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ANNELIDA AND ARTHROPODA ARE NOT SISTER TAXA: A PHYLOGENETIC ANALYSIS OF SPIRALIAN METAZOAN MORPHOLOGY

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Abstract.—Annelids and arthropods have long been considered to be each other's closest relatives, as evidenced by similarities in their segmented body plans. In the first cladistic analysis of metazoan morphology accompanied by an explicit data matrix, Schram (Meglitsch and Schram, 1991, Invertebrate zoology, 3rd edition, Oxford Univ. Press, New York) suggested tentative support for this conventional "Articulata" hypothesis. Our reanalysis of the Schram data matrix yielded weak support for an alternative "Eutrochozoa" grouping of annelids, molluscs, and certain other spiralian phyla, exclusive of arthropods. Likewise, recent 18S ribosomal RNA sequence comparisons have favored the Eutrochozoa hypothesis. This study presents a new analysis of 141 independently assembled characters, purported to represent the current state of knowledge of metazoan morphology and embryology. This maximum parsimony analysis resulted in robust support of Eutrochozoa. For this data compilation and method of analysis, the Articulata hypothesis could only be supported by adding multiple ad hoc proposals of evolutionary events. Instead, the more parsimonious Eutrochozoa hypothesis is favored as the best-supported current reconstruction of higher level animal genealogy. [Phylogeny; Metazoa; animal; Arthropoda; Annelida; Mollusca; morphology; embryology; RNA.]

An enormous literature of descriptive and experimental work on the ontogeny and morphology of animals has accumulated over the last century and a half. Much is now known about the life history, anatomy, and genetic organization of species in most of the more than 30 recognized animal phyla. Our modern interpretation of character homology rests on the results of these studies and on the understanding of animal evolution they make possible. The recent emphasis of cladistic methods in systematic biology, however, has focused new attention on the evidential basis for asserting hypotheses of homology among characters (Wiley, 1981; Patterson, 1982). The criteria for homology formalized by Remane (1956) involve evaluation of similarities of ontogeny, composition, and anatomical position on a case-by-case basis. These similarities are now regarded by many systematists to be preliminary to the test of character congruence on a cladogram (Hennig, 1966; Patterson, 1982), and only shared derived characters (synapomorphies) are considered candidates for homology. Other similarities that result from independent or parallel evolution (homoplasies) or are retained from more ancient evolutionary transformation (plesiomorphies) are not considered relevant even when they are phenotypically identical.

Studies on the early evolution of major animal lineages and on the origins of important features in their structural design have only recently begun to incorporate the character congruence approach of evaluating phenotypic similarities. One example is the investigation of genealogical relationships among spiralian metazoans, including Arthropoda, Mollusca, Annelida, and several other less speciose phyla. Spiralians comprise >90% of all living metazoan (multicellular animal) species (Barnes, 1987; Brusca and Brusca, 1990), yet the genealogical relationships among many of them remain undocumented by phylogenetic criteria. Members of the Spiralia have also been grouped as Protostomia by

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Grobben (1909) on the basis of certain embryological similarities, especially spiral cleavage of the blastomeres, derivation of the mesoderm from a single (4d mesentoblast) cell, and protostomous mouth formation.

The overt segmental arrangement of parts in the adult body plan of Annelida and Arthropoda, but not Mollusca, led Cuvier (1817:508) to erect "Les articulés" as "le troisième grande division du règne animal." Later, Haeckel (1866) popularized the use of "articulates" as an evolutionary lineage, which in his opinion rivaled the vertebrates as among the greatest of his 12 stems of the animal kingdom. Although its exact composition has varied in the eyes of subsequent authors, the "Articulata" superphylum-level grouping is still widely used. Some nomenclatural problems have arisen from the proposal of an alternative name (Hadži, 1963) as well as the use of "Articulata" in brachiopod taxonomy (Huxley, 1869). More recently, some zoologists have elevated Articulata to include most spiralian taxa, even including Mollusca (Nielsen, 1985; Ghiselin, 1988). Although we do not dispute the potential value of this suggestion, Articulata is used here in the conventional sense as a hypothesis that axial mesodermal segmentation arose but once in the shared common ancestor of Annelida and Arthropoda, perhaps also including Pogonophora, Tardigrada, and Onychophora, but not Mollusca.

The near universal use of the Articulata hypothesis in textbooks and classrooms of this century has left a deep impression on the interpretation of patterns and processes of animal evolution by zoologists. From Haeckel's perspective as an advocate of the biogenetic law, the observation that annelids and arthropods pass through an unsegmented early ontogenetic stage is evidence that segmentation is derived with respect to the molluscan condition. This, of course, reinforced his opinion that segmentation is homologous between the two groups.

The theory of recapitulation is no longer emphasized, yet a survey of recent invertebrate zoology textbooks (Lutz, 1986; Barnes, 1987; Pearse et al., 1987; Brusca and Brusca, 1990; Kozloff, 1990; Meglitsch and Schram, 1991) revealed that the Articulata hypothesis continues to dominate discussions of animal relationships. Although these texts differ in their treatment of hypotheses on the origin of character systems such as the coelom and mesoderm, there is an apparent unanimity regarding the origin of spiralian segmentation. The union of Annelida and Arthropoda, exclusive of Mollusca, is one of the few consistent elements in the diversity of their summary phylogenetic diagrams. Some of these authors (e.g., Barnes, 1987) even echo Haeckel's (1866) depiction of Arthropoda emerging directly from Annelida, although few researchers today would posit the derivation of one higher taxon from another (Ghiselin, 1974; Wiley, 1981).

The hypothesis of a monophyletic Articulata (Figs. 1a, 1b) has yet to be documented by an analysis of data using the character congruence approach. For example, Brusca and Brusca (1990:882) presented one of the first character-labeled branching diagrams for metazoans. Nonetheless, it is not clear whether the phylogenetic distribution of characters was used to build the tree. The reader is left to conclude with the authors (1990:682) that "[t]here is little argument that annelids and arthropods are closely related" and that "[t]he body plans of these two phyla are more similar to one another than to any other major protostome group." This conclusion is reinforced elsewhere (1990:765): "molluscs probably arose early in the protostome clade, soon after the origin of the coelom but *before* the origin of annelid–arthropod metamerism" (emphasis in original). In the final analysis, however, a single character diagnosing a clade including Annelida and Arthropoda (with Pogonophora) was proposed, "true segmentation arising by teloblastic growth and resulting in serial repetition of body parts," although conflicting data were not reported.

An older alternative view (Pelseneer, 1899, 1906; Naef, 1913, 1924), more re-

cently advocated by investigators who have examined 18S ribosomal RNA (rRNA) data (Field et al., 1988, 1989; Ghiselin, 1988; Patterson, 1989; Raff et al., 1989; Lake, 1990; Eernisse, unpubl. manuscript), posits that annelids, molluscs, and certain other less speciose phyla share a more recent common ancestor with one another than any do with arthropods. This assemblage (Figs. 1c, 1d) was referred to as "Eutrochozoa" by Ghiselin (1988) and approximately coincides with a long-standing grouping of those taxa with a trochophore larva in at least some marine representatives of each group (Hatschek, 1891; De Beer, 1930; Hyman, 1951; Gruner, 1982; Nielsen, 1987; Strathmann, 1987; cf. Salvini-Plawen, 1980b). Although ciliary bands have been reported from widely divergent metazoan taxa, no species of arthropod is known to possess any element of the trochophore larva.

Because of the incongruent character distribution of trochophore larvae and segmented body plans among spiralian taxa, Articulata and Eutrochozoa are mutually exclusive hypotheses of relationships. The Articulata hypothesis requires that the lack of a trochophore larva in arthropods be explained either by its independent derivation in molluscs and annelids (Fig. 1a) or by its derived loss in arthropods (Fig. 1b). If a trochophore larva and segmentation are each viewed as unitary characters, these two hypotheses are equally parsimonious, each requiring three steps.

The hypothesis of a monophyletic Eutrochozoa also requires at least three transformations (Figs. 1c, 1d). Either mesodermal segmentation is an independent derivation in annelids and arthropods or Eutrochozoa is primitively segmented, with molluscs exhibiting a derived loss of this phenotype. Members of some extant molluscan taxa do possess elements of serially arranged (or possibly metameric) structures, especially Polyplacophora, Monoplacophora, and Nautiloidea (Cephalopoda) (Naef, 1924; Wingstrand, 1985).

The third logical alternative is that molluscs and arthropods are more closely related than either is to annelids. This

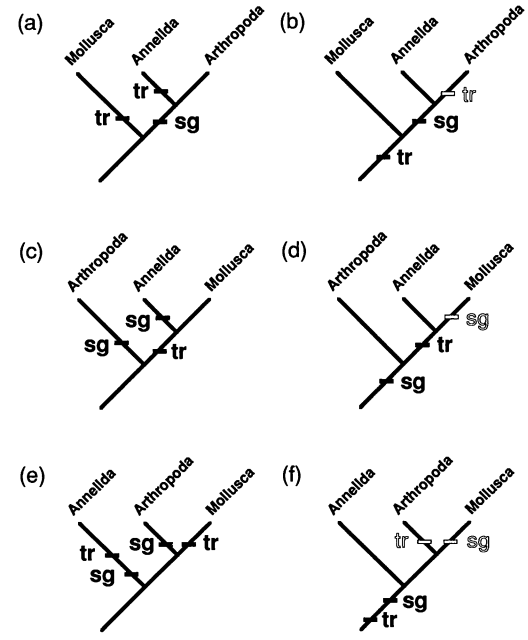


FIGURE 1. Diagrammatic cladograms of the three possible resolved hypotheses of branching relationships among Arthropoda, Annelida, and Mollusca, assuming each is monophyletic with respect to the others, with the phylogenetic distributions of two characters plotted; tr = trochophore larva, sg = mesodermal segmentation. A solid bar indicates derived presence, an outlined bar indicates derived loss. (a), (b) Articulata. (c), (d) Eutrochozoa. (e), (f) Two of the four most-parsimonious distributions of these characters required by an Arthropoda–Mollusca grouping exclusive of Annelida.

hypothesis requires at least four transformations (only two of the four possible combinations of losses or gains of trochophore larvae and segmentation are shown in Figs. 1e, 1f). Although less parsimonious for explaining these particular attributes, this hypothesis is more consistent with still other character evidence, such as the distribution of hemocyanin respiratory pigments, which is known only from Mollusca and Arthropoda (Mangum, 1985; Ghiselin, 1989). This third hypothesis was developed shortly after the discovery of living monoplacophorans and arose from observations of serial repetition in several of their organ systems (Lemche, 1959a, 1959b; Fretter and Graham, 1962). This view considers molluscs to be derived from

short-bodied segmented animals with very small coelomic sacs and an open circulatory system.

Objections to viewing molluscs as primitively metameric have come from advocates for an unsegmented, possibly acoelomate, flatwormlike ancestor (Clark, 1964, 1979; Stasek, 1972; Salvini-Plawen, 1980a, 1985), perhaps similar to the wormlike aplacophoran molluscs. Recent treatments of molluscan evolution (Runnegar and Pojetta, 1985; Salvini-Plawen, 1985; Barnes, 1987; Scheltema, 1988) have tended to replace an older archetypical gastropodlike hypothetical ancestral mollusc (HAM) with a flatwormlike HAM. Although at least two of these authors have since abandoned in part their earlier published views (Runnegar, pers. comm.; Scheltema, pers. comm.) and new studies reconstructing plesiomorphic molluscan characters are being published (Wingstrand, 1985; Eernisse and Kerth, 1988), many (e.g., Willmer, 1990) continue to believe a consensus has been reached in favor of the flatworm hypothesis.

Perhaps the publication of only a single phylogenetic analysis of metazoans (Meglitsch and Schram, 1991) accompanied by an explicit matrix of morphological character hypotheses, including all spiralian phyla and appropriate outgroups, is the result of the emphasis on taxonomic specialization in the field of zoology. It is often asserted that the tremendous divergence of these taxa prohibits studies of morphological variation from recovering phylogenetically relevant information. Patterson (1990:199), for example, suggested that higher level metazoan phylogeny "has been something of a backwater for decades, largely because all the morphological clues had been pushed beyond their limits, and mutually contradictory speculations led only to dead ends."

In our view, reproducible testing of hypotheses has only just begun. A reanalysis of Schram's data set discussed herein suggests that the connection between evidence and hypotheses deserves closer scrutiny. A reanalysis of 55 characters extracted from the text of Willmer (1990) was undertaken by Wheeler (1990), who failed to find unambiguous support for the Articulata depicted in graphical form.

Our goal in this paper was to abstract from primary reports a representative accounting of embryological and morphological evidence bearing information on spiralian metazoan phylogeny. It was not our intent to present an exhaustive survey of this literature but rather a fair approximation of the current state of knowledge in the zoological community. The main purpose of this work is to recover from the available information the best hypothesis or set of hypotheses of relationships congruent with the phylogenetic distribution of variation.

METHODS

Characters used in the present analysis were extracted from the primary and secondary literature on metazoan morphology and embryology and were screened for phenotypic similarity so that patterns in their phylogenetic distribution across taxa could be examined using maximum-parsimony algorithms. A complete accounting of published information pertaining to metazoan phylogeny was complicated by the tendency of authors to omit evidence inconsistent with their narrative interpretations of character evolution. Studies whose data are best represented in the present character matrix were those with explicit, if perhaps a priori, proposals of homology (e.g., Beklemishev, 1969; Brusca and Brusca, 1990).

Character data were extracted from multiple sources, seeking to incorporate all encodable discrete characters that appeared to show variation at an appropriate level. Character descriptions were given binary alternative states, usually present or absent, accompanied by reasonably precise character descriptions (Appendix 1). The 141 binary characters were grouped into 16 morphological categories for descriptive purposes only. For the sake of brevity, the character list in Appendix 1 includes only the major citations used. To avoid rep-

etition, a few of the more general references utilized for specific taxonomic groups are listed at the start of Appendix 1.

Two assumptions regarding character independence and weighting are central to this analysis. The independence of characters was assessed by spatial or topological segregation within an organism, temporal discoupling in ontogeny, or hierarchical distinction (e.g., organs, cells, molecules). We accepted at face value the independence of characters with unique distributions across taxa. Complex morphological systems such as "trochophore larvae" and "segmentation" were considered composite suites of many phylogenetically independent characters.

For example, we distinguished between serially repeated characters ("meres" of Bateson, 1894) and segmented body plans. This distinction can be difficult because there is no consensus regarding the mechanisms leading to the production of either the segmented body plans of spiralians or serially repeated characters. Here we refer to segmentation or metamerism as the serial arrangement of similar body parts along the bilateral axis of the animal, containing congruent and repeated patterns of tissue organization. A segment (metamere) is therefore a block or unit of a segmented organism. The pattern of serially repeated characters, on the other hand, does not necessarily correlate with those of other repeated patterns in an organism. Although repeated characters are often referred to as "metameric," we prefer to restrict this term to cases of complete body segmentation, a condition referred to as "true" or "mesodermal" segmentation by Hyman (1951:28). In many cases, the phylogenetic distribution of repeated characters in one organ system is different from that of other organ systems. We took this variation as evidence that these characters do not share an identical history. Russell-Hunter and Brown (1965), Russell-Hunter (1988), and others have argued that bilateral asymmetries in numbers of repeated characters are a clear indication of "pseudometamerism," or duplication of systems not related to metamerism. We suspect that such asymmetries, whether or not metameric in origin, could instead be due to the nondeterministic consequences of inductive cues during development (Hall, 1992).

If characters are hypotheses of phylogenetic transformation, then they are also ontologically equivalent as heritable changes that occur in the evolution of a lineage. Accordingly, we weighted all characters equally and avoided using probabilistic models of character evolution (sensu Ghiselin, 1991) to guide us in weighting characters a priori. Some, no doubt, will not accept our explicit suggestion that a seemingly minor morphological feature (e.g., character 34: cilia with one basal body) should have a weight equal to that of manifestly more important evolutionary transformations (e.g., character 96: anus with proctodeum). However, a priori weighting is an epistemological issue; no claim is made that nature produces variation in all characters with equal frequency, and equal weighting is used in the absence of compelling reasons to weight otherwise. Also, equal weighting is not intended to be a null hypothesis. We have simply decided to break down the known variation into as many encodable characters as the data will permit, and this seems as good a reason as any to weight them equally. Some characters turn out to be more useful at certain levels than others, i.e., useful because they help predict the set of relationships that carry a library of phylogenetic information.

Variation in all characters was atomized into derived and plesiomorphic states to reflect specific hypotheses of evolutionary transformation. As hypotheses of unique historical events, characters are ontologically equivalent and comparable, making it possible to combine them in the study of patterns of their phylogenetic distribution in a parsimony analysis. Another methodological consequence of character equivalence involves the use of multistate characters. Additive binary coding, computationally equivalent to ordered multistate characters (Farris et al., 1970), was em-

ployed to make explicit each hypothesis of transformation (e.g., characters 5–8).

A rather broad selection of terminal taxa was chosen for the analysis, rather than restrict the analysis to only major spiralian phyla, because the relationships of spiralian phyla to other metazoans remain highly controversial (e.g., Nielsen, 1987; Lake, 1990). Inclusion of multiple outgroup taxa increases the likelihood of resolving particular ingroup relationships of interest (Swofford and Olsen, 1990). We did not attempt to address the putative monophyly of Metazoa itself, which has been brought into question by recent authors (Field et al., 1988; Lipscomb, 1989; Christen et al., 1991). In investigating the evidence for metazoan monophyly, we assembled several potential synapomorphies (characters 24–26, 73, 100, 102, 135). We intentionally left these in our matrix, although they are uninformative for this restricted metazoan comparison, with the hope that future authors will provide more details about their condition in potential protistan outgroups. Likewise, our analysis does not speak directly to the issue of the monophyly of "Bilateria" (Fig. 4: node 1) because only one taxon, Cnidaria, was included that is outside this grouping. Thus, transformations postulated between Cnidaria and the node uniting all other taxa could either be synapomorphic for bilaterians or autapomorphic for cnidarians. We did not consider some metazoan phyla, notably Entoprocta, Bryozoa, Porifera, Placozoa, Ctenophora, Rotifera, Gastrotricha, Hemichordata, Nematomorpha, Acanthocephala, Dicyemida, Orthonectida, Chaetognatha, or Loricifera. None of these phyla, except perhaps Entoprocta, are thought to be particularly closely related to the spiralian phyla considered here. Still, their inclusion may have influenced our results.

As in any phylogenetic survey, our selection of terminal taxa was constrained by the need to sample the appropriate and informative combinations of primitive and derived characters. The decision to unify or split particular lineages was based on two criteria: (1) the lineage must be thought to be monophyletic and (2) relevant variation for the question of this study must not have been reported within its representatives unless that variation could be polarized unambiguously. No attempt was made to restrict or otherwise balance the selection of terminal taxa according to Linnaean rank, numbers of species, or ecological diversity. For example, Conchifera (sensu Wingstrand, 1985) includes the divergent monoplacophorans, gastropods, cephalopods, bivalves, and scaphopods; Pogonophora includes the hot-vent inhabiting vestimentiferans; and Clitellata includes oligochaetes and leeches. Conventional taxa that were split include Arthropoda (Crustacea, Chelicerata, and Uniramia), Platyhelminthes (Acoelomorpha and Rhabditophora, sensu Ax, 1985), Aplacophora (Caudofoveata and Solenogastres), and Annelida (Polychaeta and Clitellata).

Characters for which both states are reported in different representatives of terminal taxa were scored in one of two ways. In certain cases, the results of previous studies provided compelling evidence for establishing character polarity within a taxon, permitting the assignment of the more plesiomorphic (i.e., primitive) condition to that taxon. Without such evidence, the taxon was coded as "polymorphic." The states "missing" or "inapplicable" were distinguished in Figure 3 but were treated identically (as "missing") in all analyses.

We used PAUP version 3.0r (Swofford, 1990) for all reported phylogenetic analyses. Table 1 summarizes select analyses performed on the data in Figure 3, according to the differing options used and the resulting minimum-length tree statistics. This PAUP file is available in electronic form upon request. We used the random-addition-sequence option of PAUP for stepwise addition of taxa, with 100 replicates per search, and the MULPARS option to save all minimum-length trees. The default accelerated transformation (ACCTRAN) character optimization was specified during searches, but the inferred ancestral conditions at internal nodes reported in Appendix 2 include only those that are

consistent with all three character optimization methods available in PAUP (ACCTRAN, DELTRAN, and MIN F). The analyses were duplicated in entirety, with polymorphic terminal taxa first treated as polymorphic (Tables 1, 2) and then as uncertainties, with nearly identical results except for a proportionate increase of length estimates in the polymorphic analysis. The heuristic search algorithm of PAUP was used for all analyses because the large number of taxa precluded practical use of the branch-and-bound algorithm. Using software by Eernisse (1992), we processed the PAUP output of 39 different heuristic constrained or unconstrained searches, each with 100 random-addition-sequence replicates. On average, searches found >91% of all minimum-length trees found in the first of 100 replicate searches, >97% by replicate six, and 100% by replicate 16, suggesting that the heuristic algorithm was relatively effective for our matrix but also that the replicate searches were necessary to be reasonably confident that all minimum-length trees were found. Figure 4 is presented in the form of a PAUP "phylogram," which provides information regarding the relative proportion of apomorphic characters supporting each internal node or terminal taxon. Branch lengths are best interpreted as a measure of data available at particular levels of generality rather than as a measure of anagenetic rates.

Although commonly used, bootstrapping and similar techniques have unknown statistical properties, given likely deviations from the assumptions required to use them to estimate confidence intervals on phylogenies (Felsenstein, 1988). Instead, we used the "Constraints" option of PAUP to search for the minimum number of additional evolutionary events (steps) required for various competing hypotheses, relative to the minimum-length topologies, given the character matrix and method of analysis (Table 1: analyses 2–24). We tested various combinations of constraints that, given our data matrix, forced annelids and arthropods, along with various or no additional taxa, together as a clade (analyses 2–6). Alternatively, arthropods (with or without Onycophora, Tardigrada, or Kinorhyncha) or flatworms (either Acoelomorpha or Rhabditophora, or both and with or without Gnathostomulida) were forced into a sister-taxon relationship with molluscs or more inclusive clades (analyses 7–22). Searches were constrained (analyses 23–24) to find the minimum-length tree(s) that did not support the more or less inclusive "eutrochozoan"

TABLE 1. Summary of constraint analyses of some metazoan phyla.

Analysis	Constraint[a]	Length (no. of trees)	Consistency index
1	None	384 (6)	0.458
Articulata clade			
2	(6, 17)	400 (13)	0.440
3	(6, 16)	399 (4)	0.441
4	(6, 15)	401 (12)	0.439
5	(5, 16)	394 (2)	0.447
6	(5 + 14)	393 (2)	0.448
Arthropods as eutrochozoans			
7	(7 + 14 + 18)	400 (3)	0.440
8	(6 + 14 + 18)	400 (3)	0.440
9	(10 + Nemertea + (5, 14, 18))	397 (3)	0.443
10	(5 + 14 + 18)	394	0.447
11	(5 + 14 + Nemertea + 18)	390 (3)	0.451
12	(5, (14, (Nemertea, 18)))	390 (2)	0.451
13	(5 + 9)	387 (4)	0.454
14	(5, 9)	387 (4)	0.454
Arthropod–mollusc clade			
15	(8, 18)	407 (3)	0.432
16	(7, 18)	402 (32)	0.438
17	(6, 18)	401 (16)	0.439
18	(5, 18)	397 (13)	0.443
Flatworm–mollusc clade			
19	(Rhabditophora, 18)	396 (14)	0.444
20	(11 + 18)	390 (4)	0.451
21	(10 + 18)	390 (4)	0.451
22	((10, (Nemertea, 18)), 14)	387 (4)	0.454
Not Eutrochozoa			
23	not (12)	387 (4)	0.455
24	not (13)	385 (4)	0.457

[a] Numbers represent nodes in Figure 4 of clades that were forced into sister-group relationships, as indicated by one or more sets of enclosing parentheses. Numbered clades separated by commas were each constrained to maintain their own monophyly, whereas those separated by plus signs were not. Constrained searches found minimum-length trees that satisfied (analyses 1–22) or did not satisfy (analyses 23, 24) the indicated constraint.

TABLE 2. Comparison of previous hypotheses of metazoan relationships.

Analysis	Tree as shown		Tree resolved	
	Length[a]	Consistency index	Length[b]	Consistency index
Strict consensus[c]	390	0.444	378 (3)	0.458
Hyman, 1940–1967	497	0.354	452	0.389
Hadži, 1953, 1963	543	0.324	534	0.330
Marcus, 1958	490	0.359	452	0.389
Salvini-Plawen, 1982, 1985	469	0.375	441 (3)	0.399
Nielsen, 1985, 1987	459	0.383	424	0.415
Barnes, 1987	465	0.378	417	0.422
Pearse et al., 1987	440	0.400	419	0.420
Brusca and Brusca, 1990	437	0.403	410 (5)	0.429
Kozloff, 1990	453	0.389	419 (2)	0.420
Willmer, 1990	649	0.271	449 (2)	0.392
Meglitsch and Schram, 1991	448	0.393	443	0.397
Meglitsch and Schram[d]	481	0.366	403	0.437

[a] See Figures 4 and 5. Reported value is the length of a figured tree when optimized on the data matrix in Figure 3.
[b] Length of minimum-length tree(s) found during searches constrained to preserve all clades contained in the figured tree but allowing any polytomies to be further resolved to the one or more hypotheses requiring the fewest postulated steps when optimized on the data matrix in Figure 3. Number in parentheses is the number of equally parsimonious minimum-length trees found, if greater than one.
[c] See Figure 4.
[d] See Figures 2a and 5l.

groupings that resulted from unconstrained searches (analysis 1).

To compare the results of this study with those of previous efforts, we present certain published hypotheses in the form of cladograms, with taxa limited to those in our analyses. Because the original trees were drawn in diverse styles, we did our best to recreate the trees as cladogram interpretations of the original drawings, with the following caveats. We entered unresolved groupings as polytomies and used the accompanying text discussions to clarify uncertainties or to confirm the placement of unresolved basal taxa. Some authors clearly postulated paraphyletic concepts for certain taxa, which often led to difficulties in translating their diagrams into cladograms. Depending on the case, we either reduced resolution to a polytomy or treated them as if the authors had intended them to be monophyletic concepts. In other cases, we found no reference to certain taxa (Hyman, 1951: Tardigrada, Onychophora; Salvini-Plawen, 1982, 1985: Priapula, Tardigrada, Gnathostomulida, Pogonophora, Nematoda). To make tree-length comparisons possible, we added these taxa in a position consistent with our strict consensus tree. A special case was our replacement of Solenogastres, Caudofoveata for Brusca and Brusca's (1990) Aplacophora, Caudofoveata, which we attributed to typographical error. Once the topologies were created, we calculated tree statistics as optimized on our data matrix using Cnidaria as an outgroup. Figure 5 presents the assembled topologies, with all but one rooted with Cnidaria; Hadži (1953, 1963) (Fig. 5b) advocated Acoelomorpha as the root, but because our characters were all binary (albeit in some cases additive binary) and undirected, the tree length was independent of rooting in this case.

Table 2 presents tree lengths that were computed for Figures 5a–k and Figure 2a as depicted when optimized on our data matrix. These values are sometimes misleading because various authors often (but not always) intended polytomies to represent their uncertainties of relationships rather than explicit hypotheses of independent character evolution. To make comparison of these cases with our minimum-length results more meaningful, we also computed tree lengths of the most resolved hypothesis or hypotheses resulting from an analysis in which only the clades

they depicted were used to constrain the search for shorter length trees.

During completion of our study, Schram's (Meglitsch and Schram, 1991; Schram, 1991) analysis of metazoan phylogeny was published. Concurrent with our own analysis, we entered Schram's matrix and performed an independent analysis of his data matrix. Because the reanalysis of Schram's matrix resulted in multiple minimum-length trees, we calculated strict and 50% majority-rule consensus trees (Swofford and Olsen, 1990) as result summaries (Fig. 2).

RESULTS

Reanalysis of Schram's Morphology Matrix

Schram (Meglitsch and Schram, 1991) presented results of two cladistic analyses of the same matrix of 77 binary morphological characters. The first assumed all characters were undirected, for which Schram presented a single resulting cladogram of length 140. The second analysis, resulting in a cladogram of length 158, assumed complete irreversibility of all characters. One curious result for both cladograms was that Onychophora was nested within Arthropoda as the sister taxon to Uniramia, with Crustacea basal. Another was that Pogonophora was the sister taxon of Arthropoda. The first cladogram indicated general support for Articulata, with Annelida the sister taxon of an Arthropoda–Pogonophora clade. In contrast, the second cladogram placed Annelida with Mollusca, along with Sipuncula and Echiura, as members of a clade. Although the second analysis favors the alternative Eutrochozoa hypothesis, it is subject to the criticism that (as Schram himself points out) the assumption of character irreversibility is a burdensome one, difficult to justify and seldom used because it yields solutions that are far from the most-parsimonious ones.

Our reanalysis confirmed that with all characters treated as unordered as reported Schram's cladogram did indeed have a length of 140 as analyzed with PAUP 3.0 (Swofford, 1990). In addition, we found 1,422 alternative topologies of length 139 (and many more than 10,000 alternative trees of length 140). As expected from this large number of minimum-length trees, a strict consensus of all trees of length 139 (Fig. 2a) expressed little resolution within the spiralian phyla, leaving no support for either the Articulata or Eutrochozoa hypothesis or for the proposed placements of Onychophora and Pogonophora. A majority-rule consensus tree (Fig. 2b) showed support for the Eutrochozoa hypothesis (including Echiura and Sipuncula) in 96% of the 1,422 trees.

Morphological Analysis

For the analysis of our independently assembled matrix of morphological and embryological data, 141 characters (Appendix 1, Fig. 3) were compiled, emphasizing variation among spiralian phyla and effectively employing nonspiralian metazoans as multiple outgroups. The parsimony analysis resulted in six minimum-length trees of length 384 steps (or 342 if taxa coded as polymorphic were treated as having uncertain states), the strict consensus of which is presented in Figure 4. Topological variation observed among the six trees is less than that implied by the three unresolved polytomies represented in the strict consensus. These differences are restricted to the branching order within a clade composed of Chordata, Echinodermata, Brachiopoda, and Phoronida (Fig. 4: node 2), the placement of Priapula (Fig. 4: node 3), and the branching order of the three extant arthropod subphyla (Fig. 4: node 8). Trees 1 and 2 of the six minimum-length trees supported a "lophophorate" clade as the sister taxon to Echinodermata, (Chordata (Echinodermata (Brachiopoda, Phoronida))), whereas tree 3 supported the reverse branching order, (Brachiopoda (Phoronida (Echinodermata, Chordata))), in which lophophorates are paraphyletic. Tree 1 placed Priapula as the sister taxon to clade 4 (Fig. 4), whereas trees 2 and 3 placed Priapula as the sister taxon to a more inclusive clade, itself composed of clades 4 and 9. Trees 4–6 mirrored topological variation among the first three trees but differed in supporting a "mandibulate" ar-

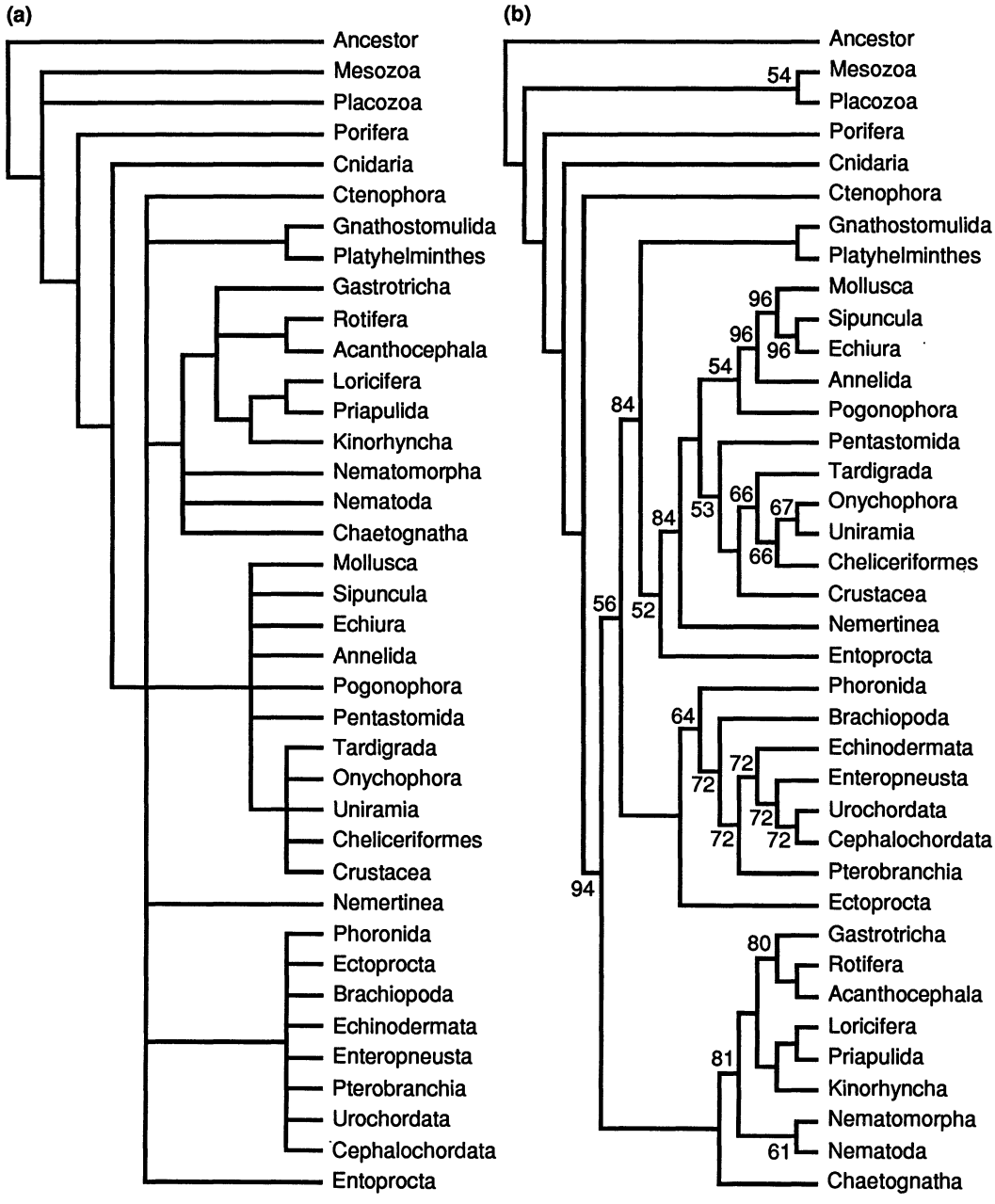


FIGURE 2. Consensus cladograms for our reanalysis of the Schram (Meglitsch and Schram, 1991) data matrix. All 77 characters for 38 taxa were binary and treated as unordered, with a hypothetical ancestor declared as an outgroup to root each otherwise unrooted network topology. (a) Strict consensus diagram for the 1,422 equal and minimum-length trees found, each with a length of 139 and a consistency index of 0.554. (b) The 50% majority-rule consensus diagram, depicting only those nodes supported in at least 50% of the minimum-length topologies. Only nodes supported in less than 100% of the 1,422 trees are labeled with the percentage value.

```
                ....*....1....*....2....*....3....*....4....*....5....*....6....*....7
Cnidaria        00100000000?0NNNNNNNNNN11100110000010000000000000000000000000000000000
Chordata        ------------11101000000---1-------------P-------1-1----------1-1-1----
Echinodermata   ------------11110100100---1----------1--P---1----------------1--------
Phoronida       ---------?--10110100100---1-0-------P1--1--11---1------------1---1----
Brachiopoda     ---------?--1111PP00100---1-0-------P1----1-1--11-?-----P----1------1-
Nematoda        -101------?-10000000001---1100-----0?-------------P---1---------------
Acoelomorpha    -101-----110-0000000000----10-11-11-?-------------------------------?
Rhabditophora   11011--P-110-0000000000---110-11-11-----1-11P-P-P-P-P-----------------
Gnathostomulida ?10?????--?-10000000000---1----1-?--1-------------P-?-----------------
Nemertea        11011-1---?-10001000000---1-0-11-?1-?-1-1-1-1-----1-1-1-1-1-----P1----
Priapula        --------??--10000000100---1?0--------------------?-1-1-?------------1-
Sipuncula       11011111--1010001000100---1-0-1-----?--11-1111--1------------------1-
Caudofoveata    ??????????-1?00??01?10---1-0-111-1-?-??????-?1-11P11---------?---11-?
Solenogastres   1101------?010001001?10---1-0-111-1-1-111--1-1111-1111-1------1---11-?
Polyplacophora  11011111--1010001011010---1-0-111-1-1-111-1--11111P111-1-1-1----1-11-1
Conchifera      11011111--1010001011010---1-0-111-1-P-11PP11-11111P111-111-1--1-1-11-1
Echiura         110-1?11--1110001000100---1-0-1-1-1-1-111111?---1-1--------------1----
Pogonophora     -10?????1---1P101000100---1-0-1-1---?-1?1--1?---111-1-P------1--11----
Polychaeta      110111111-1111101000100---1-0-1-1-1-1-11111P1111111-1111111-111111--1-
Clitellata      1101111111??-11101000100---1-0-1-1-1-1-1------11-1-1-1111111-111111----
Onychophora     ????????1-?-1010??11000---1-0-------?---------111-1-11111--1---11-1--?
Crustacea       110111-11---10?00011000---1-00-----0?-----1---11111-1111-1-1---11-1--1
Uniramia        --011---1-?-11?0001?000---1-00-----0?---------11--P-1111---1---11-1--1
Chelicerata     ????11-11-?-10?0001?100---1-00-----0?---------11111-1111---1---11-1--1
Tardigrada      ????11-????-10100011101---1100-----0?---------11--P-1111--1----1-----?
Kinorhyncha     ?????1?????-10000000001---110---1---1---------111-1-1111-------1-----?

                ....*....8....*....9....*....0....*....1....*....2....*....3....*....4.
Cnidaria        ?01111100000000000000000000011100000000000000000000000000000001000000
Chordata        ---0000--------1---P----11-1-----1-------1------P1---------1-----1-----
Echinodermata   ---0000--------1---1----11-1----1---------------1-----1---------1-----
Phoronida       ----00-------------1-----1-1--?--1--------------1-----1----11---1-----
Brachiopoda     ----0---------11---1----11-1--?--?1--------------1-----1-----1---1-----
Nematoda        -----?-11--1----------?-1----0-1-1111---?---1--1-----1---1--1---1-----
Acoelomorpha    ---0000-------------1--------0-1------1-1----111---------PP-----1-1---
Rhabditophora   ---0000-------------1------1-0-1PP-P--P-1----111---------------1-1---
Gnathostomulida ---000---------------------1??-?-?----1-?---1111----------1-1---1-----
Nemertea        ---000-------------------1----0-1---1P-?-?---1P------------11---1-----
Priapula        ----0--1---------------1-1-11-?-1-1----?-----1--------1-----11---1-----
Sipuncula       -----00----------11---1-1-1--0-1-11---?-1---1--------1-----11---1-----
Caudofoveata    -1-00??-------1---1----1--1----0-1?111-1?-1-------1---1------11---1-----
Solenogastres   -1-000?-------1---1-1-1--1----0-1?111-1?-1-------1---1------11---1-1-1-
Polyplacophora  11-000?-------11--?-111--1----0-11111-11-1-------1--11-------1---1-1-1-
Conchifera      11-000--------11----111--1----0-11111-11-1----P-P1--11-----P-1---1-1-1-
Echiura         -----0---1----------?--1-1-1--?-1-1----?-1---1-------------11---1-----
Pogonophora     --------?1---------?-----P--1-?---1----?-1---1---1---------11---1-----
Polychaeta      -----0---1---1-1-------1-1-1--0--111---1P1--11---1----1-----11---11---
Clitellata      -----0---1-------------?-1-1--0--11------1---11-1----------11---1-----
Onychophora     ---00--1---1?---------1-111--?--11--------11--1-11------1-11-1-11----
Crustacea       0--00--11-1111-111-----1-111--0--111----1-1111--1111----1111--11-11-1-1
Uniramia        0--00--11-1111-111-----1-111--0--111----1-1111--1-11----1111--11-11-1--
Chelicerata     0--00--11-1111-111-----1-111--0--111------1-11--1111------11--11-11-1-1
Tardigrada      ---00--11-?1?----------1-1----?--111-----?--?1--1-----1----1-1?1-11----
Kinorhyncha     ----0--1-?-1??----1----?-1----?--11------?---1--1-----1---?1111--1-----
```

FIGURE 3. Data matrix of 141 morphological characters (Appendix 1) for 26 taxa selected for their relevance to our investigation of higher spiralian relationships. All characters are binary, assigned states of 0, 1, P (polymorphic; 0 and 1), N (inapplicable), or ? (unknown). Inapplicable states were analyzed as if they were unknown. The 141 columns correspond to the character numbers used in Appendix 1; dashes denote a match to the observed state in the first taxon, Cnidaria.

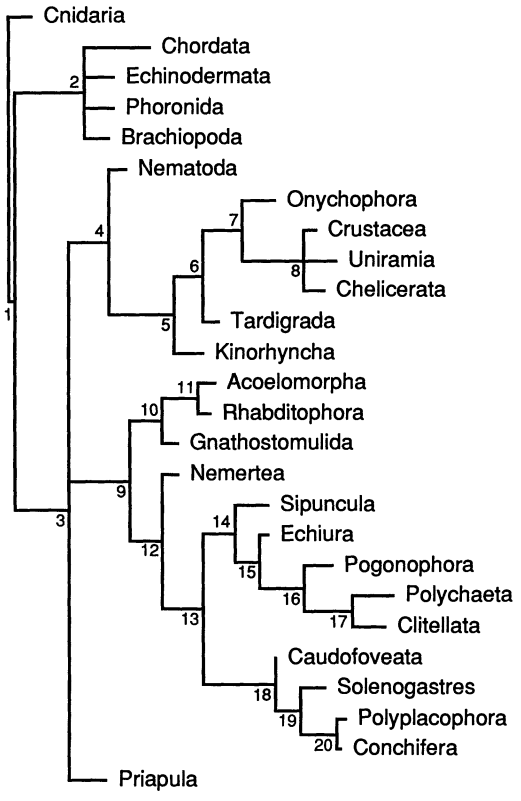


FIGURE 4. Strict consensus phylogram of the six minimum-length trees resulting from maximum parsimony analysis of the matrix in Figure 3. All 141 characters (Appendix 1) for 26 taxa were binary and treated as undirected, with Cnidaria declared as an outgroup to root each otherwise unrooted network topology. The six trees (Fig. 4) summarized in this consensus diagram each had a length of 384, a consistency index of 0.458, a homoplasy index of 0.651, and a retention index of 0.684. The ingroup (1) and all interior (2–20) nodes are labeled, and the apomorphy hypotheses supporting each interior node are presented in Appendix 2. Branch lengths correspond to the relative number of apomorphic hypotheses supporting each interior node or terminal taxon.

thropod clade (Crustacea, Uniramia) instead of a "biramous" (Crustacea, Chelicerata) clade.

The results reported by the strict consensus diagram (Fig. 4) include support for a clade (node 3) approximately consisting of a conventional grouping of protostome phyla, exclusive of deuterostome–lophophorate phyla (node 2). The monophyly of arthropods is supported (node 8) within a larger clade (node 4), which (listed in order of decreasing proximity to arthropods) includes Onychophora, Tardigrada, Kinorhyncha, and Nematoda, with priapulans as the next outgroup in two of the six most-parsimonious trees. Opposing this clade is a large sister clade that includes all other protostomous phyla, including a flatworm–gnathostomulid clade (node 10), Nemertea, and a remaining clade referred to hereafter as Eutrochozoa (node 13). Within this clade, molluscs are the sister taxon to a group that includes annelids and other allied worms. Nemertea is the sister taxon to the mollusc–annelid clade, together forming a clade that is the sister taxon to a flatworm–gnathostomulid clade.

Lists of synapomorphies that unite Eutrochozoa (Fig. 4: clade 13) are presented in Appendix 2. Three unambiguous synapomorphies (characters 40, 44, and 49) unite taxa in this node. Strong support for the Eutrochozoa hypothesis was most evident from searches constrained to keep Annelida and Arthropoda together as sister taxa (Table 1: analysis 1 vs. 2–5). These Articulata-constrained trees required 9–16 additional steps, depending on how inclusive we made "annelid" and "arthropod" clades. A 5+14 constraint (enforcing a clade including all taxa encompassed by clades 5 and 14 of Fig. 4 but not requiring clades 5 and 14 to remain monophyletic) required only nine additional steps (Table 1: analysis 6). The shortest of these trees, however, would require reversal(s) of segmentation in Sipuncula and Echiura to support a common origin for "articulate" segmentation. Searches constrained to include arthropods (as clades 5, 6, 7, or 8) as derived eutrochozoans (i.e., within clade 13) minimally required 10 additional steps (Table 1: analysis 10), and even then clades 14 and 18 (i.e., molluscs and annelids, etc.) were joined as sister taxa. Somewhat shorter trees were found when Nemertea (Table 1: analyses 11, 12) or Nemertea and clade 10 of Figure 4 (Table 1: analyses 13, 14) were included in the constraint. Constraints enforced to keep molluscs and arthropods together as sister taxa required 13–23 additional steps (Table 1: analyses

15–18), depending again on the composition of the arthropod clade. Enforcing constraints to join molluscs and flatworms as a clade (Table 1: analyses 19–22) required 3–12 extra steps, depending on the inclusiveness of "flatworms." The shortest of these trees (Table 1: analysis 22) included Nemertea as the sister taxon to molluscs, a result that was topologically equivalent to that of analysis 23 (Table 1), which was generated by imposing a converse constraint, searching for only those trees that did not satisfy the monophyly of clade 12 (Fig. 4). Shorter trees, requiring only a single step more than the minimum-length trees (Table 1: analysis 24), were found in searching for trees that did not maintain clade 13. All four resulting trees were characterized by the placement of Nemertea as the sister taxon to molluscs within an otherwise unmodified clade 13.

As in the constraint analyses, characters from our matrix optimized onto the tree topologies of previously published hypotheses (Table 2, Figs. 5a–k) required multiple additional steps. The closest alternative proposals were those of Brusca and Brusca (1990; length = 437) and Pearse et al. (1987; length = 440), requiring 53 and 56 additional steps, respectively, over our six minimum-length hypotheses (length = 384). Part of these length differences can be attributed to these authors' support of Articulata, which requires at least nine additional steps. Even Nielsen's diagram, which, based on his text statement "[t]here are so many similarities between sipunculans, echiurans, annelids and molluscs, that these phyla must be closely related" (1985:256), was interpreted as support for the Eutrochozoa clade, required an additional 75 steps. Another factor contributing to these length differences was the tendency of various authors to express uncertainties as polytomies, which as hypotheses of relationship require multiple acquisition of traits. This well-known shortcoming of consensus summaries (Miyamoto, 1985) is illustrated by variation among our six most-parsimonious trees relative to the strict consensus summary (Fig. 4). Yet, even when these authors' proposed polytomies (Figs. 5a–k) were resolved by branch swapping to minimize required steps on our matrix, these trees remained substantially longer than the best-fitting hypotheses, requiring no fewer than 26 additional steps (Table 2: Brusca and Brusca, 1990).

Discussion

In certain respects, our results were anticipated by Ghiselin (1988), whose paper should be consulted for historical aspects of the Articulata versus Eutrochozoa controversy. Ghiselin and other authors (Field et al., 1988; Lake, 1990) have argued for the consistency of morphology with their rRNA results supporting the Eutrochozoa hypothesis. The results reported here confirm these suggestions by supporting the monophyly of Eutrochozoa (Mollusca plus Annelida plus several less speciose spiralian phyla). We base our conclusion on a maximum-parsimony analysis of as many relevant, putatively independent, morphological and embryological characters as we could compile. It would take at least nine additional ad hoc hypotheses of character evolution (Table 1) to claim support for Articulata. This analysis is the first to recover evidence from congruence in the phylogenetic distribution of morphological and embryological characters, corroborating the results of the 18S rRNA sequence data (Field et al., 1988, 1989; Ghiselin, 1988; Patterson, 1989; Raff et al., 1989; Lake, 1990; Eernisse, unpubl. manuscript). The resulting placement of Nemertea in close proximity to eutrochozoans is also in accordance with recent 18S rRNA-based comparisons by Turbeville et al. (1992).

Previous hypotheses of metazoan branching patterns are all substantially less parsimonious than our own, when optimized on our data matrix. We acknowledge that comparisons of previous hypotheses are difficult to interpret for several reasons. For instance, it was often necessary to include certain terminal taxa neglected by particular authors. The translation of conventional evolutionary trees into cladograms is further complicated by the dif-

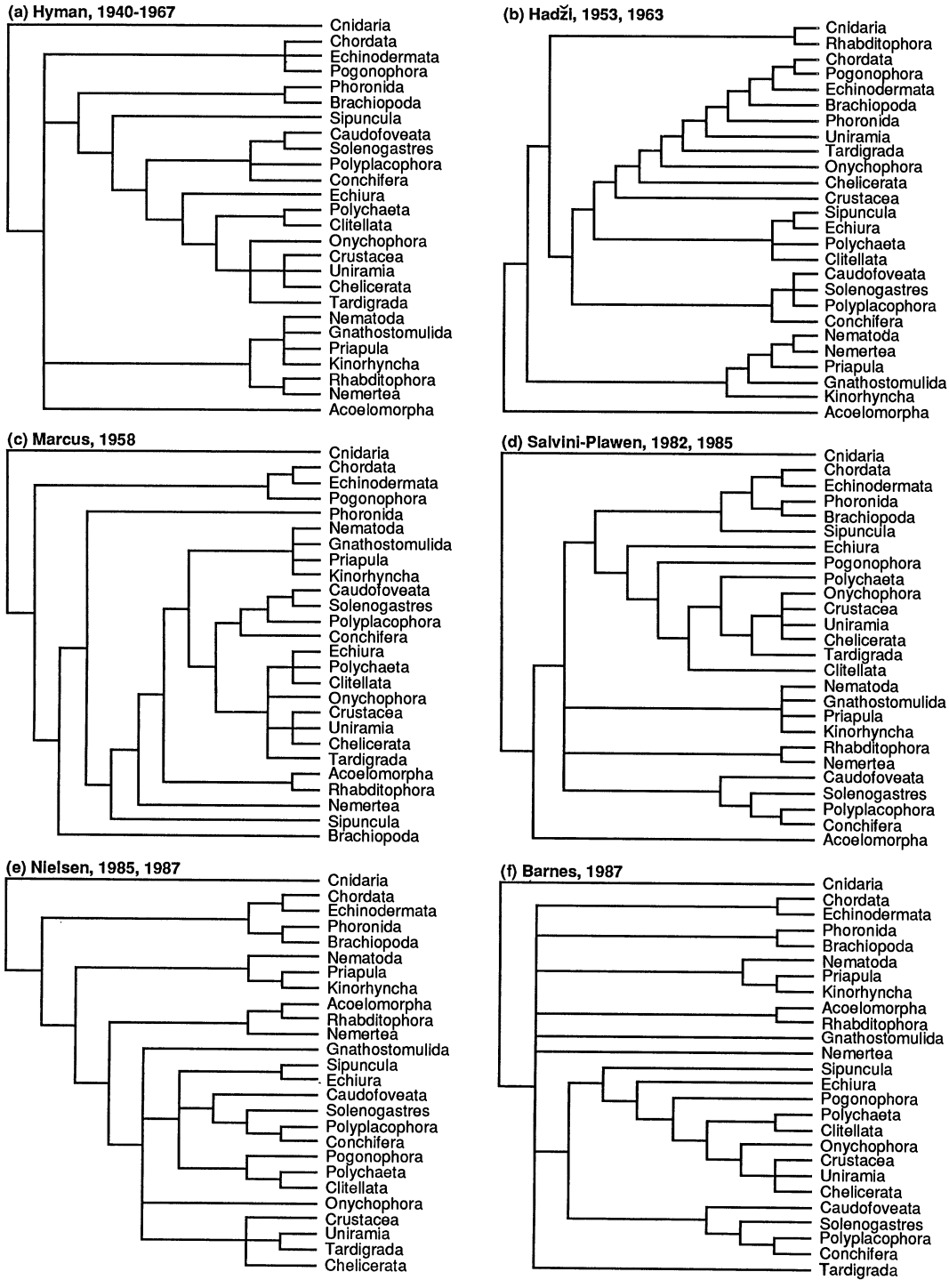
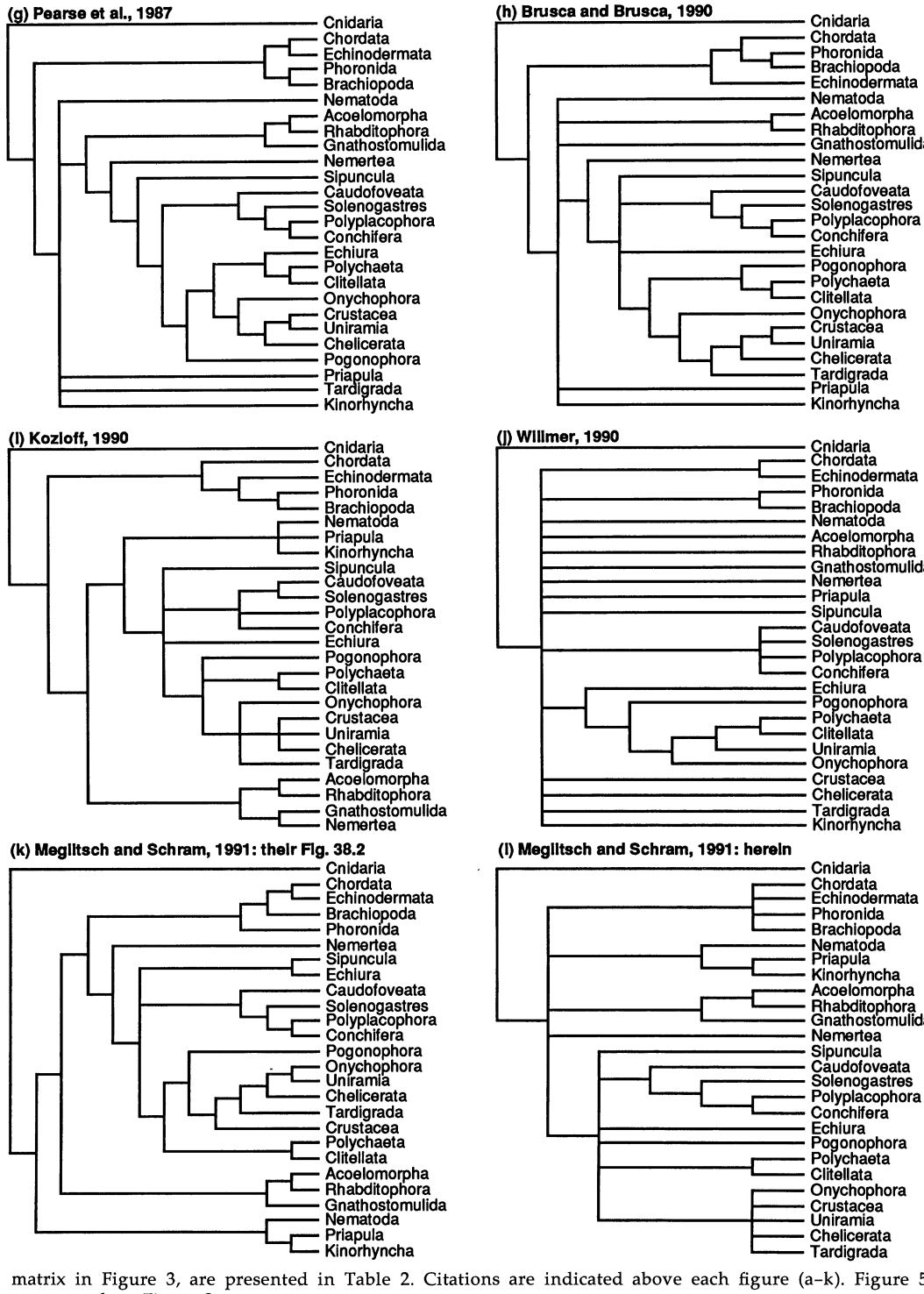


FIGURE 5. Summary of topologies translated from published figures into cladogram hypotheses. Taxon composition of each cladogram has been adjusted to compare to Figure 4. Tree statistics, optimized on the



matrix in Figure 3, are presented in Table 2. Citations are indicated above each figure (a–k). Figure 5l corresponds to Figure 2a.

ferent concepts of evolutionary entities held by each author. Taxa considered ancestral to other higher taxa were treated as terminal taxa to make comparison with our results meaningful. Because of these concerns, Figures 5a–k cannot be used to judge the relative merit or worth of these works. Rather, they are presented to make evaluation of the relative support for different tree topologies possible. These disclaimers aside, we believe we have established a more rigorous precedent for reporting support for alternative hypotheses of metazoan phylogeny.

Because we were interested in the question of spiralian interrelationships, little effort was made to collect characters lacking variation among these taxa. Moreover, beyond affirming the monophyly of Mollusca and Arthropoda, relatively little of the enormous literature bearing on these groups was included in this analysis. Our analysis was unable to resolve some important aspects of metazoan phylogeny. In particular, our lack of resolution of deuterostome–lophophorate relationships reflects the largely unsettled nature of the corresponding literature (cf. Jefferies, 1986; Nielsen, 1987; Ax, 1989; Brusca and Brusca, 1990; Kozloff, 1990), as is the case for relationships among arthropod subphyla (cf. Brusca and Brusca, 1990; Emerson and Schram, 1990). This lack of resolution may largely be due to a combination of sampling biases, especially the emphasis of variation among basal spiralian lineages and the inclusion of the particular phyla used in this study. A similar condition may apply to our results with regards to priapulans, kinorhynchs, and nematodes. In addition, these and other relationships may reflect the biases of particular sources used; for example, the characters used to diagnose clade 10 (Fig. 4) are mainly those emphasized by Ax (1985). Further research should be directed to the careful addition of more relevant character evidence, including fossil and molecular sequence data not included in the present study.

We anticipate such a combination of data recovered from morphological and molecular studies. We do not, however, encourage mere comparison of our results with those inferred from molecules. Like Kluge (1989) and Eernisse (unpubl. manuscript), we advocate that ultimately, the best estimates of branching order for metazoans will come from combined analyses of all relevant data, whether classical in nature or a site-by-site comparison of nucleic acid or peptide sequences. Ghiselin (1988) observed that molecules have morphology and can be analyzed as such. In this spirit, we have coded variation in the structure of the hemocyanin molecule as characters 71 and 72.

One consequence of character congruence analyses is that specific homoplasies required to support alternative views are highlighted. To further judge the confidence of the Eutrochozoa hypothesis, the structure of the present data could be analyzed to discern whether the homoplastic characters are randomly and independently distributed or produced by conflicting suites of characters. This issue is only beginning to be addressed in cladistic studies (Farris, 1991) but would be a natural extension of the present analysis.

Because the character congruence approach assumes character independence, this analysis is subject to the criticism that a suite of ciliary characters have been lost as a unit in the arthropods in association with yolk-rich development. For example, at least two of the three characters (40, 44) unambiguously optimized to diagnose Eutrochozoa (Fig. 4: node 13) are features associated with early larval development and the presence of cilia. Absence of these characters could be interpreted as the result of a single evolutionary reversal in Arthropoda. This hypothesis is appealing because many of the differences between arthropods and other metameric spiralians could be explained by relatively few and simple evolutionary changes. The presence of flagella in crustacean and pycnogonid sperm (Franzén, 1987) and of ciliated gonoducts and nephridial segmented organs in onycophorans (Boudreaux, 1979) indicates that cilia were not entirely lost. A similar argument could be made for the position of chordates as the sister taxon to

other deuterostomes in four of the six minimum-length trees of Figure 4 as the result of the difficulty of comparing larval characters in these taxa. Although consideration of all the data available for this analysis does not support these interpretations, close scrutiny of the characters in question could lead to a reappraisal of the present notions of phenotypic similarity.

The support for Eutrochozoa recognized here does not settle the controversy surrounding the extent to which molluscs are primitively metameric. Two important uncertainties remain regarding the identity of the immediate sister taxa to Mollusca and the optimization of overt metamerism, which may be absent in this group due to derived loss or retained plesiomorphy (Fig. 1). The lack of apparent metamerism in Solenogastres, Caudofoveata, Sipuncula, and Echiura presents difficulties for the argument of a primitively metameric eutrochozoan. Furthermore, Nemertea may be more closely aligned to Mollusca than has been generally appreciated. Of all the constraint analyses previously reported, placement of Nemertea as the sister taxon to Mollusca required only one additional step (Table 1: analysis 24), yet we see no reason to prefer this less-parsimonious arrangement to our best-fitting hypotheses.

If annelids and arthropods are not sister taxa, then similarities of their segmentation must have been much more ancient, or in many respects independently derived, or both. There is now a rapidly advancing knowledge of the genetic and developmental bases of a basic level of metamerism in metazoans as diverse as insects, vertebrates, sea urchins, brachiopods, molluscs, and leeches, especially involving homeodomain-encoding ("homeobox") gene clusters (Holland and Hogan, 1986; Gould, 1991; Wedeen et al., 1991). This burst of activity is leading to a reevaluation of the basic underlying mechanisms of segmentation and a search for likely homologies. Homeotic homologies, extending to the sequence and functional level, will probably be hypothesized for molluscs and other phyla. If further analyses and new data continue to support the notion that molluscs, sipunculans, and echiurans descended from a segmented ancestor, our understanding of the biology of these groups will be deepened. We hope that the data matrix compiled for this study will serve as a nucleus from which further discussions of animal relationships can grow.

ACKNOWLEDGMENTS

The compilation of a character matrix for spiralian metazoans and select nonspiralian outgroups was initiated in fall 1989 during a graduate seminar led by one of us (D.J.E.). We thank members of that seminar group for stimulating discussion and suggestions. Arnold G. Kluge and James N. Cather provided helpful suggestions at various stages of this project. We give special thanks to Vicki B. and John S. Pearse for their many useful observations in review of the data matrix. Rich Mooi, Malcolm Telford, and an anonymous reviewer also provided many valuable suggestions. We thank William R. Dawson, Director of the Museum of Zoology, University of Michigan, for use of facilities.

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APPENDIX 1

Sources used are listed by character with the exception of certain taxon-specific references, including those pertaining to Cnidaria (Fautin and Mariscal, 1991; Thomas and Edwards, 1991), Chordata (Gans and Northcutt, 1983), Echinodermata (Smith, 1984), Platyhelminthomorpha (Ax, 1985; Ehlers, 1985), Mollusca (Runnegar and Pojeta, 1985; Wingstrand, 1985; Haszprunar, 1988), Annelida (Giese and Pearse, 1975; Jameison, 1988), Echiura and Sipuncula (Giese and Pearse, 1975; Rice and Todorovic, 1975–1976; Strathmann, 1978), Crustacea (Schram, 1986; Brusca and Brusca, 1990), Uniramia (Daly et al., 1978; Boudreaux, 1979), and Chelicerata (Meglitsch and Schram, 1991).

Characters by System[a]

A. Cleavage
1. Spiral quartet cleavage; cells B and D with ventral transverse contact, cells A and C with dorsal saggital contact (Wilson, 1898; Cather, 1971; Anderson, 1973; Freeman and Lundelius, 1992) [a/p]
2. Spiral cleavage with nuclear migration[b] (Schleip, 1929; Costello and Henley, 1976) [a/p]
3. Radial holoblastic cleavage (Wilson, 1898; Brusca and Brusca, 1990) [p/a][b]
4. Cell fates of primary germ layers fixed by end of fifth cleavage (Anderson, 1973; Brusca and Brusca, 1990; Meglitsch and Schram, 1991) [a/p]
5. Entomesoblast cell (blastomere 4d) (Beklemishev, 1969; Boudreux, 1979, Verdonk and Biggelaar, 1983) [a/p]
6. Entomesoblast proliferation into paired anterior coelomic sacs (Brusca and Brusca, 1990) [a (noncavitated mesenchyme)/p]
7. Entomesoblast proliferation contributing to mesoderm (Wilson, 1898; Anderson, 1973) [mesoderm absent or forming from ectoderm or archenteron/p]
8. Entomesoblast proliferation into paired dorsoposterior mesodermal tissue bands (Salvini-Plawen, 1985, 1988; Brusca and Brusca, 1990) [a/p]
9. Teloblastic segmentation of mesodermal sacs with pygidial growth zone (Salvini-Plawen, 1985, 1988) [a/p]
10. Epidermal mitosis by parenchymal kineto-

[a] Character numbers 1–141 correspond to columns in Figure 3. Alternative states, in square brackets following the character description, are listed in order of states 0 and 1 with no implied polarity (a = absent; p = present)

[b] See supplemental notes following listing of characters.

some-containing cells (Ehlers, 1986; Smith et al., 1986) [a/p]
11. Apical and intermediate micromere quartet form cross (Meglitsch, 1972) [a/p]
12. Cross pattern (Meglitsch, 1972; Brusca and Brusca, 1990) [radiate/interradiate]
13. Triploblastic tissue organization (Hanson, 1977; Brusca and Brusca, 1990; Meglitsch and Schram, 1991; cf. Nelson and Weisblat, 1991) [a/p]

B. Coelom
14. Bilaterally paired coelomic anlagen[b] (Brusca and Brusca, 1990; Meglitsch and Schram, 1991) [a/p]
15. Longitudinally metameric coelomic cavities with mesodermal contribution to mesenteric partitions (Brusca and Brusca, 1990; Meglitsch and Schram, 1991) [a/p]
16. Tripartite coelom (Brusca and Brusca, 1990) [a/p]
17. Schizocoelous formation of body cavity lined with mesodermal peritoneum[b] (Brusca and Brusca, 1990; Meglitsch and Schram, 1991) [a/p]
18. Enterocoelous formation of body cavity lined with mesodermal peritoneum[b] (Brusca and Brusca, 1990; Meglitsch and Schram, 1991) [a/p]
19. Hemocoel; main body cavity unlined with lymph-filled vacuities (Brusca and Brusca, 1990; Meglitsch and Schram, 1991) [a/p]
20. Gonocoel; coelom reduced to perigonadal region (Brusca and Brusca, 1990; Meglitsch and Schram, 1991) [a/p]
21. Hydrostatic skeleton; coelomic compartment under relatively high pressures (Brusca and Brusca, 1990; Meglitsch and Schram, 1991) [a/p]
22. Pericardial excretory complex of coelomoducts connected to cloaca (Salvini-Plawen, 1985) [a/p]
23. Pseudocoelom; blastocoel persisting as unlined cavity(ies) between endoderm and mesoderm (Meglitsch and Schram, 1991) [a/p]

C. Cellular
24. Multicellular with population of gamete cells (Hanson, 1977; Nielsen, 1985) [a/p]
25. Special sense cells (Hanson, 1977; Nielsen, 1985) [a/p]
26. Gap junctions (Nielsen, 1985) [a/p]
27. Basal lamina (Nielsen, 1985) [a/p]
28. Mitosis lacking in epidermal cells (Ehlers, 1986; Smith et al., 1986) [a/p]

D. Ciliary
29. Collared ciliary units (Willmer, 1990) [a/p]
30. Ciliated somatic cells[b] (Nielsen, 1987) [a/p]
31. Multiciliary epidermis with ciliated rootlets (Smith et al., 1986; Nielsen, 1987) [a/p]
32. Ciliated ventral surface in adult (Nielsen, 1987) [a/p]
33. Monociliated cells with accessory centriole (Nielsen, 1987) [p/multiciliated without corresponding accessory centrioles]
34. Cilium with one basal body; without accessory centriole (Ehlers, 1986; Nielsen, 1987) [a/p]
35. Coordinated cilia with ciliated necklace (Nielsen, 1987) [a/p]
36. Motile somatic cilia or flagella (Nielsen, 1987) [a/p]
37. Chemoreceptor cells with paddle-shaped discocilia[b] (Haszprunar, 1985a) [a/p]

E. Larval
38. Upstream collecting bands of cilia in larvae with separate cilia on monociliate cells[b] (Nielsen, 1987) [a/p]
39. Swimming/feeding band(s) of cilia in larvae with compound cilia[b] (Nielsen, 1987) [a/p]
40. Prototroch; locomotory equatorial ciliary band(s) with two or four rows of broad cilia formed before gastrulation (Strathmann, 1987; Brusca and Brusca, 1990) [a/p]
41. Pelagic larvae with apical ciliary tuft and plate (Nielsen, 1987) [a/p]
42. Nutritive metatroch with opposed bands; postoral (segmentally added) paired ciliary bands beating in opposite directions and serving in food capture[b] (Strathmann, 1978; Salvini-Plawen, 1988) [a/p]
43. Cerebral rhabdomeric larval ocelli or integumentary pigment cups[b] (Rosen et al., 1979; Brusca and Brusca, 1990; Meglitsch and Schram, 1991) [a/p]
44. Telotroch; pelagic larvae with para- or circumanal ciliary tuft (Nielsen, 1987; Brusca and Brusca, 1990; Meglitsch and Schram, 1991) [a/p]
45. Preoral fold covering larval hyposphere (Brusca and Brusca, 1990; Meglitsch and Schram, 1991) [a/p]
46. Pericalymna; unpaired, mineralized, epispheral test enveloping larvae[b] (Salvini-Plawen, 1988; Brusca and Brusca, 1990) [a/p]

F. Bilateral symmetry
47. Paired ventral nerve bundles[b] (Beklemishev, 1969; Brusca and Brusca, 1990; Kozloff, 1990) [a/p]
48. Paired ventral lateral pedal retractor muscle bundles[b] (Wingstrand, 1985) [a/p]
49. Paired excretory organs and ducts open externally (nephridiopores) (Brusca and Brusca, 1990) [a/p]
50. Paired gills; ectodermal filamentous or lamellar respiratory surfaces (Brusca and Brusca, 1990) [a/p]
51. Paired gonads and gonoducts (or nephridiopores used as gonoducts)[b] [a/p]
52. Paired endothelium-lined pericardial diverticulae (auricle) (Wingstrand, 1985) [a/p]

G. Serial repetition
53. Serially repeated nerve collaterals; ladderlike nervous system with ventrolateral nerve cords and lateral connectives[b] (Beklemishev, 1969; Wingstrand, 1985) [a/p]
54. Serially repeated transverse discrete muscle bundles (Beklemishev, 1969; Wingstrand, 1985) [a/p]

55. Serially repeated nerve ganglia[b] (Beklemishev, 1969; Wingstrand, 1985) [a/p]
56. Serially repeated transverse discrete muscle bundles (Beklemishev, 1969; Wingstrand, 1985) [a/p]
57. Serially arranged series of excretory ducts; nephridiopores (Brusca and Brusca, 1990) [a/p]
58. Serially arranged ectodermal filamentous or lamellar respiratory surfaces[b] (Brusca and Brusca, 1990) [a/p]
59. Serially arranged series of gonads (Brusca and Brusca, 1990) [a/p]
60. Atria; serially arranged muscularized regions of a dorsal blood vessel (Salvini-Plawen, 1985; Wingstrand, 1985) [a/p]
61. Schizocoelous metamerism between preoral prostomium and nonmetameric pygidium [a/p]
62. One or more transverse coelomic septa (Brusca and Brusca, 1990) [a/p]
63. Serially repeated ventricles; branchioauricular sinuses with ctenedial pores (Ruppert and Carle, 1983; Wingstrand, 1985; Brusca and Brusca, 1990) [a/p]
64. Metamerism in associated cuticular, muscular, and nervous tissues (Brusca and Brusca, 1990; Meglitsch and Schram, 1991) [a/p]

H. Circulatory

65. Heart(s); dorsal blood vessel with contractile epithelium formed around a vascularized longitudinal lumen by fusion of coelomic walls and lined by a basal lamina[b] (Ruppert and Carle, 1983; Brusca and Brusca, 1990) [a/p]
66. Closed posterior circulation (Ruppert and Carle, 1983; Salvini-Plawen, 1985; Brusca and Brusca, 1990) [a/p]
67. Atrial ostia; muscularized opening(s) in dorsal blood vessel (Ruppert and Carle, 1983; Salvini-Plawen, 1985, 1988) [a/p]
68. Atrial ultrafiltration (Salvini-Plawen, 1985, 1988) [a/p]
69. Hemerythrin or myohemerythrin as a respiratory pigment molecule (Mangum et al., 1985; Richardson et al., 1987; Volbeda and Hol, 1989; Demuynck et al., 1991; Takagi and Cox, 1991; Yano et al., 1991) [a/p]
70. Hemocyanin as a respiratory pigment molecule hypothesized to be homologous based on spectroscopic and sequence similarities (Mangum et al., 1985, 1987; Lang, 1988; Volbeda and Hol, 1989; Voit and Feldmaier-Fuchs, 1990; Lang and Holde, 1991) [a/p]
71. Hemocyanin structure (Linzen et al., 1985; Mangum et al., 1987) [hexamer or multihexamer "boxcar" molecules with subunits of about 75,000 molecular weight, together combining to up to 3–5 million, each containing one dinuclear copper site/cylindrical molecules made up of about 10–20 "stacked petri-plate" subunits, each of about 350,000 molecular weight, containing seven or eight domains with one oxygen-binding dinuclear copper site per domain]
72. Mesodermal origin of pericardioducts (Salvini-Plawen, 1985, 1988) [a/p]

I. Integumentary

73. Cellular production of collagenous proteins[b] (Brusca and Brusca, 1990; Meglitsch and Schram, 1991) [a/p]
74. Cuticle; continuously secreted, nonliving external layer(s) containing protein[b] (Brusca and Brusca, 1990; Meglitsch and Schram, 1991) [a/p]
75. Collagenous proteins sequestered in cuticle[b] (Brown, 1975; Bereiter-Hahn et al., 1984; Brusca and Brusca, 1990; Meglitsch and Schram, 1991) [a/p]
76. Chitinous proteins invested in cuticle[b] (Brown, 1975; Bereiter-Hahn et al., 1984; Brusca and Brusca, 1990; Meglitsch and Schram, 1991) [a/p]
77. Cellular secretion of chitinous proteins[b] (Brown, 1975; Bereiter-Hahn et al., 1984; Brusca and Brusca, 1990; Willmer, 1990; Meglitsch and Schram, 1991) [a/p]
78. Cuticular covering of entire external body surface (Brusca and Brusca, 1990; Meglitsch and Schram, 1991) [a/p]
79. Sclerotinization of cuticle with tannin proteins (Brusca and Brusca, 1990; Meglitsch and Schram, 1991) [a/p]
80. Protrusible and retractable chitinous setae (Brusca and Brusca, 1990; Meglitsch and Schram, 1991) [a/p]
81. Alpha and/or beta ecdysone (Willmer, 1990) [a/p]
82. Periodic ecdysis under hormonal control (Brusca and Brusca, 1990; Meglitsch and Schram, 1991) [a/p]
83. Anterior ecdysome-producing gland (Brusca and Brusca, 1990; Meglitsch and Schram, 1991) [a/p]
84. Aciculae; tonofibrillae penetrating epidermis with muscle attachment (Brusca and Brusca, 1990; Meglitsch and Schram, 1991) [a/p]
85. Mantle; thick epidermal cuticular sheet with band(s) of glands capable of secreting a hard calcareous skeleton (Brusca and Brusca, 1990; Meglitsch and Schram, 1991) [a/p]
86. Calcified skeletal covering secreted by epidermis[b] (Brown, 1975; Tidball, 1984; Salvini-Plawen, 1988; Brusca and Brusca, 1990; Carter, 1990) [a/p]
87. Lateral tergal folds or paranotal lobes (Ghiselin, 1988; Salvini-Plawen, 1988) [a/p]
88. Anterior cephalic tagma formed from metamere(s) and the primary sensory acron (Brusca and Brusca, 1990; Meglitsch and Schram, 1991) [a/p]
89. Serially arranged mineralized ectodermal plates (Brusca and Brusca, 1990; Meglitsch and Schram, 1991) [a/p]

J. Alimentary

90. U-shaped alimentary canal (Brusca and Brusca, 1990) [a/p]

91. Crystalline stylus and associated ciliated midgut digestive organs (Brusca and Brusca, 1990) [a/p]
92. Pharyngeal diverticulae (Salvini-Plawen, 1988) [a/p]
93. Esophageal pouches (Salvini-Plawen, 1988) [a/p]
94. Terminal alimentary zones of cuticle (Boudreaux, 1979) [a/p]
95. Secondary mouth formation (Meglitsch and Schram, 1991) [a/p]
96. Anus with proctodeum; complete unidirectional alimentary canal[b] (Meglitsch and Schram, 1991) [a/p]

K. Excretory
97. Antennal gland excretory ducts; mandibular (first) pair metanephridia [a/p]
98. Metanephridia; paired mesodermal excretory ducts with ciliated funnel draining coelomic cavity(ies) (Brusca and Brusca, 1990) [a/p]
99. Protonephridia; ampullary (blind) vessels bearing multiciliated cells serving excretory/osmoregulatory function (Brusca and Brusca, 1990; Meglitsch and Schram, 1991) [a/p]

L. Nervous
100. Acetocholine[b] (Willmer, 1990) [a/p]
101. Creatine phosphotase (Willmer, 1990) [a/p]
102. Population of specialized polar neurons with neurites and synaptic terminals (Nielsen 1985; Brusca and Brusca, 1990) [a/p]
103. Orthogon; dense diffuse neural plexus with short peripheral connections and very long interganglionic connections (Beklemishev, 1969; Brusca and Brusca, 1990; Meglitsch and Schram, 1991) [a/p]
104. Subcutaneous neural plexus; subepithelial location of (at least some) epidermally derived neurons (Beklemishev, 1969; Brusca and Brusca, 1990; Meglitsch and Schram, 1991) [a/p]
105. Circumpharyngeal chain of ganglia (buccal, pharyngeal, or subenteric) attached to longitudinal ventral nerve cord(s) (Beklemishev, 1969; Kozloff, 1990) [a/p]
106. Adnate ventral nerve cords (Kozloff, 1990) [a/p]
107. Lateral nerve cords; paired longitudinal cutaneous or subepidermal axon bundles descending from an anterior commissure or ganglion (Beklemishev, 1969; Brusca and Brusca, 1990; Meglitsch and Schram, 1991) [a/p]
108. Dorsal nerve cord; median longitudinal cutaneous or subepidermal axon bundle descending from an anterior commisure or ganglion (Beklemishev, 1969; Brusca and Brusca, 1990; Meglitsch and Schram, 1991) [a/p]
109. Osphradia; chemosensory epithelial surfaces located on or near the gill(s); mechanoreceptive collar cells with eight or nine stereo microvilli (Haszprunar, 1985a, 1985b, 1987; Brusca and Brusca, 1990) [a/p]
110. Endon; median cerebral ganglion and adjacent aboral statocyst organ (Beklemishev, 1969) [a/p]
111. Three pairs of cerebral ganglia; an anterior one receiving ocular input, a second receiving palpar or antennal input, and a third contributing to circumenteric connectives (Beklemishev, 1969; Brusca and Brusca, 1990) [a/p]
112. Paired olfactory fossae of preoral lobes (Beklemishev, 1969) [a/p]
113. Compound eyes with ommatidia (Paulus, 1979; Brusca and Brusca, 1990; Meglitsch and Schram, 1991) [a/p]
114. Ommatidium consisting of a cornea with two corneagen cells, a tetrapartite eucone crystalline cone, and a retinula of eight cells (Brusca and Brusca, 1990) [a/p]
115. Prostomial sensory antennae with basal ocelli with or without lens structure (Brusca and Brusca, 1990) [a/p]
116. Unicellular sensory (tactile) setae in epidermis (Brusca and Brusca, 1990) [a/p]

M. Reproductive
117. Hermaphroditic sexual system (Brusca and Brusca, 1990) [gonochoric/present in more than isolated species]
118. Filiform morphology of sperm (Wirth, 1984; Ax, 1985; Smith et al., 1986; Franzén, 1987) [a/p]
119. Direct internal fertilization (Ax, 1985) [external/internal]

N. Respiratory
120. Gill with counter-current O_2 exchange[b] (Brusca and Brusca, 1990) [a/p]
121. Cuticle-lined tracheal tubes (Brusca and Brusca, 1990) [a/p]
122. Semi-internal lateroventral respiratory chamber (Brusca and Brusca, 1990) [a/p]

O. Oral
123. Subradular organ (Wingstrand, 1985) [a/p]
124. Radula; ribbon or plates or recurved chitinous teeth stretched over a supportive cartilaginous (or hemocoelic) basal expansion of the foregut epithelium (Wingstrand, 1985; Eernisse and Kerth, 1988) [a/p]
125. Introvert (proboscis) at anterior end of digestive tract with barbs and hooks (Nielsen, 1987; Brusca and Brusca, 1990; Meglitsch and Schram, 1991) [a/p]
126. Lophophore; anterior ring of hollow ciliated tentacles formed by coelomic evaginations (Brusca and Brusca, 1990) [a/p]
127. Mandibles; appendages of the third postacronal head somite (Brusca and Brusca, 1990) [a/p]
128. Two pairs of maxillae; appendages of postacronal head somites four and five (Brusca and Brusca, 1990) [a/p]

P. Muscular
129. Oblique striated muscle fibers (Brusca and Brusca, 1990) [a/p]

130. Cross striated muscle fibers[b] (Brusca and Brusca, 1990) [a/p]
131. Dermal circular (or "external transverse") muscular fibers (Salvini-Plawen, 1978, 1985; Brusca and Brusca, 1990) [a/p]
132. Longitudinal muscle sheet(s) or band(s) (Salvini-Plawen, 1978, 1985; Brusca and Brusca, 1990) [a/p]
133. Intersegmental tendon system (Brusca and Brusca, 1990) [a/p]
134. Locomotor coxae with extrinsic and intrinsic muscles (Brusca and Bursca, 1990) [a/p]
135. Myofilaments (Hanson, 1977; Nielsen, 1985) [a/p]
136. Smooth muscle fibers (Hanson, 1977; Nielsen, 1985; Brusca and Brusca, 1990) [a/p]
137. Scleratized terminal structure on coxae (Brusca and Brusca, 1990) [a/p]
138. Broad creeping sole or narrow hydrostatic foot in ventral furrow ("pedal groove") (Wingstrand, 1985) [a/p]
139. Segmented serially arranged locomotor appendages with basal coxite and distal telopide (Brusca and Brusca, 1990) [a/p]
140. Pedal glands; ventral and large with mucous secretion (Brusca and Brusca, 1990) [a/p]
141. Biramous appendages (Emerson and Schram, 1990; Grosberg, 1990; Meglitsch and Schram, 1991) [a/p]

Supplementary Notes by Character Number

2. Pogonophoran ontogeny poorly understood (Ivanov, 1963, 1988; Bakke, 1980), pogonophorans represented by Perviata (Meglitsch and Schram, 1991) for all characters except no. 14; Gnathostomulida (Riedl, 1969); "acoel" and "polyclad" flatworm cleavage (Boyer, 1971, 1989, respectively)
3. Solenogastres cleavage and ontogeny (reviewed by Hadfield, 1979; Salvini-Plawen, 1985), Caudofoveata ontogeny undescribed; "Turbellaria" interpreted as polyclads (Salvini-Plawen, 1988)
14. Characters of coelom and mesoderm ontogeny: mollusc references reviewed in Salvini-Plawen (1985); monoplacophoran "dorsal coeloms" not coded as present (Wingstrand, 1985); uniramian coelomic metamerism from Brusca and Brusca (1990); pogonophorans represented by Vestimentifera (Brusca and Brusca, 1990)
17. Nemertean rhynchocoel (Turbeville and Ruppert, 1985; Turbeville, 1991)
18. Tardigrades (in Nelson, 1982); priapulids (Lang, 1848, in Meglitsch, 1972)
30. Ciliary and flagellar character 30–39 (Tyler, 1979; Nielsen, 1985, 1987)
37. The discocilia's paddle shape itself may be a preservation artifact (Nielsen, 1987)
38. Characters of larval morphology: taxa with exclusive direct development coded 0 if absent
39. Lack of compound cilia in oweniids (Polychaeta) considered a derived exceptional case (Nielsen, 1987)
42. Sipunculans and nemerteans without opposed band mechanism (Strathmann, 1978)
43. Uniramia coded 1, although myriopods possess direct development (Brusca and Brusca, 1990)
46. Sipunculan "serosa larva"; polychaete "endolarva" (Salvini-Plawen, 1988)
47. Characters of paired lateral structures sharing similar relative topology and ultrastructure
48. Solenogastres (Salvini-Plawen, 1985)
51. Uniramia coded present; Diplopoda present, Chilopoda variable, Pauropoda females have single ovary, all with paired gonoducts
53. See Methods for our distinction between serial repetition and metamerism
55. Pogonophora coded polymorphic (Meglitsch and Schram, 1991)
58. Chordata (C. Gans, pers. comm.)
65. Characters of the blood vascular system (Ruppert and Carle, 1983); *Crania* (Brachiopoda) with several, *Ikeda* (Echiura) with one, neither coded as plesiomorphic (Brusca and Brusca, 1990)
73. Characters of the ectodermal integument: arthropods (Manton and Anderson, 1979), molluscs (Runnegar, 1983; Salvini-Plawen, 1985, 1988; Wingstrand, 1985)
74. Arthropod codes largely from Manton and Anderson (1979); see notes for character 75 for Cnidaria
75. Collagenous exoskeletal perisarc of gorgonians (Cnidaria) considered similar to the cuticle of insects (Goldberg, 1976)
76. Chitin found in tube but not cuticle of Phoronida (Hyman, 1958)
77. Report of chitin in Chordata (Sannasi and Hermann, 1970) rejected by Azariah (1973)
86. The calcification mechanisms of anthozoan and hydrocoral (Cnidaria) skeletons, which occurs within specialized region of epidermis, not completely understood (Fautin and Mariscal, 1991)
96. Vestimentifera have transitory alimentary canal in early ontogeny; character coded as polymorphic for Pogonophora (Meglitsch and Schram, 1991)
100. Characters of the nervous system (Beklemishev, 1969)
120. Characters of the respiratory system: arthropod terminals coded from sources providing ingroup polarizations as described in Methods.
130. Characters of the somatic musculature and locomotor apparatus: arthropods (Manton and Anderson, 1979; Emerson and Schram, 1990), molluscs (Runnegar, 1983; Salvini-Plawen, 1985, 1988; Wingstrand, 1985)

APPENDIX 2. Apomorphy hypotheses for internal nodes of metazoan tree in Figure 4.[a]

Node	Character no.
2	38, 45, 49, 62, −76, 86, 90, 95, 98, 120, 126
3	51, −101, 103, 116
4	23, 28, 55, 82, 119
5	6, 47, 48, 54, 56, 64, −103, 104, 130
6	15, 19, 20, −74, 134, 137
7	−23, −28, 60, 65, 67, 97, 98, 121, 122, −125
8	81, 84, 86, 87, 88, 113, −132, 139
9	11, 32, −74, −76, 110, 112
10	−96, 117, 118, 119
11	10, −13, 28, −77, 92, −116, −132, 138
12	7, 17, 39, 41, 43, 131
13	40, 44, 49
14	21, −32, 74, 75, 94, 98
15	12, 66, 80
16	9, 15, 62, 65, −103
17	47, 54, 56, 57, 58, 59, 61, 63, 64, 104
18	20, 22, 47, 52, 67, 68, 72, 85, 93, 109, −116, 120, 124
19	48, 54, 56, 91, 138, 140
20	19, 58, 60, 65, 86, 92, 123, −131

[a] This listing of apomorphies by labeled interior node (2–20) of the strict consensus morphology-based cladogram (Fig. 4) gives characters hypothesized to have changed prior to each node according to their numbering in Appendix 1. Those preceded by a minus sign are hypothesized to have changed from 1 to 0, whereas all other changes are from 0 to 1. Only those changes consistent with all different optimization methods available in PAUP (ACCTRAN, DELTRAN, MIN F) are included in this listing. The strict concensus cladogram differs from the six individual minimum-length hypotheses as explained in the text.