

A genome-based phylogeny for Mollusca is concordant with fossils and morphology

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Abstract:

Extreme morphological disparity within Mollusca has long confounded efforts to reconstruct a stable backbone phylogeny for the phylum. Familiar mollusc groups—gastropods, bivalves, and cephalopods—each represent a diverse radiation with myriad morphological, ecological, and behavioral adaptations. The phylum further encompasses many more unfamiliar experiments in animal body plan evolution. Here, we reconstruct the phylogeny for living Mollusca based on metazoan BUSCO (Benchmarking Universal Single-Copy Orthologs) genes extracted from 77 (13 new) genomes, including multiple members of all eight classes with two high-quality

genome assemblies for monoplacophorans. Our analyses confirm a phylogeny proposed from morphology and show widespread genomic variation. The flexibility of the molluscan genome likely explains both historic challenges with their genomes and their evolutionary success.

Main Text:

Phylum Mollusca exhibits the widest disparity of body plans in metazoan evolution, and the inter-relationships of the living lineages have been a subject of contentious debate for centuries (1–3). The eight classes of extant molluscs are each unambiguously monophyletic, but their diverse morphologies allow many plausible sister-group combinations. The three most diverse groups are more familiar—bivalves, gastropods, and cephalopods—and the other five include worm-molluscs (Solenogastres, Caudofoveata), eight-shelled chitons (Polyplacophora), tubular infaunal predators (Scaphopoda), and headless limpets (Monoplacophora). Over 500 million years of morphological evolution have produced even more transitional forms in the fossil record, many with unclear affinities (4). Most studies have focused on deep phylogenetic nodes, resolving the clades Conchifera (Bivalvia, Cephalopoda, Gastropoda, Monoplacophora, Scaphopoda) and Aculifera (Caudofoveata, Polyplacophora, Solenogastres) (2, 5, 6). Resolving the topological positions of all eight clades is critical to understanding molluscan evolution (7), but genomic resources were limited or lacking for most classes.

Early molecular phylogenetic studies of molluscs sometimes completely conflicted with previous morphology-based hypotheses (8). Phylogenomic studies using transcriptome data covered all classes but had low coverage, outdated orthology inference methods, and uncertain topological support (2, 6). Results from standard genetic markers, mitochondrial genomes, transcriptomes, morphology, and the fossil record have at times seemed to be all mutually contradictory but increasingly converged toward a potential consensus (7, 9). Beyond the question of resolving molluscan phylogeny, a larger issue is understanding the fundamental evolutionary factors that made it so difficult and controversial to resolve.

We present a total group phylogeny for Mollusca based on genome-wide markers comprising all metazoan Benchmarking Universal Single-Copy Orthologs (BUSCO) in a phylogenomic analysis that includes all major clades and multiple representatives from all eight living classes. We assembled two near-complete genomes of the enigmatic deep-sea class Monoplacophora,

and generated genomes for five additional taxonomic classes: Caudofoveata, Scaphopoda, Gastropoda, Bivalvia, and Polyplacophora. This dataset, combined with previously published genome assemblies, underpins our new phylogenetic analysis that supports the origin of the phylum in the Cambrian and a rapid split into the major clades Aculifera and Conchifera (Fig 1).
5 Congruent with the extensive fossil record of molluscs and phylogenetic hypotheses proposed from morphology (Fig 2), we find that Monoplacophora is sister to the remaining Conchifera, and Cephalopoda is sister to the clade Gastropoda + Diasoma (Scaphopoda + Bivalvia).

Selecting representative data

10 Genome-wide data have played a pivotal role in reconstructing complex histories of the tree of life; however, genomic resources for Mollusca remain significantly underrepresented and unevenly distributed, with only 300 genomes for over 100,000 living species (Fig 3A). The quality of these assemblies varies widely, in large part because of intrinsic technical difficulties
15 that begin with challenges for sample acquisition, preservation, and DNA extraction and sequencing from animals that produce copious mucopolysaccharides (10). Assembly quality is further hampered by variability in genome size, heterozygosity, and repeat content, even within particular clades (Fig 3B-D). We generated 13 de novo genomes that fill gaps for key classes (two monoplacophorans, one caudofoveate, four chitons, one scaphopod), deeply divergent or
20 contentious lineages (*Solemya*, *Verpa*, *Tectura*), and diverse morphologies (*Scintilla*, *Concholepas*) (Fig S1, Table S1, Table S2). We considered genome quality and phylogenetic diversity, retaining species in controversial branches even where genome quality was slightly lower (Fig S2, Table S3). This totals 77 species covering all eight classes, including two Caudofoveata, two Solenogastres, eight Polyplacophora, two Monoplacophora, 11 Cephalopoda,
25 22 Gastropoda, three Scaphopoda, and 27 Bivalvia.

Our phylogenomic inference relied on metazoan BUSCO genes (11). BUSCO represents a universal gene set that provides effective comparative power across the phylum. Because BUSCO genes are conserved regions, they are suitable for reconstructing deep divergences.
30 Strict quality control in the selected genomes allowed us to identify a sufficient number and more even distribution of loci across species (Fig S2-S3): each species had an average of 897 BUSCO genes (94%) identified in the genome assemblies (Table S4). In order to test the impact

of gene occupancy on topology, we compared the metazoan BUSCO set and a reduced 96-gene dataset with 100% occupancy (Fig S3–S4).

The balance of taxon sampling and character (gene) occupancy is a persistent dilemma, especially in deeply divergent organismal groups (12). High coverage of lineages, high-quality molecular markers, and sufficient molecular loci enabled us to construct the phylogeny within extant Mollusca. Coupling these factors with a more even distribution of molecular markers across species also allowed us to recover the major branches within each class and estimate the divergence times within the molluscan tree of life (Fig 1).

The resulting tree recovered a topology that harmonizes key elements from former hypotheses into one topology: Aculifera including a monophyletic Aplacophora (2, 6), Conchifera with basal branching Monoplacophora, and the Gastropoda-Bivalvia-Scaphopoda (2, 6, 13, 14) group including Diasoma (13) (Fig 2, Fig S5).

A molluscan backbone

Morphological arguments about molluscan phylogeny historically focused on two alternative topologies, one with the clades Aculifera and Conchifera, or an alternative with basal branching Aplacophora (vermiform molluscs) and a shelled clade “Testaria” (9) (Fig 2). Early studies based on several loci recovered controversial alternative topologies with a putative sister relationship between Polyplacophora and Monoplacophora, termed Serialia, and other novel combinations (8). Later phylogenomic studies, despite low taxon sampling and data quality limitations, have mostly recovered Aculifera and Conchifera, but the relationships within Conchifera remained unresolved.

The fossil record of molluscs provides important evidence to complement genomic data (Fig 1A, Table S5), and vice versa: in our topology, the origin of cephalopods pre-dates Bivalvia and Gastropoda, although both appear earlier than cephalopods in the fossil record (3). Long-standing confusion over the affinities of univalved and bivalved small shelly fossils from the early Palaeozoic may be reinterpreted in light of a stable backbone phylogeny.

The first transcriptome-based phylogeny that included all eight classes resolved Monoplacophora as the sister to Cephalopoda (6), as did another recent study (14). This hypothesis is problematic as it is not supported by morphology or the fossil record (9). Adding partial genome data for a monoplacophoran resolved them as the sister to the remaining Conchifera, as predicted by morphological studies (2). Our subset analysis of a smaller gene set reproduced the Cephalopoda + Monoplacophora relationship; however, it also failed to recover a plausible ingroup topology among some bivalves (Fig S4). Cephalopoda + Monoplacophora is an anomalous result that likely arises from limited datasets.

Among Conchifera, Rostroconchia represents the ninth, extinct class of molluscs, which appears before both gastropods and bivalves in the fossil record (3). Rostroconchs exhibit character combinations from bivalves and scaphopods and are important evidence supporting Diasoma (3, 13). Evidence for Diasoma derives from morphology and fossils, but molecular data alone are not unequivocal. All phylogenomic studies have consistently supported a Gastropoda-Bivalvia-Scaphopoda clade. Early analyses recovered a clade (Bivalvia + (Gastropoda + Scaphopoda)), but often with relatively low support for inter-class relationships (2, 6, 13, 14). The first phylogenomic analysis including scaphopods recovered Diasoma, as also found here (Fig 1). Analyses of scaphopod genomes support interpretations of incomplete lineage sorting (13). Molecular data show higher support for a Gastropoda-Bivalvia-Scaphopoda polytomy than either bifurcation (Fig 2, Fig S5). These groups likely represent the descendants of a complex and rapid radiation from a common conchiferan ancestor. This clade, here named Megalopodifera (big foot-bearing), is united by the veliger larva (although the larvae of monoplacophorans are entirely unknown), a body that can retract into the shell, a reduction in the number of foot retractor muscles, and a large foot extending beyond the shell.

The molluscan tree of life

Internal topologies and timing within each class in the reconstructed phylogeny correspond well with established phylogenetic consensus and resolve some points of ongoing debate (Supplementary text, 15). The monophyly of Aculifera is not controversial and is supported by fossil (5), anatomical (16), and molecular studies (Supplementary text, 15). A deep split between Solenogastres and Caudofoveata in the Silurian corresponds to a radiation of disparate fossils

attributed to stem aplacophorans (5). Diversification within the Aplacophora is difficult to calibrate because of the poor fossil record of shell-less molluscs; the Mesozoic splits between sampled species in each class here may indicate that biodiversity recovery after the Triassic-Jurassic extinction included a radiation in Solenogastres. The topology of sampled chitons is consistent with morphological and paleontological evidence as well as previous genetic and genomic work (17, 18).

The two species used for the first full genomes of Monoplacophora include *Veleropilina oligotropha* from the Clarion Clipperton Zone, central Pacific, and an undescribed *Veleropilina* species collected from the Aleutian Trench that was morphologically indistinguishable from *V. oligotropha* but geographically distant (Table S1). The divergence time between them was calibrated using an earlier divergence time estimate for the living monoplacophorans, which estimated the first split among living monoplacophorans in the Cretaceous (19). Genome data from additional monoplacophoran groups and regions will likely influence this result.

Within Cephalopoda, the phylogeny of Decapodiformes (squid and cuttlefish) has been much debated, with conflicting hypotheses from mitochondrial genomes (20) or nuclear transcriptome data (21). Our phylogeny confirms a topology proposed from a large-scale five-gene analysis (22). Sepiolina has a basal branching position within Decapodiformes, which also agrees with results from transcriptomes (21, 23); we recognize Sepiolina as a taxonomic order, Sepiolida, based on this topology. Sepiida resolved as sister to the squids (Oegopsida + Myopsida). This scenario is more parsimonious than the previous “omics”-based phylogenetic analyses, as it suggests only a single loss of the calcareous phragmocone in the evolution of the clade. Our time-calibrated phylogeny indicates a relatively very recent radiation of Decapodiformes in the Cretaceous, in agreement with earlier work showing accelerating diversification rates in the Cenozoic (22). The diversification of extant coleoid cephalopods has been rapid.

Within living scaphopods, the new genome for *Fustiaria* revealed a much shallower divergence than previously predicted within Dentaliida (24) and for the origin of crown group Scaphopoda. Scaphopods have a long and complex fossil record with many stem lineages, and the biology of extant species remains poorly understood (3, 25).

Gastropoda is divided into two fundamental clades: Psilogastropoda, which unites Patellogastropoda as sister to Vetigastropoda + Neomphaliones, and Adenogonogastropoda, which comprises Neritimorpha as sister to Apogastropoda (Caenogastropoda + Heterobranchia) (Supplementary text, 15). This structure corroborates transcriptome-based studies that excluded the deep-sea subclass Neomphaliones (26, 27). A subsequent transcriptome study with expanded taxon sampling recovered contradictory topologies but supported Adenogonogastropoda (28), first identified in an early molecular study (29) and later named based on morpho-cladistic analysis (30). The early Cretaceous internal split in Neomphaliones also matches the scant fossil record for the group, as it post-dates the earliest neomphaline fossil from the Late Jurassic (31).

In contrast to most morphological analyses, our results indicate that Patellogastropoda is not the earliest branching clade within gastropods. A secondarily derived Patellogastropoda is recovered in most “omics”-based reconstructions (27, 32). One recent transcriptome tree (28) could recover basal-branching Patellogastropoda when rapidly evolving sites representing more than half of the dataset were excluded. Incongruity between morphology and molecular results leaves the position of Patellogastropoda uncertain, or awaiting further explanation. The limpet form has evolved convergently more than 50 times among gastropods but has its largest radiation in the patellogastropods (33). It is well-established that patellogastropods are split into two clades Patellida + Nacellida (3), but divergence time estimates in other studies vary wildly depending on the calibration points used within the clade. Our results are concordant with the fossil record, as the earliest known crown-group patellogastropod fossil is from the Triassic of Italy (34).

Our Caenogastropoda topology recovered Neogastropoda (*Concholepas*) nested within a paraphyletic Littorinimorpha (*Oncomelania* + *Alviniconcha*), consistent with previous findings (35). Our tree also supports Cerithioidea (*Batillaria* + *Semisulcospira*) clustering with hypsogastropods, which does not contradict the morphologically supported Sorbeoconcha concept (3). Some other studies (36), by contrast, unexpectedly recovered Cerithioidea as the sister to the architaenioglossan superfamily Ampullarioidea (represented here by *Pomacea*). In Heterobranchia, we recovered a monophyletic Tectipleura as sister to Ringipleura (*Berghia*). Within Tectipleura, the observed sequence of branching in Panpulmonata first gives rise to Sacoglossa (*Elysia*), then Hygrophila (*Anisus*), and finally Ellobioidea (*Ellobium*), and Stylommatophora (*Arion*), consistent with the recent consensus (3, 37, 38).

We recovered the consensus backbone phylogeny for bivalves (39), with Protobranchia (*Solemya*) in the earliest derived position, followed by Pteriomorphia, then Palaeoheterodonta (*Sinohyriopsis*), and Anomalodesmata (*Verpa*) sister to Imparidentia. Relationships within Pteriomorphia and Imparidentia also mostly align with other molecular and morphological results (40, 41) (Supplementary text, 15). One notable deviation is that the order Adapedonta (*Panopea*, *Sinonovacula*) is paraphyletic; the split between Adapedonta + Galeommatida (represented here by *Scintilla*) formed a conflicted node noted in earlier work (39, 42). The reduced shells of many galeommatidans led to a poor fossil record and taxonomic uncertainty. The rich fossil record of bivalves provides a well-resolved chronology for the origin of major lineages; however, our time-calibrated tree does not account for the impact of mass extinctions and rapid bursts that shaped bivalve diversification (43). Within Imparidentia, all orders except Lucinida first appear in the fossil record after the end-Permian mass extinction event (40, 42), as also found here. The common ancestor of crown-group Imparidentia necessarily had an origin before the end of the Permian, and the clade extends deeper in the stratigraphic record, which is not accounted for in our timetree.

Evolution of molluscan forms

The animal biomineralization toolkit predates the evolution of molluscs (44). Molluscs are effective at repurposing ancient gene families in novel ways (45). Phylogenetic relationships of phyla within Lophotrochozoa are not well resolved, and alternative hypotheses have different implications for the evolution of mineralized skeletons. Early small shelly fossils with molluscan affinity were used to calibrate the origin of Mollusca (Table S5). Some putative early molluscs lack a solid shell, such as *Odontogriphus* and *Shisania* (4); however, the ancestor of Aculifera + Conchifera did possess a shell, and fossils lacking solid shells represent further diversity and not the condition of the common ancestor leading to Aculifera + Conchifera.

Fossil molluscs present additional character mosaics that are not found in any living groups. The early Cambrian *Pelagiella exigua*, with a coiled shell but flexible chaetae, was interpreted as a stem gastropod (46), but our timetree suggests it is more likely a stem conchiferan. The deep divergence between Aplacophora + Polyplacophora (ca. 440 Mya) is congruent with mosaic

aculiferan forms in the fossil record (*Phthipodochiton*, *Kulindoplax*) from the Ordovician to Silurian (453–422.9 Ma) and much greater disparity in fossil forms in the aculiferan stem group. Character combinations underscore the flexibility of the molluscan genome to repeatedly re-deploy morphological adaptations.

Body plans in living molluscs include evidence of extensive convergence and character shifts on shorter timescales. For example, Juliidae, gastropods with bivalved shells, were once thought to be a transitional form between gastropods and bivalves but represent a recently derived adaptation to a specialized ecological niche (47). Endolithic bivalves occur in 11 separate families; *Verpa*, included here, has morphological modifications so extreme it cannot be accommodated in morphometric comparisons (48), but this family has a Cenozoic origin (49).

Important evolutionary transitions often require the transformation of shell mineralization and associated physiology, as seen in the repeated colonization of terrestrial and freshwater environments among bivalves, gastropods, and one estuarine cephalopod. Even the few non-marine lineages included in the study here show that terrestrialization events were time-independent and not directly connected to global patterns (Fig 1). Morphological and ecological plasticity has been a constant throughout the long history of the phylum.

Evolution of molluscan genomes

Broad taxon sampling now reveals that many molluscs have extremely high heterozygosity and a high proportion of repetitive sequences (Fig 3). These factors can result in highly fragmented assemblies, with mixed diploid sequences that are difficult to separate clearly, and multiple steps are needed for each new genome (1) (Table S2). In molluscs, concurrent high heterozygosity results in most assembly sizes being larger than the estimated genome size due to the inclusion of another haplotype (Fig 3, Table S2, Table S6). Estimates of heterozygosity for six of the eight classes are above 1%, with some species above 4%; these rates easily surpass other animal groups, such as Lepidoptera (butterflies and moths), where heterozygosity is considered high (50).

Genome features are not linearly correlated to morphological or ecological adaptations. The morphologically conserved Polyplacophora show very high rates of chromosome rearrangement (17). Well-conserved syntenic regions in more recent radiations of animals, such as mammals or birds, result in relatively similar genome structures, facilitating whole genome comparisons (51, 52). A new understanding of the variation in molluscan genomes provides a foundation for further work on fundamental questions of genome evolution, including the drivers of heterozygosity. Adding chromosome-level genomes for molluscan groups (e.g., Monoplacophora) is needed for comprehensive syntenic comparisons and to understand the role of chromosome rearrangements in speciation and cladogenesis. Several major lineages still have no genomes available: the bivalve clade Archiheterodonta and, at a finer scale, the bivalve orders Nuculida and Trigoniida (*Neotrigonia* is the only living member of an order known otherwise from fossils). Adding genomes for the gastropod order Cocculinida may alter, or confirm, the position of Patellogastropoda. Increased taxon sampling, especially in scaphopods, is important to fully understand molluscan topology.

Conclusions

Extensive economic and research interests have consistently held molluscs at the forefront of diverse disciplines, including fisheries, neurobiology, ecotoxicology, and biomimetic design. However, their extraordinary diversity fundamentally hinders a confident assessment of their evolutionary history. The question of why early genetic studies struggled to recover reliable evolutionary patterns is potentially answered by the extreme variability among genomes now available for all classes and their major clades within the phylum. Large-scale genomic analyses across the tree of life have provided inconclusive evidence regarding the drivers of genetic diversity (53, 54). These analyses are currently limited by incomplete sampling; to establish general principles of organismal phenome and genome, they must also include molluscs, the most extensive experiment in animal body plans.

A stable topology for the total group Mollusca is fundamental for understanding the evolution of body plans and the fossil record. Based on this new phylogeny, we infer that the molluscan ancestor had a solid dorsal shell, a foot, multiple dorsal-ventral and oblique foot retractor muscles, a radula, and lacked eyes. Throughout the long evolutionary history of molluscs and

continuing today, aspects of a flexible genome led to a flexible phenome: endless forms of molluscs showcase the power of animal evolution.

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Data and materials availability:

The whole genome sequencing data and novel genome assemblies generated in this study are available via the National Center for Biotechnology Information (NCBI): *Chaetoderma nitidulum* under the BioProject number PRJNA1122582; *Acanthochitona discrepans* under the number PRJNA1114954, *Boreochiton ruber* under the number PRJNA1120663, *Callochiton septemvalvis* under the number PRJNA1114372, *Deshayesiella sirenkoi* under the number PRJNA1114373, *Veleroipilina oligotropha* under the number PRJNA1120392, *Veleroipilina* sp. under the number PRJNA1120412, *Fustiaria rubescens* under the number PRJNA1120416, *Scintilla philippinensis* under the number PRJNA1120792, *Verpa penis* under the number PRJNA1120794, *Solemya velum* under number PRJNA1163513, *Tectura virginea* under the number PRJNA1120664, and *Concholepas concholepas* under the number PRJNA1120782. All other data are available in the manuscript or the supplementary material.

Supplementary Material

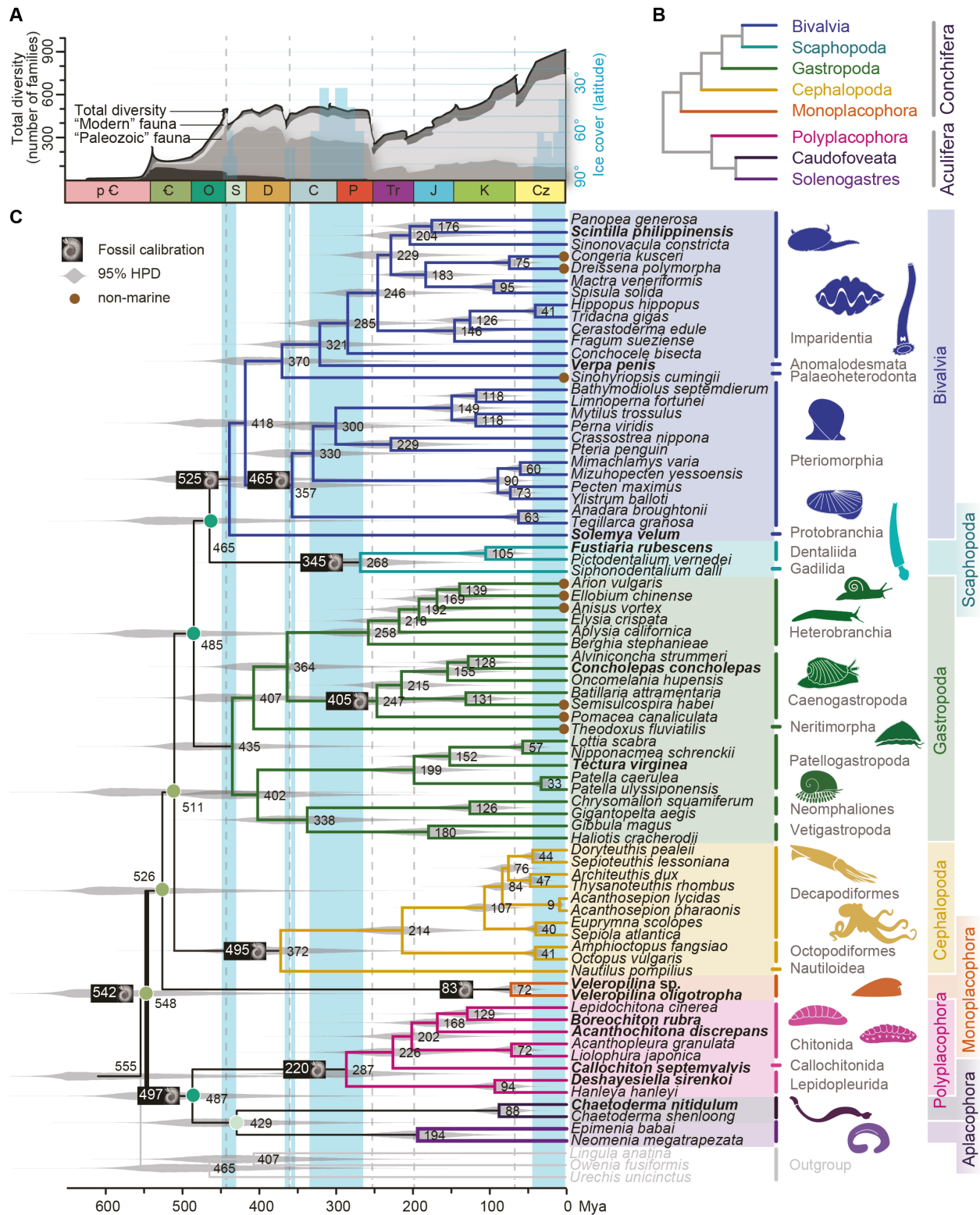
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of extensive ice cover(56) (blue bars, axis to the right). Marine invertebrate fossils, dominated by molluscs, are divided into three successive faunas in the Sepkoski curve shown here: Cambrian, Paleozoic, and Modern(55). **b.** Schematic showing the overview phylogeny of the eight living classes of Mollusca. **c.** Phylogenetic topology was computed by ASTRAL-Pro using a set of 954 BUSCO family trees from 77 species and three outgroups. Key clades are illustrated with icons (Table S7) Species from non-marine habitats are noted with brown dots on the branch tip. Divergence time estimates were obtained with MCMCTREE using the LG + Γ_4 + F model, IR. Fossil calibration times are labeled at the corresponding nodes in black boxes. Solid circles at the nodes emphasize the differentiation of eight classes, with the colors representing geological eras. The age distribution violin plot (grey) for each node shows the 95% highest posterior density intervals drawn by TreeViewer. Cz, Cenozoic; K, Cretaceous; J, Jurassic; Tr, Triassic; P, Permian; C, Carboniferous; D, Devonian; S, Silurian; O, Ordovician; C, Cambrian; p-C, pre-Cambrian.

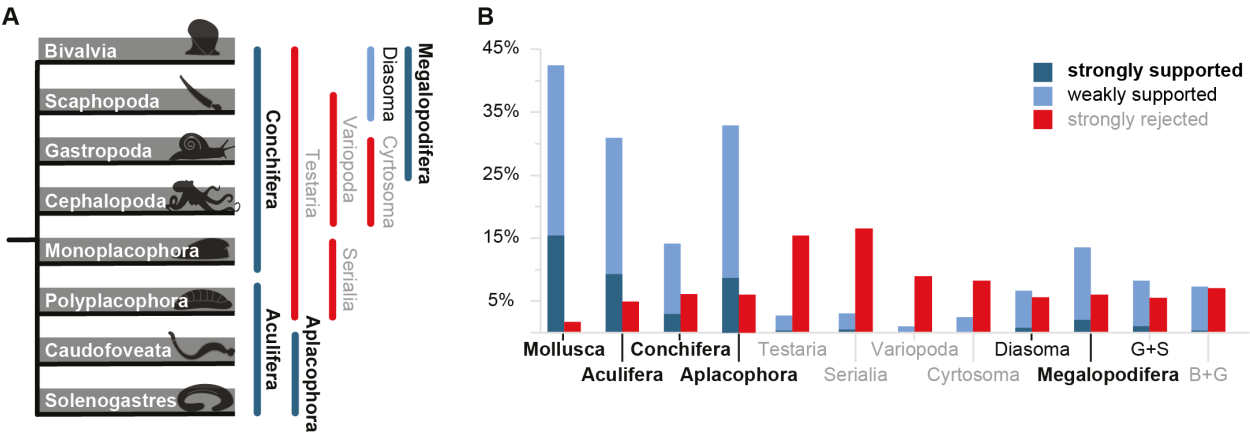


Fig. 2. Support for competing hypotheses in Molluscan relationships.

a. Schematic polytomy illustrating a subset of proposed molluscan ingroup relationships; colors illustrate the support. **b.** Bar chart(57) comparing the proportion among 945 gene trees that support (blue) and reject (red) different proposed clades within molluscs.

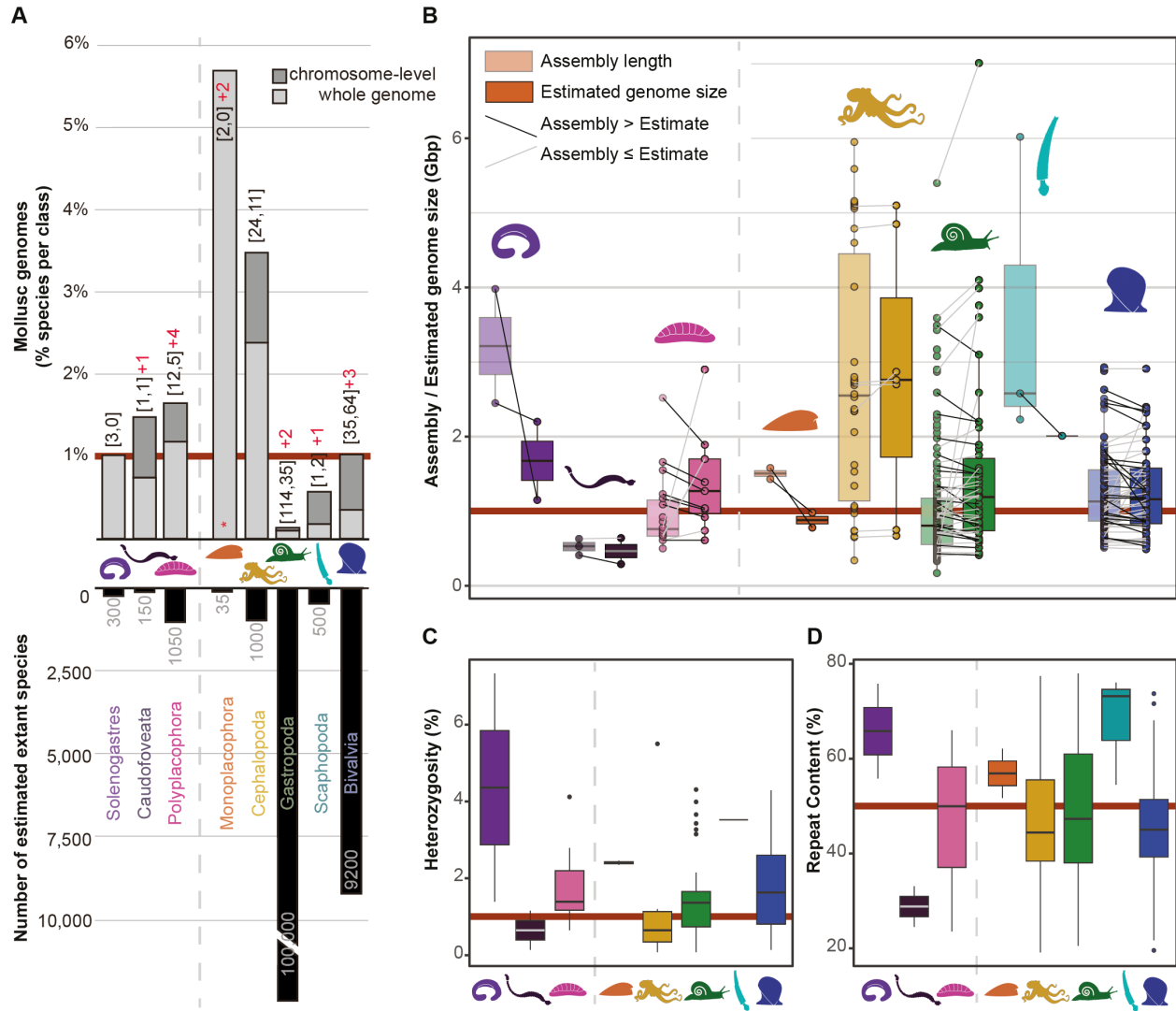


Fig. 3. Status and features of currently published molluscan genomes per class.

a. Variation in taxonomic richness and genome/chromosome-level availability. The numbers in brackets show non-chromosomal assemblies (first) and chromosomal level assemblies (second), including those in this study and all published in NCBI GenBank (20 May 2024). The numbers of new genomes are indicated in red (* in monoplacophorans notes that all available data are from the present study). A dashed line separates classes in Aculifera (left) and Conchifera (right).

b. Box plot summarizing reported assembly length (light colored) and estimated genome size (dark colored) median and inner quartile values for each class. Individual dots (without connecting lines) are values missing predicted genome size data. Darker lines highlight the common phenomenon of assembly size being higher than genome size due to very high heterozygosity. This phenomenon is not observed in cephalopods because the high percentage of repetitive sequences makes genome assemblies incomplete, combined with redundancy caused by high heterozygosity.

c. Estimated genomic heterozygosity per class. **d.** Estimated repeat content per class.



Supplementary Materials for

A genome-based phylogeny for Mollusca is concordant with fossils and morphology

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Materials and Methods

Sequencing and genome survey

Sample collection, preservation, and DNA extraction methods of the 13 new genomes are described in the supplementary data (Table S1). High molecular weight (HMW) gDNA was extracted from ethanol-preserved tissues of the monoplacophorans *Veleropilina oligotropha* and *Veleropilina* sp. using a modified protocol version of the Qiagen MagAttrack Kit (Hilden, Germany) in which the vortex mixing steps were replaced by flicking or gentle shaking of the tubes. For the chitons *Acanthochitona discrepans*, *Boreochiton ruber*, *Callochiton septemvalvis*, the gastropod *Tectura virginea*, and the bivalves *Scintilla philippinensis* and *Verpa penis* gDNA was extracted following the protocol of Mayjonade et al.(58); for the last two species mentioned, a pre-wash step with sorbitol was performed. For the scaphopod *Fustiaria rubescens* and the gastropod *Concholepas concholepas*, gDNA was extracted according to an alternative protocol(59). DNA concentration and DNA fragment length were assessed using the Qubit dsDNA BR Assay kit on the Qubit Fluorometer (Thermo Fisher Scientific) and the Genomic DNA Screen Tape on the Agilent 2200 / 4150 TapeStation system (Agilent Technologies). For the caudofoveate *Chaetoderma nitidulum*, DNA was extracted using the Circulomics Animal Tissue Big DNA Kit. For the bivalve *Solemya velum*, DNA was extracted using the MegaLong Genomic DNA Purification Kit. For the chiton *Deshayesiella sirenkoi*, DNA was extracted using the SDS method(60).

Whenever PacBio sequencing using the PacBio Low DNA input library protocol resulted in very low sequencing yields, we switched to the PacBio Ultra-Low DNA input protocol using different polymerases for PCR amplification(61) (Table S1). We prepared the low-

input PacBio HiFi libraries according to the SMRTbell Express Prep Kit v2.0 instructions. The PacBio Ultra-Low input HiFi libraries were prepared according to the manufacturer's instructions. To reduce the potential PCR bias of polymerase A/B, we used a third PCR reaction, using Polymerase C (KOD Xtreme™ Hot Start DNA Polymerase, Merck). The DNA libraries were size-selected to remove smaller fragments. A total of 28 SMRT 8M cells were sequenced in CCS mode using the PacBio Sequel II / IIe instrument. Libraries were loaded, where possible, with an on-plate concentration of up to 80 pM using adaptive/diffusion loading and the Sequel II binding kit 2.1, 2.2, 3.1 or 3.2 (Pacific Biosciences, Menlo Park, CA). Pre-extension time was 2 hours, run time was 30 hours. The PacBio Low and Ultra-low libraries for *Concholepas concholepas* and the PacBio Ultra-low library for *Velero pilina* sp. were loaded on a PacBio Revio instrument at an on-plate concentration of 200-250 pM using diffusion loading and the Revio polymerase kit (PacBio, Menlo Park, CA, USA). The run time was 24 hours.

HiFi reads were called using a pipeline, which is running PacBio's tools ccs 6.4.0 (<https://github.com/PacificBiosciences/ccs>), actc 0.3.1 (<https://github.com/PacificBiosciences/actc>), samtools 1.15 (62) and DeepConsensus 1.2.0 (63).

All commands were executed as recommended in the respective guide for DeepConsensus¹. For each species, we generated between 18 and 94 Gb of HiFi data, with sequencing depth ranging from 23 to 143X of their estimated genome size, and an average depth of 60X. For PacBio ultra-low libraries, PCR adapter sequences and duplicates were removed using lima v2.9.0 (<https://github.com/PacificBiosciences/barcoding>) and

¹ https://github.com/google/deepconsensus/blob/v1.2.0/docs/quick_start.md

pbmarkdup v1.0.3 (<https://github.com/PacificBiosciences/pbmarkdup>), and HiFiAdapterFit(64) (Table S2).

Genome size and heterozygosity were estimated from a k-mer profile of the HiFi reads (Fig S1). First, count from Jellyfish 2.3.0(65) was run with the additional parameters “-m 21 -s 100M” and all HiFi reads as input, then the GenomeScope v1.0 and v2.0(66) in combination with R 4.3.1 was executed with the Kmer size set to 21. For three species (*Acanthochitona discrepans*, *Boreochiton ruber* and *Tectura virginea*), fresh frozen tissue was available and we were able to perform genome size estimation using a flow cytometry (FCM) protocol using propidium iodid-stained nuclei(67) (Table S2).

Genome assembly

All the HiFi reads were processed for *de novo* genome assembly using hifiasm v0.16.1 with default parameters(68). We used the following three main methods (Polish, Purge_Dups, Purge Haplotigs) to remove duplicated haplotigs and overlaps (methods were adapted for each species, as shown in Table S2). Polish: The primary unphased contigs generated by hifiasm were polished using the following steps: all HiFi reads were first mapped to the draft assembly using minimap2 v2.24 with “-x map-hifi” in the parameters(69). Duplicated reads were removed from alignments using Picard v3.0.0(70), and the .bam file was sorted using samtools v1.15.1(71). Variants were called using DeepVariant v1.2(72) with default parameters, and the variants with “PASS” and “1/1” were passed to bcftools v1.13(71) to generate a consensus sequence. This was followed by Purge_Dups v1.2.6(73) and then Purge Haplotigs v1.1.0(74). Genome completeness was assessed by BUSCO v5.4.3(12), in euk_genome_met mode using the lineage dataset metazoa_odb10. We selected the optimal assembly results by taking into account the

continuity of the assemblies, the genomic integrity, and the proportion of duplicated haplotypes.

The proportion of complete BUSCOs was > 92% except for *Concholepas concholepas*, which is predicted to have a very high heterozygosity of 3.99% and a very high repetitive sequence content of 62% (Fig S1). It is worth noting that for some species there are a significant number of haplotigs that cannot be effectively removed (Fig S2). This is reflected in the very high content of duplicated BUSCOs and assembly lengths well above the estimated genome size, even when there is a very high sequencing depth (Table S1-3, Fig 3). This is the same problem faced by many other published mollusc genome assemblies (Fig 3, Fig S2, Table S3).

Data collection and phylogenetic inference

For our main coalescence-based tree analysis, we screened for 954 metazoan BUSCO genes(12). We did not include all possible samples from among available molluscan genomes with sufficient quality; for example, for species of the same genus, we retained only the one species with the highest BUSCO completeness. On the other hand, we deliberately included genomes with slightly lower quality where species represented lineages with controversial placements, deeper diverging branches, or fill important gaps. Other high-quality lophotrochozoan genomes were used as outgroups: *Lingula anatina*, *Urechis unicinctus* and *Owenia fusiformis* (Table S3).

Both coalescence-based and concatenated phylogenetic trees were inferred using sequences in the metazoa_odb10 BUSCO dataset(12, 75). Protein sequences of single-copy BUSCOs were extracted from each species. Considering the large proportion of duplicated

BUSCOs in many species, for the concatenated method, we manually selected (by length) one of the multicopy BUSCOs as single copy BUSCOs and added it to the final datasets. After screening for BUSCOs that are present in all 80 species, only 96 BUSCOs were left (Fig S3). Those genes were concatenated and aligned using MUSCLE v5.1.0 with the default parameter(76). The final alignment is 61,874 amino-acid sites. Maximum likelihood (ML) trees were inferred using IQ-TREE v2.1.3(77) using an extended model selection followed by tree inference (-m MFP), performing a full tree search for every model (--mtree) with 100 non-parametric bootstrap replicates (-b 100) (Fig S4).

For the main coalescent-based analysis, we used ASTRAL-Pro(78), which is able to integrate gene trees constructed from multicopy genes. We used all complete BUSCO genes extracted from all species, regardless of their single-copy or multi-copy status or species coverage. Each species had an average of 897 BUSCO genes (94% of 954 Complete BUSCOs from metazoa_odb10). An unrooted gene tree was inferred using RAxML v8.2.12(79) with 100 replicates for each gene with “-m PROTGAMMAAUTO”. The best-scoring tree of each gene was then selected and merged as input to ASTRAL-Pro3 v1.18.3.5 to infer the species tree with default parameters, as well as calculate the quartet support and posterior probabilities (PP)(78) (Fig S4).

Fossil constraints and divergence time estimation

Divergence time was computed using the MCMCTREE program(80) implemented in the PAML v4.10.7 package(81) using fossil calibrations that have been applied in previous time-calibrated phylogenies for Mollusca(2, 8) (Table S5). The prior was constructed with parameters $\lambda = \mu = 1$, $\rho=0$ (representing a uniform distribution of node ages given the root age). The overall substitution rate of 0.040245 per time unit (100 million years) was

estimated from the protein alignment (61,874 sites) with the 96 BUSCO set, using CODEML and the tree topology inferred by ASTRAL-Pro as input. Divergence time estimates were then obtained with MCMCTREE using the LG + Γ_4 + F model, IR, and the gamma prior was set to “G (2, 50)”. The number of iterations, the burn in and the sampling frequency were adjusted in test runs of the program to adjust to acceptance proportions close to 0.3, as suggested in the software notes.

Supplementary Text 1. Previous large phylogenomic studies within Mollusca

Sample representation

To illustrate the taxonomic coverage and main hypotheses in previous studies, and the advances provided by this present work, we compiled a table of previous large phylogenomic studies within Mollusca (Table S4). Since the evolutionary history of the mitochondrial genome differs from that of the nuclear genome, we only compared phylogenetic trees constructed from large orthologous genes based on whole-genome or transcriptome data, excluding mitogenome-based topologies. The use of orthologs from whole genomes and transcriptomes enables phylogenetic relationships to be inferred from hundreds or even thousands of loci. However, previous studies have shown that taxon sampling, the number of orthologs, compositional and rate heterogeneity, and incongruence across loci all affect phylogenetic recovery(21, 28, 82).

Among the eight classes of Mollusca, the genomic and transcriptome resources for Monoplacophora, Scaphopoda, and the two aplacophoran classes (Caudofoveata and Solenogastres) are consistently lacking and under-represented, while the more familiar classes, especially Cephalopoda, are better sampled (Main text Fig 3). Bivalves and gastropods are more often the dominant part of larger phylogenies. One recent study included seven out of eight classes of Mollusca, with a total of 134 species—116 of which were gastropods and bivalves(14). Gastropods are the clade with the most ‘omics’ data, but the taxonomic coverage is not even, with most data from the subclasses Caenogastropoda and Heterobranchia (Table S4). This has led to early phylogenomic studies unable to recover, or only poorly resolve, the full landscape of the phylogeny within the class or its constituent clades. For example, only one previous study included all six gastropod

subclasses with 25 species sampled, including representatives from the less-represented subclasses(28). Similarly, in Bivalvia, most data are from Pterimorphia and Imparidentia, with two new genomes here representing the first and so far only available genomes for other major clades (Protobranchia, Anomalodesmata). In Cephalopoda, most phylogenomic studies have focused on ingroup relationships within Decapodiformes.

Effects of ortholog number, missing data, and filtering

Transcriptome sequencing has been the most common approach for large-scale phylogenies, due to its time and cost savings(83). However, transcriptome data are influenced by factors such as lineage, developmental stage, specimen condition, and treatment, which can affect the expressed gene set. This variability may reduce the coverage of orthologous genes across species. For example, the number of orthologs dropped from 1,185 to 301 when species occupancy increased from 40% to 50% in a study of 35 mollusks covering eight classes(6). In Gastropoda, the number of orthologs dropped from 1,059 to 149 as taxon occupancy increased from 50% to 70% in a dataset of 74 species(27) (Table S4). It is also possible that the number of orthologous genes with phylogenetic utility reduces with an increasing number of taxa included(83).

Another challenge is that varying evolutionary rates among genes can affect tree inference, and can significantly alter the tree topologies as shown by a gastropod study: Uribe et al. recovered Psilogastropoda with 955 orthologs (BS = 98, LocalPP = 1) but recovered Orthogastropoda with strong support (BS = 96, LocalPP = 0.92) after discarding 30% of rapidly evolving sites(28). Previous work on cephalopods showed the order-level relationships within Decapodiformes were highly influenced by missing data and filtering methods(82), demonstrating that datasets maximizing taxonomic coverage but including

fewer orthologs were less stable than those that sacrificed taxon sampling to increase ortholog numbers. The comparison of our two main results (Fig S4, Main text) emphasizes that reduced datasets in favor of higher occupancy can lead to anomalous results.

Genomic data provide a relatively larger number of genes, and genome-based orthologues decrease less with increasing taxon occupancy, lending higher analytical power. Nonetheless, the limited availability of high-quality genomes has led whole genome-based phylogenomic studies to have more limited taxon coverage than transcriptome-based studies. For example, in a study of 22 molluscs covering five classes, 3,825 genes were recovered at 75% species occupancy, and 663 genes were recovered at 92% occupancy(13). In another study of 14 gastropods covering five subclasses, 1,610 homologous genes were identified at 80% species occupancy, and 933 genes at 94% occupancy(31) (Table S4).

Here, we utilized conserved BUSCO genes, which exhibit relatively consistent evolutionary rates. Our sampling strategy balanced BUSCO gene coverage with broad species diversity, yielding a comparable number of loci and sufficiently informative sites compared to previous studies, enabling us to resolve the deep phylogeny of Mollusca. As described in the main text, we recovered 954 genes with 93.75% species occupancy across 77 molluscs representing all eight classes (Fig 1, Fig S2). The representation of the three largest and most extensively studied classes—gastropods, bivalves, and cephalopods—in our phylogeny is comparable to previous studies in both gene and species coverage, with taxon sampling carefully selected to provide an even representation of major branches, and the addition of important new bivalve clades: We sampled 22 gastropods, covering all six subclasses, and deliberately selected a subset of available Heterobranchia and Caenogastropoda while increasing representation of the more controversial

Patellogastropoda. Only two genomes are available for the deep sea Neomphaliones and we aimed to include a minimum of two selected high-quality genomes representing Vetigastropoda. The inclusion of the only available genome from Neritimorpha exemplifies the trade-offs we made: although it retained only 77.4% of the BUSCOs (the lowest of all species), we included it in the tree with a total of 738 genes, which is still high compared to previous phylogenomic studies (Table S4). Our dataset includes 27 bivalves, covering five of the six major clades (lacking Archiheterodonta) with the major clades Imapridentia, Palaeoheterodonta, and Pteriomorpha, and adding genomes that represent two clades for the first time: Anomalodesmata and Protobranchia; there are also 11 cephalopods, covering all three subclasses: Nautiloidea, Octopodiformes, and Decapodiformes. Among the more under-studied classes we used a subset of available chiton genomes, with eight species representing all three orders: Lepidopleurida, Callochitonida, and Chitonida; and among the other classes we used all available genomes with three scaphopods representing both orders Gadilida and Dentaliida, two monoplacophorans, two solenogasters and two caudofoveates. Much important molluscan diversity remains to be sampled, but this is the first time there has ever been a pan-molluscan phylogeny with any molecular data with multiple representatives of all eight classes and comprehensive coverage of within-class diversity.

Supplementary Text 2. Internal relationships of Aculifera

Aculifera, comprising Polyplacophora and the aplacophorans, Solenogastres and Caudofoveata, is one aspect of molluscan evolution that is clearly supported by multiple lines evidence(84). The fossil record of chitons includes relatively large-bodied animals that had a seven- or eight-plate scleritome similar to modern chitons, but the body was completely enclosed by a spicule-covered cuticle: palaeoloricate chitons with no foot, or armored aplacophorans, which possibly had a solenogaster-like pedal groove(5, 84). There is also evidence that they were, like modern solenogasters, predatory(85). Our timetree, which dates the split between Solenogastres and Caudofoveata in the Silurian (Fig 1), agrees perfectly with the occurrence of the best preserved body fossils of footless chitons in the Silurian(5) and slightly earlier in the Ordovician(84), supporting the conclusion that these animals were the ancestors of modern aplacophorans.

Monophyly of Aplacophora

The shell-less, worm-like aplacophoran molluscs (Solenogastres and Caudofoveata) have been central to discussions of early molluscan evolution(86–98). Aplacophorans were historically considered plesiomorphic and “basal” within Mollusca because of their relatively simple morphology and possession of aragonitic sclerites rather than one or more shells(86–91, 97, 98). Whether these two groups constitute a monophyletic taxon, Aplacophora(99–101), or a paraphyletic grade(89, 91, 97, 98) has been widely debated(9). Morphology has been variously interpreted to suggest basal placement for caudofoveates (Scutopoda-Adenopoda hypothesis)(89), which are distinguished from other molluscs by lacking a foot, but also solenogasters (Hepagastralia hypothesis)(91, 98), which are distinguished from other molluscs by lacking distinct subdivision of the midgut, ctenidia,

and a radular membrane. Notably more recent histological and ultrastructural work has demonstrated the presence of a true radular membrane in at least some Solenogastres(103, 104). Studies of Entoprocta (=Kamptozoa), a hypothesized molluscan sister taxon(105, 106), have also been cited in support of the Hepagastralia hypothesis as the solenogaster nervous system and preoral sensory organ are similar to those of larval entoprocts(107).

In contrast to hypotheses placing the Solenogastres and Caudofoveata as a basal, paraphyletic grade, the Aplacophora hypothesis(99–101) unites the two groups in a monophyletic clade. Morphological synapomorphies of Aplacophora include vermiform body shape, dorsoterminal sensory organs, and paired gonads that open into the pericardium and exit through U-shaped pericardioducts. The presence of a radula with two teeth per row (distichous) has also been viewed as a synapomorphy for Aplacophora(108) but some aplacophorans have a radula with one, four, or many teeth per row(109) and more data are needed about the internal phylogeny of the group to confidently reconstruct the ancestral state in Solenogastres(110).

Molecular phylogenetic studies have overwhelmingly supported Aplacophora as a monophyletic group. Early molecular analyses relied heavily on nuclear ribosomal RNA (rRNA) genes but often struggled to resolve the placement of the aplacophoran taxa (and higher-level molluscan relationships in general)(111–115). These studies faced challenges, particularly with contamination of solenogaster samples by cnidarian and annelid prey DNA, sometimes leading to contaminant or chimeric rRNA sequences(116, 117). However, studies employing nuclear protein-coding genes have consistently recovered a monophyletic Aplacophora with strong support(2, 6, 118–123). These studies have employed diverse taxon sampling schemes and methods for orthology inference and data analysis (Table S4) but have consistently converged on strong support for Aplacophora.

Similarly, our analyses overwhelmingly support the monophyly of Aplacophora (Figs 1-2, Fig S5).

Genomic resources for Caudofoveata only became available in 2024, with the publication of the first genome assembly for this class from an undescribed species(124) which has subsequently been named *Chaetoderma shenloong*(125). This species is highly unusual for the group, as it is endemic to a deep-sea cold seep and is potentially chemosymbiotic. Highly adapted lineages used alone are not ideal choices for resolving deep phylogenetic divergences(1). To support the results presented here, it is important that we were able to include a second, new genome for the class from a distantly related, more “typical” species *Chaetoderma nitidulum*. Caudofoveata is generally very poorly understood and lacks any fossil record.

Supplementary Text 3. Internal relationships of Gastropoda

The internal relationships of major clades within Gastropoda have been in flux for decades. This is despite numerous analyses using morphology, genetic, or combined datasets having repeatedly recovered the same six major clades – Patellogastropoda, Vetigastropoda, Neomphaliones, Neritimorpha, Caenogastropoda, and Heterobranchia (26, 28, 126). Based on morphology, a basal split of gastropods into Eogastropoda (the only extant group being Patellogastropoda) and Orthogastropoda (all other subclasses) was proposed and reliably recovered in morphocladistic studies(127). Though this was supported in the first phylogenomic tree(123), the vast majority of “omics” analyses since then have failed to find support for the Eogastropoda/Orthogastropoda division(6, 14, 26, 27, 128). Instead, Patellogastropoda was frequently recovered in a derived position sister to Vetigastropoda (or Vetigastropoda + Neomphaliones, after the first neomphaline genome became available(45)); a clade later named Psilogastropoda(27), after the unprotected nature of their gametes, embryos, and larvae – a condition considered plesiomorphic(3). Most recent support for Orthogastropoda comes from mitogenome analyses(129, 130) as well as a transcriptomic study (noted above) which found the progressive removal of rapidly evolving sites led to differing topologies, which switched from supporting Psilogastropoda to Orthogastropoda when more than 30% of such sites were deleted(28). Some previous genome-based phylogenetic studies based on single-copy orthologous genes (SOGs) also recovered Orthogastropoda(131,132); however, it was subsequently discovered that with increasing number of SOGs, the topology changed to Psilogastropoda *s.l.* with high support (131). Our tree presented herein (Main text Fig 1) again resolves Patellogastropoda in a derived position as sister to Vetigastropoda + Neomphaliones (Psilogastropoda *s.l.*(31)).

The Apogastropoda hypothesis uniting Caenogastropoda and Heterobranchia was first proposed based on comparative anatomy(133) and then predominantly supported by early molecular or combined-data trees(29, 126) as well as reconstructions using “omics” data(2, 6). In these molecular analyses, Neritimorpha has been most recovered as the sister of Apogastropoda(29), a condition later named Adenogonogastropoda(30) (synonymous with Angiogastropoda proposed later in(27)), based on their derived reproductive traits and veliger larva. Yet, this position of Neritimorpha remains unsettled as several studies have failed to recognize Adenogonogastropoda. A couple of morphocladistic studies have proposed Neritimorpha as sister to Neomphaliones(134) or at a more basal position diverging just after Patellogastropoda(127) and a multi-loci study notably observed Neritimorpha as the earliest diverging subclass within Gastropoda(135). A more recent study using 107 SOGs recovered Neritimorpha as the sister to Psilogastropoda *s.l.*, though only one neritimorph was included(14). These anomalous relationships have not been substantiated since. The most likely alternative hypothesis for Adenogonogastropoda that has some support from both morphology and multi-locus or mitogenome phylogenies(3, 136, 137) is that Neritimorpha may be the sister group to Caenogastropoda. Our phylogenomic tree (Main text Fig 1), however, recovers a well-supported Adenogonogastropoda, adding further support to this hypothesis(26, 30). Nevertheless, as our support for this node is moderate (posterior probability = 0.852), alternative hypotheses are not completely rejected (Fig S4).

Supplementary Text 4. Internal relationships of Bivalvia

Previous molecular studies have provided strong evidence supporting the monophyly of Bivalvia, including comprehensive phylogenomic(39, 138) and more recent target capture analyses(139). Bivalve systematics has been substantially revised based on molecular results in the last 20 years, but with increasing confidence and a topology that is corroborated here. Studies using phylogenomic(39, 138) (Table S4), nuclear markers(140), and combined approaches including morphology(141) all largely agree. Work using mainly mitochondrial genes often recovered contradictory topologies(8, 142–144). Taxonomic gaps remain in genomic data, and the addition of some key lineages will be interesting for understanding evolution within the class. Nonetheless, the generalized topology of living clades appears well resolved. The backbone phylogeny for bivalves that is supported by recent analyses with different lines of evidence, and the topology recovered here, includes Protobranchia sister to Autobranchia, and within Autobranchia, Pteriomorphia sister to Heteroconchia, and within Heteroconchia a monophyletic Euheterodonta.

The main clades that we refer to here (Fig 1, Fig S4)—Anomalodesmata, Imparidentia, Palaeoheterodonta, Pteriomorphia, and Protobranchia—represent important subdivisions within the class that are informative for comparisons with other phylogenetic results, but these are not groups of the same taxonomic rank. In the current classification, Bivalvia is divided into two subclasses: Autobranchia and Protobranchia. The latter subclass is represented in our analyses by *Solemya*. Other work on the internal topology of Protobranchia has confirmed its monophyly(145), but genomic resources are lacking for the group (apart from the *Solemya* genome included here). Protobranchia includes many small-bodied, deep-sea, and taxonomically difficult groups. The more familiar and well-

studied bivalves are found within Autobranchia. Autobranchia comprises two clades that are given the rank of infraclass: Heteroconchia and Pteriomorphia. The latter clade is represented here by a selected subset of 12 species from a relatively large set of available genomes (Table S6). Pteriomorphia includes many of the economically important bivalves that contribute to large-scale fisheries, including oysters, mussels, scallops, and pearl mussels. Heteroconchia comprises the four other major clades of bivalves: Archiheterodonta (not represented in the present analysis), Anomalodesmata, Imaridentia, and Palaeoheterodonta. Anomalodesmata and Imaridentia together form a clade Euheterodonta.

The topology within each clade in our analysis agrees with previous analyses. Within Pteriomorphia, Mytilida is the sister group to Ostreida, and Arcida is the sister group to all other pteriomorphians. Pteriomorphian monophyly is robustly supported, with well-resolved major clades: Mytilidae, Ostreida (including Pinnidae, Ostreoidea, Pterioidea), Arcida, and Pectinoidea(39, 138, 140, 146). Heterodonta (Palaeoheterodonta+ Euheterodonta) is a diverse group that includes clams and cockles with both marine and freshwater representatives. This clade features diverse families including Cardiidae (*Cerastoderma*, *Fragum*, *Hippopus*, and *Tridacna*) and Dreissenidae (*Dreissena*). Veneridae, one of the largest bivalve families and perhaps the canonical heterodonts, are missing from the present analysis, but the order Venerida is represented by Mactridae (*Mactra*, *Spisula*). Recent phylogenetic analyses have provided strong support for the monophyly of Imaridentia and have introduced new hypotheses regarding the relationships within this clade, such as the grouping of Galeommatida with Adapedonta and Cardiida(39, 40, 139). Within Imaridentia, we recover families Dreissenidae and Mactridae as sister lineages, results consistent with previous phylogenomic studies(39, 40,

139). Congruently, Thyasiridae (*Conchocele*) is among the earliest diverging lineages within Imparidentia(39).

In our alternative analysis (Fig S4), while the topology of the large clades is maintained, the organization of species within Imparidentia is not recovered as expected from other phylogenetic and systematic results. The derived position of *Conchocele* in our alternative analysis is not concordant with other results(147, 148). Further, the family Cardiidae s.l. (noted above) is monophyletic in our main analysis but not monophyletic in the alternative, concatenated-based tree. The subfamilies Fraginae (*Fragum*) and Tridacninae (*Hippopus* and *Tridacna*) are photosymbiotic, unlike other cockles(149). In the alternative, concatenated-based tree, these shallow, photosymbiotic cardiids (Fraginae+Tridacninae) resolve as the sister to the deep sea symbiotic bivalve *Conchocele*. This is evidentially an artifact, potentially reflecting some convergence, and lends further support to the overall conclusion that the results of the main, coalescence-based tree provide a more robust representation of molluscan evolution.

The bivalve fossil record is extensive and has been used in modeling diversification dynamics in deep time (Fig 1)(42, 55). The total diversity of bivalves, including extinct lineages, is much larger in terms of taxon richness and morphology than what is represented by the descendent lineages that survive today. It is also clear from the fossil record that the evolution of bivalves (and other groups) has not proceeded at a constant pace of speciation, but is characterized by sequential bursts and radiations(55, 150). Among Imparidentia, the family Lucinidae (not sampled in our analysis) has a fossil record that extends to the Silurian(42, 151). This means that our result for the split between Anomalodesmata and Imparidentia occurring in the Carboniferous (later than the Silurian) is probably erroneously recent. The new timetree here is calibrated based on relatively few fossils

(Table S5), and while it is concordant with the fossil record for deep splits and major clades, it cannot capture the full information available from the fossil record.

Supplementary Figures S1-S5

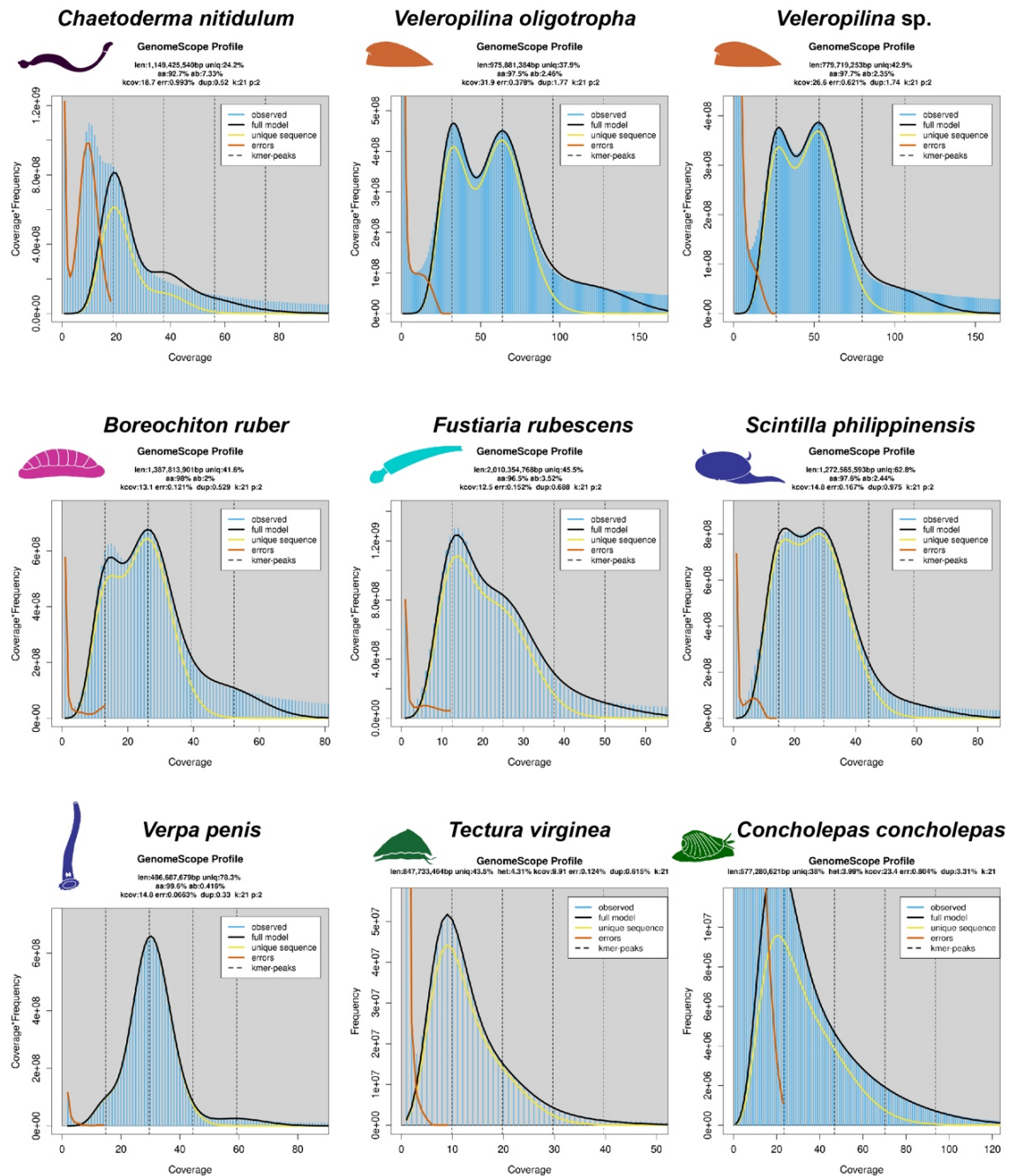


Fig S1 Estimation of genome features based on the distribution of 21-mer frequency. These represent nine of thirteen new genomes in the present study (those generated in the Translational Biodiversity Genomics Centre, Frankfurt, Germany) that are not part of other publication projects (see Table S1).

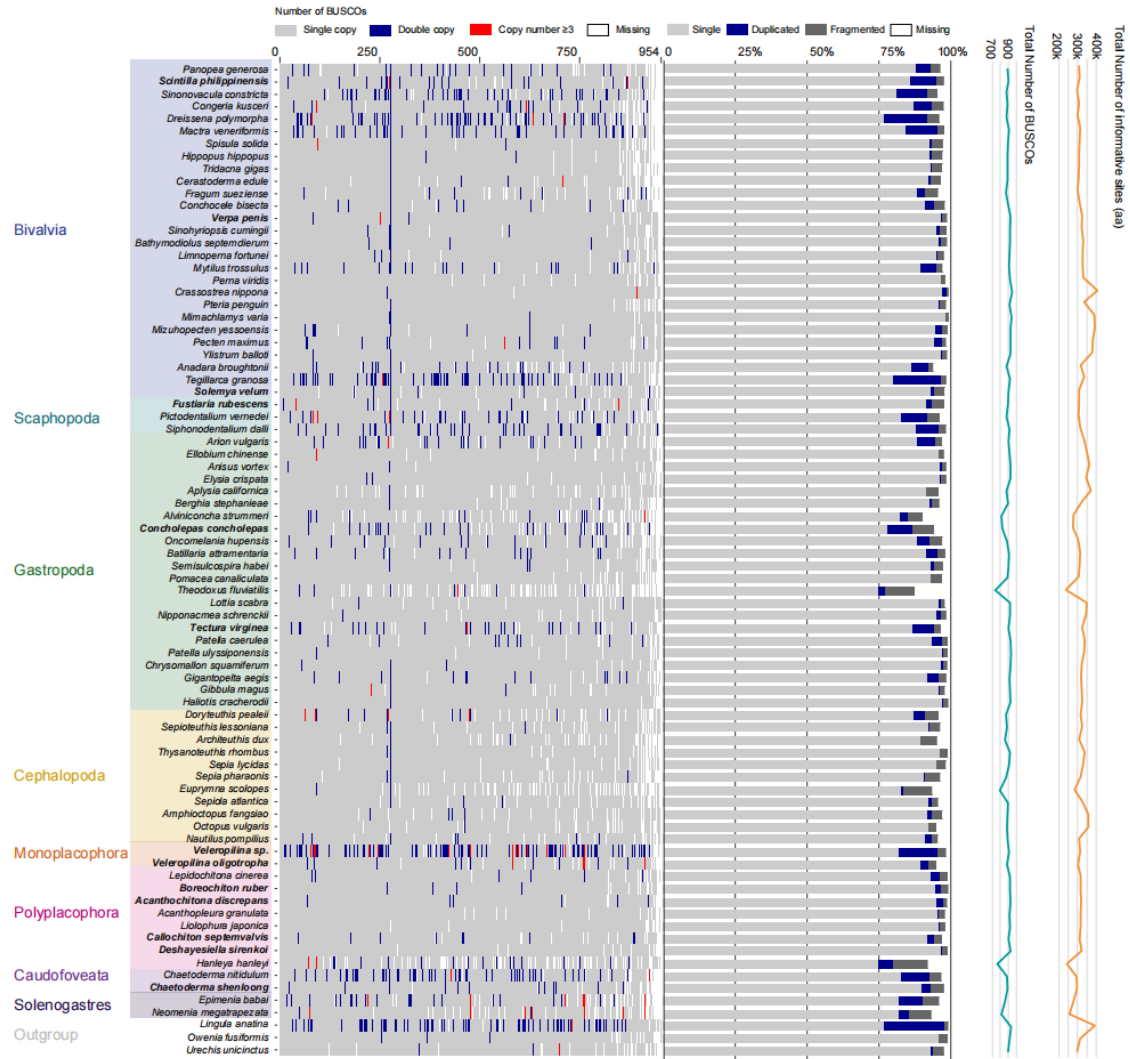


Fig S2 The BUSCO gene array (metazoa odb10 datasets) (left) used to construct the mollusc phylogenetic tree and statistics of single-copy, duplicated, fragmented, and missing BUSCOs for each species (right). Species names in bold text indicate new genomes. The green and yellow lines (far right) show the total number of genes and informative amino acid sites for each species, respectively.

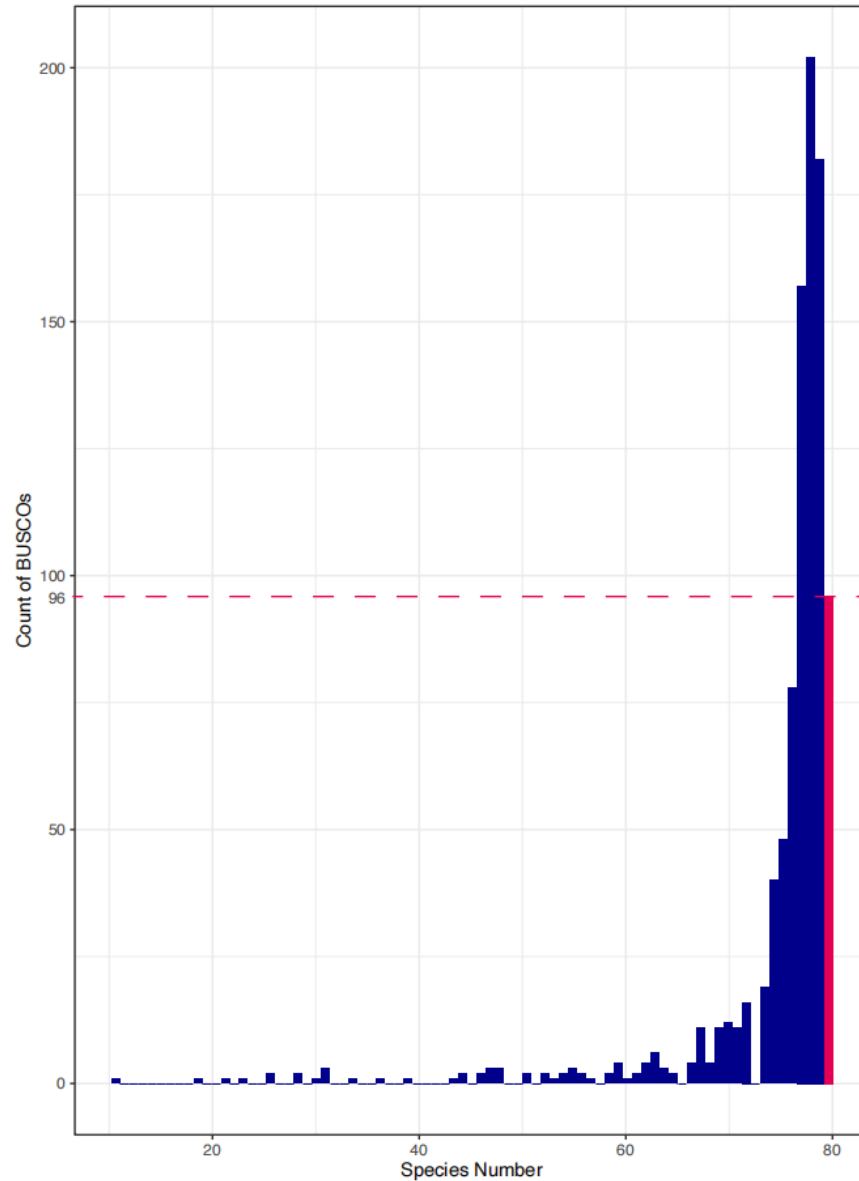


Fig S3 Frequency of species-group sizes with shared BUSCO genes. Each marker in the BUSCO gene set occurs in one or more species (maximum 80 species in this analysis). This plot shows the frequency of how many BUSCO genes were found in only one species, how many were found in two species, etc. The maximum overlap is a group of 78 species. Groups of 80 species indicate full coverage (77 ingroup species + outgroups), or 100% occupancy, which is the case for a set of 96 BUSCO genes (last column, marked in red) used in the construction of the concatenated species tree (Fig S4).

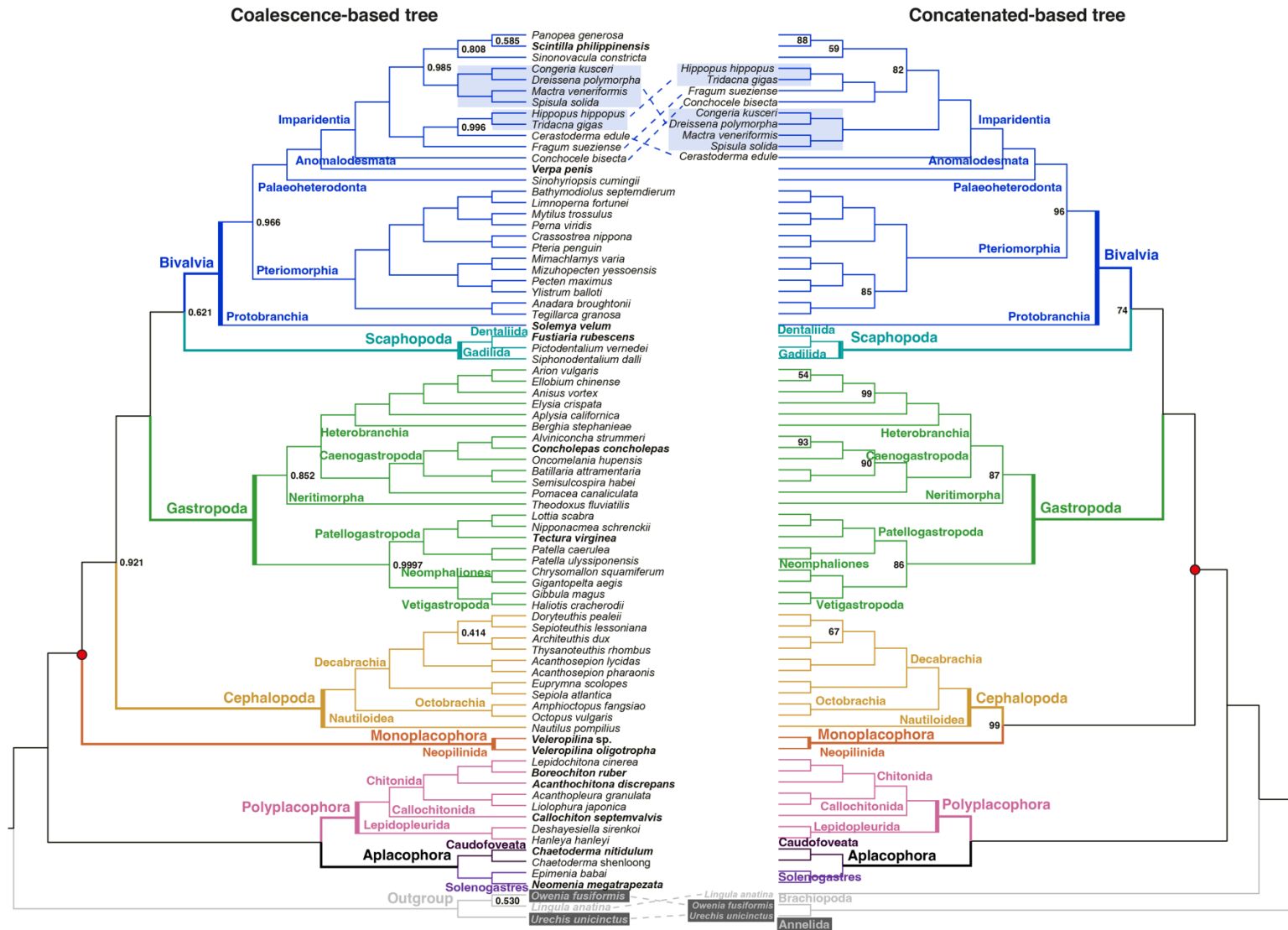


Fig S4 (previous page) Tree topology inferred from coalescence-based method from 954 BUSCOs gene trees (left) and concatenated-based method from 96 BUSCOs with 61,874 amino-acid sites covered by all 80 species (right). The numbers of the nodes represent local posterior probability that do not have a support of 1 (left), and bootstrap values that do not have a support of 100 (right). The positions of Monoplacophora differed between the two trees at the nodes indicated with red dots, and the shifting positions of several taxa are indicated with dashed lines.

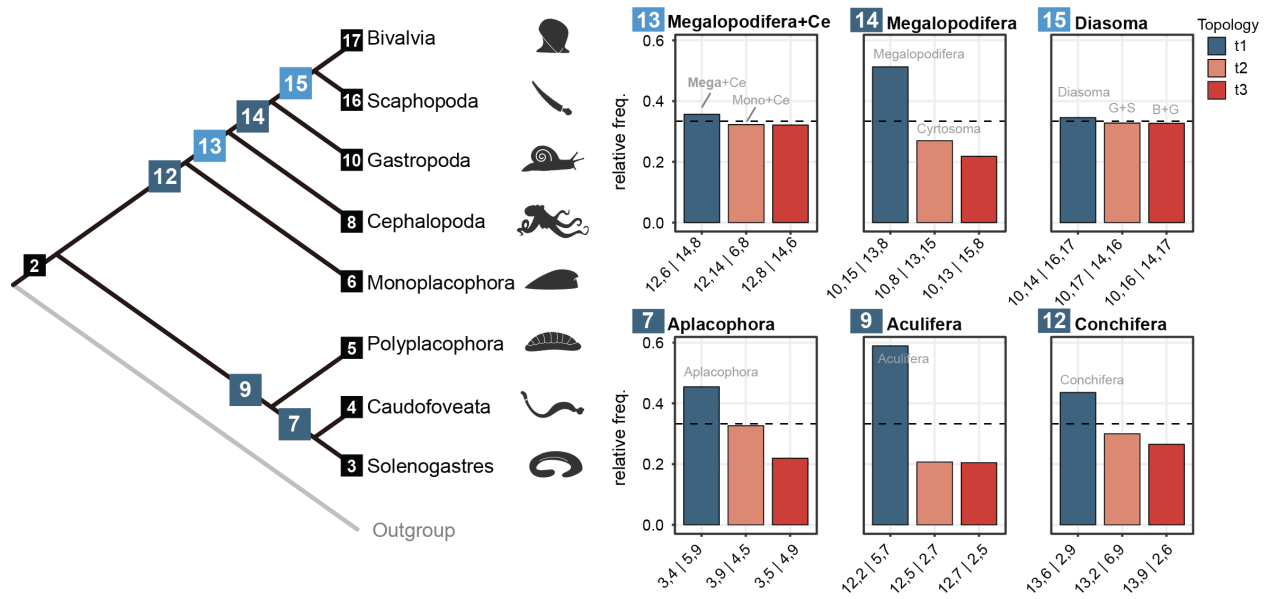


Fig S5 Frequency of three topologies around each of the controversial internal branches of molluscan phylogeny, analyzed using DiscoVista(57). **Left**, class-level cladogram based on the ASTRAL species tree (Main text Fig 1). Numbered branch labels in dark blue indicate highest support for the respective clades. **Right**, bar charts showing the relative frequency analysis for six aspects of the topology. The number at the top left of each subfigure indicates the corresponding node on the tree shown to the left. Each internal branch has four neighboring branches, which could be used to represent quartet topologies. On the x-axis of each subfigure panel, the exact definition of each quartet topology is shown using the neighboring branch labels separated by “|”. For example: branch “13” indicates the clade Megalopodifera+Cephalopoda, and the highest support is shown for the quartet 12,6 (Conchifera, Monoplacophora) | 14,8 (Meaglopodifera, Cephalopoda), with lower support for 12,14 (Conchifera, Megalopodifera) | 6,8 (Monoplacophora, Cephalopoda). The main (highest supported) topologies are shown in dark blue, and two less-supported alternative topologies are shown in light and dark red. The dotted lines indicate the 1/3 threshold (equal support for all three alternatives).

Supplementary Tables

- Table S1** Sample collection and sequencing information.
- Table S2** Genome assembly and assessment of continuity and integrity.
- Table S3** Assemblies used in the construction of the mollusc phylogenetic tree.
- Table S4** Summary of phylogenomic studies on Mollusca.
- Table S5** Fossil calibration used for divergence time estimation of Mollusca.
- Table S6** Mollusc genomic features.
- Table S7** An illustrated key to the schematic molluscs.

Table S1 Sample collection and sequencing information. Species were identified by one or more co-authors of this work, all specimens were collected as adults.

Class	Species name	Sample collection	Longitude and latitude	Preservation
Caudofoveata	<i>Chaetoderma nitidulum</i>	Barents Sea, Russia	70°25'30.0"N, 42°17'18.0"E	95% EtOH and RNAlater; partly at room temperature
Polyplacophora	<i>Acanthochitona discrepans</i> *	Northern Ireland: Kircubbin, Strangford Lough	54°29'24.0"N, 5°32'24.0"W	absolute ethanol partly at room temperature
Polyplacophora	<i>Boreochiton ruber</i>	Northern Ireland: Kircubbin, Strangford Lough	54°29'24.0"N, 5°32'24.0"W	fresh frozen
Polyplacophora	<i>Callochiton septemvalvis</i> *	Northern Ireland: Kircubbin, Strangford Lough	54°29'24.0"N, 5°32'24.0"W	frozen (liquid nitrogen, -80 °C)
Polyplacophora	<i>Deshayesiella sirenkoi</i> *	Pacific Ocean: Daikoku vent field	35°26'31.4"N, 139°42'59.9"E	fresh frozen
Monoplacophora	<i>Veleropilina oligotropha</i>	Pacific Ocean: Clarion-Clipperton Zone, Area 1 (German claim - prospective area)	11°49'12.0"N, 117°02'24.0"W	absolute ethanol partly at room temperature
Monoplacophora	<i>Veleropilina</i> sp.	Pacific Ocean: Aleutian Trench	9°24'34.7"S, 123°24'57.4"W	absolute ethanol partly at room temperature
Scaphopoda	<i>Fustiaria rubescens</i>	Spain: Roses	42°13'54.7"N, 3°11'52.6"E	absolute ethanol partly at room temperature
Bivalvia	<i>Scintilla philippinensis</i> *	Malaysia: Merambong Island	1°18'54.0"N, 103°36'36.0"E	absolute ethanol partly at room temperature
Bivalvia	<i>Verpa penis</i> *	Malaysia: Merambong Island	1°18'54.0"N, 103°36'36.0"E	absolute ethanol partly at room temperature
Bivalvia	<i>Solemya velum</i> *	Atlantic Ocean: Veckatimet Island	41°30' 31" N, 70°42' 04" W	frozen (liquid nitrogen, -80 °C)
Gastropoda	<i>Tectura virginea</i>	Northern Ireland: Kircubbin, Strangford Lough	54°29'24.0"N, 5°32'24.0"W	frozen (liquid nitrogen, -80 °C)
Gastropoda	<i>Concholepas concholepas</i>	Chile: Caleta Constitucion, Antofagasta	23°25'30.9"S, 70°35'56.7"W	absolute ethanol partly at room temperature

* Included in separate genome papers published in parallel to this study

Table S1 (continued)

Species name	Library preparation method (see Methods)	Total HiFi data (bp)	No. reads	Genome size (bp) estimated based on kmer analysis	Sequencing depth (X)
<i>Chaetoderma nitidulum</i>	low/9	64,759,040,306	7,646,630	1,149,425,540	56
<i>Acanthochitona discrepans</i> *	low/2	30,682,344,156	5,785,025	1,015,241,929	30
<i>Boreochiton ruber</i>	low/2	45,338,817,201	6,321,498	1,378,578,216	33
<i>Callochiton septemvalvis</i> *	low/2, ultral low A+B/4	39,763,935,831	8,086,367	1,040,963,215	38
<i>Deshayesiella sirenkoi</i> *	SMRTBELL library prep for HiFi sequencing kit	78,716,116,715	4,713,356	1,266,396,972	62
<i>Veleropilina oligotropha</i>	low/1, ultral low A+B/4	94,275,002,942	14,580,701	968,525,216	97
<i>Veleropilina</i> sp.	low/1, ultral low A+B/1	60,055,406,285	8,584,304	779,719,253	77
<i>Fustiaria rubescens</i>	low/2, ultral low A+B/1	61,339,970,554	8,323,485	1,989,688,191	31
<i>Scintilla philippinensis</i> *	low/3, ultral low A+B/1, ultral low C/1	69,019,919,367	10,203,359	1,265,546,934	55
<i>Verpa penis</i> *	low/2	17,812,702,580	3,273,748	486,155,513	37
<i>Solemya velum</i> *	PacBio 20 kb Template Preparation	92,857,307,651	14,926,436	1,497,672,255	62
<i>Tectura virginea</i>	low/2	19,408,332,528	3,498,911	838,738,493	23
<i>Concholepas concholepas</i>	low/1, ultral low A+B/1, ultral low C/1	82,319,625,462	12,958,611	577,280,621	143

Table S2. Genome assembly and assessment of continuity and integrity. Key to assembly methods: a: lima, pbmarkdup, a+:HiFiAdapterFit, b: hifiasm with default parameter, b+: hifiasm with disabled purge duplication (-l 0), selection of more complete haplotypes, c: picard MarkDuplicates, deepvariant polish, d: purge_dups, e: purge_haplotigs

Species name	Assembly method	Genome size (Gbp) estimated based on kmer analysis	Genome size (Gbp) estimated based on flow cytometry	Assembly length (bp)	Contig N50	No. contigs
<i>Chaetoderma nitidulum</i>	b,e,d	1.149	-	3,742,095,578	708,380	9,155
<i>Acanthochitona discrepans</i> *	b,c,d,e	1.015	1.058	1,173,574,984	4,185,741	704
<i>Boreochiton ruber</i>	b,c,d	1.379	1.578	1,660,742,690	4,032,827	825
<i>Callochiton septemvalvis</i> *	a+,b+,d	1.041	-	1,229,767,851	619,644	3,543
<i>Deshayesiella sirenkoi</i> *	a+,b,d	1.266	-	1,546,234,380	27,005,026	358
<i>Veleropilina oligotropha</i>	b,c,d	0.969	-	1,425,193,133	289,878	10,421
<i>Veleropilina</i> sp.	a,b,e	0.780	-	1,576,671,360	57,663	42,440
<i>Fustiaria rubescens</i>	b,c,d	1.990	-	2,578,751,445	846,414	5,436
<i>Scintilla philippinensis</i> *	a,b,e,d	1.266	-	1,671,121,595	497,550	6,192
<i>Verpa penis</i> *	b,d	0.486	-	507,038,511	5,325,940	522
<i>Solemya velum</i> *	d,f	1.498	-	1,497,672,255	390,153	8,353
<i>Tectura virginea</i>	b,c,d	0.839	0.969	1,020,027,926	540,487	3,253
<i>Concholepas concholepas</i>	b	0.577	-	648,998,573	105,660	9,464

* Included in separate genome papers published in parallel to this study

Table S2. (continued)

Species name	BUSCOs	Genome Accession Number	PRJ
<i>Chaetoderma nitidulum</i>	C:92.5%[S:82.8%,D:9.7%],F:4.3%,M:3.2%,n:954	JBITNJ000000000	PRJNA1122582
<i>Acanthochitona discrepans</i> *	C:97.5%[S:95.0%,D:2.5%],F:1.3%,M:1.2%,n:954	PRJNA1114954	PRJNA1114954
<i>Boreochiton ruber</i>	C:96.6%[S:94.7%,D:1.9%],F:2.7%,M:0.7%,n:954	JBEULR000000000	PRJNA1120663
<i>Callochiton septemvalvis</i> *	C:94.0%[S:90.9%,D:3.1%],F:3.0%,M:3.0%,n:954	PRJNA1114372	PRJNA1114372
<i>Deshayesiella sirenkoi</i> *	C:97.0%[S:96.2%,D:0.8%],F:2.0%,M:1.0%,n:954	PRJNA1114373	PRJNA1114373
<i>Veleropilina oligotropha</i>	C:92.4%[S:89.2%,D:3.2%],F:2.7%,M:4.9%,n:954	JBEUKZ000000000	PRJNA1120392
<i>Veleropilina</i> sp.	C:95.3%[S:81.8%,D:13.5%],F:3.1%,M:1.6%,n:954	PRJNA1120412	PRJNA1120412
<i>Fustiaria rubescens</i>	C:93.8%[S:91.3%,D:2.5%],F:4.1%,M:2.1%,n:954	JBEULQ000000000	PRJNA1120416
<i>Scintilla philippinensis</i> *	C:95.0%[S:86.2%,D:8.8%],F:2.7%,M:2.3%,n:954	JBEULU000000000	PRJNA1120792
<i>Verpa penis</i> *	C:97.2%[S:96.4%,D:0.8%],F:1.6%,M:1.2%,n:954	JBEUMR000000000	PRJNA1120794
<i>Solemya velum</i> *	C:94.3%[S:92.8%,D:1.5%],F:3.6%,M:2.1%,n:954	JBHNWI000000000	PRJNA1163513
<i>Tectura virginea</i>	C:94.2%[S:86.8%,D:7.4%],F:2.5%,M:3.3%,n:954	JBEULS000000000	PRJNA1120664
<i>Concholepas concholepas</i>	C:86.9%[S:77.8%,D:9.1%],F:7.5%,M:5.6%,n:954	JBEULT000000000	PRJNA1120782

Table S3 Assemblies used in the construction of the mollusc phylogenetic tree. Species in bold blue text indicate new genomes.

Class	Species name	BUSCOs	Assembly Accession
Caudofoveata	<i>Chaetoderma nitidulum</i>	C:92.5%[S:82.8%,D:9.7%],F:4.3%,M:3.2%,n:954	JBITNJ000000000
	<i>Chaetoderma shenloong</i>	C:93.3%[S:89.7%,D:3.6%],F:4.5%,M:2.2%,n:954	GCA_034401795.1
Solenogastres	<i>Epimania babai</i>	C:90.2%[S:81.9%,D:8.3%],F:5.8%,M:4.0%,n:954	GCA_011762755.2
	<i>Neomenia megatrapezata</i>	C:85.5%[S:82.1%,D:3.4%],F:7.8%,M:6.7%,n:954	(122)
Polyplacophora	<i>Acanthochitona discrepans</i>	C:97.5%[S:95.0%,D:2.5%],F:1.3%,M:1.2%,n:954	PRJNA1114954
	<i>Boreochiton ruber</i>	C:96.6%[S:94.7%,D:1.9%],F:2.7%,M:0.7%,n:954	JBEULR000000000
	<i>Callochiton septemvalvis</i>	C:94.4%[S:91.9%,D:2.5%],F:2.6%,M:3.0%,n:954	PRJNA1114372
	<i>Deshayesiella sirenkoi</i>	C:97.1%[S:96.4%,D:0.7%],F:1.9%,M:1.0%,n:954	PRJNA1114373
	<i>Acanthopleura granulata</i>	C:96.0%[S:95.3%,D:0.7%],F:2.1%,M:1.9%,n:954	GCA_016165875.1
	<i>Lepidochitona cinerea</i>	C:96.6%[S:93.0%,D:3.6%],F:2.4%,M:1.0%,n:954	GCA_963971465.1
	<i>Liolophura japonica</i>	C:96.4%[S:95.8%,D:0.6%],F:1.8%,M:1.8%,n:954	GCA_032854445.2
	<i>Hanleya hanleyi</i>	C:80.0%[S:74.9%,D:5.1%],F:12.1%,M:7.9%,n:954	GCA_036873755.1
Monoplacophora	<i>Veleroipilina oligotropha</i>	C:92.4%[S:89.2%,D:3.2%],F:2.7%,M:4.9%,n:954	JBEUKZ000000000
	<i>Veleroipilina</i> sp.	C:95.3%[S:81.8%,D:13.5%],F:3.1%,M:1.6%,n:954	PRJNA1120412
Cephalopoda	<i>Doryteuthis pealeii</i>	C:91.2%[S:87.3%,D:3.9%],F:4.7%,M:4.1%,n:954	GCA_023376005.1
	<i>Sepioteuthis lessoniana</i>	C:92.7%[S:92.2%,D:0.5%],F:3.7%,M:3.6%,n:954	GCA_963585895.1
	<i>Nautilus pompilius</i>	C:93.5%[S:91.2%,D:2.3%],F:2.1%,M:4.4%,n:954	GCA_018389105.1
	<i>Amphioctopus fangsiao</i>	C:93.5%[S:91.6%,D:1.9%],F:3.6%,M:2.9%,n:954	figshare.com/s/fa09f5dadcd966f020f3
	<i>Octopus vulgaris</i>	C:92.4%[S:92.0%,D:0.4%],F:2.7%,M:4.9%,n:954	GCA_951406725.2
	<i>Architeuthis dux</i>	C:89.7%[S:89.3%,D:0.4%],F:5.6%,M:4.7%,n:954	GCA_006491835.1
	<i>Thysanoteuthis rhombus</i>	C:96.5%[S:96.0%,D:0.5%],F:2.6%,M:0.9%,n:954	GCA_963457665.1
	<i>Euprymna scolopes</i>	C:83.4%[S:82.9%,D:0.5%],F:10.2%,M:6.4%,n:954	GCA_024364805.1
	<i>Sepia lycidas</i>	C:95.1%[S:94.8%,D:0.3%],F:3.2%,M:1.7%,n:954	GCA_963932145.1
	<i>Sepia pharaonis</i>	C:91.1%[S:90.7%,D:0.4%],F:5.2%,M:3.7%,n:954	GCA_903632075.3
	<i>Sepioida atlantica</i>	C:93.6%[S:92.2%,D:1.4%],F:2.1%,M:4.3%,n:954	GCA_963556195.1
Scaphopoda	<i>Fustiaria rubescens</i>	C:93.8%[S:91.3%,D:2.5%],F:4.1%,M:2.1%,n:954	JBEULQ000000000

Class	Species name	BUSCOs	Assembly Accession
	<i>Pictodentalium vernedei</i>	C:91.6%[S:82.9%,D:8.7%],F:4.7%,M:3.7%,n:954	GCA_031216915.1
	<i>Siphonodentalium dalli</i>	C:95.6%[S:87.9%,D:7.7%],F:2.8%,M:1.6%,n:954	GCA_032622095.1
	<i>Scintilla philippinensis</i>	C:95.0%[S:86.2%,D:8.8%],F:2.7%,M:2.3%,n:954	JBEULU000000000
	<i>Verpa penis</i>	C:97.2%[S:96.4%,D:0.8%],F:1.6%,M:1.2%,n:954	JBEUMR000000000
	<i>Solemya velum</i>	C:94.3%[S:92.8%,D:1.5%],F:3.6%,M:2.1%,n:954	JBHNWI000000000
	<i>Panopea generosa</i>	C:92.9%[S:88.1%,D:4.8%],F:3.6%,M:3.5%,n:954	GCA_029582155.1
	<i>Sinonovacula constricta</i>	C:91.6%[S:81.0%,D:10.6%],F:3.9%,M:4.5%,n:954	GCA_007844125.1
	<i>Cerastoderma edule</i>	C:93.3%[S:92.3%,D:1.0%],F:3.4%,M:3.3%,n:954	GCA_947846245.1
	<i>Fragum sueziense</i>	C:91.1%[S:88.5%,D:2.6%],F:4.6%,M:4.3%,n:954	GCA_963680895.1
	<i>Hippopus hippopus</i>	C:93.5%[S:92.7%,D:0.8%],F:3.7%,M:2.8%,n:954	GCA_946811185.2
	<i>Tridacna gigas</i>	C:93.5%[S:92.8%,D:0.7%],F:3.6%,M:2.9%,n:954	GCA_945859785.2
	<i>Conchocele bisecta</i>	C:94.2%[S:90.8%,D:3.4%],F:3.8%,M:2.0%,n:954	GCA_029237695.1
	<i>Congerina kusceri</i>	C:93.4%[S:87.1%,D:6.3%],F:4.2%,M:2.4%,n:954	GCA_027627225.1
	<i>Dreissena polymorpha</i>	C:91.7%[S:77.0%,D:14.7%],F:4.4%,M:3.9%,n:954	GCA_020536995.1
Bivalvia	<i>Sinohyriopsis cumingii</i>	C:96.5%[S:94.9%,D:1.6%],F:2.3%,M:1.2%,n:954	GCA_028554795.2
	<i>Mactra veneriformis</i>	C:95.2%[S:84.3%,D:10.9%],F:2.8%,M:2.0%,n:954	GCA_025267735.1
	<i>Spisula solida</i>	C:93.6%[S:92.6%,D:1.0%],F:3.8%,M:2.6%,n:954	GCA_947247005.1
	<i>Anadara broughtonii</i>	C:92.2%[S:86.6%,D:5.6%],F:1.7%,M:6.1%,n:954	(152, 153)
	<i>Tegillarca granosa</i>	C:96.5%[S:79.8%,D:16.7%],F:2.1%,M:1.4%,n:954	GCA_029721355.1
	<i>Bathymodiolus septemdierum</i>	C:96.7%[S:95.8%,D:0.9%],F:2.1%,M:1.2%,n:954	GCA_963383655.1
	<i>Limnoperna fortunei</i>	C:95.7%[S:95.1%,D:0.6%],F:2.1%,M:2.2%,n:954	GCA_944474755.1
	<i>Mytilus trossulus</i>	C:95.1%[S:89.2%,D:5.9%],F:2.1%,M:2.8%,n:954	GCA_036588685.1
	<i>Perna viridis</i>	C:96.6%[S:96.5%,D:0.1%],F:1.6%,M:1.8%,n:954	GCA_037379345.1
	<i>Crassostrea nippona</i>	C:98.9%[S:96.9%,D:2.0%],F:0.5%,M:0.6%,n:954	GCA_033439105.1
	<i>Mimachlamys varia</i>	C:98.5%[S:98.0%,D:0.5%],F:1.0%,M:0.5%,n:954	GCA_947623455.1
	<i>Mizuhopecten yessoensis</i>	C:97.3%[S:94.4%,D:2.9%],F:1.8%,M:0.9%,n:954	GCA_002113885.2
	<i>Pecten maximus</i>	C:97.2%[S:94.0%,D:3.2%],F:1.2%,M:1.6%,n:954	GCA_902652985.1

Class	Species name	BUSCOs	Assembly Accession
	<i>Pteria penguin</i>	C:96.1%[S:95.9%,D:0.2%],F:2.3%,M:1.6%,n:954	GCA_035048485.1
	<i>Ylistrum balloti</i>	C:97.2%[S:96.4%,D:0.8%],F:1.6%,M:1.2%,n:954	GCA_031769215.1
	<i>Tectura virginea</i>	C:94.2%[S:86.8%,D:7.4%],F:2.5%,M:3.3%,n:954	JBEULS000000000
	<i>Concholepas concholepas</i>	C:86.9%[S:77.8%,D:9.1%],F:7.5%,M:5.6%,n:954	JBEULT000000000
	<i>Alviniconcha strumneri</i>	C:85.0%[S:82.5%,D:2.5%],F:5.1%,M:9.9%,n:954	GCA_963584105.1
	<i>Batillaria attramentaria</i>	C:95.4%[S:91.4%,D:4.0%],F:2.8%,M:1.8%,n:954	GCA_018292915.2
	<i>Oncomelania hupensis</i>	C:92.7%[S:88.6%,D:4.1%],F:4.3%,M:3.0%,n:954	GCA_036898135.1
	<i>Pomacea canaliculata</i>	C:93.3%[S:92.9%,D:0.4%],F:3.7%,M:3.0%,n:954	GCA_036321745.1
	<i>Semisulcospira habei</i>	C:94.4%[S:93.1%,D:1.3%],F:3.0%,M:2.6%,n:954	GCA_036245525.1
	<i>Anisus vortex</i>	C:97.1%[S:96.2%,D:0.9%],F:1.4%,M:1.5%,n:954	GCA_949126835.1
	<i>Aplysia californica</i>	C:91.7%[S:91.5%,D:0.2%],F:4.2%,M:4.1%,n:954	GCA_000002075.2
	<i>Arion vulgaris</i>	C:94.5%[S:88.3%,D:6.2%],F:2.6%,M:2.9%,n:954	GCA_020796225.1
Gastropoda	<i>Berghia stephanieae</i>	C:93.7%[S:92.7%,D:1.0%],F:2.4%,M:3.9%,n:954	GCA_034508935.1
	<i>Ellobium chinense</i>	C:96.1%[S:95.6%,D:0.5%],F:1.7%,M:2.2%,n:954	GCA_033807755.1
	<i>Elysia crispata</i>	C:96.9%[S:96.1%,D:0.8%],F:1.6%,M:1.5%,n:954	GCA_963854125.1
	<i>Chrysomallon squamiferum</i>	C:97.4%[S:96.6%,D:0.8%],F:1.5%,M:1.1%,n:954	GCA_012295275.1
	<i>Gigantopelta aegis</i>	C:96.1%[S:91.7%,D:4.4%],F:2.5%,M:1.4%,n:954	GCA_016097555.1
	<i>Theodoxus fluviatilis</i>	C:77.4%[S:74.7%,D:2.7%],F:10.2%,M:12.4%,n:954	GCA_034096865.1
	<i>Lottia scabra</i>	C:96.6%[S:95.8%,D:0.8%],F:1.4%,M:2.0%,n:954	GCA_029955415.1
	<i>Nipponacmea schrenckii</i>	C:96.6%[S:95.1%,D:1.5%],F:2.0%,M:1.4%,n:954	GCA_030562195.1
	<i>Patella caerulea</i>	C:97.3%[S:93.5%,D:3.8%],F:1.8%,M:0.9%,n:954	GCA_036850965.1
	<i>Patella ulyssiponensis</i>	C:97.6%[S:97.1%,D:0.5%],F:1.5%,M:0.9%,n:954	GCA_963678685.1
	<i>Gibbula magus</i>	C:96.3%[S:95.8%,D:0.5%],F:1.7%,M:2.0%,n:954	GCA_936450465.1
	<i>Haliotis cracherodii</i>	C:97.3%[S:97.1%,D:0.2%],F:2.0%,M:0.7%,n:954	GCA_022045235.1
Outgroup: Annelida	<i>Owenia fusiformis</i>	C:96.1%[S:95.8%,D:0.3%],F:3.0%,M:0.9%,n:954	GCA_903813345.2
	<i>Urechis unicinctus</i>	C:93.8%[S:92.8%,D:1.0%],F:3.9%,M:2.3%,n:954	GCA_034190875.2
Outgroup: Brachiopoda	<i>Lingula anatina</i>	C:97.6%[S:76.8%,D:20.8%],F:1.6%,M:0.8%,n:954	GCA_001039355.2

Table S4. Summary of phylogenomic studies on Mollusca.

Source: T-Transcriptome; G-Genome; Marker: Ortho- orthologous genes, BUSCOs, Nuclear markers; spp: ingroup (molluscan) species; out: number of outgroup species; Represents: number of taxa for each sampled clade, see “clades” at far left for respective codes, missing clades are highlighted in red text. In the column "No. genes / sites / species occupancy (missing data)", the numbers only list some of the matrices, i.e., the matrices with the highest number of loci; the matrices with the highest species coverage, etc., for the matrices retained by different screening methods, please refer to the original references for details (given on later pages). Details for phylogenetic tree construction, supporting values and other technical aspects refer to the respective references. Lines in **bold blue** text indicate the present study, which contributes to total-group Molluscan topology and ingroup topology in all classes.

Clades	Source	Marker	spp	out	Clade representation	Year	Reference	Supported hypotheses
	T	Ortho	35	0	10G_14B_2Sc_4Ce_1M_1P_2So_1Ca	2011	(6)	Aculifera/Conchifera Megalopodifera/G+Sc
	G	Ortho	42	7	17G_12B_2Sc_5Ce_0M_2P_0So_2Ca	2011	(123)	Aculifera/Conchifera Aplacophora Megalopodifera/G+B
Mollusca G: Gastropoda B: Bivalvia Sc: Scaphopoda Ce: Cephalopoda M: Monoplacophora P: Polyplachophora So: Solenogastres Ca: Caudofoveata	T+G	Ortho	32	0	6G_8B_3Sc_6Ce_2M_2P_3So_2Ca	2020	(2)	Aculifera/Conchifera Aplacophora Megalopodifera/G+Sc
	G	Ortho	22	2	7G_8B_2Sc_4Ce_0M_1P_0So_0Ca	2023	(13)	Megalopodifera/Diasoma
	T+G	BUSCOs	134	1	53G_63B_1Sc_9Ce_1M_6P_3So_0Ca	2024	(14)	Aculifera/Conchifera Megalopodifera/G+Sc
	G	Ortho	56	4	16G_25B_2Sc_5Ce_0M_8P_0So_0Ca	2024	(17)	Conchifera Megalopodifera/Diasoma
	G	BUSCOs	77	3	22G_27B_3Sc_11Ce_2M_8P_2So_2Ca	this study	this study	Aculifera/Conchifera Aplacophora Megalopodifera/Diasoma

Table S4. (continued)

Clades	Source	Marker	spp	out	Represents	Year	Reference	Supported hypotheses
	T	Ortho	36	6	1P_8V_0Neo_2Ner_8C_21H	2014	(26)	Psilogastropoda
Gastropoda								
P: Patellogastropoda	T	Ortho	56	18	9P_8V_0Neo_8Ner_15C_16H	2019	(27)	Psilogastropoda
V: Vetigastropoda								
Neo: Neomphaliones	G	Ortho	14	2	1P_1V_1Neo_0Ner_4C_7H	2022	(31)	Psilogastropoda Orthogastropoda
Ner: Neritimorpha								
C: Caenogastropoda								
H: Heterobranchia	T	Ortho	25	8	4P_9V_2Neo_4Ner_3C_3H	2022	(28)	Psilogastropoda Orthogastropoda Orthogastropoda
	G	BUSCOs	22	58	5P_2V_2Neo_1Ner_6C_6H	this study	this study	Psilogastropoda
Bivalvia								
S: Protobranchia (Solemyoida)	T	Ortho	34	6	3S_6P_25H	2015	(138)	
P: Pteriomorpha								
H: Imparidentia + Anomalodesmata + Palaeoheterodonta (Heterodonta)	T	Ortho	98	9	9S_22P_68H	2019	(39)	
	G	BUSCOs	27	53	1S_12P_14H	this study	this study	

Table S4. (continued)

Clades	Source	Marker	spp	out	Represents	Year	Reference
Cephalopoda N: Nautiloidea O: Octobranchia D: Decapodiformes	G	Nuclear	26	56	1N_8O_18D	2017	(154)
	T	Ortho	26	0	1N_7O_18D	2018	(82)
	T	Ortho	31	0	0N_4O_27D	2020	(21)
	G	BUSCOs	11	69	1N_2O_8D	this study	this study
Polyplacophora L: Lepidopleurida Ca: Callochitonida Ch: Chitonida	T+G	Ortho	27	3	4L_2Ca_21Ch	2023	(18)
	G	BUSCOs	8	72	2L_1Ca_5Ch	this study	this study
Aplacophora So: Solenogastres Ca: Caudofoveata	T	Ortho	27	5	6Ca_21So	2019	(120)
Soleongastres C: Cavibelonia N: Neomeniamorpha P: Pholidoskepia S: Sterrofustia	T	Ortho	30	14	10C_2N_17P_1S	2024	(110)

Table S4. (continued)

Clades	No. genes / sites / species occupancy (%); missing data (% when given)	Bootstrap(LocalPP) at controversial/lower supported nodes	Reference (as above)
	301 / 50,930 / 50% occ. 1,185 / 216,402 / 40% occ.	M+Ce: 100/98/100/99 G+Sc: 100/70/100/89	(6)
	308 / 84,614 / 41% occ.; 74.4% missing data	G+B: 72/0.98	(123)
Mollusca	257 / 54,596 / 27.86% missing data	M (Ce+B+Sc+G): 94/0.99/0.90 M+Ce: 88 G+Sc: 66/92/1/0.36	(2)
G: Gastropoda			
B: Bivalvia			
Sc: Scaphopoda	663 / - / 92% occ.		
Ce: Cephalopoda	3,825 / - / 75% occ.	B+Sc: 0.7-1	(13)
M: Monoplacophora	6,430 / - / 50% occ.		
P: Polyplachophora			
So: Solenogastres	107 / - / 90% occ.	M+Ce: 75 G+Sc: 75	(14)
Ca: Caudofoveata			
	4,194 / 1,340,691 / 67% occ. (62species, plus 2 Solemya)	G+Sc: 97	
	4,037 / 1,222,084 / 67% occ (60 species)	B+Sc: 100	(17)
	954 / 321,310 / 90% occ. 96 / 61,874 / 100% occ.	M (Ce+B+Sc+G): 0.921; B+Sc: 74 M+Ce: 99; B+Sc: 0.621	this study

Table S4. (continued)

Clades	No. genes / sites / species occupancy (%); missing data (% when given)	Bootstrap(LocalPP) at controversial/ lower supported nodes	Reference (as above)
Gastropoda	ABA: 862 / 49,752 / 50% occ. OMA: 1245 / 190752 / 50% occ. ABA: 300 / 15,735 / 70% occ. OMA: 364 / 45084 / 70% occ.	Psilogastropoda: 0.58 Psilogastropoda: 1 Psilogastropoda: 0.11 Psilogastropoda: 1	(26)
P: Patellogastropoda	1,059 / - / 50% occ.	-	(27)
V: Vetigastropoda	149 / - / 70% occ.		
Neo: Neomphaliones	1,610 / - / 80% occ.	Psilogastropoda: 1	(31)
Ner: Neritimorpha	395 / - / 100% occ.	Orthogastropoda: 0.66	
C: Caenogastropoda	955 / 217,237 / 78.8% occ.	Psilogastropoda: 98/1	
H: Heterobranchia	323 / 72,548 / 88% occ. - / 151,606 (-30% rapid sites) / -	Orthogastropoda: 74/0.74 Orthogastropoda: 96/0.92	(28)
	954 / 321,310 / 90% occ. 96 / 61,874 / 100% occ.	Psilogastropoda: 0.9997 Psilogastropoda: 86	this study
Topology of interest			
	1,377 / 231,823 / 46.6% missing 729 / - / 35.4% missing 173 / - / 16% missing	Internal relationships of Protobranchia, Heterodonta, Neoheterodonte	(138)
Bivalvia			
S: Solemyoida	312 / 70,488 / 50% occ.	Internal relationships of Protobranchia, Pteriomorpha, Anomalodesmata	(39)
P: Pteriomorpha	102 / 22,164 / 70% occ.	Imparidentia: Adapedonta, Galeommatida and Cardiida	
H: Heterodonta			
	954 / 321,310 / 90% occ. 96 / 61,874 / 100% occ.	Imparidentia: Adapedonta, Galeommatida and Cardiida	this study

Table S4. (continued)

Clades	Bootstrap(LocalPP) at controversial/low supported nodes	Topology of interest	Reference (as above)
Cephalopoda N: Nautiloidea O: Octobranchia D: Decabrachia	180 / 36156 / 26% missing	Internal relationships of Decabrachia	(154)
	1,377 / 736,736 / 74% missing	Internal relationships of Decabrachia	(82)
	281 / 123,520 / 63% missing	Internal relationships of Decabrachia	(21)
	1,404 / 719,229 / 48% missing	Internal relationships of Decabrachia	(21)
	154 / 86,268 / 28% missing	Internal relationships of Decabrachia	(21)
	954 / 321,310 / 90% occ. 96 / 61,874 / 100% occ.	Internal relationships of Decabrachia	this study
Polyplacophora L: Lepidopleurida Ca: Callochitonida Ch: Chitonida	3,593 / 696,897 / 50% occ.	Internal relationships of Chitonida	(18)
	Best 800 / 194,356 / 50% occ.	Internal relationships of Chitonida	(18)
	954 / 321,310 / 90% occ. 96 / 61,874 / 100% occ.	-	this study
Aplacophora So: Solenogastres Ca: Caudofoveata	200 / 30,185 / 30.70% missing	Internal relationships of Aplacophora	(120)
Soleongastres C: Cavibelonia N: Neomeniamorpha P: Pholidoskepia S: Sterrofustia			
	949 / 33,352 / 22.6% missing	Internal relationships of Solenogastres	(110)

Table S5 Fossil calibration used for divergence time estimation of Mollusca

Calibration node	Fossil calibration	Date range (Mya)	Reference
Mollusca	First shell record	542	(8)
Aculifera	<i>Matthevia</i>	497	(155)
Polyplacophora	<i>Leptochiton davolii</i>	220	(8)
Monoplacophora	Living Monoplacophora	83	(19)
Cephalopoda	<i>Plectronoceras</i>	495	(8)
Scaphopoda	<i>Dentalium</i>	345	(8)
Origin of Pteriomorpha	Cyrtodontidae	465	(8)
Bivalvia	<i>Fordilla</i>	525	(8)
Origin of Caenogastropoda	Subulitoidea	405	(8)

Table S6 Mollusc genomic features. Het: heterozygosity (%), Rep (% repeats), Assem. (assembly size), Genome size estimation methods include: FAID: Feulgen image analysis densitometry, BFA: Bulk fluorometric assay.

Organism Name	Method for: Genome size / Het./ Repeat	Estimated genome size (Gbp)	Assem. Size (Gbp)	Chr	Het%	Rep%	Assembly Accession	Additional references
Caudofoveata								
<i>Chaetoderma nitidulum</i>	kmer	1.15	3.98	-	7.33	75.80	JBITNJ0000000000	this study
<i>Chaetoderma shenloong</i>	kmer	2.20	2.45	-	1.39	55.81	GCA 034401795.1	(124)
Solenogastres								
<i>Epimenia babai</i>	kmer:0.54, FAID:0.75	0.64	0.63	-	1.16	24.53	GCA 011762755.2	(121)
<i>Neomenia megatrapezata</i>	kmer, FAID	0.29	0.41	-	0.14	33.11	doi.org/ 10.6084/m9.figshare.25111997.v1	(121)
<i>Wirenia argentea</i>	-	-	0.53	-	-	-	GCA 025802215.1	-
Polyplacophora								
<i>Boreochiton ruber</i>	kmer	1.39	1.66	-	2.00	58.40	JBEULR0000000000	this study
<i>Acanthochitona discrepans</i>	kmer	1.02	1.17	8	2.79	54.00	PRJNA1114954	(17)
<i>Callochiton septemvalvis</i>	kmer	1.04	1.23	13	4.12	58.20	PRJNA1114372	(17)
<i>Deshaysiella sirenkoi</i>	kmer	1.27	1.55	11	0.97	45.90	PRJNA1114373	(17)
<i>Acanthochitona rubrolineata</i>	kmer	0.92	1.09	8	1.49	40.10	PRJNA1114370	(17)
<i>Acanthopleura granulata</i>	Flow cytometry	0.74	0.61	-	0.65	23.56	GCA 016165875.1	(156)
<i>Cryptochiton stelleri</i>	BFA	2.90	0.78	-	-	-	GCA 031471745.1	(157)
<i>Hanleya hanleyi</i>	kmer	1.89	2.52	-	1.29	66.00	GCA 036873755.1	(158)
<i>Katharina tunicata</i>	FIAD	1.70	0.68	-	-	-	GCA 032466195.1	(157)
<i>Lepidochitona cinerea</i>	-	-	0.71	15	-	-	GCA 963971465.1	-
<i>Lepidozona retiporosa</i>	-	-	0.50	-	-	-	GCA 032360285.1	-
<i>Liolophura japonica</i>	kmer	0.61	0.61	13	1.24	27.89	GCF 032854445.1	(159)
<i>Mopalia ciliata</i>	-	-	0.66	-	-	-	GCA 034783835.1	-
<i>Mopalia kennerleyi</i>	-	-	0.74	-	-	-	GCA 034783855.1	-
<i>Mopalia muscosa</i>	FIAD	1.70	0.66	-	-	-	GCA 031763545.1	(157)
<i>Mopalia swanii</i>	-	-	0.86	-	-	-	GCA 030265315.1	-

Organism Name	Method for: Genome size / Het./ Repeat	Estimated genome size (Gbp)	Assem. Size (Gbp)	Chr	Het%	Rep%	Assembly Accession	Additional references
<i>Mopalia vespertina</i>	-	-	0.68	-	-	-	GCA 030265115.1	-
<i>Tonicella lineata</i>	-	-	0.92	-	-	-	GCA 034781015.1	-
Monoplacophora								
<i>Veleroipilina oligotropha</i>	kmer	0.98	1.43	-	2.46	62.10	JBEUKZ000000000	this study
<i>Veleroipilina</i> sp.	kmer	0.78	1.58	-	2.35	51.70	PRJNA1120412	this study
Cephalopoda								
<i>Acanthosepion esculentum</i>	-	-	5.14	46	-	-	GCA 964036315.1	-
<i>Acanthosepion lycidas</i>	-	-	5.16	46	-	-	GCA 963932145.1	-
<i>Acanthosepion pharaonis</i>	kmer	4.85	4.79	-	0.35	77.30	GCA 903632075.3	(160)
<i>Amphioctopus burryi</i>	-	-	1.03	-	-	-	GCA 026873925.1	-
<i>Architeuthis dux</i>	kmer	2.70	2.69	-	-	49.17	GCA 006491835.1	(161)
<i>Ascarosepion bandense</i>	-	-	5.95	-	-	-	GCA 037127315.1	-
<i>Doryteuthis pealeii</i>	SNP /annotation	-	4.60	46	1.20	61.00	GCA 023376005.1	(162)
<i>Eledone cirrhosa</i>	-	-	3.00	27	-	-	GCA 964016885.1	-
<i>Enteroctopus dofleini</i>	-	-	0.74	-	-	-	GCA 027564315.1	-
<i>Euprymna scolopes</i>	-	-	5.12	48	-	50.00	GCA 024364805.1	(162, 163)
<i>Hapalochlaena maculosa</i>	SNP /annotation	-	4.01	-	0.95	37.00	GCA 015501135.1	(164)
<i>Idiosepius hallami</i>	-	-	0.97	-	-	-	GCA 034780655.1	-
<i>Japetella diaphana</i>	-	-	0.66	-	-	-	GCA 029822975.1	-
<i>Muusoctopus eicomar</i>	-	-	0.34	-	-	-	GCA 027122395.1	-
<i>Muusoctopus leioderma</i>	-	-	1.53	-	-	-	GCA 027124095.1	-
<i>Muusoctopus longibrachus</i>	-	-	1.07	-	-	-	GCA 027122495.1	-
<i>Nautilus pompilius</i>	kmer	0.75	0.73	-	-	30.95	GCA 018389105.1	(165)
<i>Octopus americanus</i>	-	-	2.53	-	-	-	GCA 027568995.1	-
<i>Octopus bimaculoides</i>	kmer /SNP /annotation	2.87	2.34	30	0.08	43.00	GCF 001194135.2	(166)
<i>Octopus insularis</i>	-	-	1.62	-	-	-	GCA 029874955.1	-

Organism Name	Method for: Genome size / Het./ Repeat	Estimated genome size (Gbp)	Assem. Size (Gbp)	Chr	Het%	Rep%	Assembly Accession	Additional references
<i>Octopus jollyorum</i>	-	-	2.57	-	-	-	GCA 034509045.1	-
<i>Octopus maya</i>	-	-	2.34	-	-	-	GCA 027122535.1	-
<i>Octopus minor</i>	kmer	5.10	5.09	-	-	44.43	10.5524/100503	(167)
<i>Octopus mimus</i>	-	-	2.06	-	-	-	GCA 027568975.1	-
<i>Octopus rubescens</i>	-	-	2.76	-	-	-	GCA 027564515.1	-
<i>Octopus sinensis</i>	kmer	2.76	2.72	30	0.34	39.80	GCF 006345805.1	(168)
<i>Octopus vulgaris</i>	-	-	2.80	30	-	64.87	GCA 951406725.2	(169)
<i>Onykia robusta</i>	-	-	2.37	-	-	-	GCA 037178445.1	-
<i>Sepiola atlantica</i>	-	-	5.59	41	-	-	GCA 963556195.1	-
<i>Sepioteuthis lessoniana</i>	-	-	5.06	44	-	-	GCA 963585895.1	-
<i>Thysanoteuthis rhombus</i>	-	-	2.74	46	-	-	GCA 963457665.1	-
<i>Vulcanoctopus hydrothermalis</i>	-	-	1.34	-	-	-	GCA 036023795.1	-
<i>Watasenia scintillans</i>	kmer	0.67	0.65	-	5.50	19.20	GCA 015471945.1	(170)
<i>Xipholeptos notoides</i>	-	-	1.05	-	-	-	GCA 036873565.1	-
Scaphopoda								
<i>Fustiaria rubescens</i>	kmer	2.01	2.58	-	3.52	54.50	JBEULQ000000000	this study
<i>Pictodentalium verneidei</i>	-	-	6.02	10	-	73.18	GCA 031216915.1	(13)
<i>Siphonodentalium dalli</i>	-	-	2.23	9	-	76.05	GCA 032622095.1	(13)
Bivalvia								
<i>Scintilla philippinensis</i>	kmer	1.27	1.68	-	2.44	37.20	JBEULU000000000	(61)
<i>Solemya velum</i>	kmer	-	1.50	11	0.86	35.5	JBHNWI000000000	this study
<i>Verpa penis</i>	kmer	0.49	0.51	19	0.42	21.70	JBEUMR000000000	(171)
<i>Anadara broughtonii</i>	kmer	0.70	0.88	19	1.83	46.10	10.5524/100607	(152)
<i>Anadara kagoshimensis</i>	-	-	1.12	19	-	-	GCA 021292105.1	-
<i>Anodontia alba</i>	-	-	1.86	18	-	-	GCA 964016985.1	-
<i>Archivesica marissinica</i>	kmer	1.60	1.54	19	0.41	42.20	GCA 014843695.1	(172)
<i>Arcuatula senhousia</i>	-	-	1.04	15	-	-	GCA 963971305.1	-
<i>Argopecten irradians concentricus</i>	kmer	0.97	0.87	-	-	47.19	GCA 004382765.1	(173)

Organism Name	Method for: Genome size / Het./ Repeat	Estimated genome size (Gbp)	Assem. Size (Gbp)	Chr	Het%	Rep%	Assembly Accession	Additional references
<i>Argopecten irradians irradians</i>	kmer	1.00	0.84	-	-	46.43	GCA 004382745.1	(173)
<i>Atrina japonica</i>	-	-	0.84	-	-	-	GCA 030159075.1	
<i>Azorinus chamasolen</i>	-	-	1.72	19	-	-	GCA 963576725.1	-
<i>Bathymodiolus brooksi</i>	-	-	2.00	16	-	-	GCA 963680875.1	-
<i>Bathymodiolus septemdierum</i>	-	-	1.43	15	-	-	GCA 963383655.1	-
<i>Botula fusca</i>	-	-	1.33	-	-	-	GCA 028566455.1	-
<i>Cerastoderma edule</i>	-	-	0.86	19	-	-	GCA 963989375.1	-
<i>Chlamys farreri</i>	kmer	1.00	0.78	19	0.80	32.10	mgb.ouc.edu.cn/cfbase/html	(174, 175)
<i>Chlamys rubida</i>	-	-	0.73	-	-	-	GCA 032359455.1	-
<i>Clinocardium nuttallii</i>	-	-	0.87	-	-	-	GCA 031471495.1	-
<i>Conchocele bisecta</i>	kmer	2.14	1.90	17	0.82	66.96	GCA 029237695.1	(148)
<i>Congerius kusceri</i>	-	-	1.55	17	-	-	GCA 027627225.1	-
<i>Corbicula fluminea</i>	kmer	1.64	0.66	-	2.41	64.55	GCA 001632725.1	(176)
<i>Corbicula japonica</i>	-	-	1.07	-	-	-	GCA 038501625.1	-
<i>Crassadoma gigantea</i>	-	-	0.90	-	-	-	GCA 032360845.1	-
<i>Crassostrea (Magallana) gigas</i>	kmer	0.59	0.61	10	2.95	48.87	GCA 025765685.3	(177)
<i>Crassostrea (Magallana) angulata</i>	kmer	0.57	0.58	10	2.60	48.02	GCA 025765675.3	(177)
<i>Crassostrea (Magallana) ariakensis</i>	kmer	0.62	0.61	10	0.47	54.14	GCA 020567875.1	(178)
<i>Crassostrea (Magallana) hongkongensis</i>	kmer	0.61	0.61	10	2.65	64.00	GCA 015776775.1	(179)
<i>Crassostrea (Magallana) nippona</i>	-	-	0.53	10	-	-	GCA 033439105.1	-
<i>Crassostrea virginica</i>	-	-	0.68	10	-	40.30	GCF 002022765.2	(148)
<i>Ctena decussata</i>	-	-	1.66	18	-	-	GCA 963989385.1	-
<i>Cyclina sinensis</i>	kmer	0.86	0.90	19	1.53	43.14	GCA 012932295.1	(180)
<i>Cyrtodaria siliqua</i>	-	-	2.88	-	-	-	GCA 037043455.1	-
<i>Dreissena polymorpha</i>	kmer	1.30	1.80	16	2.13	47.40	GCF 020536995.1	(181)
<i>Dreissena rostriformis</i>	kmer	1.60	1.24		2.40	31.90	GCA 007657795.1	(182)

Organism Name	Method for: Genome size / Het./ Repeat	Estimated genome size (Gbp)	Assem. Size (Gbp)	Chr	Het%	Rep%	Assembly Accession	Additional references
<i>Fragum fragum</i>	kmer	1.24	1.15	19	3.12	34.53	GCA 946902895.1	(183)
<i>Fragum sueziense</i>	-	-	1.21	19	-	-	GCA 963680895.1	-
<i>Fragum whitleyi</i>	kmer	1.18	1.22	18	3.60	41.00	GCA 948146395.1	(184)
<i>Gari tellinella</i>	-	-	1.60	19	-	-	GCA 922989275.2	(185)
<i>Geukensia demissa</i>	-	-	1.02	-	-	-	GCA 034783895.1	-
<i>Geukensia granosissima</i>	-	-	1.18	-	-	-	GCA 032357055.1	-
<i>Gigantidas platifrons</i> (as ' <i>Bathymodiolus</i> ')	kmer	1.64	1.70	-	1.24	47.90	GCA_002080005.1	(175)
<i>Gigantidas platifrons</i>	-	-	1.66	-	-	48.31	GCA 002080005.1	(148)
<i>Hippopus hippopus</i>	kmer	1.10	1.07	18	0.95	36.65	GCA 946811185.2	(186)
<i>Limnoperna fortunei</i>	kmer	1.30	1.34	15	2.40	43.00	GCA 944474755.1	(187)
<i>Lithophaga antillarum</i>	-	-	1.42	-	-	-	GCA 028566495.1	-
<i>Lithophaga nigra</i>	-	-	1.49	-	-	-	GCA 028566555.1	-
<i>Loripinus fragilis</i>	-	-	1.99	18	-	-	GCA 964019695.1	-
<i>Luciniscia nassula</i>	-	-	2.93	18	-	-	GCA 963580285.1	-
<i>Lutraria lutraria</i>	-	-	0.82	19	-	-	GCA 964023205.1	-
<i>Lutraria rhynchaena</i>	kmer	0.55	0.54	-	1.63	29.40	GCA 008271625.1	(188)
<i>Macoma nasuta</i>	-	-	1.40	-	-	-	GCA 032359355.1	-
<i>Mactra quadrangularis</i>	kmer	0.87	0.98	19	3.34	51.79	GCA 025267735.1	(189)
<i>Mactromeris polynyma</i>	-	-	0.79	-	-	-	GCA 032209605.1	-
<i>Magallana gigas</i>	kmer	0.53 kmer 0.64 flow	0.65	10	3.20	43.00	GCF 902806645.1	(190)
<i>Magallana hongkongensis</i>	-	-	0.76	-	-	41.12	GCA 016163765.1	(191)
<i>Margaritifera margaritifera</i>	kmer	2.35	2.45	-	0.14	57.32	GCA 029931535.1	(192)
<i>Megalonias nervosa</i>	-	-	2.37	-	-	-	GCA 016617855.1	-
<i>Mercenaria mercenaria</i>	fluorometric	1.96	1.86	19	-	45.00	GCA 021730395.1	(193)
<i>Mimachlamys varia</i>	kmer	2.40	0.98	19	3.57	73.72	GCA 947623455.1	(194)
<i>Mizuhopecten yessoensis</i>	kmer /SNP /annotation	1.43	0.99	-	1.04	39.00	GCF 002113885.1	(195)

Organism Name	Method for: Genome size / Het./ Repeat	Estimated genome size (Gbp)	Assem. Size (Gbp)	Chr	Het%	Rep%	Assembly Accession	Additional references
<i>Modiolus philippinarum</i>	kmer	2.38	2.63	-	2.02	62.00	GCA 002080025.1	(175)
<i>Mya arenaria</i>	snp	-	1.22	17	3.10	35.00	GCF 026914265.1	(196)
<i>Mytilisepta virgata</i>	flow cytometry	1.08	1.03	14	-	-	GCA 028015205.1	(197)
<i>Mytilus californianus</i>	kmer	1.57	1.65	-	2.73	-	GCF 021869535.1	(198)
<i>Mytilus coruscus</i>	kmer	1.51	1.57	14	1.39	47.37	GCA 017311375.1	(199)
<i>Mytilus edulis</i>	kmer	1.00	1.83	14	3.69	56.33	GCA 905397895.1	(200)
<i>Mytilus galloprovincialis</i>	snp	1.48	1.41	14	1.73	44.36	GCA 037788925.1	(201)
<i>Mytilus trossulus</i>	-	-	1.30	14	-	-	GCA 036588685.1	-
<i>Nuttallia obscura</i>	-	-	1.29	-	-	-	GCA 032358795.1	-
<i>Ostrea denselamellosa</i>	kmer	0.58	0.64	-	0.80	50.51	GCA 024699665.1	(202)
<i>Ostrea edulis</i>	kmer	0.88	0.95	10	1.02	57.30	GCA 032173915.1	(203)
<i>Ostrea lurida</i>	-	-	1.14	-	-	-	GCA 903981925.1	-
<i>Panopea generosa</i>	kmer	1.47	1.47	19	1.37	68.08	GCA 029582155.1	(204)
<i>Pecten maximus</i>	kmer	1.15	0.92	19	1.71	27.00	GCF 902652985.1	(205)
<i>Perna viridis</i>	kmer	0.72	0.72	15	0.63	19.57	GCA 037379345.1	(206)
<i>Pinctada fucata</i>	kmer	1.03	0.93	14	3.26	53.30	GCA 028142955.1	(207)
<i>Pinctada imbricata</i>	-	-	0.99	14	-	44.78	GCA 033119305.1	(148)
<i>Pinctada margaritifera</i>	-	-	1.31	-	-	-	GCA 964021215.1	-
<i>Pinna nobilis</i>	kmer	0.64	0.59	-	1.16	36.00	GCA 016161895.1	(208)
<i>Pododesmus macrochisma</i>	-	-	0.77	-	-	-	GCA 032359645.1	-
<i>Potamilus streckersoni</i>	kmer	1.81	1.78	-	0.57	51.03	GCA 016746295.1	(209)
<i>Pteria penguin</i>	kmer	0.80	0.84	14	1.02	43.89	GCA 035048485.1	(210)
<i>Ruditapes philippinarum</i>	kmer	1.37	1.12	19	-	26.38	GCA 009026015.1	(211)
<i>Rugalucina vietnamica</i>	-	-	1.09	17	-	-	GCA 964017185.1	-
<i>Saccostrea cucullata</i>	-	-	1.17	-	-	-	GCF 032062105.1	-
<i>Saccostrea echinata</i>	-	-	0.92	-	-	-	GCF 033153115.1	-
<i>Saccostrea glomerata</i>	kmer /SNP /annotation	0.78	0.79	-	0.51	45.03	GCA 003671525.1	(212)

Organism Name	Method for: Genome size / Het./ Repeat	Estimated genome size (Gbp)	Assem. Size (Gbp)	Chr	Het%	Rep%	Assembly Accession	Additional references
<i>Saxidomus gigantea</i>	-	-	0.92	-	-	-	GCA 032359765.1	-
<i>Saxidomus purpurata</i>	kmer	0.84	1.16	-	2.61	50.81	GCA 022818135.1	(213)
<i>Serripes groenlandicus</i>	-	-	1.76	-	-	-	GCA 031761405.1	-
<i>Sinohyriopsis cumingii</i>	kmer	2.91	2.91	19	0.92	50.86	GCA 028554795.2	(214)
<i>Sinonovacula constricta</i>	kmer /SNP /annotation	1.33	1.22	19	0.71	39.79	GCA 007844125.1	(215)
<i>Solen grandis</i>	-	-	1.32	19	-	-	GCA 021229015.1	-
<i>Spisula solida</i>	kmer	0.88	0.93	19	2.31	39.85	GCA 947247005.1	(216)
<i>Spisula subtruncata</i>	kmer	0.92	0.93	19	4.29	39.56	GCA 963678985.1	(217)
<i>Tegillarca granosa</i>	kmer	0.81	0.80	19	1.17	53.75	GCA 013375625.1	(218)
<i>Tridacna crocea</i>	kmer	1.39	1.05	18	-	71.60	GCA 943736015.1	(219, 220)
<i>Tridacna derasa</i>	-	-	1.06	18	-	-	GCA 963210305.1	-
<i>Tridacna gigas</i>	kmer	1.17	1.18	17	0.75	37.25	GCA 945859785.2	(221)
<i>Unio delphinus</i>	kmer	2.32	2.51	-	0.64	52.83	GCA 029339505.1	(222)
<i>Unio pictorum</i>	kmer	2.15	2.43	-	1.68	49.98	GCA 030141615.1	(223)
<i>Venustaconcha ellipsiformis</i>	kmer	1.80	1.59	-	0.63	37.81	GCA 003401595.1	(175)
<i>Ylistrum balloti</i>	-	-	0.65	-	-	-	GCF 031769215.1	-
Gastropoda								
<i>Concholepas concholepas</i>	kmer	0.58	0.65	-	3.99	62.00	JBEULS000000000	this study
<i>Tectura virginea</i>	kmer	0.85	1.02	-	4.31	56.50	JBEULS000000000	this study
<i>Aegires albopunctatus</i>	-	-	0.68	-	-	-	GCA 032466395.1	-
<i>Aeolidia papillosa</i>	-	-	0.17	-	-	-	GCA 032466315.1	-
<i>Alviniconcha marisindica</i>	-	-	0.83	-	-	-	GCA 018857735.1	-
<i>Alviniconcha strummeri</i>	-	-	0.45	18	-	-	GCA 963584105.1	-
<i>Ampullaceana balthica</i>	-	-	1.11	-	-	-	GCA 944989445.1	-
<i>Anatoma sagamiana</i>	-	-	0.48	-	-	-	GCA 032362125.1	-
<i>Ancula gibbosa</i>	-	-	0.64	-	-	-	GCA 031833985.1	-
<i>Anentome helena</i>	-	-	0.33	-	-	-	GCA 009936545.3	-

Organism Name	Method for: Genome size / Het./ Repeat	Estimated genome size (Gbp)	Assem. Size (Gbp)	Chr	Het%	Rep%	Assembly Accession	Additional references
<i>Anisus vortex</i>	kmer	0.81	0.87	18	1.73	40.14	GCA 949126835.1	(224)
<i>Aplysia californica</i>	-	0.85	0.93	17	-	32.01	GCF 000002075.1	(148)
<i>Ariolimax columbianus</i>	-	-	2.29	-	-	-	GCA 036924085.1	-
<i>Arion rufus</i>	-	-	1.08	-	-	-	GCA 032362185.1	-
<i>Arion vulgaris</i>	kmer	1.50	1.54	26	1.55	75.09	GCA 020796225.1	(131)
<i>Babylonia areolata</i>	-	-	1.11	-	-	-	GCA 011634625.1	-
<i>Batillaria attramentaria</i>	kmer	0.64	0.72	-	-	43.87	GCA 018292915.2	(225)
<i>Berghia stephanieae</i>	kmer	0.76	1.12	15	0.69	46.68	GCA 034508935.3	(226)
<i>Biomphalaria glabrata</i>	FAID	0.92	0.91	18	1.52	44.80	GCF 000457365.2	(227–229)
<i>Biomphalaria pfeifferi</i>	Flow cytometry, kmer	0.92	0.77	18	1.01	40.00	GCA 030265305.1	(229)
<i>Biomphalaria straminea</i>	-	-	1.00	-	-	40.68	GCA 021533235.1	(230)
<i>Biomphalaria sudanica</i>	SNP	-	0.94	-	0.32	40.30	GCA 036873155.1	(231)
<i>Bulinus truncatus</i>	kmer	-	1.22	-	1.36	51.03	GCA 021962125.1	(232)
<i>Bullina lineata</i>	-	-	0.65	-	-	-	GCA 039654405.1	-
<i>Calliostoma canaliculatum</i>	-	-	1.18	-	-	-	GCA 032361085.1	-
<i>Candidula unifasciata</i>	kmer	0.99	1.36	-	1.09	61.10	GCA 905116865.2	(233)
<i>Cepaea hortensis</i>	-	-	3.17	22	-	-	GCA 963921405.1	-
<i>Cepaea nemoralis</i>	kmer	3.10	3.49	-	1.43	77.00	GCA 014155875.1	(234)
<i>Chromodoris orientalis</i>	-	-	0.73	-	-	-	GCA 028571245.1	-
<i>Chrysomallon squamiferum</i>	kmer	0.44	0.40	15	1.38	25.00	GCA 012295275.1	(45)
<i>Clione limacina</i>	-	-	1.26	-	-	-	GCA 032362205.1	-
<i>Clypeosectus delectus</i>	-	-	0.40	-	-	-	GCA 032355325.1	-
<i>Colubraria reticulata</i>	-	-	0.67	-	-	-	GCA 900004695.1	-
<i>Concholepas concholepas</i> (preliminary)	kmer	1.83	0.83	-	-	66.00	GCA 034780235.1	(235)
<i>Conus betulinus</i>	kmer	3.99	3.43	-	-	38.56	GCA 016801955.1	(236)
<i>Conus canariensis</i>	kmer	3.60	2.87	35	-	55.80	GCA 033310375.1	(237)

Organism Name	Method for: Genome size / Het./ Repeat	Estimated genome size (Gbp)	Assem. Size (Gbp)	Chr	Het%	Rep%	Assembly Accession	Additional references
<i>Conus consors</i>	-	-	2.05	-	-	-	GCA 004193615.1	-
<i>Conus tribblei</i>	-	-	2.16	-	-	-	GCA 001262575.1	-
<i>Conus ventricosus</i>	Flow cytometry	4.10	3.59	35	1.07	53.36	GCA 018398815.1	(238)
<i>Corambe burchi</i>	-	-	0.81	-	-	-	GCA 031762895.1	-
<i>Cyathernia naticoides</i>	-	-	0.52	-	-	-	GCA 033042655.1	-
<i>Dendronotus rufus</i>	-	-	0.47	-	-	-	GCA 032358575.1	-
<i>Dendronotus venustus</i>	-	-	0.68	-	-	-	GCA 031834005.1	-
<i>Depressigra globulus</i>	-	-	0.84	-	-	-	GCA 032274625.1	-
<i>Diodora aspera</i>	-	-	0.78	-	-	-	GCA 031471655.1	-
<i>Dirona albolineata</i>	-	-	0.52	-	-	-	GCA 031763155.1	-
<i>Dirona pellucida</i>	kmer	2.59	0.56	-	2.00	56.48	GCA 030265205.1	(239)
<i>Doris montereyensis</i>	-	-	0.52	-	-	-	GCA 037039925.1	-
<i>Doris verrucosa</i>	-	-	0.60	-	-	-	GCA 031762655.1	-
<i>Dracogyra subfusca</i>	kmer	1.20	1.16	-	-	56.00	GCA 016106625.1	(132)
<i>Ellobium chinense</i>	kmer	0.82	0.95	-	2.15	37.05	GCA 033807755.1	(240)
<i>Elysia chlorotica</i>	kmer	0.58	0.56	-	3.66	32.60	GCA 003991915.1	(241)
<i>Elysia crispata</i>	kmer	0.71	0.79	15	0.88	29.85	GCA 033675545.1	(242)
<i>Elysia marginata</i>	-	-	0.79	-	-	-	GCA 019649035.1	-
<i>Gastropteron pacificum</i>	-	-	0.55	-	-	-	GCA 031471215.1	-
<i>Gibbula magus</i>	-	-	1.47	18	-	-	GCA 936450465.1	-
<i>Gigantopelta aegis</i>	kmer	1.21	1.15	15	0.50	50.00	GCF 016097555.1	(132)
<i>Goniobranchus coi</i>	-	-	0.68	-	-	-	GCA 025762795.1	-
<i>Goniobranchus fidelis</i>	-	-	0.93	-	-	-	GCA 028565935.1	-
<i>Goniobranchus geometricus</i>	-	-	0.91	-	-	-	GCA 028565895.1	-
<i>Goniobranchus kuniei</i>	-	-	1.19	-	-	-	GCA 025770095.1	-
<i>Goniobranchus leopardus</i>	-	-	0.43	-	-	-	GCA 028566475.1	-
<i>Haliotis asinina</i>	kmer	1.10	1.14	16	0.66	38.42	GCA 037392515.1	(243)
<i>Haliotis cracherodii</i>	kmer	1.18	1.18	-	1.56	-	GCA 022045235.1	(244)

Organism Name	Method for: Genome size / Het./ Repeat	Estimated genome size (Gbp)	Assem. Size (Gbp)	Chr	Het%	Rep%	Assembly Accession	Additional references
<i>Haliotis laevigata</i>	kmer	1.54	1.76	-	0.68	-	GCA 008038995.1	(245)
<i>Haliotis rubra</i>		-	1.38	-	-	-	GCF 003918875.1	-
<i>Haliotis rufescens</i>	kmer	1.28	1.33	-	1.37	46.03	GCF 023055435.1	(148, 246)
<i>Hermisenda crassicornis</i>	-	-	0.48	-	-	-	GCA 031471375.1	-
<i>Hyalogyrina grasslei</i>	-	-	0.46	-	-	-	GCA 034780735.1	-
<i>Lanistes nyassanus</i>	kmer	0.51	0.51	-	0.60	28.87	GCA 004794575.1	(247)
<i>Lepetodrilus cristatus</i>	-	-	0.42	-	-	-	GCA 032361305.1	-
<i>Lepetodrilus elevatus</i>	-	-	0.42	-	-	-	GCA 034780755.1	-
<i>Lepetodrilus fucensis</i>	-	-	0.49	-	-	-	GCA 032361325.1	-
<i>Lepetodrilus galriftensis</i>	-	-	0.52	-	-	-	GCA 032361785.1	-
<i>Lepetodrilus gordensis</i>	-	-	0.43	-	-	-	GCA 034780695.1	-
<i>Lepetodrilus ovalis</i>	-	-	0.58	-	-	-	GCA 032361185.1	-
<i>Lepetodrilus pustulosus</i>	-	-	0.45	-	-	-	GCA 032355345.1	-
<i>Lepetodrilus schrolli</i>	-	-	0.43	-	-	-	GCA 032361225.1	-
<i>Lepetodrilus tevnianus</i>	-	-	0.42	-	-	-	GCA 032361465.1	-
<i>Limacina bulimoides</i>	-	-	2.90	-	-	-	GCA 009866985.1	-
<i>Littorina saxatilis</i>	karyology	1.35	1.26	17	1.50	-	GCA 037325665.1	(248)
<i>Lissachatina immaculata</i>	kmer /SNP /annotation	1.75	1.65	31	0.24	57.70	GCA 009760885.1	(249)
<i>Lissachatina fulica</i>	kmer	2.12	1.85	31	0.47	71.00	10.5524/100647	(250)
<i>Lomanotus vermiformis</i>	-	-	0.90	-	-	-	GCA 037974715.1	-
<i>Lottia gigantea</i>	kmer	3.76	0.36	-	3.15	21.23	GCF 000327385.1	(148, 156)
<i>Lottia scabra</i>	-	-	0.41	-	-	-	GCA 029955415.1	-
<i>Lymnaea stagnalis</i>	Flow cytometry	1.22	0.94	-	-	37.89	GCA 964033795.1	(251)
<i>Marisa cornuarietis</i>	kmer	0.51	0.54	-	0.08	30.82	GCA 004794655.1	(247)
<i>Megaustenia siamensis</i>	kmer	2.20	2.59	-	0.36	60.69	GCA 032433765.1	(252)
<i>Meghimatium bilineatum</i>	kmer	1.50	1.61	25	1.05	43.69	GCA 034231615.1	(253)

Organism Name	Method for: Genome size / Het./ Repeat	Estimated genome size (Gbp)	Assem. Size (Gbp)	Chr	Het%	Rep%	Assembly Accession	Additional references
<i>Melanodrymia aurantiaca</i>	-	-	0.42	-	-	-	GCA 032355825.1	-
<i>Melanodrymia brightae</i>	-	-	0.34	-	-	-	GCA 032273805.1	-
<i>Melanoides tuberculata</i>	-	-	0.97	-	-	-	GCA 028565955.2	-
<i>Melibe bucephala</i>	-	-	0.83	-	-	-	GCA 039654525.1	-
<i>Melibe leonina</i>	-	-	0.57	-	-	-	GCA 031471865.1	-
<i>Melibe viridis</i>	-	-	0.81	-	-	-	GCA 039654465.1	-
<i>Monoplex corrugatus</i>	-	-	3.08	35	-	-	GCA 030674185.1	-
<i>Nipponacmea schrenckii</i>	-	-	0.42	9	-	-	GCA 030562195.1	-
<i>Nodopelta heminoda</i>	-	-	0.45	-	-	-	GCA 032362005.1	-
<i>Omphiscola glabra</i>	-	-	0.74	18	-	-	GCA 964017045.1	-
<i>Onchidella celtica</i>	-	-	1.14	18	-	-	GCA 963931925.1	-
<i>Oncomelania hupensis</i>	kmer	1.46	1.45	17	1.69	64.16	GCA 036898135.1	(254)
<i>Oreohelix idahoensis</i>	kmer	7.01	5.40	-	0.51	74.90	GCA 024509875.1	(255)
<i>Pachydermia laevis</i>	-	-	0.56	-	-	-	GCA 032355525.1	-
<i>Patella caerulea</i>	kmer	0.62	0.75	-	2.06	-	GCA 036850965.1	(256)
<i>Patella depressa</i>	kmer	0.71	0.68	9	1.56	39.57	GCA 948474765.1	(257)
<i>Patella pellucida</i>	kmer	0.79	0.71	9	1.89	42.84	GCA 917208275.1	(258)
<i>Patella ulyssiponensis</i>	kmer	0.61	0.69	8	3.27	77.96	GCA 963678685.1	(259)
<i>Patella vulgata</i>	kmer	0.73	0.70	9	1.10	36.28	GCF 932274485.2	(260)
<i>Peltoospira delicata</i>	-	-	0.62	-	-	-	GCA 034780715.1	-
<i>Peltoospira operculata</i>	-	-	0.48	-	-	-	GCA 032361585.1	-
<i>Peltoospira smaragdina</i>	-	-	0.38	-	-	-	GCA 032361425.1	-
<i>Peronia peronii</i>	-	-	1.31	-	-	-	GCA 039654565.1	-
<i>Peronia verruculata</i>	-	-	1.21	-	-	-	GCA 038406235.1	-
<i>Phorcus lineatus</i>	kmer	1.03	0.96	18	1.06	29.38	GCA 921293015.1	(261)
<i>Phyllaplysia taylori</i>	-	-	0.81	-	-	-	GCA 032273245.1	-
<i>Phymorhynchus buccinoides</i>	Annotation	-	2.12	34	-	73.40	GCA 017654935.2	(262)
<i>Physella acuta</i>	-	-	0.70	-	-	-	GCF 028476545.1	-

Organism Name	Method for: Genome size / Het./ Repeat	Estimated genome size (Gbp)	Assem. Size (Gbp)	Chr	Het%	Rep%	Assembly Accession	Additional references
<i>Plakobranthus ocellatus</i>	-	-	0.93	-	-	-	GCA 019648995.1	-
<i>Plocamopherus ceylonicus</i>	-	-	1.09	-	-	-	GCA 039654545.1	-
<i>Plocamopherus imperialis</i>	-	-	0.76	-	-	-	GCA 039654505.1	-
<i>Plocamopherus tilesii</i>	-	-	0.80	-	-	-	GCA 037974505.1	-
<i>Pomacea canaliculata</i>	kmer	0.44	0.45	14	1.41	20.53	GCA 004794335.1	(247)
<i>Pomacea maculata</i>	kmer	0.41	0.43	-	1.22	21.25	GCA 004794325.1	(247)
<i>Pseudorimula midatlantica</i>	-	-	0.42	-	-	-	GCA 032361205.1	-
<i>Radix auricularia</i>	Flow cytometry	1.58	0.91	-	-	70.00	GCA 002072015.1	(263)
<i>Rapana venosa</i>	kmer	2.20	2.30	35	1.41	67.04	GCA 028751875.1	(264)
<i>Scyllaea pelagica</i>	-	-	0.65	-	-	-	GCA 031833965.1	-
<i>Semisulcospira habei</i>	kmer	1.89	1.98	-	-	76.00	GCA 036245525.1	(265)
<i>Sinotaia purificata</i>	kmer	0.94	0.98	8	-	47.93	GCA 028829895.1	(266)
<i>Steromphala cineraria</i>	kmer	1.40	1.27	18	3.43	54.55	GCA 916613615.1	(267)
<i>Stramonita haemastoma</i>	-	-	2.24	35	-	-	GCA 030674155.1	-
<i>Symmetromphalus regularis</i>	-	-	0.63	-	-	-	GCA 032355785.1	-
<i>Theodoxus fluviatilis</i>	kmer	1.29	1.04	-	0.97	48.38	GCA 034096865.1	(268)
<i>Tritonia festiva</i>	-	-	0.91	-	-	-	GCA 030265255.1	-
<i>Tritonia tetraquetra</i>	-	-	0.74	-	-	-	GCA 030265355.1	-
<i>Tylodina fungina</i>	-	-	0.67	-	-	-	GCA 031762815.1	-
<i>Urosalpinx cinerea</i>	-	-	0.73	-	-	-	GCA 034780015.1	-

Supplementary Table 7. An illustrated key to the schematic molluscs. The list below includes the clades labelled on the phylogenetic reconstruction in Main text Fig 1, and the mollusc icons that illustrate each clade. The icon used to represent its taxonomic class is indicated with a box. These icons were first used in an earlier study(8), but the set has been expanded here. In some cases the cartoons depict individual species, most of which reflect new genomes produced in this study, while others are nonspecific schematic depictions of a group. All drawings were made by JD Sigwart.

Bivalvia		
Imparidentia	<i>Scintilla philippinensis</i> <i>Tridacna</i>	
Anomalodesmata	<i>Verpa penis</i>	
Archiheterodonta	[not shown]	
Pteriomorpha	<i>Pteria</i>	
Protobranchia	<i>Solemya velum</i>	
Scaphopoda		
Dentaliida	Scaphopoda schematic	
Gadilida	[not shown]	
Gastropoda		
Heterobranchia	Acochliidae	
	Helicidae	
Caenogastropoda	<i>Concholepas concholepas</i>	
Neritimorpha	[not shown]	
Patellogastropoda	Patellidae	
Neomphaliones	<i>Chrysomallon squamiferum</i>	
Vetigastropoda	[not shown]	
Cephalopoda		
Decapodiformes	squid schematic	
	octopus schematic	
Nautiloidea	[not shown]	
Monoplacophora	monoplacophoran schematic	
Polyplacophora	chiton schematic	
Chitonida	<i>Acanthochitona discrepans</i>	
Callochitonida	[not shown]	
Lepidopleurida	[not shown]	
Caudofoveata	caudofoveate schematic	
Solenogastres	solenogaster schematic	

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