

# The evolution of the Ecdysozoa

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Ecdysozoa is a clade composed of eight phyla: the arthropods, tardigrades and onychophorans that share segmentation and appendages and the nematodes, nematomorphs, priapulids, kinorhynchs and loriciferans, which are worms with an anterior proboscis or introvert. Ecdysozoa contains the vast majority of animal species and there is a great diversity of body plans among both living and fossil members. The monophyly of the clade has been called into question by some workers based on analyses of whole genome datasets. We review the evidence that now conclusively supports the unique origin of these phyla. Relationships within Ecdysozoa are also controversial and we discuss the molecular and morphological evidence for a number of monophyletic groups within this superphylum.

**Keywords:** Ecdysozoa; Arthropoda; Introverta; Protostomia; phylogeny; evolution

## 1. INTRODUCTION

The Ecdysozoa is a widely accepted clade that encompasses the Euarthropoda (Insecta, Crustacea, Myriapoda and Chelicerata), the arthropod-like Onychophora and Tardigrada and five phyla of introvert bearing worms: the Nematoda, Nematomorpha, Priapulida, Kinorhyncha and Loricifera. In terms of species numbers and niche diversity, the Ecdysozoa is far and away the most significant clade of animals with over a million described species and an estimated total of more than 4.5 million living species (Chapman 2005). The extraordinary number of insects is well known—there are estimated to be more than 10 times as many species of insects than there are of all the deuterostomes and lophotrochozoans put together—yet even had the founder of the insect lineage been eaten by a passing frog, the nematodes and the rest of the arthropods (myriapods, chelicerates and crustaceans) would still easily outnumber all other living animals by close to a quarter of a million species (Chapman 2005). Their characteristic tough cuticle also means that ecdysozoans are well represented in the fossil record adding further wonderful forms to the diversity of the phylum.

Despite the huge numbers of species and great niche diversity, the basic body plans of the Ecdysozoa are rather conservative, being either insect-like with a segmented body and jointed appendages or worm-like with an anterior circum-oesophageal nerve ring and a terminal mouth usually found on an introvert. All groups lack a primary larva as generally conceived and possess a moulted cuticle with concomitant lack of locomotory cilia; it is this periodic moulting or ecdysis that gives the assemblage its name. Although the morphological diversity of ecdysozoan phyla may be seen as fairly restrained when compared to the diversity of shapes seen

among, for example, Lophotrochozoa, these two ecdysozoan body plans happen to manifest themselves in the two most extensively and intensively studied invertebrates on the planet, the nematode *Caenorhabditis elegans* and the fruitfly *Drosophila melanogaster*.

Prior to 1997, the prevalent view of arthropod relationships linked them, via the onychophorans, to the annelid worms. This annelid–arthropod clade is called Articulata in recognition of the principal character uniting these phyla: a segmented body. Articulata was generally thought to be part of a larger assemblage of animal phyla linked by the possession of a coelomic cavity and called Coelomata. Although the alternative concept of a relationship between arthropods and pseudocoelomate worms such as nematodes and priapulids existed much earlier (discussed in Schmidt-Rhaesa 1998), the first support from molecular sequence data for such a relationship, and indeed the first reference to the Ecdysozoa, date to a paper by Aguinaldo *et al.* (1997).

This discussion is predicated on the assumption that the Ecdysozoa is a natural, monophyletic group. However, the existence of the Ecdysozoa is not yet universally accepted and so we will consider the evidence that has amassed in support of the monophyly of this group in the 10 years since the paper by Aguinaldo *et al.* The relationships among the introvertan worms, their position relative to Panarthropoda (Onychophora, Tardigrada and Euarthropoda) and several aspects of the phylogeny within Panarthropoda and Euarthropoda themselves are all still controversial and we will consider recent arguments around each of these.

## 2. THE ECDYSOZOA IS A MONOPHYLETIC GROUP

The initial support for the Ecdysozoa came from a study of small subunit (18S) ribosomal RNA (SSU rRNA) that specifically addressed one problem encountered when using molecular data derived from

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the *C. elegans* genome and many other nematodes (Aguinaldo *et al.* 1997). This problem derives from the fact that the *C. elegans* genome is rapidly evolving relative to those of other animals and is therefore perceived to be susceptible to the systematic phylogenetic error of long-branch attraction (LBA; Felsenstein 1978). This artefact of tree reconstruction would tend to cause the branch leading to the fast-evolving nematodes to be shifted towards the root of a tree. The use of short-branched nematodes in the analysis of Aguinaldo *et al.* resulted in the nematodes moving from their position close to the root of the bilaterian tree (one also supported by consideration of their morphology, in particular their lack of a coelomic cavity) to a close relationship with the arthropods and priapulid worms in a clade which the authors named, the Ecdysozoa (Aguinaldo *et al.* 1997).

Subsequent analyses of rRNAs have extended membership of the Ecdysozoa beyond Nematoda and Priapulida to include three further phyla of worms; Nematomorpha, Kinorhyncha and Loricifera. This clade had been anticipated by various authors who had already linked these five worm phyla in a group called the Cycloneuralia (Ahlrichs 1995) or Introverta (Nielsen 2001).

The monophyletic Ecdysozoa has been replicated by other taxonomically well-sampled datasets including combined small and large subunit (LSU) rRNAs (Mallatt & Winchell 2002; Mallatt & Giribet 2006) and myosin heavy-chain sequences (Ruiz-Trillo *et al.* 2002) as well as *Hox* gene signature peptides (de Rosa *et al.* 1999) and the (somewhat puzzling) shared presence, in the nervous system of all studied ecdysozoans, of an unidentified antigen recognized by the anti-horseradish peroxidase (HRP) antibody (Haase *et al.* 2001). The multimeric beta-thymosin gene found in flies and nematode worms (Manuel *et al.* 2000) has been shown not to be a synapomorphy of the Ecdysozoa (Telford 2004).

Despite these congruent results, there exists a powerful series of papers arguing against the close relationship of nematodes and arthropods and supporting instead the traditional view of the monophyletic Coelomata linking arthropods such as *D. melanogaster* to humans rather than to nematode worms (Blair *et al.* 2002; Wolf *et al.* 2004; Philip *et al.* 2005; Ciccarelli *et al.* 2006; Rogozin *et al.* 2007). This specific phylogenetic question has the attraction of being approachable with the largest possible molecular datasets, the completely sequenced genomes of flies, worms and humans. What almost all studies that have used this approach have found is that the evidence is strongly in favour of the Coelomata hypothesis and against Ecdysozoa.

The counter argument, naturally, is that these whole genome studies suffer from precisely the problem that the Aguinaldo *et al.* paper addressed; the systematic artefactual attraction of the nematode branch towards the root of the Bilateria due to LBA. This contention does seem to be borne out by a number of publications in the past few years. Copley *et al.* (2004) compared the presence or absence of 1712 genes or distinct combinations of protein domains specific either to flies and humans or to

flies and nematode worms. There were many more of the former giving apparently strong support to the Coelomata hypothesis. However, they were able to show that this strong signal was an artefact resulting from a strong tendency towards secondary loss of genes in the nematode, a feature of its high rate of genomic evolution (Copley *et al.* 2004). In parallel, Philippe *et al.* (2005) used large 'phylogenomic' datasets (whole genomes combined with data from expressed sequence tag projects and hence having much broader taxon sampling) and showed that experiments designed to reduce potential long-branch effects—using less distant out-groups, selecting slowly evolving nematodes and discarding the more unevenly evolving genes—supported Ecdysozoa while Coelomata was supported without these efforts. Finally, Irimia *et al.* (2007) have used a similar approach to show that claims of an excess of identical, rarely changing amino acids shared by flies and humans and lacking in nematode worms (Rogozin *et al.* 2007), are biased by the use of distant out-groups and by the rapid evolution of *C. elegans*. They show that, when these biases are accounted for, there is significantly more support for Ecdysozoa than for Coelomata from this source of evidence (Irimia *et al.* 2007).

The phylogenomic approach has recently been extended to the slowly evolving Priapulida that are strongly supported as ecdysozoans (Webster *et al.* 2006; also true of Nematomorpha: T. Juliusdottir, R. Jenner, M. Telford & R. Copley 2007, unpublished results). This result was further strengthened by analyses of the very arthropod-like mitochondrial genome of *Priapulid caudatus*. Perhaps even more strikingly, the priapulid mitochondrial gene order can be reconciled with that of the arthropods by a single inversion (Webster *et al.* 2007).

We would also like to highlight a further very convincing synapomorphy supporting the monophyly of Protostomia and hence, we believe, definitively ruling out the Coelomata hypothesis. Papillon *et al.* (2004) used the presence of a dozen rarely changing amino acids in the mitochondrial *nad5* gene of protostomes as a striking indication that the chaetognaths were protostomes and not deuterostomes as traditionally believed. The signatures constitute a very complex, conserved, derived character defining a monophyletic group of Protostomia. We have extended this analysis of the *nad5* gene, which appears to have undergone a significant burst of evolution within the lineage leading to the protostomes. Almost all of these signature amino acids are found in nematodes and priapulids as well as in other controversial protostomes including rhabditophoran and catenulid flatworms and lophophorates. A monophyletic Protostomia, while not specifically proving the existence of Ecdysozoa, is at least incompatible with a monophyletic group of coelomate animals and therefore contradicts the results from whole genome studies supporting Coelomata.

We strongly support the notion of a monophyletic Ecdysozoa and feel that the only opposing evidence—the whole genome support for Coelomata—is flawed by systematic error, which has been addressed successfully

by much better taxon sampling, in particular the use of a close out-group (Philippe *et al.* 2005). In addition to the molecular systematic support, the monophyly of Ecdysozoa is supported by a number of synapomorphies including ecdysis of a trilayered cuticle (consisting of epi-, exo- and endocuticle), lack of locomotory cilia, lack of primary larva, terminal mouth and the HRP antigen in nervous system and conserved mitochondrial gene order that have been mentioned (see also Schmidt-Rhaesa 1998).

### 3. CYCLONEURALIA, INTROVERTA, SCALIDOPHORA AND NEMATOIDA

While we do not have an equivalent of the *nad5* rare genomic change to support the monophyly of the Ecdysozoa within the Protostomia, as we have seen, we do have strong evidence from phylogenomic datasets of tens to hundreds of genes for the monophyly of Arthropoda plus Nematoda, Nematomorpha and Priapulida (Philippe *et al.* 2005; Webster *et al.* 2006). These three worm phyla had previously been linked to two further phyla, Kinorhyncha and Loricifera in a group collectively known as the Cycloneuralia (Ahrichs 1995). The name refers to their collar-shaped, circum-oral brain; something similar is seen in Gastrotricha which are, however, lophotrochozoans not ecdysozoans (Telford *et al.* 2005). These phyla (but not Gastrotricha) also share an eversible anterior end, or introvert, which terminates in the mouth and gives the alternative name of Introverta (Nielsen 1995), although the introvert is only seen in the larvae of Nematomorpha and in isolated examples of Nematoda.

What we still do not have is much reliable information on the relationships between these phyla or their relationships to the Panarthropoda. This may be explained in part by the difficulty in working on the minute and hard to study Kinorhyncha and Loricifera.

The two groupings that do seem credible are a close relationship between Nematoda and Nematomorpha and between Priapulida and Kinorhyncha. Nematodes and nematomorphs share a number of characters, including the reduced circular muscles in the body wall, the cloaca seen in both sexes, the aflagellate sperm, the cuticle (collagenous not chitinous) and the ectodermal ventral and dorsal nerve cords and were grouped by Nielsen (1995) and named Nematoida by Schmidt-Rhaesa (1996). This clade has weak support from SSU analyses (Peterson & Eernisse 2001), combined analyses of LSU and SSU rRNA (Mallatt *et al.* 2004) and from a small-scale phylogenomic analysis of 50 nuclear protein-coding genes (T. Juliusdottir, R. Jenner, M. Telford & R. Copley 2007, unpublished results).

Morphologists have also united Priapulida, Kinorhyncha and Loricifera as the Scalidophora (Schmidt-Rhaesa 1998) or Cephalorhyncha (Nielsen 2001), on the basis of a shared introvert with scalids and the presence of two rings of retractor muscles on the introvert. The close relationship between priapulids and kinorhynchs at least seems to hold-up based on the three gene sequences available from kinorhynchs but the single phylogenetic study

including the Loricifera showed their position to be ambiguous (Park *et al.* 2006).

There is no clear indication that the Cycloneuralia is a monophyletic group and in fact most evidence points to Priapulida (and therefore Kinorhyncha too) being the earliest branch and the Nematoida being the sister group of the Panarthropoda (e.g. Mallatt & Giribet 2006; Webster *et al.* 2006).

### 4. PANARTHROPODA: EUARTHROPODA, TARDIGRADA AND ONYCHOPHORA

The monophyly of the Euarthropoda plus Onychophora and Tardigrada seems, on the face of it, uncontroversial. They are linked by a number of features, most important of which is the segmentally repeated limbs with terminal claws; Onychophora translates as 'claw-bearer'. The limbs in all three groups straddle parasegmental boundaries marked by the expression of the segment polarity gene *engrailed* (Patel *et al.* 1989; Gabriel & Goldstein 2007; J. Eriksson 2007, personal communication). Their segmental paired, saccate nephridia (reduced in number and functioning as excretory organs in euarthropods) and open circulatory system also seem to be valid synapomorphies although both are missing in the miniaturized tardigrades (Hejnol & Schnabel 2005). The circulatory system is characteristically formed as a fusion of both the coelomic cavities and the primary body cavity/embryonic blastocoel (i.e. a mixocoel) and there is a dorsal heart with characteristic openings (ostia) into the open circulatory system (Nielsen 2001).

Despite the characters in common with Panarthropoda, not all molecular studies support their monophyly, grouping Tardigrada with the nematodes rather than with Euarthropoda (see Lartillot & Philippe 2008). However, if we assume that Panarthropoda is monophyletic, three trees could unite them: Euarthropoda with either Tardigrada or Onychophora, or Tardigrada and Onychophora as sister groups. The branching order of these three taxa is not resolved by molecules or morphology. However, if we assume that the small size of tardigrades is derived and accounts for the absence of mixocoel, heart and nephridia (Schmidt-Rhaesa 2001; characteristics found in Onychophora and Euarthropoda), we suggest that the similarities of cuticle, ganglionated ventral nerve chord and limbs in tardigrades and Euarthropoda may indicate a sister group relationship.

### 5. EUARTHROPODA: MYRIOCHELATA VERSUS MANDIBULATA

The relationships of the four euarthropod clades—Chelicerata, Myriapoda, Crustacea and Hexapoda (Hexapoda = Insecta, plus the basally branching groups Diplura, Protura and Collembola) have long been disputed. A decade or so ago there were even serious arguments over the single versus multiple origins of arthropodization, and therefore over the monophyly versus polyphyly of euarthropods (Fryer 1997). Molecular analyses have emphatically supported the monophyly of euarthropods and a unique origin of their cuticularized body and jointed appendages and in the

past years, the attention has been focused more on the relationships between these four groups. One common feature of morphology based interpretations of arthropod phylogeny was the close relationship between Myriapoda and Hexapoda in a clade called the Atelocerata (which means malformed horns and refers to their common lack of a second antennal segment) defined additionally by unbranched (uniramous) appendages, malpighian tubules and tracheal breathing (Telford & Thomas 1995). According to the polyphyleticists, the Atelocerata is grouped with the Onychophora in a clade called the Uniramia. Arguably, the clearest result to date in arthropod phylogeny shows that the insects are not most closely related to the myriapods but to the crustaceans (Boore *et al.* 1995) and, in all likelihood, constitute a subgroup within the Crustacea. This clade of crustaceans plus insects has been referred to as the Pancrustacea or as the Tetraconata due to the tetrapartite crystalline cones of the ommatidia (Dohle 1997, 2001; Harzsch 2004).

More controversial though, is the true position of Myriapoda which share numerous similarities of head organization not only with the insects (as discussed) but also with crustaceans, most notably the presence of a mandible on the third, appendage-bearing head segment. The common head structure of myriapods, crustaceans and insects with two pairs of antennae (at least primitively), paired gnathobasic mandibles and two pairs of maxillae strongly supports their monophyly. This group is named the Mandibulata, reflecting the particular importance of detailed similarities seen between the mandibles of Pancrustacea and Myriapoda in terms of segmental identity, positioning relative to other body parts, gene expression, detailed similarities in terminal differentiation and, of course, in function (Scholz *et al.* 1998; Edgecombe *et al.* 2003; Harzsch *et al.* 2005).

Surprisingly, a number of molecular studies using rRNA, nuclear protein-coding genes and complete mitochondrial genome sequences do not support the mandibulate clade, instead linking the myriapods to the chelicerates in a group called the Paradoxopoda or the Myriochelata (Mallatt *et al.* 2004; Negrisolo *et al.* 2004; Pisani *et al.* 2004). An analysis of nuclear coding genes (Regier *et al.* 2005), however, did not find strong support for either Myriochelata or Mandibulata, suggesting that there is great uncertainty over the affinity of myriapods. Recognizing that the distinction between the two possibilities comes down to the position of the root of the euarthropod tree, we have reanalysed the complete mitochondrial genome sequences of various arthropods using a priapulid as a short-branched, phylogenetically close relative of the arthropods. We find that, in contrast to previous studies that had used more distant out-groups (lophotrochozoans), our mitochondrial tree narrowly supports Mandibulata over Myriochelata (figure 1; see also Pisani 2004). We have also analysed a number of nuclear, protein-coding genes and have reached the same conclusion (figure 2). While our bias in support of a return to the Mandibulata is probably obvious, it is clear that this question remains to be resolved one way or the other.

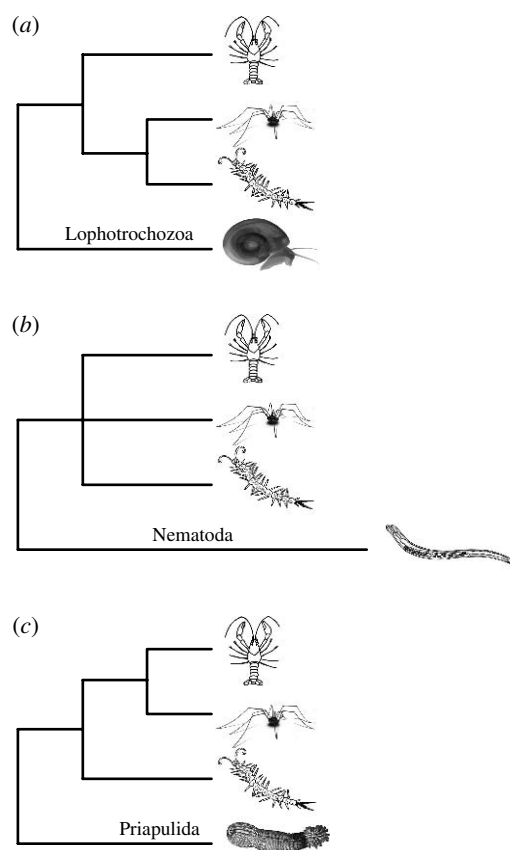


Figure 1. Different out-groups support different positions of the root of the Euarthropoda. (a) Support for Myriochelata (Myriapoda plus Chelicerata) is strong using mitochondrial when the Euarthropod tree is rooted using phylogenetically distant lophotrochozoans (Paradoxopoda). (b) Support is equivocal for either rooting using long branch but phylogenetically closer ecdysozoan nematodes (unresolved Euarthropoda). (c) The tree switches to supporting a monophyletic Mandibulata (Myriapoda with Crustacea and Hexapoda) when using the phylogenetically close and short-branched priapulid as an out-group (O. Rota-Stabelli & M. J. Telford 2007, unpublished results).

While there are specific characteristics shared by myriapods and chelicerates yet absent from Pancrustacea (Dove & Stollewerk 2003; Kadner & Stollewerk 2004; Stollewerk & Simpson 2005), it is difficult to demonstrate these as synapomorphies as we have insufficient data from an out-group and the suspicion is that the Myriapoda/Chelicerata character state may be plesiomorphic and uninformative (Harzsch 2004; Harzsch *et al.* 2005). While the same criticism may be made of some of the characters supporting Mandibulata, the chelicerate homologue of the mandible (the first walking leg; Telford & Thomas 1998) seems likely to represent the plesiomorphic condition as it strongly resembles adjacent, serially homologous, walking appendages. This implies that the mandible itself is a shared derived character uniting the mandibulates.

## 6. PYCNOGONIDS ARE CHELICERATES

The pycnogonids or sea spiders have long been associated with the chelicerates due to the shared character of chelicerae (cheliferes in pycnogonids) on

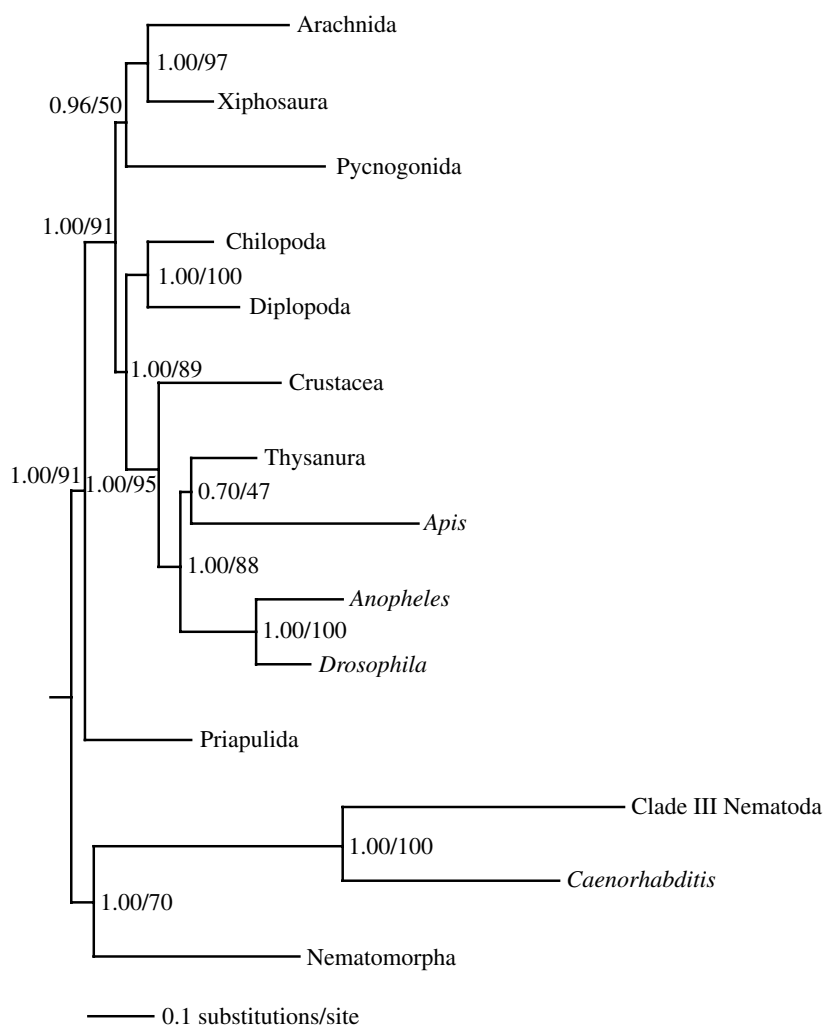


Figure 2. Phylogeny of the Ecdysozoa. Bayesian analysis using SSU and LSU ribosomal RNA sequences, complete mitochondrial genomes and eight nuclear protein-coding genes (Vacuolar ATP-synthase, enolase, Glyceraldehyde 3-phosphate dehydrogenase, carnitine palmitoyltransferase, Na/K ATPase, RNA Pol II, Dyskerin and EF 1-alpha). Some taxa with missing data have been merged into composite sequences. Support values shown as Bayesian posterior probabilities and non-parametric bootstrap.

the first limb-bearing segment. This phylogenetic link was questioned recently both by studies of their nervous systems and by molecular systematic analyses. The larval nervous system of a pycnogonid from the genus *Anoplodactylus* was studied and the chelifere appeared to be innervated from the frontmost portion of the brain (the protocerebrum), suggesting that this appendage was therefore not homologous to the chelicerae of other chelicerates which is innervated from the second portion of the brain—the deutocerebrum (Budd & Telford 2005; Maxmen *et al.* 2005). This tied in with a molecular phylogenetic study placing the Pycnogonida at the base of Euarthropoda and not with Chelicerata (Regier *et al.* 2005). Subsequent analysis of *Hox* expression patterns have disproved the protocerebral position of the cheliferes showing that they are indeed in the same deutocerebral position as chelicerae (Jager *et al.* 2006) and most molecular data imply that the chelicerates including Pycnogonida is a monophyletic group and that the contrary result is most likely derived from the rapid evolutionary rate of the Pycnogonida (Mallatt & Giribet 2006).

## 7. THE POSITION OF THE HEXAPODA WITHIN THE PANCRUSTACEA

The support for the monophyly of Crustacea plus Hexapoda, which came most emphatically from the evidence of a shared mitochondrial genome rearrangement, has been bolstered by numerous subsequent molecular phylogenetic analyses (Friedrich & Tautz 1995; Hwang *et al.* 2001; Delsuc *et al.* 2003; Nardi *et al.* 2003; Regier *et al.* 2005). Consideration of various aspects of morphology, in particular of nervous system ontogeny and structure, gives further weight to the integrity of this clade. Harzsch (2004) and Harzsch *et al.* (2005) list neuroblasts, two pairs of serotonergic neurons per hemineuromere, a fixed number of excitatory motoneurons per limb muscle and aspects of lateral eye ultrastructure in support of the Crustacea plus Hexapoda clade, which they term Tetraconata.

More controversial has been the placement of Hexapoda within Crustacea and the monophyly versus polyphyly of Hexapoda with a number of studies separating the Collembola from the Insecta. While it is generally agreed that Crustacea is paraphyletic rather than being the sister group of the Hexapoda (and that

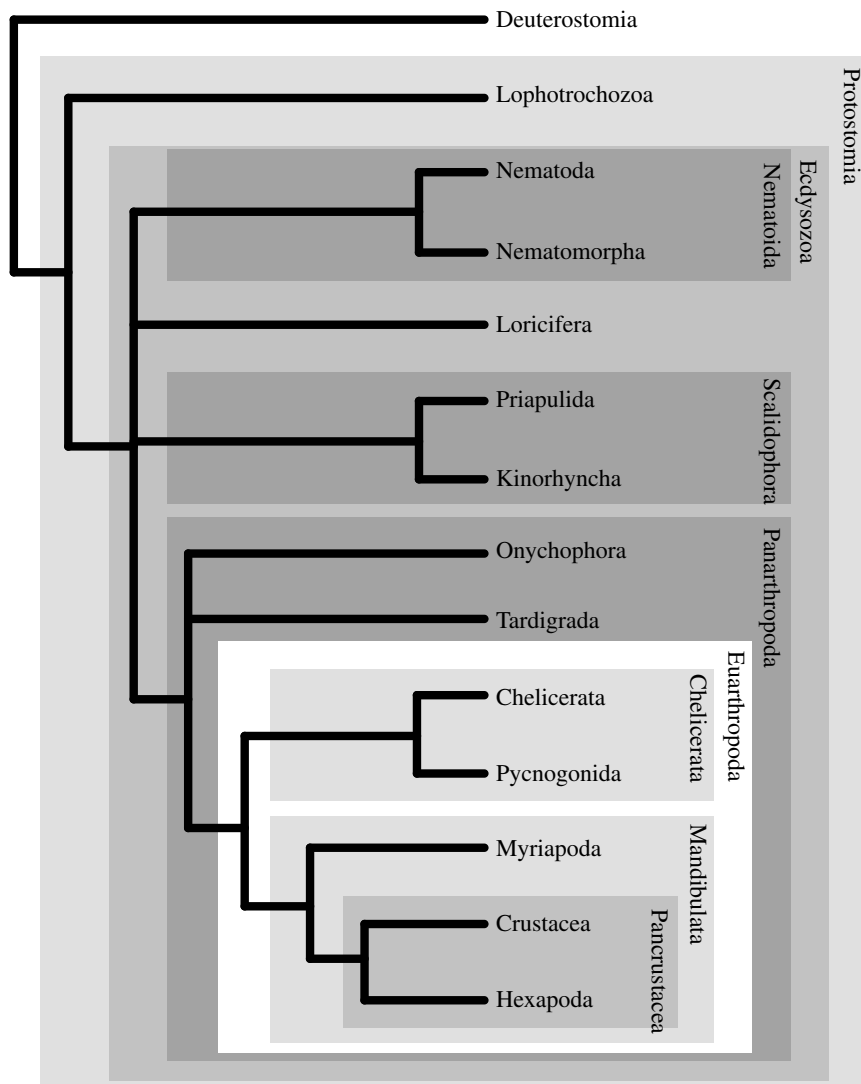


Figure 3. The phylogeny of the Ecdysozoa espoused in this manuscript. Names of probable monophyletic groups are given for each box. Unresolved portions of the tree are shown as multifurcations. We have shown Mandibulata (including Myriapoda) as monophyletic and Panarthropoda (including Tardigrada) as monophyletic groups despite some uncertainty over these groups as we feel the morphological evidence particularly convincing for these clades.

Hexapoda is in effect a terrestrial group of crustaceans), the closest crustacean sister group of the hexapods has been debated. Ignoring for the moment the little studied Cephalocarida and Remipedia, there are two contenders among the main crustacean classes: the Malacostraca that includes familiar species such as crabs and mantis shrimps and Branchiopoda such as *Artemia* the brine shrimp and *Daphnia* the water flea. The Hexapoda–Malacostraca clade is supported by various features of brain anatomy; specifically, members of these two groups share the presence of three brain neuropils joined by chiasmata where other crustaceans have two neuropils linked by parallel fibres (Harzsch 2002). Analyses of complete mitochondrial genome sequences on the other hand support a monophyletic Malacostraca and Branchiopoda clade as a sister group to Hexapoda (Cook *et al.* 2005). Most other molecular analyses, however, support a sister group relationship between Hexapoda and Branchiopoda (Regier *et al.* 2005; Mallatt & Giribet 2006).

We have recently gathered all available data from rRNAs, mitochondrial genomes and various nuclear protein-coding genes, and our analyses support the

close relationship between Hexapoda and Branchiopoda. This relationship is of great interest to the many workers interested in the evolution of the insects as it shows that *Daphnia*, a crustacean with a completely sequenced nuclear genome, is a relatively close sister group of insects. Our analyses also include data from Cephalocarida and Remipedia and the placement of these two groups is less certain. Both taxa are derived in terms of numbers of substitutions. While the remipede consistently groups close to the hexapods, the position of the cephalocarid is very unstable (A. Economou & M. J. Telford 2007, unpublished results).

Although the relationships within the Hexapods are beyond the scope of this paper, the controversy over the placement of the Collembola is worth mentioning. While rRNA and nuclear coding gene phylogenies recover the expected monophyly of the hexapods (Insecta, Diplura, Protura and Collembola), analyses using complete mitochondrial genomes recover a diphylectic Hexapoda with the Insecta separated from the Collembola (Nardi *et al.* 2003); sequences for Diplura and Protura were not available. The basis of this result has been questioned by subsequent authors

and one must conclude that, although monophyly of Hexapoda ultimately seems the most likely result, this needs to be tested with larger datasets.

## 8. CONCLUSION

In figure 3, we summarize our best current estimate of ecdysozoan phylogeny. The first thing that is obvious from this tree and from the preceding discussion is that, while it seems clear that Ecdysozoa is a monophyletic group, the relationships between phyla and major classes within the clade are often uncertain.

While the pattern of relationships of the Ecdysozoa has its own great intrinsic interest, the phylogeny should also be viewed as the basis for a further understanding of the evolution of the Ecdysozoa. The mapping of character states onto a phylogeny allows us to go beyond the relationships of organisms to the evolution of characters and ultimately a fuller understanding of the process of evolution. The characteristics of the common ancestor of Ecdysozoa is of particular interest and it can be safely assumed to have possessed the synapomorphies of the group; Budd has tentatively reconstructed the common ancestor as a large worm-like form with a terminal mouth (Budd 2001) and to these characteristics we can add the shared characters discussed previously. The monophyly versus paraphyly of the Cycloneuralia becomes important now as, if paraphyletic, then their common ancestor becomes synonymous with the ecdysozoan ancestor and suggests that it also possessed a cycloneuralian brain (not unreasonable considering the similar situation seen in onychophorans; Eriksson *et al.* 2003), and an introvert.

More controversial is the possibility that the ecdysozoan ancestor was segmented. While the kinorhynch metameres are generally referred to as zonites rather than segments, this seems a rather pointless distinction and is one indication that segmentation may be primitive in the group (Müller & Schmidt-Rhaesa 2003; Schmidt-Rhaesa & Rothe 2006). The similar deployment of homologous genes ('segment polarity' or 'pair rule' genes) in arthropods and kinorhynchs would be a more direct indication of homology and hence common ancestry of segmentation within the group as would the demonstration that arthropod segmentation can be convincingly homologized with that of annelids (Prud'homme *et al.* 2003) or even vertebrates (Damen 2007).

Through comparison of the completely sequenced genomes of *D. melanogaster* and *C. elegans*, there is also the theoretical possibility of learning something about the genome of the ecdysozoan common ancestor, or perhaps something close to it depending on the position of the Arthropoda/Nematoida common ancestor. One significant conclusion from comparative genomics to date has been the secondary loss of large numbers of genes in the two model ecdysozoans (Copley *et al.* 2004; Putnam *et al.* 2007). The problem, of course, is that the two model species appear to have much derived genomes making comparisons particularly difficult to interpret—are they different from other animals due to common ecdysozoan gene losses or through convergent gene losses in these two derived

models? The ecdysozoan genome projects ongoing or recently announced, in particular that of the priapulid *P. caudatus*, are very exciting for the purpose of reconstructing the ancestral ecdysozoan genome, and should also add further to our understanding of the evolutionary relationships of this huge, diverse and fascinating group.

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