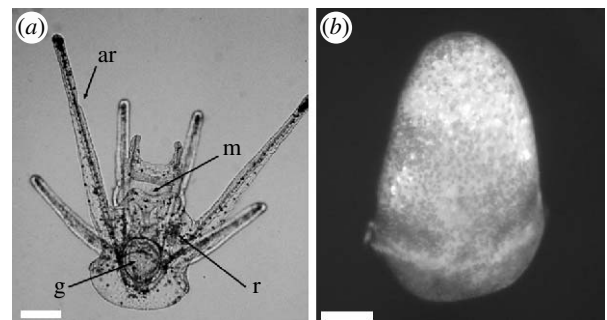


Figure 4. Rapid evolution of larvae shown by two congeneric sea urchins, 4 Myr diverged. (a) Planktotrophic pluteus larva of the indirect developer *Heliocidaris tuberculata*. The notable features are: the arms (ar), each supported by a skeletal rod and bearing a ciliary band; the large gut (g); the mouth (m); and the developing adult rudiment (r) that will grow to become the juvenile sea urchin released at metamorphosis (approx. six weeks post-fertilization). (b) Non-feeding direct-developing larva of *Heliocidaris erythrogramma*. All internal features are those of the developing adult. Metamorphosis is 3–4 days post-fertilization (scale bar, 100 μm).

planktotrophic larvae have not yet been detected, or that the full suite of planktonic feeding features were acquired slowly, and included convergences among related lineages (Rouse 2000). Similarly, the dipleurula larva links the pentamerous echinoderms with the bilaterian worm-like hemichordates, indicating that the origins of this larval form occurred after the split of this clade from chordates. Basal chordates and echinoderms are present in Mid-Cambrian strata.

8. CONTINUING GENE CO-OPTION IN LARVAL EVOLUTION

Larvae did not cease evolving in the Cambrian with the establishment of the basal diversity of feeding larval body plans. First, novel features evolved in planktonic larvae after the initial evolution of a larval stage. This kind of evolution has been inferred by Rouse (2000) for downstream feeding in trochophore-like larvae by analysis of the distribution of features in a phylogeny of lophotrochozoan clades. Among deuterostomes, we have analysed the arms of the sea urchin pluteus larva (figure 4). This is an indirect-developing feeding planktonic larva derived from the basal dipleurula-type larva of echinoderms. The echinopluteus has, since the split of sea urchins from other crown echinoderm

classes of approximately 450 Myr ago, evolved long arms that contain a novel rigid calcium-carbonate skeleton and bear a circumoral ciliary band (Bottjer *et al.* 2006). These arms evolved somewhere between the Late Ordovician and the Permian, and thus followed the initial evolution of the dipleurula. Love *et al.* (2007) have found that the pluteus arm is a novel larval organ. The arms consist of an ectoderm bearing a ciliary band and an underlying mesoderm consisting of skeletogenic mesenchyme cells. Expression of particular genes occurs in the growing arm tips (e.g. tetraspanin in ectoderm and advillin and carbonic anhydrase in mesenchyme). These genes are also expressed in various adult tissues. Their role in larval arms indicates that they have been recruited for expression in these structures following the origin of the dipleurula. This recruitment serves as an accessible proxy for the more remote events of the Cambrian.

A second type of larval evolution is that of the various non-planktotrophic derivatives of larvae in various clades (e.g. snails, Collin 2004; starfish and sea urchins, Raff & Byrne 2006). In many taxa, planktonic feeding larvae have given rise to non-feeding direct-developing planktonic or brooded larvae, and even viviparous larvae. These modified larvae rapidly evolve distinct morphologies, as seen in the larvae of the congeneric sea urchins *Heliocidaris tuberculata* and *Heliocidaris erythrogramma* (figure 4), which diverged approximately 4 Myr ago (Zigler *et al.* 2003). *Heliocidaris tuberculata* takes approximately six weeks of feeding in the water column to reach metamorphosis. *Heliocidaris erythrogramma* takes 3 days, and does not feed. The *H. erythrogramma* egg is 100 times the volume of that of indirect-developing sea urchins and supports development through post-metamorphic development of the adult mouth. At first glance, it would appear that *H. erythrogramma* is simplified by loss of larval features, but retains adult ontogeny. Some feeding structures, such as the larval arms and gut, are lost; nonetheless, developmental features retain a high degree of complexity and dramatic novel features have appeared. These include changes in oogenesis and spermatogenesis, maternal embryonic axis determination, cleavage pattern, cell embryonic lineages, and heterochronies in larval gene expression and morphogenetic events (Raff & Byrne 2006). The details, described elsewhere, show that rapid and profound evolutionary changes in larval development occur. In fact, they occur frequently with, for example, several clades of sea urchins independently having evolved larvae similar to that of *H. erythrogramma* (Sly *et al.* 2003). The evolutionary lability of larvae suggests that evolution of primary larval features would have been rapid in the face of selection under the new ecological regime of the Late Cambrian and Early Ordovician. It is also likely that the developmental regulatory features of living larval clades give us strong clues to those of early larval forms.

9. DEVELOPMENTAL INNOVATIONS AND THE METAZOAN RADIATION

The origin of the ancestral benthic bilaterian body plan was an immense evolutionary-developmental innovation

that produced a shift from the cnidarian frond-dominated world of the Late Proterozoic to the diversified bilaterian-dominated world of the Cambrian. However, the evolution of novel developmental features depends both on the appearance of variation in development and selection on developmental stages and processes. Developmental features of early metazoans may have been less constrained by a looseness of ecological fit that allowed more experimentation with body plans, i.e. adaptive peaks were present, but in a fairly flat landscape where few deep valleys of low fitness were yet present. The rapid diversification of basal taxa related to living phyla was probably the result of ecological pressures and opportunities that selected for development of novel morphologies among bilaterians of relatively simple morphology. Acoelomorph bilaterian ancestors would have possessed a large suite of developmental regulatory genes that could be recruited for evolution of new structures. The possibilities for body plan innovation in acoelomorph grade animals would have in many respects been easier than for proposed schemes that suggest divergence from more derived ancestors. Thus, the dorsal–ventral inversion of organs of protostomes and deuterostomes would have been of little consequence at the acoelomorph grade of organization, but could have become a fixed element of body plan later. Segmentation, another feature of importance, also may be a product of convergence in emerging lineages (Seaver 2003).

The evolution of planktonic larvae followed the origins of basal bilaterian phyla by approximately 100 Myr. Again, it is less likely that developmental novelties *per se* drove this evolutionary innovation. Instead, larvae bearing features arising from novel expression of genes used in adults were selected upon as agents of exploitation of greater ranges of ecological possibility for increasing planktonic food resources, escape from benthic filter-feeding predators and a vastly improved dispersal than that offered by large direct-developing embryos. The evolutionary flexibility of larval development allowed diverse and rapid responses to selection. Selection on expression of existing genes in new contexts may underlie much of the evolution of novelties in development.

REFERENCES

- Arenas-Mena, C., Martinez, P., Cameron, R. A. & Davidson, E. H. 1998 Expression of the Hox gene cluster in the indirect development of a sea urchin. *Proc. Natl Acad. Sci. USA* **95**, 13 062–13 067. (doi:10.1073/pnas.95.22.13062)
- Arendt, D., Technau, U. & Wittbrodt, J. 2001 Evolution of the bilaterian larval foregut. *Nature* **409**, 81–85. (doi:10.1038/35051075)
- Baguñà, J. & Riutort, M. 2004 The dawn of bilaterian animals: the case of acoelomorph flatworms. *BioEssays* **26**, 1046–1057. (doi:10.1002/bies.20113)
- Bolker, J. A. 1995 Model systems in developmental biology. *BioEssays* **17**, 451–455. (doi:10.1002/bies.950170513)
- Bottjer, D. J., Davidson, E. H., Peterson, K. J. & Cameron, R. A. 2006 Paleogenomics of echinoderms. *Science* **314**, 956–959. (doi:10.1126/science.1132310)
- Briggs, D. E. G. 2003 The role of decay and mineralization in the preservation of soft-bodied fossils. *Annu. Rev. Earth Planet. Sci.* **31**, 275–301. (doi:10.1146/annurev.earth.31.100901.144746)

- Budd, G. E. & Jensen, S. 2000 A critical reappraisal of the fossil record of the bilaterian phyla. *Biol. Rev.* **75**, 253–295. (doi:10.1017/S000632310000548X)
- Cohn, M. J. & Tickle, C. 1999 Developmental basis of limblessness and axial patterning in snakes. *Nature* **399**, 474–479. (doi:10.1038/20944)
- Collin, R. 2004 Phylogenetic effects, the loss of complex characters, and the evolution of development in calyptraeid gastropods. *Evolution* **58**, 1488–1502.
- Conway Morris, S. & Caron, J.-B. 2007 Halwaxiids and the early evolution of the lophotrochozoans. *Science* **315**, 1255–1258. (doi:10.1126/science.1137187)
- Davidson, E. H., Peterson, K. J. & Cameron, R. A. 1995 Origin of adult bilaterian body plans: evolution of developmental regulatory mechanisms. *Science* **270**, 1319–1325. (doi:10.1126/science.270.5240.1319)
- Donoghue, P. C. J., Kouchinsky, A., Waloszek, D., Bengtson, S., Dong, X., Val'kov, A. K., Cunningham, J. A. & Repetski, J. E. 2006 Fossilized embryos are widespread but the record is temporally and taxonomically biased. *Evol. Dev.* **8**, 232–238. (doi:10.1111/j.1525-142X.2006.00093.x)
- Duboc, V. & Lepage, T. 2006 A conserved role for the nodal signaling pathway in the establishment of dorso-ventral and left-right axes in deuterostomes. *J. Exp. Zool. B: Mol. Dev. Evol.* **306**, 1–13.
- Dunn, E. F., Moy, V. N., Angerer, L. M., Angerer, R. C., Morris, R. L. & Peterson, K. J. 2007 Molecular paleoecology: using gene regulatory analysis to address the origins of complex life cycles in the Late Precambrian. *Evol. Dev.* **9**, 10–24.
- Erwin, D. H. 2006 The developmental origins of animal bodyplans. In *Neoproterozoic geobiology and paleobiology* (eds S. H. Xiao & A. J. Kaufman), pp. 159–197. New York, NY: Plenum.
- Erwin, D. H. & Davidson, E. H. 2002 The last common bilaterian ancestor. *Development* **129**, 3021–3032.
- Gilbert, S. F., Loredó, G. A., Bruckman, A. & Burke, A. C. 2001 Morphogenesis of the turtle shell: the development of a novel structure in tetrapod evolution. *Evol. Dev.* **3**, 47–58. (doi:10.1046/j.1525-142x.2001.003002047.x)
- Ishii, M., Mitsunaga-Nakatsubo, K., Kitajima, T., Kusunoki, S., Shimada, H. & Akasaka, K. 1999 *Hbox1* and *Hbox7* are involved in pattern formation in sea urchin embryos. *Dev. Growth Differ.* **41**, 241–252. (doi:10.1046/j.1440-169X.1999.413426.x)
- Jägersten, G. 1972 *Evolution of the metazoan life cycle*. London, UK: Academic Press.
- Jenner, R. A. 2000 Evolution of animal body plans: the role of metazoan phylogeny at the interface between pattern and process. *Evol. Dev.* **2**, 208–221. (doi:10.1046/j.1525-142x.2000.00060.x)
- Jenner, R. A. & Wills, M. A. 2007 The choice of model organisms in evo-devo. *Nat. Rev. Genet.* **8**, 311–319. (doi:10.1038/nrg2062)
- Kulakova, M. *et al.* 2007 Hox gene expression in larval development of the polychaetes *Nereis virens* and *Platynereis dumerilii* (Annelida, Lophotrochozoa). *Dev. Genes Evol.* **217**, 39–54. (doi:10.1007/s00427-006-0119-y)
- Love, A., Andrews, M. & Raff, R. A. 2007 Pluteus larval arm morphogenesis and evolution: gene expression patterns in a novel animal appendage and their transformation in the origin of direct development. *Evol. Dev.* **9**, 51–68.
- Love, A. C., Lee, A. E., Andrews, M. E. & Raff, R. A. 2008 Co-option and dissociation in larval origins and evolution: the sea urchin larval gut. *Evol. Dev.* **10**, 74–88.
- Matus, D. Q., Pang, K., Marlow, H., Dunn, C. W., Thomsen, G. H. & Martindale, M. Q. 2006 Molecular evidence for deep evolutionary roots of bilaterality in animal development. *Proc. Natl Acad. Sci. USA* **103**, 11 195–11 200. (doi:10.1073/pnas.0601257103)
- Nielsen, C. 1995 *Animal evolution. Interrelationships of the living phyla*. Oxford, UK: Oxford University Press.
- Nielsen, C. & Norrevang, A. 1985 The trochaea theory: an example of life cycle phylogeny. In *The origins and relationships of lower invertebrates* (eds S. Conway-Morris, J. D. George, R. Gibson & H. M. Platt), pp. 297–309. Oxford, UK: Clarendon Press.
- Nützel, A., Lehnert, O. & Fryda, J. 2006 Origin of planktotrophy—evidence from early mollusks. *Evol. Dev.* **8**, 325–330. (doi:10.1111/j.1525-142X.2006.00105.x)
- Peterson, K. J. 2005 Macroevolutionary interplay between planktonic larvae and benthic predators. *Geology* **33**, 929–932. (doi:10.1130/G21697.1)
- Peterson, K. J., McPeck, M. A. & Evans, D. A. 2005 Tempo and mode of early animal evolution: inferences from rocks, Hox, and molecular clocks. *Paleobiology* **31**(Suppl.), 36–55. (doi:10.1666/0094-8373(2005)031[0036:TAM OEA]2.0.CO;2)
- Raff, R. A. & Byrne, M. 2006 The active evolutionary lives of echinoderm larvae. *Heredity* **97**, 244–252. (doi:10.1038/sj.hdy.6800866)
- Raff, E. C., Villinski, J. A., Turner, F. R., Donoghue, P. C. & Raff, R. A. 2006 Experimental taphonomy: feasibility of fossil embryos. *Proc. Natl Acad. Sci. USA* **103**, 5846–5851. (doi:10.1073/pnas.0601536103)
- Ramachandra, N. B., Gates, R. D., Ladurner, P., Jacobs, D. K. & Hartenstein, V. 2002 Embryonic development in the primitive bilaterian *Neochildia fusca*: normal morphogenesis and isolation of POU genes *Brn-1* and *Brn-3*. *Dev. Genes Evol.* **212**, 55–69. (doi:10.1007/s00427-001-0207-y)
- Rouse, G. W. 2000 The epitome of hand waving? Larval feeding and the hypotheses of metazoan phylogeny. *Evol. Dev.* **2**, 222–233. (doi:10.1046/j.1525-142x.2000.00063.x)
- Ruiz-Trillo, I., Riutort, M., Fourcade, H. M., Bagaña, J. & Boore, J. L. 2004 Mitochondrial genome data support the basal position of Acoelomorpha and the polyphyly of the Platyhelminthes. *Mol. Phylogenet. Evol.* **33**, 321–332. (doi:10.1016/j.ympev.2004.06.002)
- Seaver, E. C. 2003 Segmentation: mono- or polyphyletic? *Int. J. Dev. Biol.* **47**, 583–595.
- Sempere, L. F., Martínez, P., Cole, C., Bagaña, J. & Peterson, K. J. 2007 Phylogenetic distribution of microRNAs supports the basal position of acoel flatworms and the polyphyly of Platyhelminthes. *Evol. Dev.* **9**, 409–415. (doi:10.1111/j.1525-142X.2007.00180.x)
- Signor, P. W. & Vermeij, G. J. 1994 The plankton and the benthos: origins and early history of an evolving relationship. *Paleobiology* **20**, 297–319.
- Sly, B. J., Snoke, M. S. & Raff, R. A. 2003 Who came first? Origins of bilaterian metazoan larvae. *Int. J. Dev. Biol.* **47**, 623–632.
- Sodergren, E. *et al.* 2006 The genome of the sea urchin *Strongylocentrotus purpuratus*. *Science* **314**, 941–952. [Sea Urchin Genome Sequencing Consortium] (doi:10.1126/science.1133609)
- Steiner, M., Zhu, M., Li, G., Qian, Y. & Erdtmann, B.-D. 2004 New Early Cambrian bilaterian embryos and larvae from China. *Geology* **32**, 833–836. (doi:10.1130/G20567.1)
- Valentine, J. W. & Collins, A. G. 2000 The significance of moulting in ecdysozoan evolution. *Evol. Dev.* **2**, 152–156. (doi:10.1046/j.1525-142x.2000.00043.x)
- Xiao, S. & Knoll, A. H. 2000 Phosphatized animal embryos from the Neoproterozoic Doushantuo formation at Weng'an, Guizhou, South China. *J. Paleontol.* **74**, 767–788. (doi:10.1666/0022-3360(2000)074<0767:PAEFTN>2.0.CO;2)
- Zigler, K. S., Raff, E. C., Popodi, E., Raff, R. A. & Lessios, H. E. 2003 Adaptive evolution of *bindin* in the genus *Helicoidaris* is correlated with the shift to direct development. *Evolution* **57**, 2293–2302.