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Opinion

Time to say "Bye-bye Pulmonata"?

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For more than a century, euthyneuran gastropods (> 30 000 species) were formally divided into Opisthobranchia (sea slugs and related snails) and Pulmonata, which are aquatic or predominately terrestrial slugs and snails, (most) with a "lung". The typical pulmonary cavity, however, was regarded as homologous with opisthobranch mantle (or pallial) cavities (Ruthensteiner 1997). General morphoanatomical approaches did not regularly recover or recognize monophyly of these classical but not sharply differentiated textbook taxa (e. g. Haszprunar 1985, Smith & Stanisic 1998, Dayrat & Tiller 2002), nor did some broader molecular systematic studies (e. g. Klussmann-Kolb et al. 2008, Dinapoli & Klussmann-Kolb 2010). The first study to formally reclassify euthyneuran gastropods according to multi-locus trees was Jörger et al. (2010). These authors established Euopisthobranchia for several traditional "opisthobranch orders" such as Cephalaspidea and Anaspidea, and Panpulmonata for pulmonates plus opisthobranch acochlidians and sacoglossans, and even pyramidellids, among others (Fig. 1). The combined clade of Euopisthobranchia and Panpulmonata was called Tectipleura later (Schrödl et al. 2011a), the name referring to the inferred sistergroup relationship with Nudipleura.

Changes of phylogenetic tree hypotheses can be of utmost impact, e. g. on evolutionary reconstructions. For example, recovering the supposedly derived Nudipleura in a basal euthyneuran position but the supposedly basal Cephalaspidea in a derived position (Brenzinger et al. 2013b) may turn traditionally inferred character evolution (e. g. Wägele & Klussmann-Kolb 2005) up-side down. Jörger et al. (2010) translated molecular trees into a new classification, which was initially tolerated rather than truly considered by parts of the opisthobranch community. Calling "Bye bye Opisthobranchia!", Schrödl et al. (2011a) thus wanted to stimulate open discussion about the new scenarios and potential consequences. They also wanted to revitalize older ambitions of bringing together researchers specialized on different taxa (e. g. Dayrat & Tillier 2002), to face new comprehensive challenges. The new euthyneuran tree as e. g. presented by Brenzinger et al. (2013a) was essentially supported by recent phylogenomic and nuclear rDNA dominated studies (Kocot et al. 2013, Stöger et al. 2013). Alternative topologies from mitogenomics (e. g. Medina et al. 2011, White et al. 2011) were obviously misrooted (see Schrödl et al. 2011b, Stöger & Schrödl 2013). This new euthyneuran tree displaying paraphyletic opisthobranchs thus has gained considerable acceptance within the large and active opisthobranch research community (see Wägele et al. 2014). Opisthobranchia is still used as a taxon in many collections and databases, such as WORMS (http://www.marinespecies.org/), but an obviously decreasing number of authors still believe in the old paradigm of monophyletic Opisthobranchia. According to a very rough Google Scholar search, Opisthobranchia appears as a valid taxon in the title of at least 25 papers in 2013, but so far by none in 2014 (as of September). Ultimately, evidence against Opisthobranchia has become compelling: A recent phylogenomic paper on gastropods by Zapata et al. (2014) using massive sequence data on a quite representative euthyneuran taxon sampling confirms Tectipleura, Euopisthobranchia and Panpulmonata, conclusively rejecting Opisthobranchia, but also rejecting traditional Pulmonata (see Fig. 1).

The case of Pulmonata is more problematic. On one hand, the Panpulmonata have been confirmed beyond reasonable doubt (e. g. Jörger et al. 2010, 2014, Kocot et al. 2013, Zapata et al. 2014), rendering traditional Pulmonata paraphyletic (Fig. 1). The new euthyneuran classification with Panpulmonata as an essential innovation thus far has passed the test of time on the molecular side, and the double-rooted rhinophoral nerve has been suggested as a synapomorphy (Brenzinger et al. 2013a), though not unique to panpulmonates. The new euthyneuran classification is not yet complete or definite, but based on phylogenetic evidence, and thus it should be used. On the other hand, the taxon Pulmonata still is omnipresent (but see e. g. Salvini-Plawen & Haszprunar 2013). There are numerous recent scientific studies displaying Pulmonata in the title, even in the most recent issues of major malacological journals. Are they all tolerating paraphyletic taxa and retaining an outdated evolutionary concept? The question is, whether or not well-supported Panpulmonata is strictly exclusive to a (sub)taxon Pulmonata, and how far the community is willing to adapt Pulmonata to changing topologies and evolutionary scenarios.

In their phylogenomic study using a small euthy-

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neuran taxon set, Kocot et al. (2013) recovered monophyletic Panpulmonata, with opisthobranch Sacoglossa sister to Pulmonata (Siphonarioidea sister to Hygrophila plus Stylommatophora). Most recent phylogenomic analyses by Zapata et al. (2014), which are based on a considerably broader and more representative euthyneuran taxon sampling, and multi-locus studies on a dense euthyneuran sampling (Jörger et al. 2014) consistently recover highly supported Panpulmonata, and some support for Sacoglossa as first panpulmonate offshoot. None of the latter studies recovers Pulmonata, but deep inner panpulmonate topologies are weakly supported and partly incompatible. Panpulmonate phylogeny thus is unresolved in the illustrated consensus tree (Fig. 1), and the taxon Panpulmonata is likely, but not yet securely, exclusive to a (sub)taxon Pulmonata, in a traditional sense.

In their multi-locus (3 genes) study, Dayrat et al (2011) recovered a tree with some pulmonate clades associated with non-pulmonate ones, essentially recovering Panpulmonata (but not indicating the node as such). However, the authors proposed preserving the old taxon Pulmonata, including real pulmonates having a contractile pneumostome and Siphonarioidea having a non-contractile pneumostome, or extending the Pulmonata even to include Sacoglossa. The first option would be compatible with Pulmonata sensu Kocot et al. (2013), but would tolerate Pulmonata containing Glacidorbidae, Pyramidellidae and Acochlidia besides of traditional pulmonate groups. Modern defined Eupulmonata (Fig. 1) contain most of the pulmonate groups with contractile pneumostome, but not Hygrophila. There is no molecular evidence yet for relationships of these taxa, and thus no evidence for an apomorphy-based definition of Pulmonata restricted to Eupulmonata and Hygrophila. In the second option, Pulmonata would be used as a synonym of Panpulmonata, containing even more non-pulmonate groups. So, what exactly are Pulmonata now? A taxon worth to be defended against all odds? And how inclusive (or blurred) should a Pulmonata concept become, in an e.g. evolutionary or ecological sense?

In my opinion, traditional Pulmonata most likely has failed from a phylogenetic point of view, and thus should be avoided in a systematic, taxonomic and classificatory context. In the light of numerous, ecologically and morphologically heterogeneous historical lower heterobranch and opisthobranch taxa clustering with pulmonate taxa (Fig. 1), I also see no particular, somehow coherent "pulmonate" evolutionary pattern. For example, even a "typical pulmonate" taxon, the limnic Hygrophila, may not have had an ancestral lung but rather retained a plesiomorphic mantle cavity for oxygen uptake from water. The latter can be assumed also for other basal aquatic panpulmonates (Fig. 1). Air-breathing in Hygrophila seems limited to rather big and derived species, and thus is likely an adaptation to oxygen-poor warm waters. If there is any "typical pulmonate" evolutionary pattern, it is the considerable ecological and thus evolutionary flexibility in several (but not all) panpulmonate subgroups. Adapting a statement by Vogel & Wainwright (1969) to this situation, the traditional pulmonate concept is a corpse, and newly constructed ones are ghosts.

As happened with opisthobranchs, parts of the even larger Pulmonata research community may feel uncomfortable with questioning or abandoning their familiar taxon concept, or simply may not see any necessity to change customs. While I do respect (or even like) controversial hypotheses and opinions in systematic science, I do not see the point of an almost entire research community retaining traditional (or vaguely modified) Pulmonata. Zapata et al. (2014) indicated that the traditional taxon Pulmonata and its inner classification are obsolete and it may make little sense to adapt an outdated concept to new, yet currently not fully settled inner panpulmonate topologies. It is important to note that all these unresolved panpulmonate lineages share a common evolutionary history that is most probably incompatible with conventional concepts on pulmonate evolution, if any. Basal panpulmonates are ecologically heterogeneous, some clades (in particular, pyramidellids) are highly species diverse, and none is to be neglected, since all these groups may retain e.g. morphoanatomical or molecular signatures of early panpulmonate evolution, which is especially important in groups lacking a significant fossil record. In more general, I think there is no place for non-monophyletic taxa in phylogenetic systematics and taxonomy, which should strive to infer natural relationships, reconstruct evolution, and classify biological diversity; new names for newly recognized clades generate clarity and stability, and flag changing concepts rather than hiding them. It is the task of professional systematists to evaluate and communicate changing knowledge and paradigms to the amateurs and to the public, after all. Perhaps it is time for authors, referees and editors dealing with manuscripts on "Pulmonata" to reconsider their traditions and replace rejected beliefs by a system that is based on evidence?

Admittedly, there is no stable and complete inner panpulmonate classification available yet. Traditional Pulmonata were consistently but not yet fully convincingly rejected. Avoiding Pulmonata thus may not (yet?) be a "must", but a recommendation. Some easy measures, such as putting Pulmonata into quotation marks, would already signal serious doubts on its strict meaning as a (monophyletic) taxon, still guaranteeing for being found as a key word by peers. The same is true if using the next higher, uncontroversial taxon, Panpulmonata. Personally, I would recommend the descriptive use of the adjective term pulmonate, or pulmonates, for pulmonate Euthyneura. This is analogous to the nowadays familiar usage of the informal term opisthobranchs.

While Panpulmonata is robustly recovered, deep inner panpulmonate relationships are controversial and poorly supported (e.g. Dayrat et al. 2011, Dinapoli & Klussmann-Kolb 2010, Jörger et al. 2010, 2014, Kocot et al. 2013, Zapata et al. 2014), and thus early panpulmonate evolution is basically unresolved (Fig. 1). However, some consistent topological patterns emerge from these

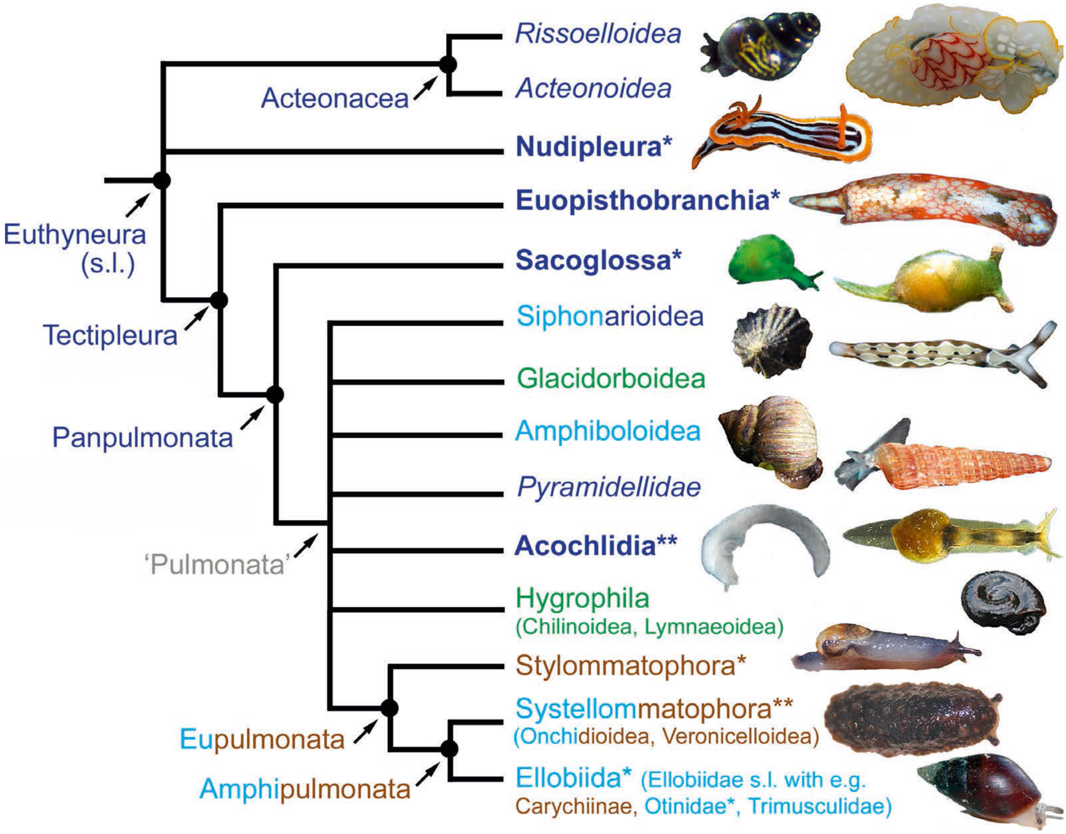

Fig. 1. Euthyneuran consensus tree from recent multi-locus (Jörger et al. 2014) and phylogenomic (Zapata et al. 2014) studies. Dots indicate robust node support. "Lower heterobranch" taxa are in italics, opisthobranchs in bold face, and traditional pulmonates in normal face. Note that siphonariids sometimes were regarded as opisthobranchs, and Glacidorbidae as basommatophoran pulmonates. Habitats are colour coded; taxa in blue are (inferred to be ancestrally) marine, taxa in light blue are intertidal, taxa in green are limnic, and taxa in brown are terrestrial; mixed colours indicate ambiguity. Eupulmonata, Amphipulmonata and Systellommatophora are inferred by parsimony to be ancestrally terrestrial or intertidal, the "pulmonate?" ancestor of unresolved non-sacoglossan panpulmonates was aquatic, likely marine or intertidal. An asterisk indicates taxa with snails (or limpets) and slugs, two asterisks mark taxa with (sea) slugs only.

molecular studies, answering several old and disputed questions in pulmonate systematics. As a consensus, the monophyly of traditional e.g. Archaeopulmonata and Basommatophora is rejected, while the monophyly of e.g. Hygrophila or Eupulmonata (sensu Smith and Stanisic 1998), comprising Stylommatophora, Systellommatophora and Ellobioidea with Otinidae and Trimusculidae, and of each of these subclades, is likely (see Fig. 1). Within Eupulmonata, rRNA gene dominated studies using 4 loci (but see Dayrat et al. 2011, using 3 loci) reject the Geophila (Systellommatophora plus Stylommatophora) concept, robustly recovering Stylommatophora sister to a clade of Systellommatophora and Ellobiida (Fig. 1). The latter combined clade, here called Amphipulmonata, comprises morphologically and ecologically heterogeneous groups, i.e. marine intertidal or terrestrial systellommatophoran slugs with eyes on stalks and mainly marine intertidal ellobiid snails with eyes at the base of tentacles, but also terrestrial carychiid snails, and marine intertidal trimusculid limpets and smeagolid slugs. It is still possible to suspect multiple adaptations to the land rather than secondary invasions of the sea (Dayrat et al. 2011), but this option becomes less likely if considering the putative sister group relationship of Amphipulmonata to terrestrial Stylommatophora (Fig. 1).

Concluding, the origin of several major pulmonate taxa is unclear and ancestral states, of e.g. ecology or tentacles of Eupulmonata and subclades, are largely unknown also. Conchologists working on derived terrestrial groups, e.g. certain stylommatophoran genera, may not be too worried about future topological hypotheses at the base of (pan)pulmonates. However, the understanding of morphoanatomy even of land pulmonates already is affected by changing homology assumptions on important organs. For example, Koller et al. (2014) suggested partial homology of apogastropod tentacles and opisthobranch rhinophores, and the latter are likely homologous to stylommatophoran and systellommatophoran "eye-stalks". Investigators dealing with pulmonate family level or higher taxa, especially if aquatic, may want to consider potential effects on their groups from recent and future molecular revolutions. These have to do with usually marine taxa such as lower heterobranchs and opisthobranchs and were performed by researchers mainly working with those groups. The pulmonate community may embrace these exciting advances and their newly gained colleagues, and should combine efforts towards a better understanding of pulmonate phylogeny and evolution. Let's say "good bye" to Pulmonata, and "Welcome" to challenges and chances imposed by the joint exploration of Panpulmonata.

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