
Test of the monophyly of Odostomiinae and Turbonilliinae (Gastropoda, Heterobranchia, Pyramidellidae) based on 16S mtDNA sequences

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While gastropod phylogeny has received much recent attention, relationships within some major gastropod clades have still not been studied. The Pyramidellidae is one such group, comprising more than 6000 named species in more than 350 genera. We sequenced part of the mitochondrial 16S gene from 32 species in an attempt to clarify pyramidellid phylogeny and employed a successive alignment approach that allowed us to maximize the phylogenetic signal of the data. Neighbour-joining, maximum parsimony and likelihood analyses recovered two distinct clades. One clade consisted of *Noemiamea* which nested within *Odostomia* (*sensu stricto*). The inclusion of *Brachystomia*, *Megastomia*, *Jordaniella* and *Liostomia* within *Odostomia* is not supported. The second clade comprised *Spiralinella*, *Brachystomia*, *Boonea*, *Jordaniella*, *Liostomia* and *Parthenina*. Our results further suggest that *Turbonilla*, as interpreted by most authors, is polyphyletic. This study shows that the 16S gene is useful in unravelling pyramidellid phylogeny but needs to be combined with other data (including molecular, morphological and developmental) to fully clarify the evolutionary relationships.

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Introduction

While gastropod phylogeny has received much recent attention (Tillier *et al.* 1992; Ponder & Lindberg 1997; Rosenberg *et al.* 1997; Thollessen 1999) relationships within some major clades are still poorly understood. An example is Heterobranchia, which includes the Pyramidellidae, comprising more than 6000 named species distributed among more than 350 genera. The basal heterobranchs, including the pyramidellids, have characteristics of both basal and more derived gastropod taxa (Bieler 1992). Until recently, most systematic studies (e.g. Peñas & Rolán 1997a,b, 1998, 1999) of pyramidellids have been based almost entirely on shell characters. A probable reason is the small size of these gastropods, often < 3 mm, and their lack of a radula. The Pyramidellidae are represented in all oceans; they are commonly ectoparasites on other molluscs and on a range of invertebrates. Although anatomical studies (Wise 1993, 1996, 2000; Hori & Okutani 1995, 1996; Peterson 1998; Hori & Nakamura 1999; Schander

et al. 1999b and references therein) have demonstrated morphological variation within this taxon, the data have for the most part not been used in phylogenetic classifications.

The taxonomy of the Pyramidellidae is unsettled when it comes to the classification within the family (e.g. Dall & Bartsch 1904, 1909; Laws 1937a,b,c,d, 1938, 1939a,b). Older classifications are still the most widely used, although there have been recent attempts (Wise 1996; Schander *et al.* 1999b) to analyse the family within a phylogenetic framework. At present, morphological data are limited and molecular data may therefore offer new insights into its evolutionary history. We test the monophyly of the genera *Odostomia* and *Turbonilla sensu lato* (Table 3) based on 16S rDNA gene sequences and present a hypothesis of some of the relationships within the Pyramidellidae. The 16S rDNA gene was chosen since previous studies (e.g. Lieberman *et al.* 1993; Thollessen 1999; Koufopanou *et al.* 1999; Medina & Walsh 2000) have shown it to be useful in reconstructing gastropod phylogenies.

Table 1 List of species included in the analysis, sampling localities, and GenBank accession numbers.

Species	Collection site	GenBank accession number	Comments
<i>Acteon tornatilis</i> (Linné, 1758)		AJ223405	Thollesson 1999
<i>Babella caelator</i> (Dall & Bartsch, 1909)	Kuwabara, Tokuyama City, Yamaguchi prefecture, Japan	AF355173	
<i>Brachystomia</i> sp.	Oregon margin 40°40'15" N 125°05'91" W, 590 m. Alvin Dive 3452	AF355161	Undescribed
<i>Brachystomia sitkaensis</i> (Clessin, 1900)	Horseshoe cove, Bodega Bay, Sonoma County, California, USA	AF355162	
<i>Boonea seminuda</i> (C. B. Adams, 1839).	Woods Hole, Massachusetts, USA	AF355163	
<i>Cingulina terebra</i> (Dunker, 1860)	Akushima, Abu-chô, Yamaguchi Prefecture, Japan	AF355178	
<i>Egilina mariellaeformis</i> (Nomura, 1938)	Ôkora, Shimonoseki City, Yamaguchi Prefecture, Japan.	AF355174	
<i>Eulimella laevis</i> (Brown, 1827)	Hågarnskär, Gullmar area, Lilleskärlätten and SW of Yttre Vattenholmen, Koster area, Sweden	AF355179	sensu Warén 1991
<i>Jordaniella nivos</i> a (Montagu, 1803)	Menaver, Helford river, Cornwall, UK	AF355160	
<i>Liostomia afzelii</i> (Warén, 1991)	Hellsöarna, Koster area, Sweden	AF355157	
<i>Liostomia afzelii</i> (form B) (Warén, 1991)	Hågarnskär and Hellsöarna, Gullmar area, Lilleskärlätten, Koster area, Sweden	AF355158	Possibly corresponding to <i>L. robusta</i> Chaster, 1898, here considered a form
<i>Liostomia clavula</i> (Lovén, 1846)	Hågarnskär, Gullmar area, Sweden	AF355159	
<i>Megastomia conoidea</i> (Brocchi, 1814)	Humlesäcksrännan, Gullmar area, Sweden	AF355166	
<i>Megastomia corimbensis</i> (Schander, 1993)	Corimbo, Luanda	AF355167	
<i>Odostomia</i> (<i>Noemiamea</i>) <i>dolioliformis</i> (Jeffreys, 1848)	Harwich, Essex, UK	AF355170	
<i>Odostomia acuta</i> (Jeffreys, 1848)	Hellsöarna and Humlesäcksrännan, Gullmar area, Lilleskärlätten, Koster area, Sweden	AF355165	
<i>Odostomia angusta</i> (Jeffreys, 1867)	Torallia Island, Vigo, Spain	AF355172	
<i>Odostomia hirotamurana</i> (Nomura, 1938)	Sanshigito Island, Hishi-Izu, Shizuoka Prefecture, Japan; Usami, Ito, Izo Peninsula, Shizuoka Prefecture, Japan	AF355171	
<i>Odostomia kromi</i> (van Aartsen, Menkhorst & Gittenberger, 1984)	Sicily, Italy	AF355169	
<i>Odostomia turrita</i> (Hanley, 1844)	Håleströmmarne, Gullmar area, Klinken, Koster area, Sweden	AF355168	
<i>Ondina diaphana</i> (Jeffreys, 1848)	Hellsöarna, Gullmar area, Sweden	AF355180	
<i>Ondina divisa</i> (J. Adams, 1797)	Lilleskärlätten, Koster area, Sweden	AF355182	
<i>Ondina perez</i> i (Dautzenberg & Fisher, 1925)	Between Spättan and Flatholmen, Gullmar area, Swedish west coast	AF355181	
<i>Parthenina affectuosa</i> (Yokoyama, 1927)	Kuwabara, Tokuyama City, Yamaguchi prefecture, Japan	AF355156	
<i>Parthenina interstincta</i> (J. Adams, 1797)	Håleströmmarna, Gullmar area, Klinken, Koster area, Sweden	AF355154	sensu Warén 1991
<i>Parthenina sarsii</i> (Nordsieck, 1972)	Hågarnskär, Gullmar area, Sweden	AF355155	comb. nov., previously called <i>Chrysallida sarsii</i>
<i>Longchaeus schanderi</i> (van Aartsen, Gittenberger & Goud, 1998)	Cape Verde Islands	AF355175	
<i>Spiralinella pellucida</i> (Dillwyn, 1817)	Lilleskärlätten, Koster area, Sweden	AF355164	
<i>Pyrgiscus crenatus</i> (Brown, 1827)	Lilleskärlätten, Koster area, Sweden	AF355184	As figured in Graham 1988
<i>Pyrgiscus rufa</i> (Philippi, 1836)	S.W. Yttre Vattenholmen, Koster Area, Sweden	AF355183	As figured in Peñas & Rolán 1997b
<i>Turbonilla kerstinae</i> (inc. sed.) Schander, 1994	Morro de Areia, Boavista Island, Cape Verde Islands	AF355185	
<i>Turbonilla lactea</i> (Linné, 1758)	Marazion, Cornwall, UK	AF355176	
<i>Turbonilla subulina</i> (Monterosato, 1889)	Corimbo, Luanda	AF355177	

Materials and methods

Specimens, DNA extraction and PCR amplification

Specimens of 32 pyramidellid species were collected at 28 sites (Table 1), placed in 70–90% ethanol, and stored at 4 °C

until DNA extraction. The choice of taxa was purely pragmatic, in that it included all the species we were able to obtain and sequence. *Acteon tornatilis* was used as an outgroup; recent studies by Mikkelsen (1996) and Thollesson (1999)

indicated that it is the sister group of, or is holding a basal position within, Euthyneura. Euthyneura is a clade nested within Heterobranchia not comprising Pyramidellidae (Ponder & Lindberg 1997).

DNA was extracted using either of two methods. In the first method, whole animals (except in the case of *Longchaeus schanderi*, where only a part of the foot was used) were placed in a tube containing 50 μ L 5% chelex 100 solution. They were then ground with a Teflon pestle, 1 μ L proteinase K (50 μ g/mL) was added, the sample was vortexed, and incubated at 56 °C for 1 h. An additional 1 μ L Proteinase K was added and incubation continued for 1–2 h until the tissue was completely dissolved. After vortexing for 10 s, the sample was boiled for 8 min to inactivate the proteinase K, and centrifuged at 13 000 r.p.m. (16 000 g) for 2 min. The supernatant was removed and kept at –20 °C prior to amplification. In the second method, whole animals were placed in 600 μ L 2 \times CTAB (= 2% hexadecyltrimethylammonium bromide (Sigma), 100 mM Tris-HCL pH 8.0, 1.4 M NaCl, 20 mM EDTA, 0.2% 2-mercaptoethanol) and were ground with a pestle in a 1.5-mL Eppendorf tube. The samples were then incubated for 30 min at 55–60 °C and briefly vortexed. A phenol extraction was performed followed by a chloroform–isoamylalcohol extraction. DNA was precipitated by adding 60 μ L 3 M NaOAc and 1000 μ L 100% ice-cold ethanol to the final aqueous phase. The samples were placed in a –20 °C freezer for 10 min to overnight. DNA was pelleted by centrifugation. After a 70% ethanol rinse, the samples were resuspended in 30–50 μ L 1 \times TE or distilled water and kept at –20 °C prior to amplification.

An approximately 450 bp section of the 16S rDNA mitochondrial gene was amplified by PCR using the primers 16Sar (CGCCTGTTTATCAAAAACAT) and 16Sbr (CCGGTCTGAACTCAGATCACGT) (Palumbi 1996) or 16LRN13398 (CGCCTGTTTAAACAAAACAT) and 16RTHB (ACGCCGTTTGAAGTCCAGGATC) (Koufopanou *et al.* 1999). PCR reactions were carried out in a final volume of 50 μ L [template DNA 1–50 ng; TMAC 0.0001%; Mg²⁺ 2.0 mM; primer 10–30 pmol; 1 \times reaction buffer (10 mM Tris-HCL pH 8.3, 50 mM KCL); dNTP 0.4 mM (0.1 mM each); *Taq* polymerase (from Perkin Elmer) 2.0 units]. The PCR temperature profile was as follows: 94 °C 2 min; (94 °C 30 s, 47–55 °C 30 s, 70–72 °C 1 min) \times 40; 72 °C 7 min; 4 °C hold. Amplification products were purified with Promega Wizard or QIAquick (QIAGEN). Sequence data were collected using Cy5 Auto-Cycle, ALFexpress (Pharmacia Biotech), Big Dye and ABI Prism 377 (PE Applied Biosystems) sequencing kits. All 16S fragments were sequenced in both directions.

Phylogenetic analyses

The sequences were combined with data for the outgroup from GenBank to produce three different aligned data sets:

(1) all-taxa, (2) Liostomini clade and (3) *Odostomia* clade (see Table 1). Initially, all the taxa were used in a single alignment. Analyses of this revealed two larger subclades for which we hoped to obtain additional resolution by realigning less inclusive subsamples of taxa (see Halanych 1998). All alignments employed Clustal X (Thompson *et al.* 1994) and were manually proofread; they are available at TREEBASE (<http://www.treebase.org/treebase>). Only unambiguously aligned regions were included.

All data sets were subject to Neighbor-joining (NJ), maximum parsimony (MP) and maximum likelihood (ML) analyses, using PAUP 4.0b4a (Swofford 2000). MacClade 3.06 (Maddison & Maddison 1996) was used for character and tree analyses. NJ trees were reconstructed under Jukes–Cantor, Kimura-2-parameter, Tamura–Nei, General-Time-Reversible, and Log/Det models (all except Log/Det were examined under equal rates of among site rate variation and using the empirically derived gamma shape parameter, α , of 0.3). All of these models are easily incorporated in PAUP and were chosen to represent a variety of assumptions commonly employed in nucleotide correction models (see Swofford *et al.* 1996 for a summary of different assumptions used in these models). A Kishino–Hasegawa (1989) likelihood evaluation of the resulting topologies revealed no significant differences between models for any of the alignments. Likelihood evaluations estimated a 6 substitution-type rate matrix for which nucleotide base frequencies were set to the observed values and α was estimated. For the purposes of presentation, we arbitrarily chose to report the results based on a Log/Det model because of its desirable properties (Lake 1994; Lockhart *et al.* 1994). NJ bootstraps consisted of 1000 iterations.

For the all-taxa and Liostomini clade alignments, parsimony analysis consisted of a heuristic search with 100 random sequence additions and Tree-Bisection-Reconnection (TBR) branch swapping. Due to the number of taxa, an exhaustive search was feasible for the *Odostomia* clade alignment. Transitions (Ti) and transversions (Tv) were given equal weighting. ML evaluation of parsimony topologies was the same as for NJ topologies; 1000 iterations were used for bootstrap analyses. ML analyses were limited by computation time. We used a nucleotide model with two substitution types where the Ti/Tv ratio was set to the value estimated for the best parsimony tree; base frequencies and rate variation among sites were assumed to be equal. The all-taxa alignment employed a heuristic search using the best parsimony trees as a start point whereas the Liostomini clade and *Odostomia* clade alignments used 10 heuristic replicates. ML bootstraps employed the Faststep option with 100 iterations.

Results

The all-taxa alignment consisted of 483 characters for which 200 could be unambiguously aligned (of these 67 [34%] were

parsimony informative). Table 2 lists mean character distance and total character difference based on the unambiguously aligned data. All three reconstruction methods yielded similar results in that well-supported (as judged by bootstrap values) associations tended to be restricted to species within recognized genera, whereas the most basal relationships varied between topologies. For comparative purposes, Fig. 1 shows parsimony and Log/Det NJ topology (Ln likelihood = -1820.423). Of particular interest is the difference in the placement of *Megastomia* species and *Odostomia acuta*. Parsimony produced 12 trees (287 steps) that were not significantly different from each other based on ML evaluation. The best of these trees (Ln likelihood = -1636.005) also contained a *Megastomia/O. acuta* grouping as in the ML topology (but differed in basal groupings). Based on the variation in placement of *Megastomia* and *O. acuta*, we explored the possible long-branch effects of the *Megastomia*. We repeated the analysis of the all-taxa alignment excluding the two *Megastomia* taxa. NJ, parsimony and ML all grouped *O. acuta* within the *Odostomia* clade, but never with greater than 65% support.

Regardless of whether *Megastomia* was included or excluded from the analyses, we consistently recovered several minor clades and two distinct major clades (Figs 1 and 2). *Babella caelator* and *Egilina mariaelliformis* always grouped together with 100% bootstrap support. For the unambiguously aligned data, these taxa show 4.0% mean character difference; a value comparable to many interspecific comparisons of recognized genera (e.g. *Parthenina* spp. 1.5–6.6%, *Liostomia clavula*–*L. afzelii* 8.0%, *Ondina* spp. 0.5–9.0%). Although *Longchaeus schanderi* consistently clustered with *B. caelator* and *E. mariaelliformis*, bootstrap support of greater than 72% was never found. A second clade of interest, here called Turbonilliinae, involved *Turbonilla lactea*, *T. subulina*, *Cingulina terebra*, and *Eulimella laevis*. This clade received moderate bootstrap support (including *Megastomia* in the alignment, 87%, 70% and 54% for NJ, parsimony and ML, respectively; excluding *Megastomia*, 88%, 76% and 58%), and was consistently resolved in the analyses. *Turbonilla kerstinae* (inc. sed.) was never recovered within this clade, nor was any of the *Pyrgiscus* species. This suggests that *Turbonilla* in the sense of Dall & Bartsch (1904, 1909) is not monophyletic. Wise (1996) came to similar conclusions based on studies of the morphology of *Pyrgiscus neisa*.

The largest consistently resolved clade was the Liostomina clade (Figs 1 and 2). Although the most basal node of this grouping received low bootstrap support (including *Megastomia*, 65%, 50% and > 50% for NJ, parsimony, and ML, respectively; excluding *Megastomia*, 80%, 64% and 54%), this clade was repeatedly recovered in the 'best' trees. Therefore, in an attempt to increase phylogenetic signal we realigned and reanalysed this subclade. This alignment

had 459 positions of which 331 were unambiguously aligned (161 variable and 130 parsimony informative characters [28%]). Based on the previous analyses, *Turbonilla kerstinae* (inc. sed.) was used to root this subclade. Parsimony yielded similar results with one of the two most parsimonious trees (424 steps, CI = 0.6392) matching the ML tree (the other placed *Jordaniella nivosa* one node more basal; Fig. 3). The results from the realignment are clearly more robust than for this subclade in the all-taxa alignment (compare bootstrap values in Fig. 1 vs. Fig. 2). However, in the realignment, *Brachystomia* rather than *Spiralinella* is basal.

A fourth clade revealed from the all-taxa analyses includes all the *Odostomia* species, except *O. acuta* (see above), and *Noemiamea dolioliformis*.

The *Odostomia* clade alignment had 435 characters, of which 363 were unambiguously aligned, with 191 variable and 137 parsimony informative characters (31%). Figure 4 shows the results of the Log/Det NJ and ML analyses (Ln likelihood = -2165.686) for this subclade. Based on previous results, *Pyrgiscus crenata* and *Pyrgiscus rufa* were chosen as outgroups. In contrast to the all-taxa alignment, the *Odostomia* alignment suggested that *O. acuta* was not basal to other *Odostomia* species. *Noemiamea* is most commonly regarded as a genus (e.g. Peñas & Rolán 1999). We here find it to be nested within *Odostomia*. Because it is easily distinguishable from most other *Odostomia* by its shell morphology, we here consider it a subgeneric taxon.

Constraint analyses

In order to compare our results with previously published classifications, we performed two likelihood analyses between topologies constrained to match given mutually exclusive hypotheses. The constraints were designed to test the taxa Turbonilliinae/*Turbonilla s.l.* and *Odostomia s.l.* as described in the literature (e.g. Dall & Bartsch 1904, 1909). We performed the Shimodaira–Hasegawa 1999 likelihood evaluation to test the monophyly of the Turbonilliinae clades (*Turbonilla kerstinae* (inc. sed.), *Pyrgiscus rufa*, *P. crenata*, *T. lactea*, *T. subulina*, *Cingulina terebra*) and *Odostomia s.l.* (*Brachystomia* n. sp., *B. sitkenisis*, *Megastomia conoidea*, *M. corimbensis*, *Odostomia acuta*, *O. turrita*, *O. kromi*, *O. birotamurana*, *O. angusta*, *Liostomia afzelii*, *L. afzeli* (form B), *L. clavula*, *Jordaniella nivosa*, *Noemiamea dolioliformis*). *Ondina* was not included in this analysis since strong arguments have been forwarded for its removal from *Odostomia* (e.g. van Aartsen 1987). Using the constraints option in PAUP, we conducted parsimony heuristic searches (specifics same as above) to find the best trees that were consistent and inconsistent with the monophyly of these clades. The sets of trees consistent and inconsistent with the constraint were then compared using Kishino–Hasegawa. The 16S data significantly reject the monophyly of the Turbonilliinae (all 18 'best' trees supporting monophyly

Table 2 Pairwise differences between taxa. Below diagonal: total character differences. Above diagonal: mean character differences (adjusted for missing data). Abbreviation in #17. *Na.* = *Noemiamea*.

	1	2	3	4	5	6	7	8	9	10	11	12	13	14	15	16	17
1. <i>Parthenina interstincta</i>		0.015	0.050	0.050	0.050	0.085	0.065	0.095	0.101	0.090	0.095	0.136	0.177	0.199	0.146	0.149	0.176
2. <i>Parthenina sarsii</i>	3		0.066	0.066	0.066	0.102	0.082	0.112	0.117	0.107	0.112	0.152	0.194	0.215	0.158	0.165	0.189
3. <i>Parthenina affectuosa</i>	10	13		0.070	0.070	0.080	0.065	0.095	0.095	0.101	0.111	0.141	0.204	0.220	0.141	0.138	0.161
4. <i>Liostomia afzelii</i> *	10	13	14		0.000	0.080	0.060	0.111	0.111	0.080	0.095	0.141	0.156	0.183	0.141	0.144	0.176
5. <i>Liostomia afzelii</i> (form B)*	10	13	14	0		0.080	0.060	0.111	0.111	0.080	0.095	0.141	0.156	0.183	0.141	0.144	0.176
6. <i>Liostomia clavula</i>	17	20	16	16	16		0.070	0.106	0.121	0.080	0.111	0.173	0.188	0.220	0.171	0.181	0.191
7. <i>Jordaniella nivosa</i>	13	16	13	12	12	14		0.095	0.101	0.065	0.090	0.157	0.188	0.199	0.141	0.149	0.171
8. <i>Brachystomia</i> n. sp.	19	22	19	22	22	21	19		0.025	0.060	0.080	0.136	0.194	0.210	0.151	0.144	0.156
9. <i>Brachystomia sitkaensis</i>	20	23	19	22	22	24	20	5		0.065	0.075	0.131	0.188	0.199	0.141	0.133	0.151
10. <i>Boonea seminuda</i>	18	21	20	16	16	16	13	12	13		0.065	0.141	0.183	0.188	0.141	0.144	0.151
11. <i>Spiralinella pellucida</i>	19	22	22	19	19	22	18	16	15	13		0.105	0.167	0.172	0.126	0.133	0.146
12. <i>Odostomia acuta</i>	26	29	27	27	27	33	30	26	25	27	20		0.195	0.211	0.084	0.107	0.136
13. <i>Megastomia conoidea</i>	33	36	38	29	29	35	35	36	35	34	31	36		0.059	0.188	0.183	0.194
14. <i>Megastomia corimbensis</i>	37	40	41	34	34	41	37	39	37	35	32	39	11		0.210	0.204	0.210
15. <i>Odostomia turrita</i>	29	31	28	28	28	34	28	30	28	28	25	16	35	39		0.032	0.070
16. <i>Odostomia kromi</i>	28	31	26	27	27	34	28	27	25	27	25	20	34	38	6		0.074
17. <i>O. (Na.) dolioliformis</i>	35	37	32	35	35	38	34	31	30	30	29	26	36	39	14	14	
18. <i>Odostomia hirotamurana</i>	31	33	30	34	34	36	32	28	26	29	23	23	34	35	11	10	14
19. <i>Odostomia angusta</i>	34	36	32	34	34	39	35	30	27	31	31	26	41	44	20	19	24
20. <i>Babella caelator</i>	31	34	33	29	29	34	34	30	32	26	27	35	36	36	30	28	29
21. <i>Egilina mariaelliformis</i>	31	34	35	33	33	33	33	30	32	27	28	34	38	38	30	30	29
22. <i>Longchaeus schanderi</i>	24	25	25	25	25	26	23	22	22	19	22	24	35	33	23	22	23
23. <i>Turbonilla lactea</i>	30	31	32	35	35	35	31	28	28	29	24	30	39	37	28	26	26
24. <i>Turbonilla subulina</i>	27	30	30	30	30	31	28	23	24	24	19	26	33	32	25	23	23
25. <i>Cingulina terebra</i>	27	30	27	30	30	27	30	23	24	26	23	27	36	36	26	23	25
26. <i>Eulimella laevis</i>	26	29	26	32	32	32	28	25	26	27	22	28	34	33	23	22	22
27. <i>Ondina diaphana</i>	21	21	27	27	27	27	26	23	23	23	20	21	33	31	22	21	26
28. <i>Ondina perezii</i>	25	28	31	31	31	31	30	27	27	27	24	26	38	36	24	25	28
29. <i>Ondina divisa</i>	28	29	30	32	32	32	31	26	26	27	27	27	38	35	27	25	30
30. <i>Pyrgiscus rufa</i> **	19	22	24	21	21	25	23	23	23	22	19	19	31	34	18	17	24
31. <i>Pyrgiscus crenata</i> **	19	22	24	21	21	25	23	23	23	22	19	19	31	34	18	17	24
32. <i>Turbonilla kerstinae</i>	23	26	23	24	24	27	27	20	19	21	17	22	34	37	22	21	25
33. <i>Acteon tornatilis</i>	33	34	33	36	36	41	38	31	31	36	27	28	32	32	30	27	34

	18	19	20	21	22	23	24	25	26	27	28	29	30	31	32	33
1. <i>Parthenina interstincta</i>	0.157	0.171	0.156	0.156	0.126	0.151	0.136	0.136	0.140	0.109	0.131	0.141	0.095	0.095	0.116	0.166
2. <i>Parthenina sarsii</i>	0.169	0.184	0.173	0.173	0.133	0.158	0.153	0.153	0.156	0.111	0.147	0.149	0.112	0.112	0.133	0.173
3. <i>Parthenina affectuosa</i>	0.152	0.161	0.166	0.176	0.131	0.161	0.151	0.136	0.140	0.141	0.162	0.152	0.112	0.112	0.133	0.166
4. <i>Liostomia afzelii</i>	0.172	0.171	0.146	0.166	0.131	0.176	0.151	0.151	0.172	0.141	0.162	0.162	0.106	0.106	0.121	0.181
5. <i>Liostomia afzelii</i> (form B)	0.172	0.171	0.146	0.166	0.131	0.176	0.151	0.151	0.172	0.141	0.162	0.162	0.106	0.106	0.121	0.181
6. <i>Liostomia clavula</i>	0.182	0.196	0.171	0.166	0.136	0.176	0.156	0.136	0.172	0.141	0.162	0.162	0.106	0.106	0.136	0.206
7. <i>Jordaniella nivosa</i>	0.162	0.176	0.171	0.166	0.120	0.156	0.141	0.151	0.151	0.135	0.157	0.157	0.116	0.116	0.136	0.191
8. <i>Brachystomia</i> n. sp.	0.141	0.151	0.151	0.151	0.115	0.141	0.116	0.116	0.134	0.120	0.141	0.131	0.116	0.116	0.101	0.156
9. <i>Brachystomia sitkaensis</i>	0.131	0.136	0.161	0.161	0.115	0.141	0.121	0.121	0.140	0.120	0.141	0.131	0.116	0.116	0.095	0.156
10. <i>Boonea seminuda</i>	0.146	0.156	0.131	0.136	0.099	0.146	0.121	0.131	0.131	0.120	0.141	0.136	0.111	0.111	0.106	0.181
11. <i>Spiralinella pellucida</i>	0.116	0.156	0.136	0.141	0.115	0.121	0.095	0.116	0.118	0.104	0.126	0.136	0.095	0.095	0.085	0.136
12. <i>Odostomia acuta</i>	0.121	0.136	0.183	0.178	0.130	0.157	0.136	0.141	0.151	0.114	0.137	0.142	0.099	0.099	0.115	0.147
13. <i>Megastomia conoidea</i>	0.184	0.220	0.194	0.204	0.197	0.210	0.177	0.194	0.183	0.184	0.204	0.205	0.167	0.167	0.183	0.172
14. <i>Megastomia corimbensis</i>	0.189	0.237	0.194	0.204	0.185	0.199	0.172	0.194	0.177	0.173	0.194	0.189	0.183	0.183	0.199	0.172
15. <i>Odostomia turrita</i>	0.056	0.101	0.151	0.151	0.120	0.141	0.126	0.131	0.124	0.115	0.126	0.136	0.090	0.090	0.111	0.151
16. <i>Odostomia kromi</i>	0.053	0.101	0.149	0.160	0.122	0.138	0.122	0.122	0.118	0.116	0.133	0.134	0.090	0.090	0.112	0.144
17. <i>O. (Na.) dolioliformis</i>	0.071	0.121	0.146	0.146	0.120	0.131	0.116	0.126	0.118	0.135	0.147	0.152	0.121	0.121	0.126	0.171
18. <i>Odostomia hirotamurana</i>		0.116	0.131	0.136	0.137	0.141	0.126	0.136	0.124	0.126	0.137	0.142	0.116	0.116	0.121	0.146
19. <i>Odostomia angusta</i>	23		0.191	0.201	0.147	0.161	0.146	0.141	0.167	0.151	0.162	0.162	0.136	0.136	0.146	0.171
20. <i>Babella caelator</i>	26	38		0.040	0.099	0.146	0.126	0.131	0.151	0.135	0.141	0.141	0.156	0.156	0.146	0.166
21. <i>Egilina mariaelliformis</i>	27	40	8		0.105	0.151	0.131	0.136	0.161	0.135	0.141	0.146	0.156	0.156	0.156	0.176
22. <i>Longchaeus schanderi</i>	26	28	19	20		0.094	0.079	0.089	0.107	0.068	0.071	0.068	0.084	0.084	0.089	0.120
23. <i>Turbonilla lactea</i>	28	32	29	30	18		0.030	0.070	0.075	0.099	0.120	0.131	0.121	0.121	0.116	0.156

Table 2 Continued

	18	19	20	21	22	23	24	25	26	27	28	29	30	31	32	33
24. <i>Turbonilla subulina</i>	25	29	25	26	15	6		0.045	0.059	0.078	0.099	0.111	0.101	0.101	0.090	0.141
25. <i>Cingulina terebra</i>	27	28	26	27	17	14	9		0.075	0.078	0.099	0.101	0.095	0.095	0.101	0.131
26. <i>Eulimella laevis</i>	23	31	28	30	19	14	11	14		0.078	0.102	0.114	0.118	0.118	0.108	0.129
27. <i>Ondina diaphana</i>	24	29	26	26	13	19	15	15	14		0.005	0.016	0.057	0.057	0.083	0.094
28. <i>Ondina perezii</i>	26	31	27	27	13	23	19	19	19	1		0.021	0.079	0.079	0.105	0.115
29. <i>Ondina divisa</i>	28	32	28	29	13	26	22	20	21	3	4		0.091	0.091	0.101	0.111
30. <i>Pyrgiscus rufa</i>	23	27	31	31	16	24	20	19	22	11	15	18		0.000	0.080	0.121
31. <i>Pyrgiscus crenata</i>	23	27	31	31	16	24	20	19	22	11	15	18	0		0.080	0.121
32. <i>Turbonilla kerstinae</i>	24	29	29	31	17	23	18	20	20	16	20	20	16	16		0.136
33. <i>Actaeon tornatilis</i>	29	34	33	35	23	31	28	26	24	18	22	22	24	24	27	

*Differs in 1 position in the excluded areas. **Differs in 9 positions in the excluded areas.

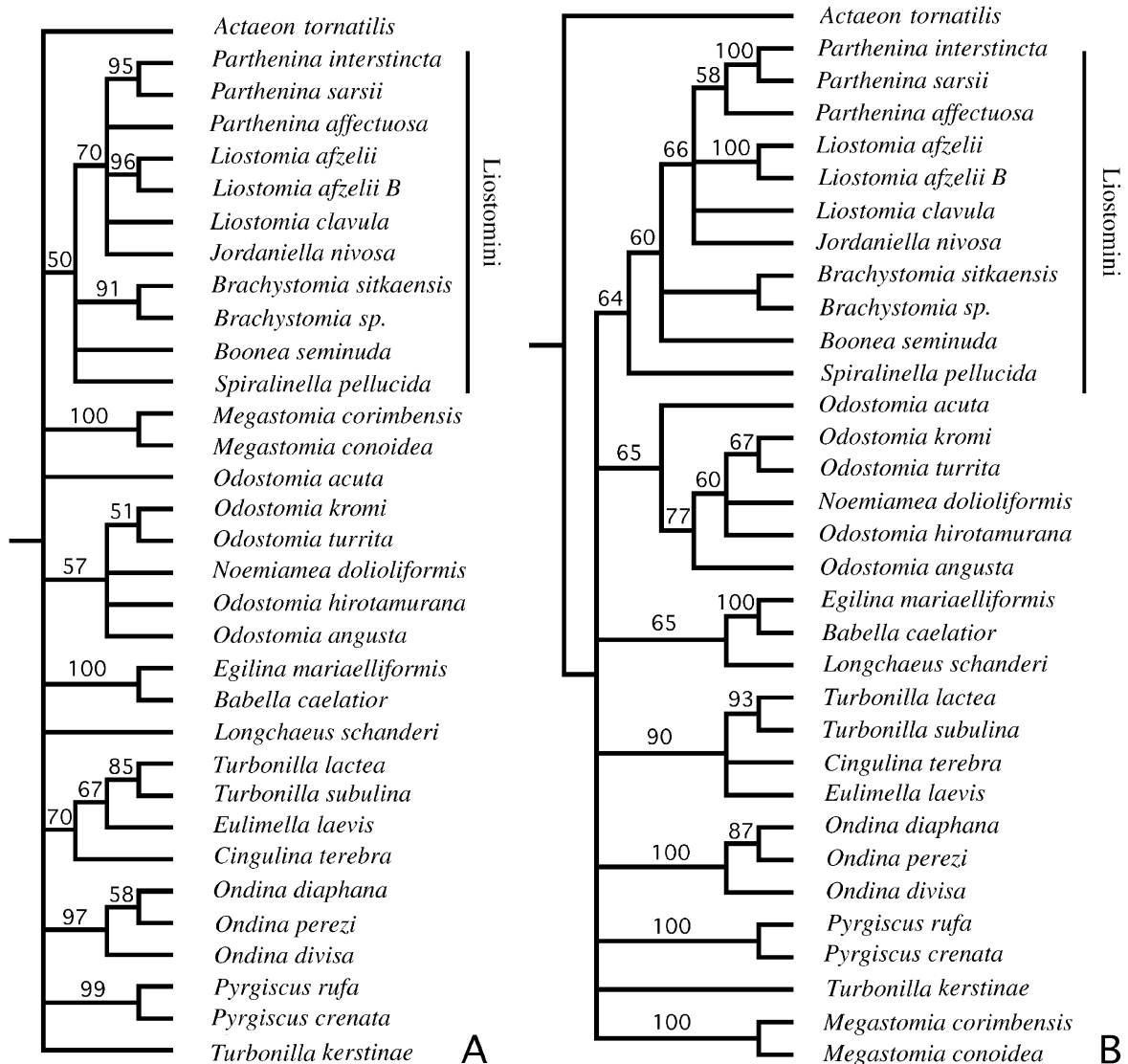


Fig 1 A, B. All-taxa alignment results. —A. Strict consensus tree of parsimony analysis including all taxa. —B. Log/Det tree of analysis including all taxa. Numerals above branches denominate bootstrap value.

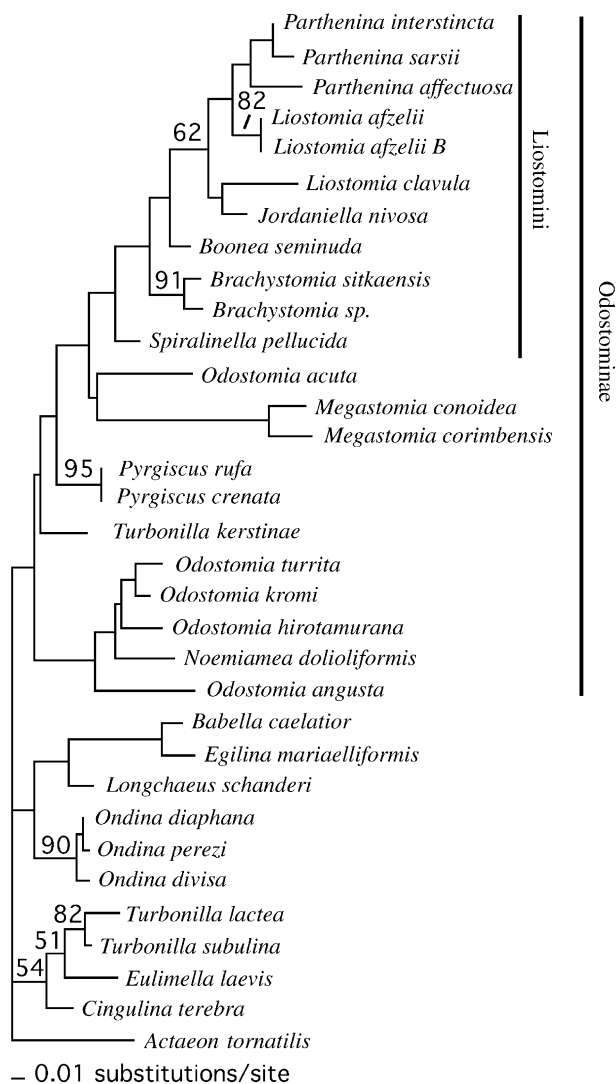


Fig. 2 All-taxa alignment maximum likelihood results. Numerals above branches denote bootstrap value.

were significantly worse than the 12 (genuinely) best trees, $P < 0.008$). For *Odostomia*, the 30/47 'best' trees were significantly worse ($P < 0.05$) than the 12 trees which did not match the constraint. Trees that were not significantly worse had a nearly significant P -value of < 0.08 . We consider the monophyly of *Odostomia s.l.* to be suspect. These results suggest that characters traditionally used to group these taxa (e.g. shell height/width ratio, axial sculpture) need to be re-evaluated.

Discussion

Our results indicate that many taxa in traditional classifications of pyramidellids (e.g. Dall & Bartsch 1904, 1909; Kuroda & Habe 1949; Bartsch 1955; Saurin 1961; Nordsieck

1972; Schander *et al.* 1999a) are nonmonophyletic. Because Turbonilliinae, Cingulininae and Eulimellinae in a Linnean system are considered to be equal entities, problems associated with ranks and synonymy arise (see de Queiroz & Gauthier 1990, Schander & Tholleson 1995; Schander 1998a).

The 16S gene is admittedly not optimal for addressing questions related to systematics, but still serves to delineate taxonomic problems with several traditionally recognized subfamilies. The commonly accepted placement of genera included in this study is found in Table 3 and additional taxonomical groupings in Fig. 5. The clade Odostomiinae was defined by Schander *et al.* (1999b) as 'the least inclusive clade comprising *Chrysalidina cincta*, *Odostomia didyma*, *O. babylonica* and *O. seminuda*.' Our analysis identifies this clade and suggests it also comprises *Parthenina*, *Liostomia*, *Jordaniella*, *Brachystomia*, *Noemiamea*, *Pyrgiscus* and *Turbonilla kerstinae* (inc. sed.). Because none of these taxa were included in the morphological study of Schander *et al.* (1999b), the results are not contradictory and there is no need to emend any definitions. Emendation of phylogenetic names should be avoided (Schander 1998b). This clade differs from *Odostomia* as interpreted by Dall & Bartsch (1904, 1909) in that it includes *Pyrgiscus* and *T. kerstinae* and excludes *Babella*, *Egilina*, *Ondina* and *Megastomia*. The genus *Odostomia* in the more modern restricted sense appears to be monophyletic if *Noemiamea* is included. Definitive placement of *O. acuta* is, however, problematic in that it alternately clusters with the long branched *Megastomia* or is basal to other *Odostomia* species depending on the phylogenetic analysis. Therefore, its placement within *Odostomia* must be questioned. Previously recognized subgenera — *Brachystomia*, *Liostomia*, *Jordaniella* and *Megastomia* — are not part of *Odostomia*.

The clade comprising *Parthenina*, *Liostomia*, *Brachystomia*, *Boonea* and *Spiralinella* is supported in all of our trees. We find that since it comprises some, but not all, representatives of the Chrysalidinae and Odostomiinae in the traditional sense, it cannot be framed within any of these names. We introduce the node-based informal name Liostomini for this clade, defining it as the least inclusive clade comprising *Liostomia clavula* (Lovén 1846) and *Spiralinella pellucida* (Dillwyn 1817). It consists of species with an 'intorted' protoconch (type C in the scheme of van Aartsen 1987). Our results indicate that the protoconch phylogeny of pyramidellids has a phylogenetic explanation. However, we note that an intorted protoconch also occurs in some, but not all, species of *Ondina*, which suggests that both character and genus need to be studied further. Interestingly, Liostomini includes both sculptured (*Parthenina*, *Jordaniella*, *Boonea*, *Spiralinella*) and un-sculptured shells (*Liostomia*, *Brachystomia*). It would have been useful to compare the morphological characters used in the analyses by Wise (1996) and Schander *et al.* (1999b) as well as

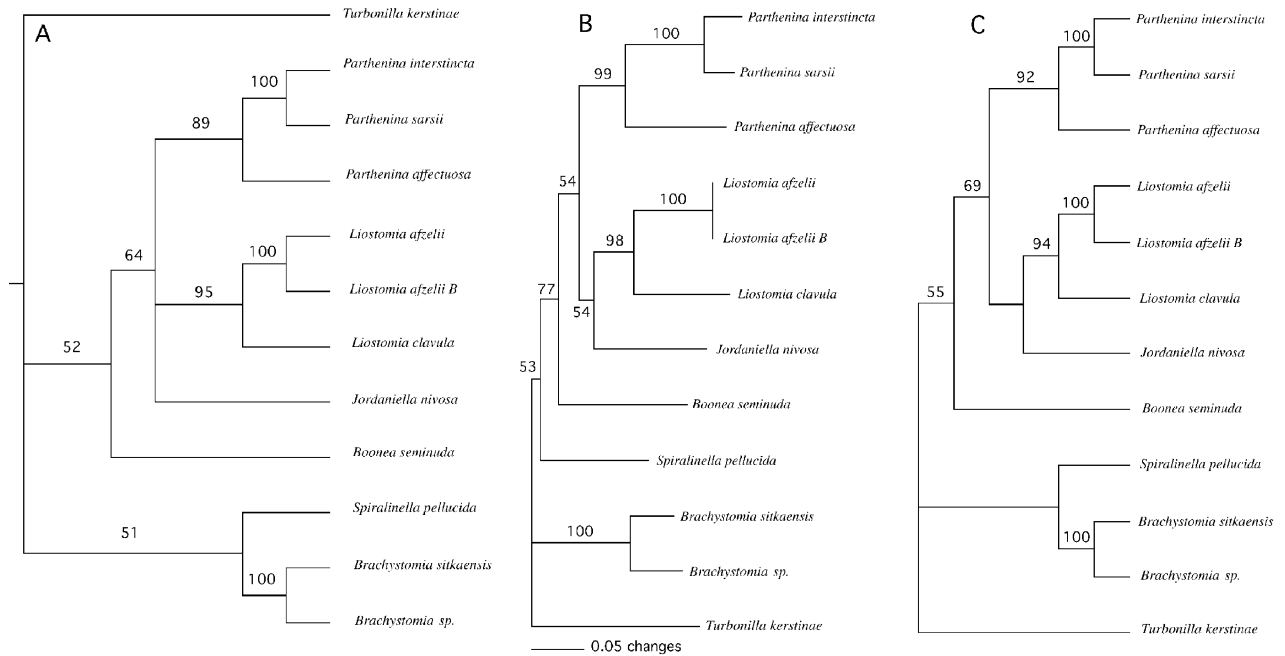


Fig. 3 Liostomini clade alignment. Results of (A) parsimony, (B) maximum likelihood and (C) Neighbor-joining analyses. Numerals above branches denominate bootstrap value.

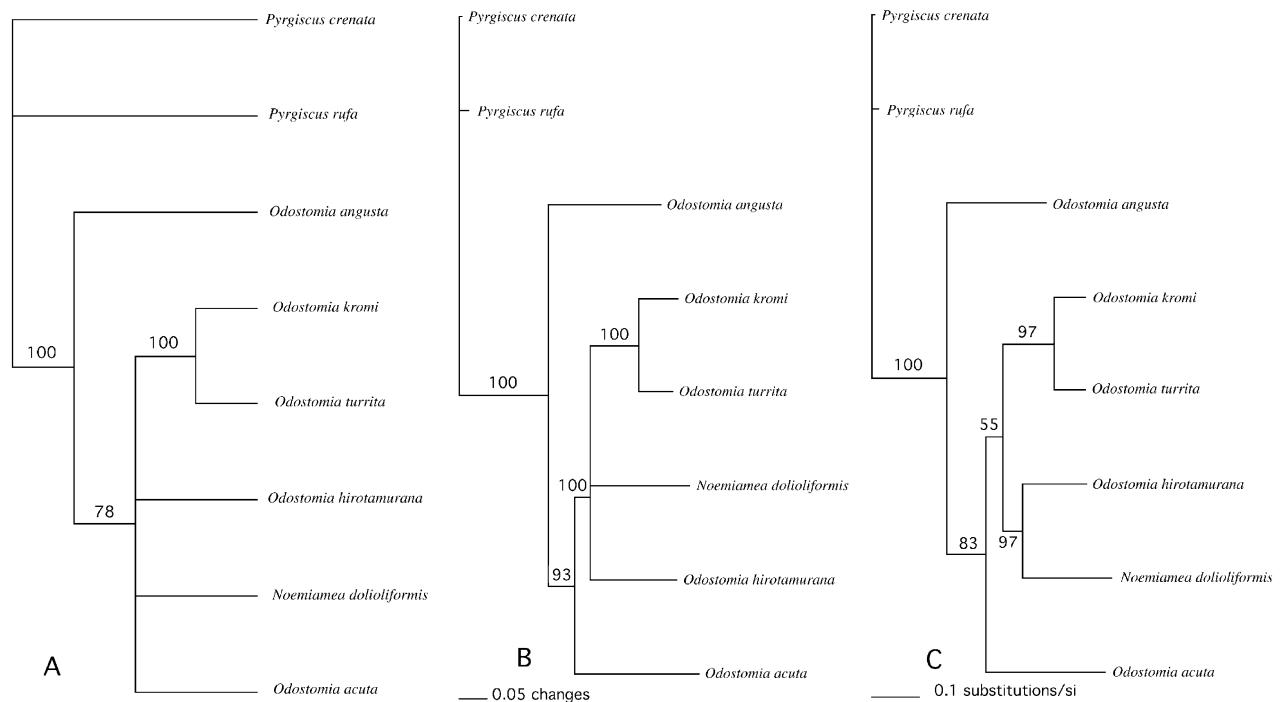


Fig. 4 *Odostomia* clade alignment. Results of (A) parsimony, (B) maximum likelihood and (C) Neighbor-joining analyses of *Odostomia*. Numerals above branches denominate bootstrap value.

Table 3 Commonly accepted placement of the genera in this study, and the traditional placement by Dall & Bartsch (1904, 1909).

Subfamily	Genera	<i>sensu</i> Dall & Bartsch (1904, 1909) and Autt.
Odostomiinae	<i>Brachystomia</i> , <i>Jordaniella</i> , <i>Liostomia</i> , <i>Megastomia</i> , <i>Noemiamea</i> , <i>Odostomia</i> , <i>Ondina</i>	<i>Odostomia</i>
Chrysalidinae	<i>Babella</i> , <i>Boonea</i> , <i>Egilina</i> , <i>Parthenina</i> , <i>Spiralinella</i>	<i>Odostomia</i>
Turbonilliinae	<i>(Cingulina)</i> , <i>Pyrgiscus</i> , <i>Turbonilla</i>	<i>Turbonilla</i>
Cingulininae	<i>Cingulina</i>	<i>Turbonilla</i>
Eulimellinae	<i>Eulimella</i>	<i>Pyramidella</i>
Pyramidellinae	<i>Longchaeus</i>	<i>Pyramidella</i>
Cyclostremellinae	Not represented	—
Odostomellinae	Not represented	—
Sayellinae	Not represented	—
Syrnoloninae	Not represented	—
Tiberiinae	Not represented	—

additional characters listed in Schander (1997) by plotting these onto the phylogenetic trees obtained here. Unfortunately this is not possible, since the anatomy is more or less unknown for the majority of the species included in this study.

This study does not refute the suggestion by Schander & Sundberg (2001) that shell characters are phylogenetically informative. The soft part anatomy is only properly described for *Boonea seminuda* (Wise 1996) and we therefore refrain from discussing morphological features of the clade. Comparison with the two morphological cladistic analyses that have been published (Wise 1996; Schander *et al.* 1999b) shows some agreement in hypothesized evolutionary relationships. However, it is difficult to make a direct comparison between these studies and the analysis presented here since the species and taxa included differ. In this study, Pyramidellinae is represented by only one species; it is the sister group of the clade formed by *Babella* and *Egilina*. Wise (1996) placed Pyramidellinae as sister to *Sayella* and *Petitilla*. In contrast, Schander *et al.* (1999b) concluded that only *Sayella*, and not *Petitilla*, is the sister clade of the Pyramidellinae. Since the results of these two studies contradict each other it would be desirable to include both *Sayella* and *Petitilla* in future molecular analyses.

The Heterobranchia has a rich and complicated fossil record (e.g. Bandel 1994; Schröder 1995 and references therein). The first species that can be identified with certainty as true pyramidellids occur in the Campanian and Maastrichtian Gulf Coast Fauna of the USA within the genera *Creonella* Wade, 1917 and *Lacrimiforma* Sohl, 1960 (Bandel 1994). Some authors (e.g. Schröder 1995) suggest that some species assigned to *Kleinella* A. Adams, 1860 from the Lower and Middle Jurassic of Northern Germany are members of Pyramidellidae, but the systematic position is uncertain and they may belong to the family Donaldinidae (Bandel 1994). A possible sister group of Pyramidellidae, Amathinae Ponder,

1987, occurs in the Upper Cretaceous of North America and Europe (Dockery 1993) with the genus *Damesia* Holzapfel, 1888. In order to understand the evolution of this important clade, it is necessary to fully understand the relationships of the Recent representatives. This study has shown that the 16S gene can be a useful tool in such research.

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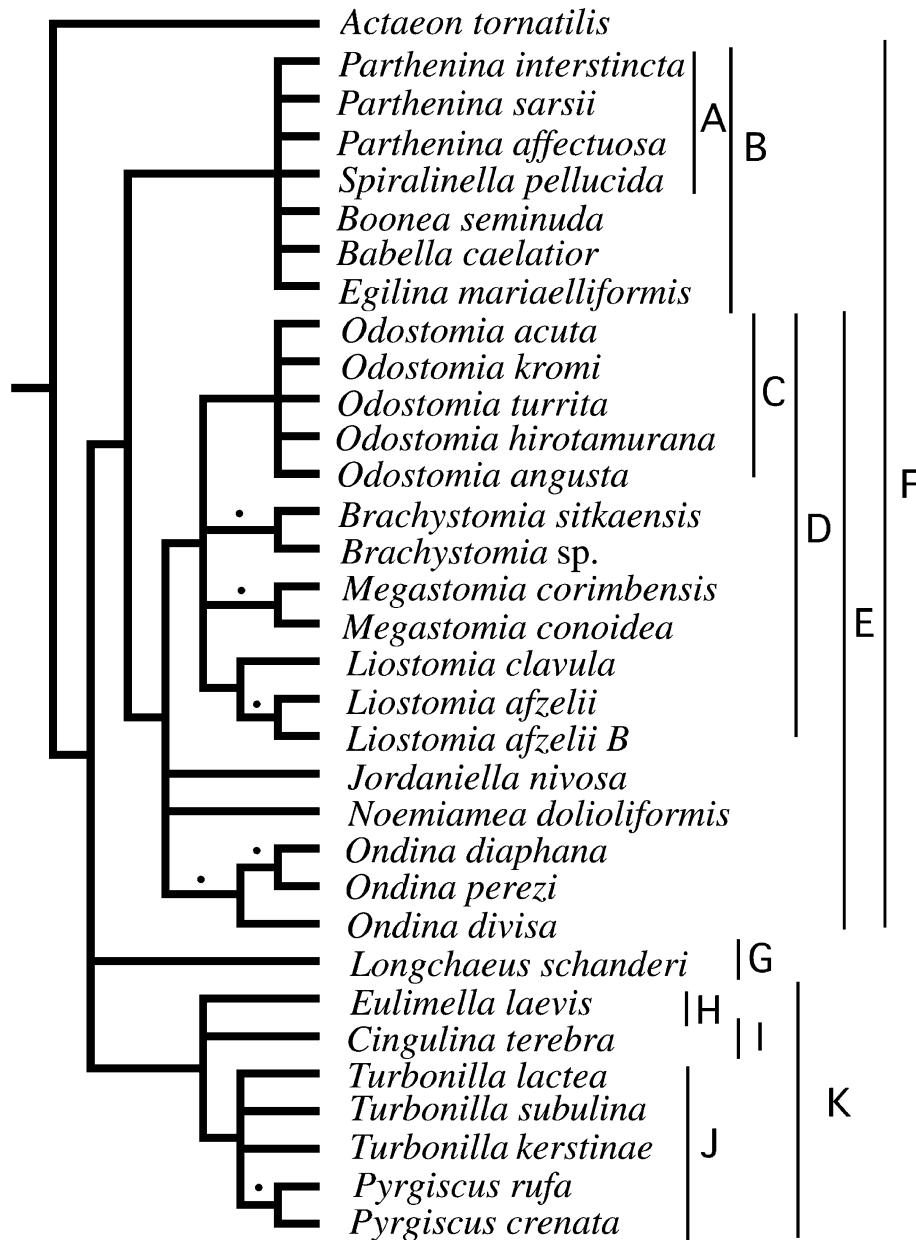


Fig. 5 Traditional arrangement of the included species. —A. *Chrysallida* Carpenter, 1856 *sensu lato*. —B. Chrysallidinae Saurin, 1958. —C. *Odostomia* Flemming, 1813 *sensu stricto*. —D. *Odostomia* Flemming, 1813 *s.l.* —E. Odostomiinae Pelseener, 1928. —F. Odostomiidae Pelseener, 1928. —G. Pyramidellinae Gray, 1840. H. Eulimellinae Saurin, 1958. —I. Cingulininae Saurin, 1959. —J. *Turbonilla* Risso, 1826 *s.l.* —K. Turbonillidae Bronn, 1849. Compiled from van Aartsen 1977, 1981, 1987, 1994, Peñas & Rolán (1997a,b, 1999), Schander *et al.* (1999a) and other sources. Branches not contradicted in any of the current analyses are indicated by dots.

References

- van Aartsen, J. J. (1977). European Pyramidellidae: I *Chrysallida*. *Conchiglie*, 14, 49–64.
- van Aartsen, J. J. (1981). European Pyramidellidae: II *Turbonilla*. *Bollettino Malacologico*, 17, 61–88.
- van Aartsen, J. J. (1987). European Pyramidellidae. III. *Odostomia* and *Ondina*. *Bollettino Malacologico*, 23, 1–34.
- van Aartsen, J. J. (1994). European Pyramidellidae: IV The Genera *Eulimella*, *Anisocycla*, *Syrnola*, *Cingulina*, *Oscilla* and *Careliopsis*. *Bollettino Malacologico*, 30, 85–110.
- Bandel, K. (1994). Triassic Euthyneura (Gastropoda) from St. Cassian Formation (Italian Alps) with a discussion on the evolution of the heterostropha. *Freiberger Forschungsheft*, 452, 79–100.

- Bartsch, P. (1955). The pyramidellid molluscs of the Pliocene deposits of North St. Petersburg, Florida. *Smithsonian Miscellaneous Collections*, 125, 1–102.
- Bieler, R. (1992). Gastropod phylogeny and systematics. *Annual Review of Ecology and Systematics*, 23, 311–338.
- Chaster, G. W. (1898). A report upon the Mollusca (excluding the Cephalopoda and Nudibranchia) obtained by the Royal Irish Academy Cruises of 1885, 1886, and 1888. *Proceedings of the Royal Irish Academy*, 5, 1–33.
- Dall, W. H. & Bartsch, P. (1904). Synopsis of the genera, subgenera and sections of the family pyramidellidae. *Proceedings of the Biological Society of Washington*, 17, 1–16.
- Dall, W. H. & Bartsch, P. (1909). A monograph of West American pyramidellid molluscs. *United States National Museum Bulletin*, 68, 1–257.
- Dockery, D. T. III (1993). The streptoneuran gastropods, exclusive of the Stenoglossa, of Northeastern Mississippi. *Bulletin of Mississippi Department of Environmental Quality Office of Geology (Jackson)*, 129, 1. 191.
- Graham, A. (1988). Molluscs: prosobranch and pyramidellid gastropods. *Synopses of the British Fauna*, 2, 2nd edn. Leiden: Brill/Backhuys.
- Halanych, K. M. (1998). Considerations for reconstructing metazoan history: signal, resolution, and hypothesis testing. *American Zoologist*, 38, 929–941.
- Hori, S. & Nakamura, Y. (1999). Two new species of the Pyramidellidae (Orthogastropoda: Heterobranchia) parasitic on bivalves in Yamaguchi prefecture. *Venus*, 58, 165–174.
- Hori, S. & Okutani, T. (1995). A new pyramidellid gastropod ectoparasitic on *Umbonium (Suchium) moniliferum* (Lamarck) from Tomioka Bay, Amakusa, Western Kyushu. *Venus*, 54, 247–258.
- Hori, S. & Okutani, T. (1996). A new pyramidellid Gastropod ectoparasitic on *Conus*. *Venus*, 55, 7–14.
- Kishino, H. & Hasegawa, M. (1989). Evaluation of the maximum likelihood estimate of the evolutionary tree topologies from DNA sequence data, and the branching order in Hominoidea. *Journal of Molecular Evolution*, 29, 170–179.
- Koufopanou, V., Reid, D. G., Ridgway, S. A. & Thomas, R. H. (1999). A molecular phylogeny of the patellid limpets Gastropoda: Patellidae) and its implications for the origins of their antitropical distribution. *Molecular Phylogenetics and Evolution*, 11, 138–156.
- Kuroda, T. & Habe, T. (1949). On the systematic position of Pyramidellidae. *Venus*, 15, 55–58.
- Lake, J. A. (1994). Reconstructing evolutionary trees from DNA and protein sequences: Paralinear distances. *Proceedings of the National Academy of Sciences, USA*, 91, 1455–1459.
- Laws, C. R. (1937a). Review of the tertiary and recent neozelandic pyramidellid molluscs, 1 — the genus *Turbonilla*. *Transactions of the Royal Society of New Zealand, Zoology*, 66, 402–422.
- Laws, C. R. (1937b). Review of the tertiary and recent neozelandic pyramidellid molluscs, 2 — the genus *Chemnitzia*. *Transactions of the Royal Society of New Zealand, Zoology*, 67, 47–70.
- Laws, C. R. (1937c). Review of the tertiary and recent neozelandic pyramidellid molluscs, 3 — further turbonillid genera. *Transactions of the Royal Society of New Zealand, Zoology*, 67, 166–184.
- Laws, C. R. (1937d). Review of the tertiary and recent neozelandic pyramidellid molluscs, 4 — the syrnelid genera. *Transactions of the Royal Society of New Zealand, Zoology*, 67, 303–315.
- Laws, C. R. (1938). Review of the tertiary and recent neozelandic pyramidellid molluscs, 5 — the eulimellid genera. *Transactions of the Royal Society of New Zealand, Zoology*, 68, 51–59.
- Laws, C. R. (1939a). Review of the tertiary and recent neozelandic pyramidellid molluscs, 6 — the genus *Odotomia*. *Transactions of the Royal Society of New Zealand, Zoology*, 69, 191–209.
- Laws, C. R. (1939b). Review of the tertiary and recent neozelandic pyramidellid molluscs, 7 — further odostomid genera. *Transactions of the Royal Society of New Zealand, Zoology*, 69, 191–209.
- Lieberman, B. S., Allmon, W. D. & Eldredge, N. (1993). Levels of selection and macroevolutionary patterns in the turritellid gastropods. *Paleobiology*, 19, 205–215.
- Lockhart, P. J., Steel, M. A., Hendy, M. D. & Penny, D. (1994). Recovering evolutionary trees under a more realistic model of sequence evolution. *Molecular Biology and Evolution*, 11, 605–612.
- Maddison, W. P. & Maddison, D. R. (1992). *MacClade. Analysis of Phylogeny and Character Evolution*. Sunderland, MA: Sinauer Associates, Inc.
- Medina, M. & Walsh, P. J. (2000). Molecular phylogeny of the order Anaspeidea based on mitochondrial DNA sequence (12S, 16S and COI). *Molecular Phylogenetics and Evolution*, 15, 41–58.
- Mikkelsen, P. M. (1996). The evolutionary relationships of cephalaspeidea S. L. (Gastropoda: Opisthobranchia): a phylogenetic analysis. *Malacologia*, 37, 375–442.
- Nordsieck, F. (1972). *Die Europäischen Meeresschnecken*, Vol. 3. Stuttgart: Gustav Fischer Verlag.
- Palumbi, S. R. (1996). Nucleic acids II: The polymerase chain reaction. In D. M. Hillis & C. Mortiz & B. K. Mable (Eds) *Molecular Systematics* (pp. 245–248). Sunderland, MA: Sinauer Associates.
- Peñas, A. & Rolán, E. (1997a). La familia Pyramidellidae Gray, 1840 (Mollusca, Gastropoda, Heterostropha) en África Occidental. 1. El género *Sayella* Dall, 1855. *Iberus*, 15, 35–40.
- Peñas, A. & Rolán, E. (1997b). La familia Pyramidellidae Gray, 1840 (Mollusca, Gastropoda, Heterostropha) en África Occidental. 2. Los géneros *Turbonilla* y *Eulimella*. *Iberus, Supplement*, 3, 1–105.
- Peñas, A. & Rolán, E. (1998). La familia Pyramidellidae Gray, 1840 (Mollusca, Gastropoda, Heterostropha) en África Occidental. 3. El género *Chrysallida* s. 1. *Iberus Supplement*, 4, 1–73.
- Peñas, A. & Rolán, E. (1999). La familia Pyramidellidae Gray, 1840 (Mollusca, Gastropoda, Heterostropha) en África Occidental. 4. Los géneros *Megastomia*, *Odotomia*, *Ondina*, *Noemiamea* y *Syrnola*. *Iberus, Supplement*, 5, 1–150.
- Peterson, B. J. (1998). The morphology, ultrastructure and function of the feeding apparatus of *Sayella fusca* (C. B. Adams, 1839 (Gastropoda: Pyramidellidae). *Journal of Molluscan Studies*, 64, 281–296.
- Ponder, W. F. & Lindberg, D. R. (1997). Towards a phylogeny of gastropod molluscs: an analysis using morphological characters. *Zoological Journal of the Linnean Society*, 119, 88–265.
- de Queiroz, K. & Gauthier, J. (1990). Phylogeny as a central principle in taxonomy: Phylogenetic definitions of taxon names. *Systematic Zoology*, 39, 307–322.
- Rosenberg, G., Tillier, S., Tillier, A., Kuncio, G. S., Hanlon, R. T., Masselot, M. & Williams, C. J. (1997). Ribosomal RNA phylogeny of selected major clades in the mollusca. *Journal of Molluscan Studies*, 63, 301–309.
- Saurin, E. (1958). Pyramidellidae de Pho-Hai (Sud Viet-Nam). *Annales de la Faculté Des Sciences de Saïgon*, 35, 63–86.
- Saurin, E. (1959). Pyramidellidae de Nha-Trang (Viet-Nam). *Annales de la Faculté Des Sciences de Saïgon*, 40, 223–283.
- Saurin, E. (1961). Pyramidellidae du Golfe de Thaïlande. *Annales de la Faculté Des Sciences de Saïgon*, 1961, 231–266.

- Schander, C. (1997). *Taxonomy and phylogeny of the Pyramidellidae (Mollusca, Gastropoda, Heterobranchia)*. PhD-thesis Gothenburg University.
- Schander, C. (1998a). Mandatory categories and impossible hierarchies — a reply to Sosef. *Taxon*, 47, 407–410.
- Schander, C. (1998b). Types, emendations and names — a reply to Lidén et al. *Taxon*, 47, 401–406.
- Schander, C., Hori, S. & Lundberg, J. (1999b). Anatomy and phylogeny of *Odostomella* and *Herviera* (Mollusca, Heterogastropoda, Pyramidellidae), with a description of a new species of *Odostomella*. *Ophelia*, 51, 39–76.
- Schander, C. & Sundberg, P. (2001). Shell characters in gastropod phylogeny — soft information or hard facts? *Systematic Biology*, 50, 9–13.
- Schander, C. & Thollessen, M. (1995). Phylogenetic Taxonomy — some comments. *Zoologica Scripta*, 24, 263–268.
- Schander, C. & van Aartsen, J. J. & Corgan, J. X. (1999a). Families and genera of the Pyramidelloidea (Mollusca: Gastropoda). *Bollettino Malacologico*, 34, 9–12.
- Schröder, M. (1995). Fröontogenetische Schalen jurassischer und unterkretazischer Gastropoden aus Norddeutschland und Polen. *Paläontographica*, 238, 1–95.
- Shimodaira, H. & Hasegawa, M. (1999). Multiple comparisons of Log-likelihoods with applications to phylogenetic inference. *Molecular Biology and Evolution*, 16, 1114–1116.
- Swofford, D. L. (2000). *PAUP* 4.0: Phylogenetic Analysis Using Parsimony (*and Other Methods)*. Sunderland, MA: Sinauer Associates.
- Swofford, D. L., Olsen, G. J., Waddell, P. J. & Hillis, D. M. (1996). Phylogenetic inference reaction. In D. M. Hillis & C. Mortiz & B. K. Mable (Eds) *Molecular Systematics* (pp. 407–514). Sunderland, MA: Sinauer Associates.
- Thollessen, M. (1999). Phylogenetic analysis of Euthyneura (Gastropoda) by means of the 16S rRNA gene: use of a 'fast' gene for 'higher-level' phylogenies. *Proceedings of the Royal Society of London Series B*, 266, 75–83.
- Thompson, J. D., Higgins, D. G. & Gibson, T. J. (1994). CLUSTAL W: improving the sensitivity of progressive multiple sequence alignment through sequence weighting, position specific gap penalties and weight matrix choice. *Nucleic Acids Research*, 22, 4673–4680.
- Tillier, S., Masselot, M., Philippe, H. & Tillier, A. (1992). Phylogenie moleculaire des Gastropoda (Mollusca) fondée sur le séquençage partiel de l'ARN ribosome 28 S. *Comptes Rendus de l'Académie Des Sciences Paris*, 314, 79–85.
- Warén, A. (1991). New and little known Mollusca from Iceland. *Sarsia*, 76, 1–28.
- Wise, J. B. (1993). Anatomy and functional morphology of the feeding structures of the ectoparasitic gastropod *Boonea impressa* (Pyramidellidae). *Malacologia*, 35, 119–134.
- Wise, J. B. (1996). Morphology and phylogenetic relationships of certain pyramidellid taxa (Heterobranchia). *Malacologia*, 37, 443–511.
- Wise, J. B. (2000). Anatomy and systematic placement of *Sayella laevigata* (Gastropoda: Pyramidellidae: Sayellinae). *Journal of Molluscan Studies*, 66, 119–124.