

Dating the origin of metazoan body plans

Andrew B. Smith

Department of Palaeontology, The Natural History Museum, Cromwell Road, London SW7 5BD, UK
(email: a.smith@nhm.ac.uk)

INTRODUCTION

The appearance of the great majority of animal phyla over a short period in the early Cambrian is viewed either as the most spectacular evolutionary radiation in the history of life, or as a woeful example of the inadequacy of the fossil record, depending upon how the data are interpreted. On the one hand, the fossil record provides a pattern of stepwise increasing complexity, consistent across continents, and telescoped into a very narrow time span of no more than 20 million years (my) (Bowring and Erwin 1998). On the other hand, molecular dating estimates anything up to half the history of metazoan animals is missing from the fossil record, and that the major groups diverged well before the first appearance of body fossils (Fig. 1).

Conflict between palaeontological and molecular data is not confined to the origination of phylum-level body plans in the Cambrian, but is a problem that recurs at other times, notably with the origin of mammalian and avian orders in the Tertiary (e.g., Bromham et al. 1999). Faced with such mismatch there are just three explanations.

1. MOLECULAR CLOCK ESTIMATES ARE WRONG

Deriving accurate tree topology and branch length estimates from single genes is notoriously difficult (e.g., Maley and Marshall 1998; Abouheif et al. 1998). Paralogy, high levels of homoplasy, substitutional saturation, sequence alignment ambiguity, base compositional biases, and compensatory changes, as well as breadth of taxon sampling, all affect tree topology. Accurate estimation of branch lengths is particularly difficult, since hidden homoplasy tends to shift internal nodes toward the termini, making sister taxa appear more closely (and hence more recently) related than they really are. Are molecular clocks therefore so prone to error as to be invalid?

Initial attempts to date the origin of major metazoan clades using molecules suffered from statistical problems and a poor choice of genes (Ayala et al. 1998). Furthermore, the inability to resolve deep branches in the molecular phylogenies of Metazoa with any degree of statistical confidence was taken as evidence that the various phyla had branched

over a relatively short time (Philippe et al. 1994). However, it is now clear that the observed lack of resolution in rRNA based trees is most likely an artifact created by rate heterogeneity among sites and taxa (Abouheif et al. 1998).

Over the past 3 years, the range of genes and statistical techniques that have been used to date the origin of metazoan clades is impressive. Initial estimates based on a small number of genes (e.g., Wray et al. 1996) have been improved by turning to genes that demonstrate clock-like evolution in the Phanerozoic (Nikoh et al. 1997), or by using large numbers of taxa to minimize lineage specific biases (Bromham et al. 1998). Alternatively, others (e.g., Doolittle et al. 1996; Gu 1998; Wang et al. 1998) have used large numbers of gene sequences and kilobases of sequence data to smooth out stochastic variation in individual clock-rate estimates and thus derive more accurate estimates. Various statistical techniques have been used for calculating divergence times, a series of Phanerozoic branching points have been used for calibration, and a range of correction factors have been applied. With one possible exception (Ayala et al. 1998), all place metazoan divergences deep in the Neoproterozoic era (Fig. 1). Thus, as long as mutation rates have not changed significantly over geological time, it seems improbable that the molecular estimates are seriously in error.

But what if molecular evolution was much faster when body plans were evolving, and has since slowed down? Then molecular rates calculated from post-Cambrian divergence dates would be inappropriate for calibrating pre-Phanerozoic time.

There are certainly cases where molecular rates can be shown to have changed dramatically over geological time. In the initial diversification of dipteran insects, there was a 20-fold increase in the rate of rDNA nucleotide substitution (Freidrich and Tautz 1997). This, however, left a very recognizable change in base composition (dipteran rDNA became markedly AT enriched). No such signature is evident in metazoans: what small base compositional differences there are cut across accepted classifications and seem associated with postdivergence rate heterogeneity (Abouheif et al. 1998).

Several studies have identified a slowing of evolutionary change in amino acid substitution in higher vertebrates (Iwabe et al. 1996; Hoshiyama et al. 1998), with amniote

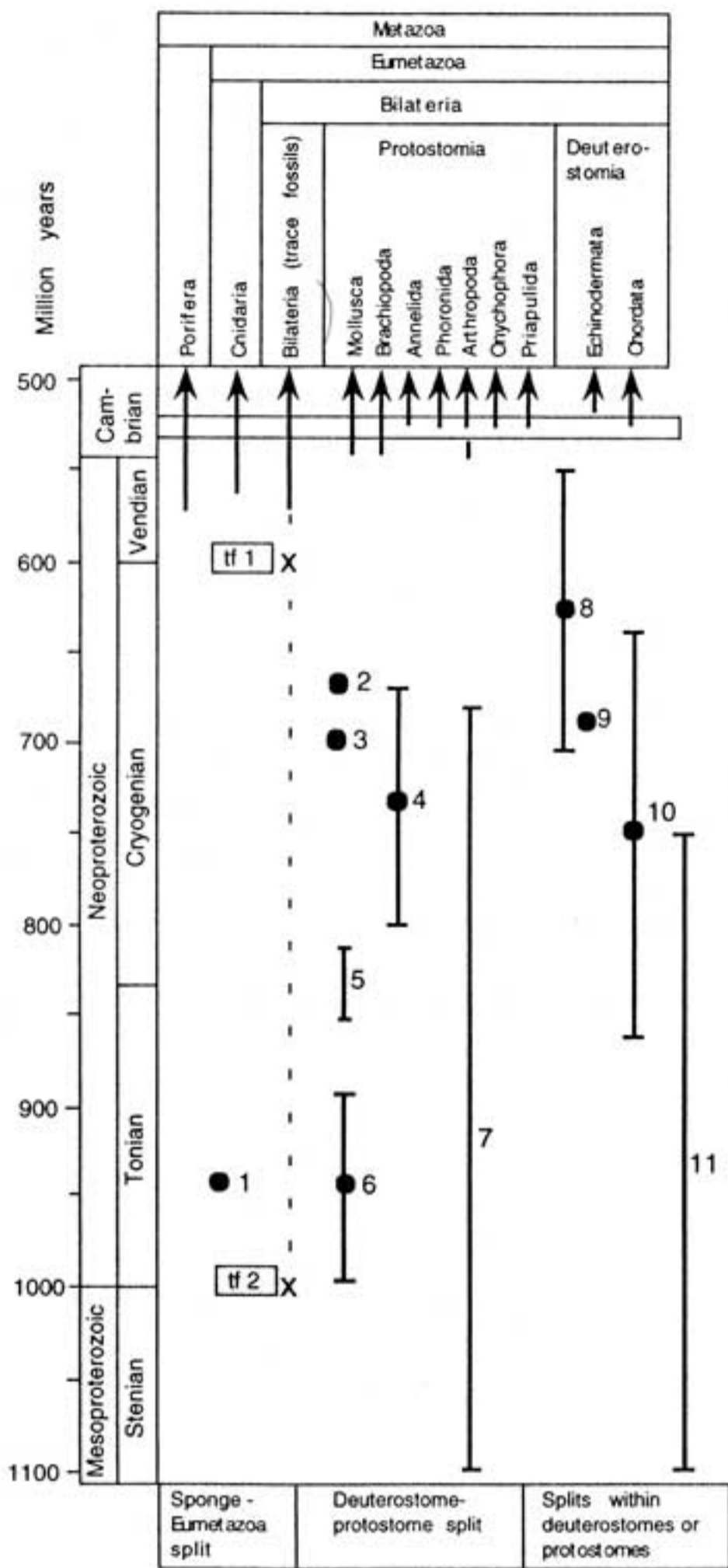


Fig. 1. Mismatch between the fossil record of metazoans and estimates of divergence times from molecular data. Solid lines, known fossil record of metazoans. Stippled band, Cambrian explosion (520–530 Ma). tf1, tf2, possible bilaterian trace fossils: tf1, Brasier and McIlroy (1998); tf2, Seilacher et al. (1998). 1–11, estimates of divergence times derived from molecular data. 1, porifera-eumetazoa split; Nikoh et al. (1997); 2–7, deuterostome-protostome split; 2, protein coding genes, Doolittle et al. (1996); 3, protein coding genes, Nikoh et al. (1997); 4, various, Ayala et al. (1998); 5, protein coding genes, Gu (1998); 6, protein coding genes, Wang et al. (1998); 7, mitochondrial protein coding genes, Bromham et al. (1998). 8–11, Divergences of phyla within either the deuterostomes or protostomes. 8, echinoderm-vertebrate, various, Ayala et al. (1998); 9, vertebrate-amphioxus, protein coding genes, Nikoh et al. (1997); 10, brachiopod-mollusc, 18S rRNA, Cohen et al. (1998). 11, echinoderm-vertebrate, mitochondrial protein coding genes, Bromham et al. (1998).

rates for *Pax* genes around one-third those of other bilateria and maybe one-tenth of those characterizing the internal branch leading to the sponge-eumetazoan split (although rate estimates for Precambrian internal branches are extremely sensitive to the calibration points used, and thus prone to massive error). If rates of genetic change have slowed over time across many gene families within amniotes, molecular clock estimates would place divergences erroneously deep in time when calibrated using dates for post-Cambrian amniote branching points. Fortunately, recent estimates for the timing of deuterostome-protostome divergence use statistical tests to remove, a priori, genes that display marked rate heterogeneity and so are immune to this problem.

Thus, although some gene families demonstrate marked rate heterogeneity, there is no compelling evidence that mutation rates in general were faster when metazoan body plans were evolving. A divergence of metazoan clades deep in the Neoproterozoic seems inescapable based on molecular data.

2. THERE ARE MAJOR GAPS IN THE FOSSIL RECORD

Knowledge of the late pre-Cambrian and Cambrian has improved tremendously over the last 10 years. Radiometric dating has advanced to a level where ages can be determined with an accuracy of ± 1 my (Bowring and Erwin 1998) and our time scale for the Vendian–Cambrian is now fairly robust. Extensive late pre-Cambrian outcrops have been scoured on all five continents for traces of life, and yet the pre-Cambrian record of metazoans remains pitifully small. Neoproterozoic sediments are just depressingly barren.

Prior to 600 million years ago (Ma), prokaryotic microbes and sheet-like or ribbon-like algae are all that are found, but from about 600 Ma, impressions of soft-bodied animals start to appear. This so-called Ediacaran fauna is now known from 30 localities on five continents but only became diverse in the last 20 my of the pre-Cambrian (Narbonne 1998). The fauna is composed of three groups of organisms: worm-like/bilaterian trace fossils, lower metazoan body fossils (mostly cnidarian-grade discs, with rare sponges and possible bilateria) and Vendobionta, quilted organisms of uncertain affinity. This fauna continues, at least in part, into the Cambrian (Jensen et al. 1998). In the Lower Cambrian the first shelly fossils (Mollusca and Brachiopoda) appear, and are quickly joined by Arthropoda. The late Lower Cambrian fossil deposits of Chengjiang, dated at around 520 Ma, provide the earliest window on soft-bodied metazoan biodiversity and show that many other phyla, including vertebrates, were present (Chen and Zhou 1997).

However, before accepting the fossil record at face value, we need to consider two possibilities. Were metazoans present but unmineralized and simply not preserved, or were

they in living in restricted habitats that have not been sampled/preserved in the fossil record?

The first possibility is easily dismissed. If metazoans were present but soft-bodied, then one would expect bilaterian-grade trace fossils to be more common than they are in the Neoproterozoic. In fact the fossil record of trace fossils is in remarkably good accord with the body fossil record. Ediacaran faunas (565–545 Ma) include small (1–3 mm) subsurface burrows that are widely accepted as the product of bilaterians. From the earliest Cambrian, the diversity and size of burrows increase significantly. Earlier records have been claimed but need confirmation (determining the biological affinities of a trace fossil maker is far from easy). Brasier and McIlroy (1998) reported 600 Ma traces attributable to coelomate or pseudocoelomate bilaterians, while Seilacher et al. (1998) recorded bilaterian traces from 1100 Ma rocks in India. Debate rages about these latter traces, with uncertainty about both the age of these beds (Azmi 1998; Kerr 1998b) and their bilaterian credentials (Kerr 1998a).

So, in rocks available for study, evidence strongly points to an absence of large-bodied bilaterians until the very end of the Neoproterozoic. But is our sample of Neoproterozoic rocks adequate in terms of the palaeohabitats it preserves? Productivity in Proterozoic oceans was likely extremely patchy, and the broad interior of shallow-water platforms widely developed in the late Proterozoic may well have been massively oligotrophic, with productivity so low as to be able to sustain only (?) autotrophic ediacaran-type organisms. The Cambrian radiation might then be a reflection of changing oceanographic chemistry and sharply increasing nitrification of shelf seas (Martin 1996; Bartley et al. 1998), allowing the spread of metazoan fauna from their original restricted habitats. Increasing nutrient levels may have triggered the widespread adoption of deposit feeding for the first time, creating the burst of trace fossil diversification.

This scenario is more difficult to disprove. A broad spectrum of Neoproterozoic habitats have been sampled for microfossils (e.g., Butterfield and Chandler 1992) without a hint of metazoans being discovered. However, available surface outcrop represents such a miniscule amount of the ancient sea floor that once existed in the Neoproterozoic. The crux is whether there is evidence for major taphonomic or sampling biases in the Neoproterozoic. If there is, this would certainly weaken the case that metazoans evolved rapidly in the Cambrian. Such evidence exists, at least for the latter part of the Neoproterozoic.

First, sponges existed in the late Neoproterozoic, but their remains are remarkably rare before 520 Ma. Rare triaxial spicules are recorded from 580 Ma beds (Xiao et al. 1998), but other sponge body fossils reported of this age (Li et al. 1998) turn out to be protists (Zhang et al. 1998). Otherwise the earliest records are from the latest Ediacaran of Mongolia, 545 Ma (Brasier et al. 1997) and possibly Australia (Gheling and

Rigby 1996). Clearly the late pre-Cambrian sponge record has major sampling gaps.

Second, early Neoproterozoic acritarch diversity has recently been shown to be very much greater than predicted, pointing to “a severe undersampling of contemporaneous fossil diversity” (Butterfield and Rainbird 1998) in 900–800 Ma rocks.

Third, if either of the pre-Ediacaran trace fossils identified as bilaterian is correct then there is a major sampling problem affecting bilaterian trace fossils in the late Proterozoic.

Fourth, trilobites occur more or less simultaneously on different continents in the mid-Lower Cambrian, but are already biogeographically and taxonomically diverse, implying a long hidden history (Fortey et al. 1996).

Fifth, the order in which small shelly fossils appear is highly variable because of strong environmental and/or taphonomic controls (Brasier et al. 1996).

Thus, although the mismatch between fossil and molecular data cannot simply be explained by the absence of mineralized skeletons in pre-Cambrian faunas, it is almost certainly exaggerated by sampling problems.

3. BOTH THE FOSSIL RECORD AND MOLECULAR CLOCK ESTIMATES ARE ACCURATE

A compromise view, first expounded by Davidson et al. (1995) and becoming widely accepted (e.g., Fortey et al. 1996; Conway Morris 1997; Valentine et al. 1999), holds that the Cambrian explosion is real although metazoan lineages diverged deep in time. According to Davidson et al.’s theory, pre-Cambrian metazoans were small larval-like forms whose mode of embryogenesis prevented them from growing beyond a few thousand cells. These evolved and diversified in the pre-Cambrian until some environmental trigger allowed a change in embryogenesis which led to the origination and proliferation of adult metazoan body forms across a variety of pre-existing larval-like clades. The development of genetic regulatory circuitry which creates set-aside cells with unlimited division capacity is seen as the key to allowing the development of large bodied forms (Cameron et al. 1998). The basic body patterning genes evolved to control metazoan “larval” morphology were sequestered for adult body patterning at that time. One suggestion is that atmospheric oxygen levels may have reached a critical threshold near the Cambrian–pre-Cambrian boundary, allowing metazoans to evolve into large-bodied forms (Conway Morris 1997).

Amazingly, although small and soft-bodied, simple larval-like bilateria, as predicted by Davidson et al. (1995), have indeed been recovered from 580 Ma phosphates (Xiao et al. 1998). Furthermore, this model explains why so many invertebrate phyla undergo indirect development. One major

stumbling block remains, however, and that is why "major deep-structural changes in the genetic control" (Cameron et al. 1998) should evolve synchronously and independently across 15 phylum-level lineages within 10 million years. Thus, although most phyla may have evolved initially as larva-like forms, the evidence for when adult body form arose relies on a literal reading of the fossil record. Neoproterozoic metazoa may have been small but were they necessarily exclusively larval-like? After all, trace fossils show that small burrowing bilaterals were present in the late Neoproterozoic, although no body fossils are preserved.

FINAL THOUGHTS

The consistency with which the succession of fossil organisms is repeated in Namibia, Siberia, Mongolia, and other key localities around the world is impressive and argues for the pattern being real. But whether it is recording an evolutionary event in the origin of body plans or an ecological event triggered by oceanographic changes is more debatable. Based on present evidence, the reality of an extended pre-Cambrian history for metazoan lineages seems undeniable, based on molecular data, and for part of this time the various clades may well have existed as small larval-like forms. Nevertheless, sampling artifact undoubtedly makes the Cambrian explosion of adult body plans appear more sudden than it really was, and large-bodied bilaterians probably evolved over a longer time interval than the 10 million year "Cambrian explosion."

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