

## The Lesser-Known Protostome Taxa: An Introduction and a Tribute to Robert P. Higgins<sup>1</sup>

JAMES R. GAREY<sup>2</sup>

*Department of Biology, University of South Florida, 4202 East Fowler Avenue SCA110, Tampa, Florida 33620*

**SYNOPSIS.** Most biologists are familiar only with a few of the approximately 40 extant animal phyla. The purpose of this symposium was to renew interest in the lesser-known invertebrate taxa, encourage their use in research and teaching and to promote the relevance of high-level systematic studies. This paper reviews the two major views of metazoan evolutionary relationships with particular attention to the lesser-known taxa and to some of the new and/or conflicting terminology used in current animal phylogenetic study. The current use of lesser-known taxa in research is briefly described, and the discussion that followed the symposium is summarized. The paper concludes with a brief history of the symposium and a tribute to Robert P. Higgins, who organized three “Symposia on the Lesser-Known Invertebrates” over the past 25 yr.

### INTRODUCTION

Most biologists are familiar with only a few of the approximately 40 extant animal phyla such as annelids, arthropods, molluscs, echinoderms and chordates. How many are familiar with the more obscure groups such as gnathostomulids, kinorhynchs, priapulids and cycliophorans? This symposium was designed to play a role in renewing interest in the lesser-known animal groups, encourage their use in research and teaching and promote the relevance of high-level systematic studies. The symposium was restricted to protostomes simply to narrow the scope so that it could be completed in a single day. The order of papers in this volume is random and not related to the phylogenetic position of the taxa. The participants of the symposium are shown in Figure 1.

Current advances in evolutionary, developmental and ecological studies make it apparent that the lesser-known animal groups are important subjects of study. Now that we have the tools to study both molecular and morphological aspects of animal evolution it is clear that we cannot understand animal evolution without paying attention to the lesser-known taxa (Garey and Schmidt-Rhaesa, 1998). Many lesser-known groups are far more important ecologically than is generally known, particularly the phyla represented by mostly meiofaunal (microscopic) species. The small body size of many of these species has confounded their placement in metazoan phylogeny. Advances in ultrastructure-level morphology and molecular analysis show promise in determining the relationships of these groups to metazoans as a whole, and it is certain that the lesser-known taxa will play an important role in future studies of metazoan phylogeny.

### THE PLACEMENT OF THE LESSER-KNOWN GROUPS WITHIN METAZOA

This section reviews the morphological and molecular evidence used to define evolutionary relationships

among the Metazoa. Some of the conflicting terminology and definitions of various monophyletic groups used in other papers from these proceedings will be clarified as well. For reviews of morphological analyses, see the recent works of Nielsen (2001) and Ax (1996, 2000). Molecular and combined analyses are reviewed in Garey (2001), Giribet *et al.* (2000), Peterson and Eernisse (2001), and Zrzavy *et al.* (1998).

In the traditional view, segmented protostomes are placed together in a clade known as Articulata (Cuvier, 1812), and lophophorates are considered to be basal deuterostomes (Nielsen, 2001). In recent years, a new hypothesis of evolution has been proposed in which all molting animals fall into a single clade called Ecdysozoa (Aguinaldo *et al.*, 1997) and in which the lophophorate groups are considered to be protostomes rather than deuterostomes (Halanych *et al.*, 1995; Mackey *et al.*, 1996). Although controversial, this new hypothesis is gaining significant support in the scientific community (*e.g.*, see Adoutte *et al.*, 2000). These unresolved conflicting views of metazoan phylogeny cause considerable confusion in the interpretation of the current literature. To complicate matters, terminology used to describe the higher taxonomic groupings of metazoans is somewhat fluid, and the names used by American and European workers differ. Table 1 defines some of these terms.

Two contrasting hypotheses of animal phylogeny are shown in Figure 2, one based on the Articulata concept, the other on Ecdysozoa. Articulata (Cuvier, 1812) is based on the hypothesis that segmentation arose only once and unites the annelids and arthropods. It should be noted that the idea that animals evolved as a series of grades, with acoelomate, pseudocoelomate and eucoelomate animals each forming a monophyletic clade is an issue separate from Articulata, and has never been seriously considered by morphologists. Unfortunately it is still commonly taught in introductory biology and invertebrate zoology courses, partly because Libbie Hyman organized her treatise on the invertebrates along those lines (Hyman, 1951). The Ecdysozoa hypothesis, originally discovered from the analysis of gene sequences (Aguinaldo *et al.*, 1997) is that molting evolved a single time and

<sup>1</sup> From the Symposium *Lesser-Known Protostome Taxa: Evolution, Development, and Ecology* presented at the Annual Meeting of the Society for Integrative and Comparative Biology, 3–7 January 2001, at Chicago, Illinois.

<sup>2</sup> E-mail: garey@chuma1.cas.usf.edu



FIG. 1. The participants of the 2001 SICB Symposium on the Lesser-known Protostome Taxa. Top row (standing), left to right: Ken Halanych (Annelida); Claus Nielsen (Entoprocta, Ectoprocta, Phoronida, Brachiopoda); Jen Grenier (Onychophora); Diane Nelson (Tardigrada); Reinhardt Kristensen (Cycliophora, Loricifera, Micrognathozoa); Tom Shirley (Priapulida); Birger Neuhaus (Kinorhyncha); Bottom row (sitting), left to right: Clint Turbeville (Nemertea), Andreas Schmidt-Rhaesa (Nematomorpha, Gastrotricha), Jim Garey (Symposium Organizer), Bob Wallace (Rotifera), Tom Near (Acanthocephala). Photo courtesy of Michael S. Robeson.

so all molting animals are united by the character of growth by ecdysis. Detailed discussions of the morphological characters related to segmentation and molting can be found in Schmidt-Rhaesa *et al.* (1998) and Garey (2001). Molecular studies have also shown that the lophophorates, long thought to be related to the deuterostomes, are clearly protostomes, and fall within a clade of the non-molting protostomes named Lophotrochozoa (Halanych *et al.*, 1995; Mackey *et al.*, 1996). The main difference between the morphological tree and the molecular tree is the placement of the lophophorates relative to the deuterostomes and protostomes and the relative position of the annelids and arthropods. The details of the topology within each of the two phylogenies in Figures 1 and 2 are continuously debated, and the two trees shown should be considered as examples of each. Despite the differences between the morphology- and molecular-based trees, some remarkable congruencies highlight the power of using both approaches.

Bilateria can be divided into two major groups in several ways (reviewed more thoroughly in Nielsen, 2001, pp. 82–83). The most common is a division based on the fate of the blastopore during embryonic development. In Deuterostomia, the blastopore most commonly becomes the anus, and a secondary opening forms the mouth. In Protostomia, the blastopore becomes the mouth, although more commonly, the blastopore fuses laterally, forming both the mouth and the

anus. Another way to divide Bilateria is based on the cleavage pattern of the early embryo. In Radialia, cleavage follows a radial pattern, while in Spiralia, cleavage follows a spiral pattern. The terms Radialia and Deuterostomia are often considered synonyms, as are Spiralia and Protostomia, but this is an oversimplification (see Table 1). Both the fate of the blastopore and early embryonic cleavage patterns vary considerably among the Bilateria and have not been assessed experimentally in many species. Molecular analyses concur with the protostome/deuterostome split except that the lophophorates are protostomes, not basal deuterostomes.

The lophophorates comprise three phyla, Brachiopoda, Phoronida, and Ectoprocta (also known as Bryozoa) that share a ciliated feeding organ known as the lophophore. Historically they have been considered to be a monophyletic group positioned within the deuterostomes, or immediately basal to the deuterostomes. Molecular analyses have demonstrated very clearly that the lophophorates are not deuterostomes (Halanych *et al.*, 1995; Mackey *et al.*, 1996), but instead are protostomes. They also indicate that although brachiopods are closely related to phoronids, the ectoprocts are not related to the brachiopod/phoronid clade. Nielsen's morphological classification also excludes ectoprocts from Lophophorata and places them with the protostomes, but he still considers brachiopods/phoronids as basal deuterostomes. Nielsen's close associ-

TABLE 1. *Notes on terminology used in metazoan phylogeny.*


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Annelida = Polychaeta + Oligochaeta + Hirundinea + Pogonophora + Echiura <sup>1</sup>
Articulata = Arthropoda + Annelida (sometimes Onychophora + Mollusca are added)
Aschelminthes = polyphyletic assemblage of mostly microscopic animals. Because it is not a monophyletic group this term should be avoided although it is still used extensively. Essentially it is Cycloneuralia + Gnathifera and sometimes includes Entoprocta and a few other taxa.
Brachiopoda = Brachiopoda + Phoronida (phoronids as shell-less brachiopods)
Bryozoa = Ectoprocta
Cycloneuralia = animals with a toroidal shaped brain that circles the pharynx or gut <i>sensu</i> Ehlers <i>et al.</i> : (Priapulida, Loricifera, Kinorhyncha, Nematoda, Nematomorpha) <i>sensu</i> Nielsen: (Priapulida, Loricifera, Kinorhyncha, Nematoda, Nematomorpha, Gastrotricha) <sup>2</sup>
Deuterostomia = Notoneuralia <sup>3</sup> ≈ Radialia
Ecdysozoa = molting protostomes (Panarthropoda + Cycloneuralia <i>sensu</i> Ehlers <i>et al.</i> )
Gnathifera = animals with cuticular jaws composed of tube-like rods (Gnathostomulida, Micrognathozoa, Syndermata)
Introverta = Cycloneuralia <i>sensu</i> Ehlers <i>et al.</i>
Kamptozoa = Entoprocta
Lophophorata (traditional) = Brachiopoda + Phoronida + Ectoprocta
Lophophorata ( <i>sensu</i> Nielsen) = Brachiopoda + Phoronida
Lophotrochozoa = Spiralia <sup>4</sup> = non-molting protostomes + Lophophorata
Nemathelminthes = Cycloneuralia <i>sensu</i> Nielsen, or Cycloneuralia <i>sensu</i> Ehlers <i>et al.</i> + Gastrotricha or Nemathelminthes = Aschelminthes in some older literature.
Nematoidea = Nematoda + Nematomorpha
Nemertea = Nemertini = Rhynocoela
Notoneuralia <sup>3</sup> = Deuterostomia ≈ Radialia
Panarthropoda = Arthropoda, Onychophora, Tardigarda
Parenchymia = Platyhelminthes + Nemertea
Plathelminthes = Platyhelminthes
Platyzoa = Syndermata + Gnathifera + Platyhelminthes + Gastrotricha + Cycliophora <sup>5</sup>
Pogonophora = Frenulata + Vestimentifera
Protostomia = Gastroneuralia <sup>3</sup> ≈ Spiralia <sup>4</sup>
Syndermata = animals with a syncytial epidermis (Rotifera + Acanthocephala)

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<sup>1</sup> The placement of all these groups within Annelida is still somewhat controversial (see article in this volume by Halanych *et al.*).

<sup>2</sup> Nielsen uses Introverta as the equivalent to Cycloneuralia *sensu* Ehlers *et al.*

<sup>3</sup> Nielsen used Notoneuralia and Gastroneuralia in the first edition of his book (Nielsen, 1995) but not in the second edition (Nielsen, 2001).

<sup>4</sup> Spiralia is often used as a synonym for Protostomia but more recently it has been used as a synonym for Lophotrochozoa because some authors claim that all ecdysozoans lack true spiral cleavage (see Schmidt-Rhaesa *et al.*, 1998). Nielsen (2001) defines Spiralia as Protostomia minus Cycloneuralia.

<sup>5</sup> The position of cycliophorans remains controversial. The first report of cycliophorans (Funch and Kristensen, 1995) suggested they were related to entoprocts, but molecular evidence to date suggests a close affinity to rotifers (Winnepeninckx *et al.* 1998; see article in this volume by Kristensen).

ation of entoprocts and ectoprocts is not supported by molecular data (Mackey *et al.*, 1996) although both groups fall within the Lophotrochozoa clade.

Most arthropods do not have spiral cleavage, and it has been suggested that spiral cleavage is not present in Arthropoda (see Table 1) at all (Schmidt-Rhaesa *et al.*, 1998). A number of other protostome taxa, most notably onychophorans, tardigrades, and all the cycloneuralians (see Table 1) also lack true spiral cleavage. Molecular studies support the monophyly of all these taxa as Ecdysozoa, so the term Spiralia is sometimes used as a synonym for the non-molting protostomes, Lophotrochozoa.

Cycloneuralia is a group that was identified only recently by morphologists (Ehlers *et al.*, 1996; Nielsen, 2001). Cycloneuralia as defined by Ehlers *et al.* (1996) includes all the molting animals that are not members of Panarthropoda (see Table 1). Combining Panarthropoda and Cycloneuralia (*sensu* Ehlers *et al.*) results in the clade of molting animals known as Ecdysozoa. It is difficult to relate the primarily meiofaunal cycloneuralians to much larger panarthropodans using morphology. In earlier molecular studies (*e.g.*, Aguinaldo *et al.*, 1997) it was unclear if Cycloneuralia was the sister group to Panarthropoda, or if the rela-

tionship between those two groups was more complicated, and a more recent 18S rRNA gene based molecular study suggests that Cycloneuralia is paraphyletic with Panarthropoda as the sister group to Nematoidea (see Fig. 2 and Garey, 2001).

Although Ecdysozoa is well defined by the presence of growth by molting, Lophotrochozoa is not as clear, as no single character identifies it. Instead, Lophotrochozoa has a node-based definition, and is comprised of the “last common ancestor of the three traditional lophophorate taxa, the molluscs, and the annelids, and all of the descendants of that common ancestor” (Halanych *et al.*, 1995). This clade also includes the flatworms and their related taxa (Aguinaldo *et al.*, 1997). More recently it has been noted that Lophotrochozoa might be composed of two clades, one containing annelids, molluscs, sipunculans, lophophorates, nemerteans and entoprocts, the other called Platyzoa, which contains flatworms, gastrotrichs and Gnathifera. The term Platyzoa was coined by Cavalier-Smith (1998) and is recognized by some using combined analyses (*e.g.*, Garey *et al.*, 1998; Giribet *et al.*, 2000) but not all (*e.g.*, Peterson and Eernisse, 2001; Zrzavy *et al.*, 1998). Gnathifera is a group that has a very clear morphological basis, but at this point only rudimentary

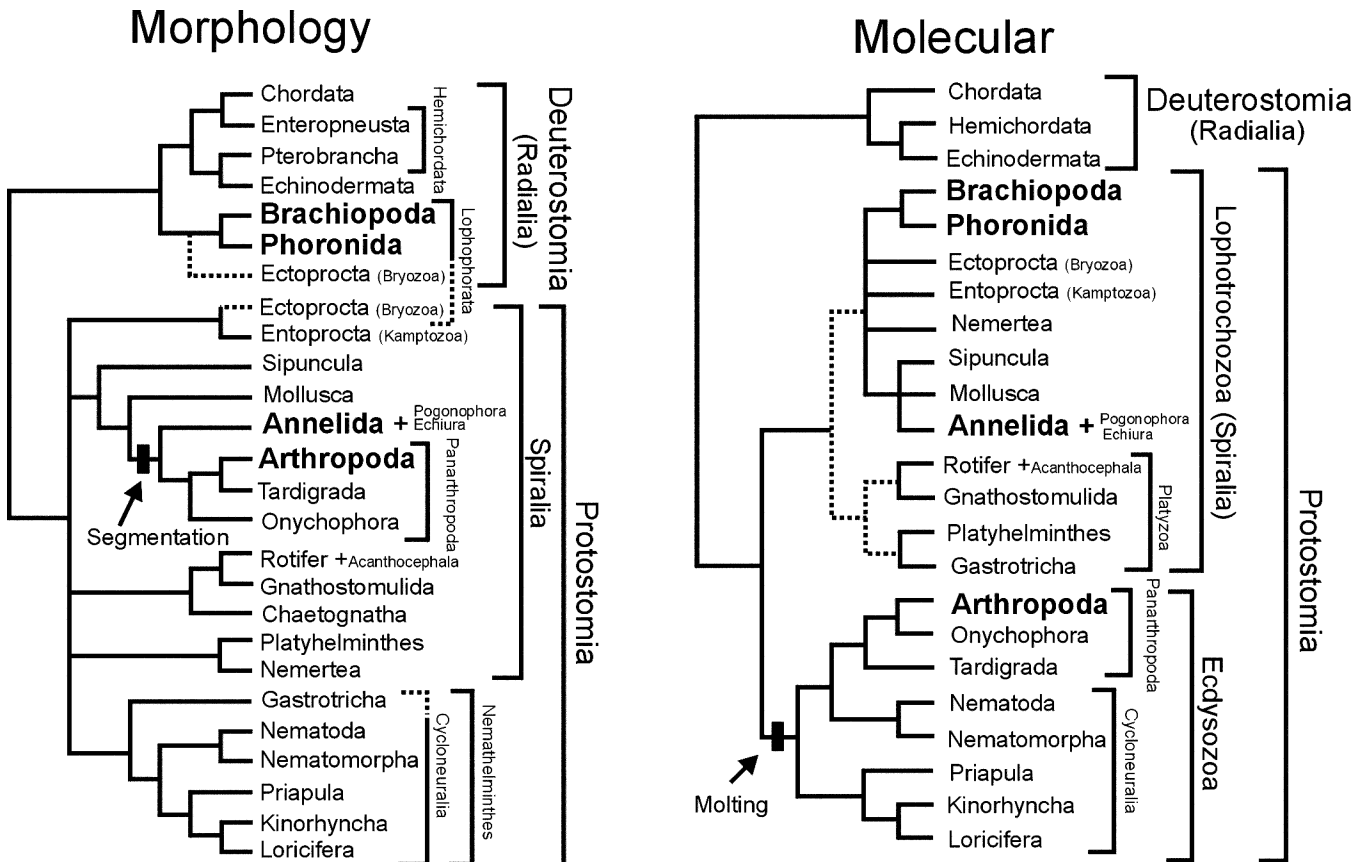


FIG. 2. Examples of conflicting trees depicting the phylogeny of the bilateral Metazoa. The main differences are in the relative locations of arthropods, annelids, brachiopods, and phoronids shown in bold in both trees. Left Panel (Morphology): Based on Nielsen (2001) in which the presence of segmentation unites Annelida and Arthropoda to form Articulata. In more traditional trees, the lophophorates (ectoprocts, phoronids, brachiopods) are a monophyletic group at the base of the deuterostomes while Nielsen places the ectoprocts within the protostomes. These differences are highlighted by dashed lines in the tree. See Table 1 for additional explanation. Names in parentheses are alternate names for the preceding taxon. Names following a taxon with the “+” sign are taxa now thought to be included within that taxon. Right Panel (Molecular): Based on Garey (2001) in which the presence of molting unites Panarthropoda and Cycloneuralia to form Ecdysozoa. Platyzoa is not completely supported by molecular analyses so is shown with dashed lines (see text for details). Names in parentheses are alternate names for the preceding taxon. Names following a taxon with the “+” sign are taxa now thought to be included within that taxon.

molecular support. Gnathifera are animals with cuticular jaws composed of tube-like rods (Sørensen, 2000), and includes Syndermata (Rotifera + Acanthocephala), Gnathostomulida, and the new taxon Micrognathozoa (see the article by Kristensen in this volume).

The placement of Nemertea continues to be controversial. Traditionally, it has been grouped with Platyhelminthes because both groups have been thought to lack a coelom. More recent molecular and morphological evidence (e.g., Turbeville *et al.*, 1992; Peterson and Eernisse, 2001) suggests that the two groups are both protostomes but not closely related, while Nielsen (2001) continues to place nemerteans as the sister group to the flatworms as the clade Parenchymia.

The old term “Aschelminthes” refers to a polyphyletic assemblage of mostly meiofaunal size animals which includes those in Cycloneuralia, Gnathifera, Gastrotricha and sometimes additional taxa as well.

Neither molecular nor morphological studies support “Aschelminthes” as a legitimate taxon, so “Aschelminthes” is an obsolete term and should no longer be used (Ruppert, 1991).

#### THE USE OF LESSER-KNOWN TAXA IN RESEARCH

The importance of the lesser known taxa is becoming apparent in testing some of the ideas mentioned in the previous section. For example, developmental biologists have examined the hox gene clusters in a brachiopod and a priapulid. These were compared to hox genes in representatives of more well known phyla with results that support the clade Ecdysozoa and the hypothesis that lophophorates are likely to be protostomes rather than deuterostomes (deRosa *et al.*, 1999). Similarly, onychophorans have been used in developmental studies of the evolution of appendages (Panganiban *et al.*, 1997) and of body segmentation (Grenier and Carroll, 2000).

With the completion of the *Drosophila* and *Caenorhabditis elegans* genome projects, it is inevitable that some genome researchers will turn their attention toward some of the lesser-known taxa. In the area of ecology, the importance of meiofaunal taxa is being realized (e.g., Austen *et al.*, 2002; Shirley, 1990), and molecular techniques offer the potential for new ways of identifying and studying these microscopic animals. Advances in biochemistry have led to the discovery and extensive study of entire communities of animals that are based on sulfide metabolism in the marine hydrothermal vent systems. For example, bacterial endosymbionts as found in the giant vent Pogonophora (Jones, 1981) are also present in many meiofaunal oligochaetes, nematodes and flatworms (Dubilier *et al.*, 2001; Hentschel *et al.*, 1999; Ott *et al.*, 1982). In recent years, several taxa of animals thought to be separate phyla allied in some way to annelids have been shown to be derived annelids (e.g., Echiura and Pogonophora, see McHugh, 1997). This has led to some confusion as to how they should be treated in invertebrate zoology classes, and this symposium proved to be an excellent venue to discuss these problems.

#### DISCUSSION AT THE SYMPOSIUM

At the end of the symposium a lengthy discussion concerning lesser-known taxa lasted until the room had to be vacated to make room for another scheduled event. The discussion emphasized increased interest in utilizing lesser-known groups, and it was agreed that the symposium did a good job bringing together systematists who study the taxa, and researchers who focus on evolutionary-developmental problems. Systematists need to make an effort to publish their findings in more widely read journals. For example, many important papers on metazoan phylogeny have been published in highly specialized journals or as articles in volumes that are not included in literature indexes (e.g., Dougherty *et al.*, 1963; Ehlers *et al.*, 1996; Ruppert, 1991) and are often overlooked. Systematists also should collaborate more with other researchers who use their animals, and it would be helpful if cultures of these animals could be made available. Many cannot currently be cultured and there should be more effort in developing ways of culturing more of them. There are a number of ways that investigators can get in touch with people who work with these organisms other than the traditional literature. SICB is currently developing the Bio-Portal (<http://sibc.org/faq.php3>) to put researchers in touch with each other. The Census of Marine Life sponsored by the Sloan Foundation (<http://www.coreocean.org/censhome.html>) and the Fauna Europaea (<http://www.faunaeur.org>) project also provide information about researchers studying lesser-known invertebrates.

The importance of educating students and the faculty who teach invertebrate zoology cannot be overlooked. One problem is finding drawings and photographs of the lesser-known taxa for educational use. Jon Houseman at the University of Ottawa is involved

in BIODIDAC, a bank of digital images, video, and animations that can be used and adapted for teaching without charge. Anyone with images to contribute can contact Jon Houseman through the website (<http://biodidac.bio.uottawa.ca/>). Many of the lesser-known groups that used to be dumped into the old "Aschelminthes" can now be related to other groups such as Syndermata, Platyzoa, Cycloneuralia, Panarthropoda and Ecdysozoa. This makes it much easier for students to learn about them, and many of these new ideas are presented in current introductory biology texts (e.g., Campbell and Reece, 2002), invertebrate zoology texts such as the fourth edition of *Biology of the Invertebrates* by Pechenik (2000) and the upcoming seventh edition of *Invertebrate Zoology* by Ruppert and Barnes.

Another suggestion was that some terms, particularly "Aschelminthes" as noted above, should be dropped from use since it does not represent a monophyletic group. The term Polychaeta may no longer be valid because Polychaeta is likely a synonym for Annelida (see the paper by Halanych *et al.*, in this volume). If the lophophorates are indeed polyphyletic, and phoronids are simply "shell-less" brachiopods, then use of the term Lophophorata may also need to be discontinued. Clearly, many taxon names are currently in flux, and the use of the conventional taxonomic levels of absolute hierarchy such as Phylum, Class, Order and Family is inconsistent and troublesome.

With all the changes and new ideas in phylogeny, new molecular tools, and with the continued discovery of new taxa (for example, Reinhardt Kristensen introduced a new group of animals, the micrognathozoans, at this symposium that were discovered in a freshwater spring in Greenland), interest appears strong in continuing symposia on this topic at approximately five year intervals, perhaps rotating different taxa at different meetings.

#### HISTORY OF THE SYMPOSIUM AND A TRIBUTE TO ROBERT P. HIGGINS

This symposium was organized in response to requests and inquiries from a number of people at the 1998 SICB symposium on Metazoan evolution (McHugh and Halanych, 1998). The symposium has a history going back to 1976 when Robert Higgins organized a refresher course on the lesser-known invertebrates at a joint meeting of the American Institute of Biological Sciences and the American Microscopical Society (AMS) that took place in New Orleans in 1976, and most recently in 1990 at a joint meeting of the American Society of Zoologists and the AMS in San Antonio that was also organized by Higgins. He has been instrumental in promoting the study of lesser-known taxa over the years. As a graduate student, he was present at the well-known symposium on the "Lower Metazoa" in Berkeley (see Dougherty *et al.*, 1963), which took place in 1961 and included such noted scientists as Adolf Remane, Peter Ax, and others



FIG. 3. Left to right: Otto Steinböck, Peter Ax (with binoculars), Adolf Remane, Tor Karling, Robert Higgins. This photo was taken near Pacific Grove, CA in September of 1960 during the Second Annual Symposium on Comparative Biology of the Kaiser Foundation Research Institute. See Dougherty *et al.* (1963) for the published proceedings. Photo courtesy of Robert Higgins.

(Fig. 3). Higgins had a long career at the Smithsonian Institution, was very active in the American Society of Zoologists (now SICB) and played a major role in the discovery of the phylum Loricifera. Despite being retired, Bob Higgins was invited to speak at this symposium, and although he was unable to attend, he co-authored a contribution with Birger Neuhaus in these proceedings on kinorhynchs. Bob Higgins provided a short biography/history of his career, which is of interest to anyone who studies the lesser-known animal groups so I have included it here in its entirety:

**How the “lesser knowns” became “better-known” by Robert P. Higgins.**

My professional introduction to invertebrates occurred in 1953 during an ecology course taught by Robert W. Pennak at the University of Colorado, Boulder. Each student had to design a research project. During this time, I was working in the university’s cryptogamic botany lab—processing mosses and lichens for the museum at 35 cents an hour. I asked Dr. Pennak for some ideas for a project and he told me to look at the washings from the mosses and lichens—he assured me that if done carefully, I would find some very interesting invertebrates occupying this habitat. Sure enough, I found tardigrades, thousands of tardigrades, along with bdelloid rotifers, nematodes, water mites, chironomid larvae, and a few others long forgotten.

The next year, I enrolled in Pennak’s two-semester course in Invertebrate Zoology. I was now a junior at the university and by the end of that year I decided to study tardigrades as a thesis subject for my Master’s degree. Dr. Pennak was very pleased and went on to tell me that there were numerous major taxa that were not as uncommon as were scientists interested in studying them. And that I should study tardigrades. And so I did.

I was awarded a James B. Duke Fellowship at Duke University in the fall of 1958. I had planned on working on marine tardigrades and possibly tardigrade embryology, but along the way, thanks to an NSF Summer Fellowship at Friday Harbor Laboratories, I found kinorhynchs, thousands of kinorhynchs, and decided that since even fewer people had studied them, they would be a good addition to my academic interests. And so I did.

And now to connect all of this to today’s symposium. At Duke University, my doctoral advisor, C. G. Bookhout, thought that a 1-semester-hour seminar course in “lesser-known” invertebrates might be useful to the half-dozen students working with him at the time. It turned out to be every bit as interesting as he predicted. I was assigned the Kinorhyncha, Tardigrada and Priapulida as topics to present. And so I did.

Over the next fifteen years or so I remained focused on the above groups. Several interesting an-

ecdotal stories connect me historically with the discoveries of *Tubiluchus* and *Macabbeus* in the 1960s, both aberrant priapulids. Both genera qualified for inclusion as meiofauna and their obvious relationship with the Kinorhyncha suggested that they would be interesting to add to my repertoire. And so I did.

In May of 1974, I saw the first species (an adult) of what was to become the phylum Loricifera about ten years later. The following year Reinhardt Kristensen also found a representative (a larva) and once we got our act in order, Dr. Kristensen published the discovery. During my doctoral thesis defense I had rather overconfidently talked about theoretical kinorhynch-related meiofaunal taxa, kinds of beasts we were likely to encounter someday if we looked carefully enough and/or in the right places. The loriciferan I saw in 1974, *Pliciloricus enigmaticus*, was the first of these wild predictions come true. About this same time, several colleagues and I decided it would be of great interest if we assembled experts in “lesser-known” invertebrates in order to present a “refresher course” on this subject at the annual meeting of the American Institute of Biological Science and the American Microscopical Society (AMS) to be held in New Orleans in 1976. I was asked to put it all together. And so I did.

The primary purpose of the “refresher-course” was to fill in the gaps left by invertebrate textbooks or lectures on minor phyla and to present up-to-date information on these taxa as the basis for updating the teaching of invertebrate zoology. At the time, most courses covered about twelve phyla adequately and simply mentioned or ignored the remaining ones thereby giving the student the impression that these were either rare, difficult to find, difficult to work with, or that no experts were available as mentors.

The first presentation on the “lesser-known invertebrates” took place on 31 May 1976 in New Orleans. Speakers and subjects included William Hummon (Gastrotricha), Bradford Calloway (Priapulida), Curtis Swanson (Nematomorpha), Wolfgang Sterrer (Gnathostomulida), Al Grigarick (Tardigrada), Edward Cutler (Sipuncula), and me (Kinorhyncha). There may have been others, but I no longer have the information in my personal files, just some names and whatever in my little black book for that year. The morning session had several empty seats. But by afternoon, word had gotten around and the place was overflowing. I was told that I should repeat this type of presentation every so often. And so I did.

The next time, December, 1984, the “Refresher course on lesser-known invertebrates” was presented in conjunction with the annual meetings of the American Society of Zoologists (ASZ) and AMS in Denver. Here, I have a more accurate list of participants: Edward Ruppert (Gnathostomulida), William Hummon (Gastrotricha), Steve Gardner (Archian-

nelida), Bradford Calloway (Priapulida), Reinhardt Kristensen (Tardigrada and Loricifera), Mary Rice (Sipuncula), John Pilger (Echiura), Meredith Jones (Pogonophora), Susan Lester (Pterobranchia), Patricia Morse (Aplacophora) and me (Kinorhyncha). Another success. The highlight of the event was the presentation on then, the newest phylum in the animal kingdom, the Loricifera, by my post-doctoral student, Reinhardt Kristensen. “Well, Bob,” they said, “better start thinking about the next workshop in another six to eight years.” And so I did.

The last of my efforts was a presentation at the annual meetings of ASZ and AMS at San Antonio in 1990. I remember only a few of the colleagues who made presentations, among them: William Hummon (Gastrotricha), Bradford Calloway (Priapulida), and me (Kinorhyncha). A few years later, 1993, I told myself it was time to retire. And so I did . . . or so I thought I did.

#### ACKNOWLEDGMENTS

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#### REFERENCES

- Adoutte, A., G. Balavoine, N. Lartillot, O. Lespinet, B. Prud'homme, and R. de Rosa. 2000. The new animal phylogeny: Reliability and implications. *Proc. Natl. Acad. Sci. U.S.A.* 97:4453–4456.
- Aguinaldo, A. M. A., J. M. Turbeville, L. S. Linford, M. C. Rivera, J. R. Garey, R. A. Raff, and J. A. Lake. 1997. Evidence for a clade of nematodes, arthropods and other moulting animals. *Nature* 387:489–493.
- Austen, M. C., P. J. D. Lamshead, P. A. Hutchings, G. Boucher, P. V. R. Snelgrove, C. Heip, G. King, I. Koike, and C. Smith. 2002. Biodiversity links above and below the marine sediment-water interface that may influence community stability. *Biodivers. Cons.* 11:113–136.
- Ax, P. 1996. *Multicellular animals*, Vol. I. *A new approach to the phylogenetic order of nature*. Springer-Verlag, Berlin.
- Ax, P. 2000. *Multicellular animals*, Vol. II. *The phylogenetic system of the Metazoa*. Springer-Verlag, Berlin.
- Campbell, N. A. and J. B. Reece. 2002. *Biology*, 6th ed. Benjamin Cummings, New York.
- Cavalier-Smith, T. 1998. A revised six-kingdom system of life. *Biol. Rev.* 73:203–266.
- Cuvier, G. B. 1812. Sur un nouveau rapprochement à établir entre les classes qui composent le Règne Animal. *Ann. Mus. Hist.* 19:73–84.
- de Rosa, R., J. K. Grenier, T. Andreeva, C. E. Cook, A. Adoutte, M.

- Akam, S. B. Carroll, and G. Balavoine. 1999. Hox genes in brachiopods and priapulids and protostome evolution. *Nature* 399:772–776.
- Dougherty, E. C., Z. N. Brown, E. D. Hanson, and W. D. Hartman. (eds.) 1963. *The Lower Metazoa, comparative biology and phylogeny*. University of California Press, Berkeley.
- Dubilier, N., C. Mulders, T. Ferdelman, D. de Beer, A. Pernthaler, M. Klein, M. Wagner, C. Erseus, F. Thiermann, J. Krieger, O. Giere, and R. Amann. 2001. Endosymbiotic sulphate-reducing and sulphide-oxidizing bacteria in an oligochaete worm. *Nature* 411:298–302.
- Ehlers, U., W. Alrichs, C. Lemburg, and A. Schmidt-Rhaesa. 1996. Phylogenetic systematization of the Nematelminthes (Aschelminthes). *Verh. Dtsch. Zool. Ges.* 89.1:8.
- Funch, P. and R. M. Kristensen. 1995. Cycliophora is a new phylum with affinities to Entoprocta and Ectoprocta. *Nature* 378:711–714.
- Garey, J. R. 2001. Ecdysozoa: The relationship between Cycloneuralia and Panarthropoda. *Zool. Anz.* 240:321–330.
- Garey, J. R. and A. Schmidt-Rhaesa. 1998. The essential role of “minor” phyla in molecular studies of animal evolution. *Amer. Zool.* 38:907–917.
- Garey, J. R., A. Schmidt-Rhaesa, T. J. Near, and S. A. Nadler. 1998. The evolutionary relationships of rotifers and acanthocephalans. *Hydrobiologia* 387/388:83–91.
- Giribet, G., D. L. Distel, M. Polz, W. Sterrer, and W. C. Wheeler. 2000. Diploblastic relationships with emphasis on the acoelomates and the position of Gnathostomulida, Cycliophora, Plathelminthes, and Chaetognatha: A combined approach of 18S rDNA sequences and morphology. *Syst. Biol.* 49:539–562.
- Grenier, J. K. and S. B. Carroll. 2000. Functional evolution of the Ultrabithorax protein. *Proc. Natl. Acad. Sci. U.S.A.* 97:704–709.
- Halanych, K. M., J. D. Bacheller, A. M. A. Aguinaldo, S. M. Liva, D. M. Hillis, and J. A. Lake. 1995. Evidence from 18S ribosomal DNA that the lophophorates are protostome animals. *Science* 267:1641–1643.
- Hentschel, U., E. C. Berger, M. Bright, H. Felbeck, and J. A. Ott. 1999. Metabolism of nitrogen and sulfur in ectosymbiotic bacteria of marine nematodes (Nematoda, Stilbonematinae). *Mar. Ecol. Prog. Ser.* 183:149–158.
- Hyman, L. H. 1951. *The Invertebrates*, Vol. I. McGraw-Hill, New York.
- Jones, M. L. 1981. *Riftia pachyptila* Jones: Observations on the vestimentiferan worm from the Galapagos rift. *Science* 213:333–336.
- Mackey, L. Y., B. Winnepeninckx, I. Backeljau, R. De Wachter, P. Emschermann, and J. R. Garey. 1996. 18S rRNA suggests that Entoprocta are Protostomes, unrelated to Ectoprocta. *J. Mol. Evol.* 42:552–559.
- McHugh, D. 1997. Molecular evidence that echiurans and pogonophorans are derived annelids. *Proc. Natl. Acad. Sci. U.S.A.* 94:8006–8009.
- McHugh, D. and K. M. Halanych. 1998. Introduction to the symposium: Evolutionary relationships of metazoan phyla: Advances, problems, and approaches. *Amer. Zool.* 38:813–817.
- Nielsen, C. 1995. *Animal evolution*, 1st ed. Oxford University Press.
- Nielsen, C. 2001. *Animal evolution*, 2nd ed. Oxford University Press.
- Ott, J., G. Rieger, R. Rieger, and F. Enderes. 1982. New mouthless interstitial worms from the sulfide system: Symbiosis with prokaryotes. *P.S.Z.N.I: Mar. Ecol.* 3:313–333.
- Panganiban, G., S. M. Irvine, C. Lowe, H. Roehl, L. S. Corley, B. Sherbon, J. K. Grenier, J. F. Fallon, J. Kimble, M. Walker, G. A. Wray, B. J. Swalla, M. Q. Martindale, and S. B. Carroll. 1997. The origin and evolution of animal appendages. *Proc. Natl. Acad. Sci. U.S.A.* 94:5162–5166.
- Pechenik, J. 2000. *Biology of the invertebrates*, 4th ed. McGraw-Hill, New York.
- Peterson, K. J. and D. J. Eernisse. 2001. Animal phylogeny and the ancestry of bilaterians: Inferences from morphology and 18S rDNA gene sequences. *Evol. Dev.* 3:170–205.
- Ruppert, E. E. 1991. Introduction to the aschelminth phyla: A consideration of mesoderm, body cavities, and cuticle. *In* F. W. Harrison and E. E. Ruppert (eds.), *Microscopic anatomy of invertebrates*, Vol. 4, *Aschelminthes*, pp. 1–17. Wiley-Liss, New York.
- Shirley, T. C. 1990. Ecology of *Priapulius caudatus* Lamarck, 1816 (Priapulida) in an Alaskan subarctic ecosystem. *Bull. Mar. Sci.* 47:149–158.
- Schmidt-Rhaesa, A., U. Ehlers, T. Bartolomaeus, C. Lemburg, and J. R. Garey. 1998. The phylogenetic position of the Arthropoda. *J. Morphol.* 238:263–285.
- Sørensen, M. V. 2000. An SEM study of the jaws of Haplognathia rosea and Rastrognathia macrostoma (Gnathostomulida), with a preliminary comparison with the rotiferan trophi. *Acta Zool.* 81:9–16.
- Turbeville, J. M., K. G. Field, and R. A. Raff. 1992. Phylogenetic position of phylum Nemertini, inferred from 18S rRNA sequences: Molecular data as a test of morphological character homology. *Mol. Biol. Evol.* 9:235–249.
- Winnepeninckx, B. M. H., T. Backeljau, and R. M. Kristensen. 1998. Relations of the new phylum Cycliophora. *Nature* 393:636–637.
- Zrzavy, J., S. Mihulka, P. Kepka, A. Bezdek, and D. Tietz. 1998. Phylogeny of the Metazoa based on morphological and 18S ribosomal DNA evidence. *Cladistics* 14:249–285.