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MORPHOLOGICAL CHARACTERS ANALYSIS, THE INTERGROUP PHYLOGENETIC RELATIONSHIPS AND POSSIBLE OUTGROUPS OF THE FAMILY SPHAERIIDAE (MOLLUSCA, BIVALVIA)

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Morphological Characters Analysis, the Intergroup Phylogenetic Relationships and Possible Outgroups of the Family Sphaeriidae (Mollusca, Bivalvia). Korniushin A. V. — Scenaria of the morphological characters transformations in freshwater molluscs traditionally included in the family Sphaeriidae are studied; they are based on the phylogenetic analysis of 69 characters in 57 taxa. It is shown, that the whole group is distinguished by synapomorphies in mantle musculature, as well as by presence of complicated nephridia with many apomorphic features. At the same time, many organs and structures (hinge, ligament, siphons, gills and stomach) demostrate signs of reduction. Two traditionally recognized subfamilies (Euperinae and Sphaeriinae), sometimes treated as families, differ in their reproductive strategies (ovoviviparity and viviparity); monophyletic status of the groups characterized by sequencial and synchronous brooding (*Sphaerium* s. l. and *Pisidium* s. l.) is also very probable. Some terminal clades correspond to the earlier suggested genera and subgenera, and relationships between them are briefly discussed. Apomorphies of mantle musculature characterizing Sphaeriidae were found also in *Polymesoda* (*Geloina*) — a taxon traditionally placed in Corbiculidae (one of the outgroups), suggested as a possible ancestor of the studied freshwater bivalves.

Key words: Bivalvia, Sphaeriidae, Corbiculidae, *Geloina,* morphological characters, phylogenetic analysis.

Анализ морфологических признаков, внутригрупповые филогенетические отношения и возможные внешние группы семейства Sphaeriidae (Mollusca, Bivalvia). Корнюшин А. В. — Рассмотрены сценарии эволюционных преобразований морфологических признаков пресноводных моллюсков, традиционно включаемых в семейство Sphaeriidae, основанные на результатах филогенетического анализа 69 признаков в 57 таксонах. Показано, что группа в целом характеризуется синапоморфиями в строении мантийной мускулатуры, а также наличием сложно устроенных нефридиев. В то же время многие органы и структуры (замок, лигамент, сифоны, жабры и желудок) демонстрируют черты редукции. Два традиционно выделяемых подсемейства (Euperinae и Sphaeriinae), иногда рассматриваемых в ранге семейств, характеризуются различными репродуктивными стратегиями (яйцеживорождение и живорождение); монофилия групп, характеризующихся последовательной или одновременной инкубацией молоди (*Sphaerium* s. l. и *Pisidium* s. l.), также весьма вероятна. Ряд терминальных клад соответствуют ранее предложенным родам и подродам, кратко обсуждаются их филогенетические отношения. Апоморфные черты мантийной мускулатуры, характеризующие Sphaeriidae, выявлены также у *Polymesoda* (*Geloina*) — таксона, традиционно относимого к семейству Corbiculidae (одна из внешних групп), который, таким образом, может быть предком анализируемой группы.

Ключевые слова: Bivalvia, Sphaeriidae, Corbiculidae, *Geloina,* морфологические признаки, филогенетический анализ.

Introduction

The family Sphaeriidae is one of the major freshwater bivalve groups represented on all continents (Cox et al., 1969; Kuiper, 1983). Its total species diversity cannot be defined at this state of knowledge, because of descrepancies between taxonomic approaches accepted in different countries (see Korniushin, 1998a for review), ongoing revisions (Ituarte, 1996, 1999, 2000) and absence of modern reviews for many regions (e. g. Southeast Asia, New Guinea, Central and tropical South America, etc). However, according to our preliminary estimations based on the species-level taxonomy accepted by the majority of specialists worldwide, the total number of valid species is about 150 (tabl. 1). Most of the known sphaeriids are strictly freshwater and only several species tolerate slightly brackish water conditions in estuaries (Kuiper, Wolf, 1970).

Table 1. Species diversity of Sphaeriidae in different regions

Таблица 1. Видовое разнообразие сфериид в различных регионах

Region	Number of species/endemics	Sources
Europe and Palearctic Asia	39/31	Korniushin, 1999, 2001
North America (native)	33/23	Burch, 1975
Africa (South of Sachara) and Madagascar	30/30	Kuiper, 1966 a; Mandahl-Barth, 1988, Korniushin, 1995
India and Nepal	11/10	Subba Rao, 1989; Nesemann et al., 2001
SE Asia and New Guinea	19/18	Odhner, 1940; Kuiper, 1983
Australia	16/15	Kuiper, 1983; Korniushin, 2000
New Zealand	3/3	Kuiper, 1966 b; Korniushin, Glushchenko, 1999
South America (Euperinae)	29/28	Mansur, Meier-Brook, 2000
South America (Sphaeriinae)	20/19	Kuiper, Hinz, 1984; Ituarte, 1996, 1999

Because of the worldwide distribution, considerable diversity and deep specialization to the freshwater environment, the group is rather interesting for evolutionary studies. However its origin, relationships with other bivalve families and internal phylogenetic relationships are still poorly understood.

Most of the modern reviewers (Burch, 1975; Kuiper, 1983; Mansur, Meier-Brook, 2000; Cooley, Ó Foighil, 2000 et al.) arrange Sphaeriidae in five genera: *Eupera*, *Byssanodonta*, *Sphaerium, Musculium* and *Pisidium*, but disagree about the number and names of subgenera. Some authors of the former USSR (Alimov, Starobogatov, 1968; Pirogov, Starobogatov, 1974; Stadnichenko, 1984; Starobogatov, Korniushin, 1986; Korniushin, 1996 a) divided *Sphaerium* and *Pisidium* into several genera. Accordingly, Sphaeriidae in its traditional understanding was divided into four families (Korniushin, 1992, 1996 a). However, such a splitting was criticized by the West European reviewers (Meier-Brook, 1993). Later on, Korniushin (1999, 2001) suggested a compromizing approach recognizing only those new taxa, which were supported by sets of reliable anatomical characters.

There are also discrepancies between phylogenetic reconstructions based on different sets of characters. While the recent morphological studies confirmed monophyly of *Pisidium* s. l. (Korniushin, 1998 b) and suggested its sister relationship to *Musculium* (Mansur, Meier-Brook, 2000), molecular works (Park, Ó Foighil 2000; Cooley, Ó Foighil, 2000) showed more close affinity between *Musculium* and *Sphaerium,* and paraphyly of *Pisidium*. Traditionally, Sphaeriidae are considered closely related to the fresh- and brackish-water family Corbiculidae, but Starobogatov (1992) suggested its direct origin from the primitive marine Astartidae. Neither of these hypotheses was confirmed by molecular study (Park, Ó Foighil, 2000) which showed Sphaeriidae to be an independent lineage without close relationship to any other studied marine or brackish water group. Noteworthy, some of the rather diverse generic/ subgeneric taxa currently included in Corbiculidae (Cox et al., 1969) were not included in any molecular or morphological phylogenetic study, thus polyphyletic status of the latter family and affinity of some its subgroupings to Sphaeriidae cannot be excluded.

Discrepancy between the phylogenetic reconstructions based on morphological and molecular characters can be at least partly explained by the restricted number of characters and taxa available for a phylogenetic analysis. In order to enlarge the morphological data set, Korniushin and Glaubrecht (in press) carried out an extensive search for the phylogenetically informative anatomical characters based on the data from published descriptions and original observations. As a result, the matrix including 69 characters and 57 taxa (54 species of Sphaeriidae, two corbiculids and a venerid) was compiled. This data set was tested then by PAUP (Swofford, 1998) under different assumptions. It appeared, that the consensus trees obtained without any constraints and by enforcement monophyly of the *Sphaerium* + *Musculium* (= *Sphaerium* s. l.) clade, as suggested by molecular works (Park, Ó Foighil, 2000; Cooley, Ó Foighil, 2000), differed in one step only; some taxonomic and biogeographic implications derived from the both analyses (with and without constraints) were discussed (Korniushin, Glubrecht, in press).

The aim of this paper is studying scenaria of the morphological characters transformations in Sphaeriidae derived from the above mentioned analyses and comparing them to the existing classifications. We focused on the key features of the groups well supported by the parsimony analysis (the whole family, its two basic partitions corresponding to the subfamilies Euperinae and Sphaeriinae, and the genus *Pisidium* in its traditional understanding), but transformations supporting some smaller terminal clades, viz. *Amesoda*, *Sphaerinova*, *Neopisidium* and some other are also discussed. We checked also some more outgroups for presence of advanced states characterizing Sphaeriidae, and a relationship between this family and *Polymesoda* (*Geloina*) — a taxon traditionally placed in Corbiculidae, is hypothesized.

Material and methods

This work is based on the morphological data matrix compiled by Korniushin & Glaubrecht (in press); the list of characters and states is provided in the Attachment 1. Additional material on Corbiculidae obtained from the Museum fьr Naturkunde (Berlin) included *Polymesoda* (*Geloina*) *erosa* (Solander, 1786) (ZMB 49532, Indian Ocean, no locality given, leg. M. Weber, undated, original identification — *Cyrena* *suborbicularis*) and *Batissa violacea* (Lamarck, 1797) (ZMB 103031, Angkona River, Sulawesi, Indonesia, leg. M. Glaubrecht, T. van Rhinthelen and F. Koehler 1999).

Here we use traditional nomenclature of hinge teeth (Cox et al., 1969); anatomical therminology is based on our previous works (Korniushin, 2000, 2001).

As already specified, there is no generally accepted system of Sphaeriidae. The taxa included in this phylogenetic analysis (see Attachment 2) were preliminarily arranged according to the latest versions of our system (Korniushin, 1999–2001), with some additions and modifications commented below. The genus *Amesoda* was restored by Alimov and Starobogatov (1968); Korniushin (2001) conservatively treated is as a subgenus of *Sphaerium* restricted to North America, but tentatively suggested close relationship of the European species *Sphaerium rivicola* to this group; here we follow a compromizing view, recognizing *Amesoda* as a genus with two subgenera — North American *Amesoda* s. str. and European *Rivicoliana*. Subgenera of *Sphaerium* in its present (strict) understanding (Alimov, Starobogatov, 1968; Falkner et al., 2001) are not observed here. *Herringtonium* is traditionally included in *Sphaerium* as a monotypic subgenus (Burch, 1975), but Heard (1977) showed its intermediate position between *Sphaerium* and *Musculium*, while Starobogatov and Korniushin (1986) treated is as a separate genus. *Sphaerinova* and *Paramusculium* are tentatively assigned in this work to *Musculium*, as suggested by Korniushin (1998 c, 2000), but their generic rank may be also questioned (Korniushin, 1996). The status of *Afromusculium* is defined according to Korniushin (1998 c).

The group comprizing all larger sphaeriids included by earlier authors into the genus *Sphaerium* (e. g. *Amesoda*, *Sphaerium*, *Herringtonium* and *Musculium* in the present understanding) is defined below as *Sphaerium* s. l., while the traditional genus *Pisidium* — as *Pisidium* s. l. Present division of the latter group into four genera is based on suggestions of Korniushin (1998 a, 1999). Taxonomic status of *Euglesa* was recently fixed by selecting a neotype for its type species *Euglesa henslowiana* Jenyns, 1832, the taxon is treated in some works as a subgenus of *Pisidium* (Korniushin, 2000; Falkner et al., 2001) and corresponds to the subgenus *Cyclocalyx* in the sense of modern North American reviewers (Burch, 1975). Most of the subgeneric groups assigned here to the genus *Euglesa* are treated as separate subgenera of *Pisidium* by Falkner et al. (2001). The genus *Neopisidium* in the broad understanding suggested by Korniushin (1999) cannot be accepted because of confusion about the type species (G. Falkner, pers. comm.), terefore the name is restricted here to "*Pisidium conventus*" species group (=*Conventus* auct.); all three groups of the so called "neotenic pisidia" (e. g. distinguished by reductions in many organs) are treated as separate genera, while the taxon traditionally defined as *Pisidium moitessierianum* is tentatively included in *Odhneripisidium*. Several species included until now in *Pisidium* (Burch, 1975; Korniushin, 1998 d, 2000; Nesemann et al., 2001), are assigned here to *Euglesa* on the base of their anatomical characters, but their subgeneric belonging is not defined. The status of *Eupera* and *Byssanodonta* is defined according to Mansur and Meier-Brook (2000).

Maximum parsimony trees were obtained by PAUP* 4.0b4a (Swofford, 1998) on Macintosh Performa using the procedure of heuristic search with stepwise addition and tree-bisection-reconnection (TBR) algorithms. A venerid and two representatives of Corbiculidae were treated as outgroups. All characters were unordered and had equal weight; the delayed transformation option (DELTRAN) giving preference to parallelisms over reversions (Swofford, 1998) was used. The multi-state taxa were treated as polymorphic (option "mstaxa=polymorph"). Searching under topological constraints derived from the molecular studies (Park, Ó Foighil, 2000; Cooley, Ó Foighil, 2000) was also performed.

Support values were obtained by a bootstrap analysis with 500 replicates (Swofford, 1998). The further study was focused on the clades with bootstrap support more than 50%. Noteworthy, some of these clades did not appear on the strict consensus trees. Reconstruction of character transformations given below is based on the apomorphy lists obtained in PAUP* (Swofford, 1998).

Abbreviations, used in the figures: af — anterior fold of stomach; as — anal (exhalant) siphon; bo — branchial opening; bs — branchial (inhalant) siphon; c1, c3 — inner and outer cardinal teeth of right valve; c2, c4 — inner and outer cardinal teeth of left valve; co — coecum; id — inner demibranch; irm — inner radial mantle muscles; l — ligament; mg — midgut; od — outer demibranch; odd — descending lamella of outer demibranch; oe — oesophagus; ps — pedal slit; sr — integrated siphonal muscles; sr1 — retractors of anal siphon; sr2, sr3 — upper and lower retractors of branchial siphon; t1 — major typhlosole; t2 — minor typhlosole.

Results

Character transformations

The most probable phylogenetic relationships on generic and subgeneric level derived from the results of the phylogenetic analysis carried out by Korniushin, Glaubrecht (in press) are provided in the figs 1, 2; synapomorphies supporting the most disputable clades are listed in the table 2. Below we observe the trends in evolution of different organs and structures, as suggested by these results.

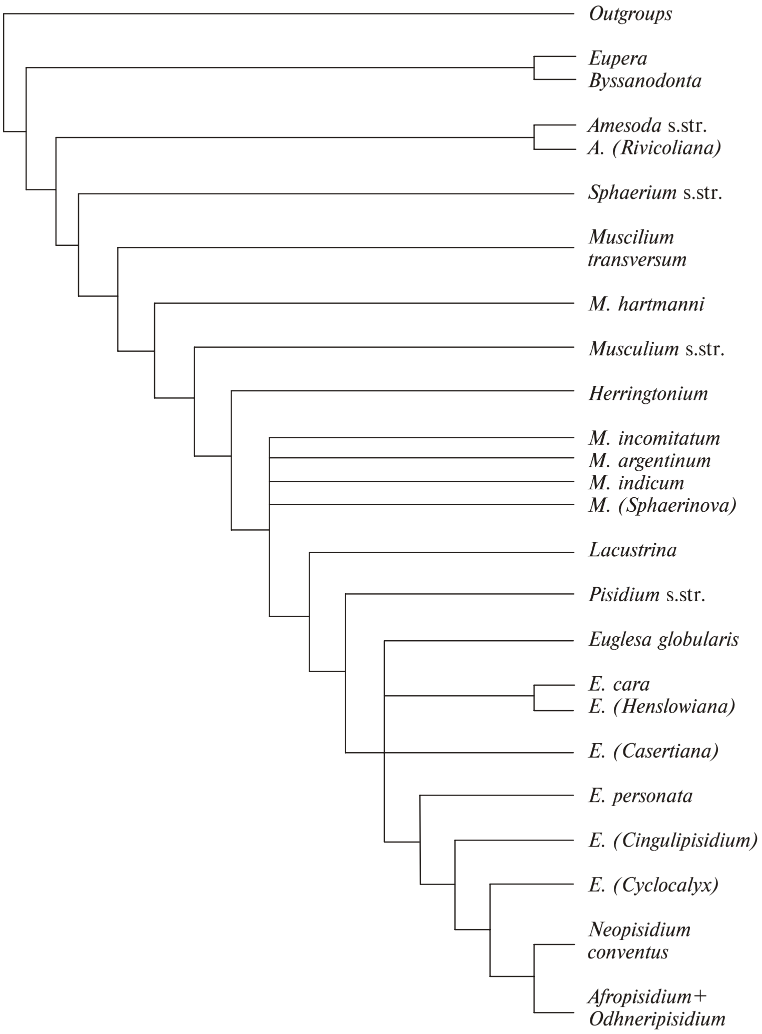

Fig. 1. Phylogenetic relationships within Sphaeriidae on generic and subgeneric levels; cladogram based on the majority rule consensus of 752 mpt (length 154) obtained in the phylogenetic analysis without constraints (Korniushin, Glaubrecht, in press).

Рис. 1. Филогенетические отношения в семействе Sphaeriidae на уровне родов и подродов; кладограмма основана на консенсусе (правило большинства) 752 наиболее экономных деревьев (длина 154), полученных в филогенетическом анализе без ограничений (Korniushin, Glaubrecht, in press).

Shell form. While both observed consensus trees (fig. 1, 2) showed that Euperinae retained the ancestral (anterior) position of umbo, the direction of evolution in the other subfamily remained unclear. In the tree obtained without constraints (fig. 1), the central position of umbo was supposed to be initial state in Sphaeriinae, while its posterior shift — a synapomorphy for *Pisidium* s. l. and the most advanced *Musculium* species (*Sphaerinova, Afromusculium* and several *Musculium* species with uncertain placement). However, inforcing monophyly of the *Sphaerium* + *Musculium* clade made the mentioned subgrouping of *Musculium* also monophyletic, by this condition it occupied basal position in *Sphaerium* s. l. ckade.

Sculpture. Only the well recognizable patterns of sculpture were included in the data matrix; both analyses showed that the presence of prominent periostracum folds was a synapomorphy for Euperinae, while peculiar periumbonal striae distinguished the group bearing the name *Cingulipisidium.*

Hinge teeth (fig. 3). Results of our study show, that the common ancestor of sphaeriids had a hinge without the tooth c1, with a single bent or dome-like c2, single c3 (occasionally divided in c3a and c3b in *Amesoda rivicola*) and c4. This condition could derive from the typical veneroid hinge (c1, c3a and c3b in the right valve; c2a, c2b and c4 in the left one) by means of reduction. Hinge apparently suffered a further reduction — the lost of c4 — in Euperinae. The straight form of c2 (as in *S. corneum, M. lacustre* and *E. subtruncata*) is apparently an advanced state, which evolved independently in several clades within *Sphaerium*, *Musculium* and *Euglesa*.

Ligament. The outgroups selected for this analysis, as well as the majority of heterodont bivalves, are characterized by a parivincular ligament, which is supported by a peculiar projection of hinge plate — nympha. This condition is also shared by

Table 2. Synapomorphies, supporting the most important clades distinguished by the phylogenetic analysis in Sphaeriidae

Таблица 2. Синапоморфии, поддерживающие важнейшие клады, выделенные в ходе филогенетического анализа семейства Sphaeriidae

Clade	Synapomorphies
Sphaeriidae	**5.1**; 6.1; **7.1**; **17.1**; 20.1**; **29.1**; 37.1*; 41.1; **42.1**; **47.1**; **49.1**; **51.1**; **57.1**
Euperinae (Eupera and Byssanodonta)	**8.1**; **11.1**; **15.1**; **16.1**; 20.1*; **21.1**; 23.1; 41.1**; 44.1; **66.1**
Sphaeriinae (*Sphaerium s.* l and *Pisidium* s. l.)	1.1; 4.1**; 10.1**; 20.2*; **21.2**; 24.1; **34.1**; 35.2*; **36.1**; 38.1**; 39.1; **40.1**; 45.1**; 48.1**; **52.1**; 54.1*; **55.1**; **59.1**; **60.2**; 61.1*; **62.1**; **67.1**; 68.1*
Musculium+Herringtonium+ Pisidium s. l.	48.1*; **69.0***
Sphaerium s. l.**	35.2**; 61.1**; 68.1**
*Sphaerium+Amesoda***	48.0**; 69.1**
Amesoda	4.0 (reversion)**; 28.0**; 38.0 (reversion)**; 45.0 (reversion)**; 56.1; 65.0**
Sphaerium s. str.	41.0; 58.0
Musculium s. str.: *M. lacustre* and *M. securis*	2.1; 6.2
Sphaerinova s. l._**: *M. incomitatum, M. argentinum, M. indicum, M. tasmanicum* and *M. novaezelandiae*	**19.1****
Pisidium s. l.	**14.1**; **18.1**; 32.1; 35.1**; **46.1**; **50.1**; 61.0 (reversion)*; 63.1; 68.0 (reversion)*
Pisidium s. str.: *P. amnicum* and *P. dubium*	35.1*; 63.2; **64.1**
***Euglesa* (*Henslowiana*)**: *E. lilljeborgi, E. henslowana* and *E. supina*	35.1; 54.1
Henslowiana + E. cara	26.1
E. (*Casertiana*): *E. casertana, E. compresa, E. keniana, E. viridaria, E. etheridgei, E. atkinsoniana*	32.2
E. (*Cingulipisidium*): *E. hibernica, E. langleyana, E. nitida, E. pseudosphaerium*	12.1 (secondarily lost in *E. pseudosphaerium*); 26.1
E. (*Cyclocalyx*): *E. obtusalis, E. ovampicum, E. milium, E. pulchella* and *E. subtruncata*	27.1
E. pulchella and *E. subtruncata* (***Pseudeupera***)	**27.2**
„Neotenic“ pisidia (*Neopisidium, Odhneripisidium* and *Afropisidium*)	**13.1**; **31.1**
“*Neotenic pisidia*” except *Neopisidium conventus*	6.1; **22.1**; 56.2; 63.2; **64.2** (traditional subgenus *Odhneripisidium* supported by **3.1**)

Notes. * — shown only in the trees obtained without constraints; ** — shown only in the trees obtained under condition of monophyly of *Sphaerium* s. l. clade; clades with bootstrap support more than 50% and unambiguous synapomorphies (in bold); characters and states numbers correspond to those in the Attachment 1.

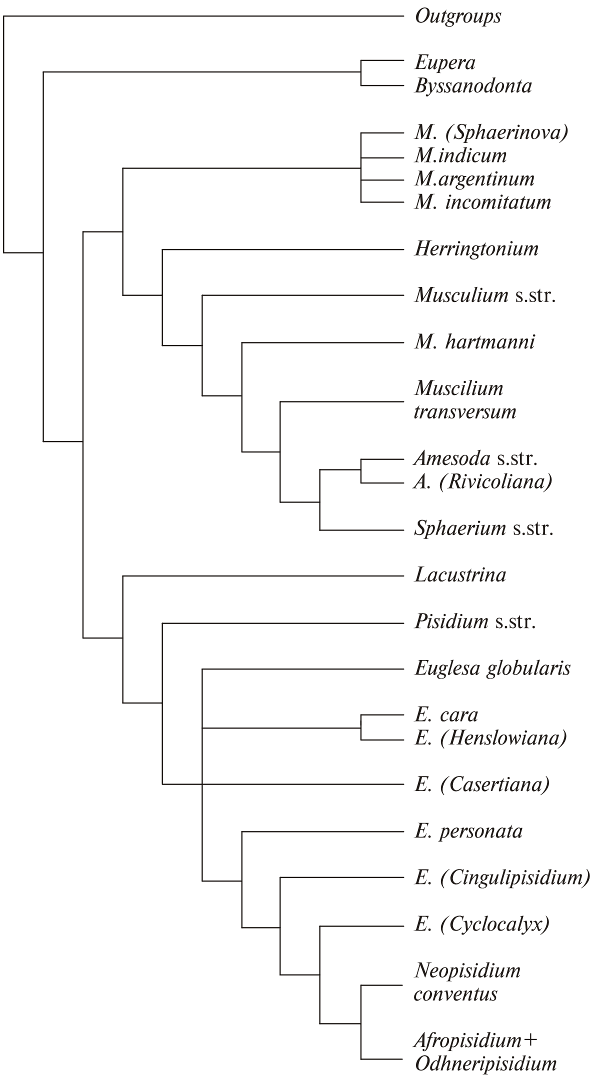

Fig. 2. Phylogenetic relationships within Sphaeriidae on generic and subgeneric levels; cladogram based on the majority rule consensus of 126 mpt (length 155) obtained in the phylogenetic analysis with enforced monophyly of the clade characterized by sequencial brooding (*Sphaerium* s. l.) (Korniushin, Glaubrecht, in press).

Рис. 2. Филогенетические отношения в семействе Sphaeriidae на уровне родов и подродов; кладограмма основана на консенсусе (правило большинства) 126 наиболее экономных деревьев (длина 155), полученных в филогенетическом анализе при условии монофилии клады, характеризующейся последовательной инкубацией молоди (*Sphaerium* s. l.) (Korniushin, Glaubrecht, in press).

Euperinae and some taxa of Spheriinae, viz. *Amesoda* and *M. hartmanni* (fig. 4, *A*, *B*), but it is still not clear, whether the mentioned groups inherited this state from the common ancestor of the subfamily, or it was first lost and then restored (as suggests the analysis under constraints). In the majority of sphaeriids the nympha was reduced, and a highly modified (introverted) ligament was developed in *Odhneripisidium* (fig. 4, *C*).

Mantle edge and its musculature. While the outgroups have integrated siphonal muscles (fig. 5, *A*), sphaeriids are distinguished in having separate retractors of exhalant and inhalant siphons (fig. 5, *B–E*). Both analyses show this feature as a synapomorphy of the family. Two subfamilies are characterized by different arrangements of lower muscles of the inhalant siphon: in Euperinae they are organized in several bundles, while in Sphaeriinae — in a single pair of retractors. Such characters as sepa-

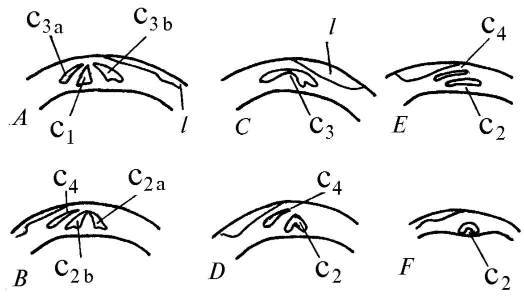

Fig. 3. Arrangement of cardinal teeth (diagrammatic): *A* — Veneridae and Corbiculidae, right valve; *B* — Veneridae and Corbiculidae, left valve; *C* — typical hinge of Sphaeriinae, right valve; *D* — typical hinge of Sphaeriinae, left valve; *E* — Sphaeriinae with straight cardinal teeth (left valve); *F* — Euperinae (left valve).

Рис. 3. Расположение кардинальных зубов (схематизировано): *A* — Veneridae и Corbiculidae, правая створка; *B* — Veneridae и Corbiculidae, левая створка; *C* — типичный замок Sphaeriinae, правая створка; *D* — типичный замок Sphaeriinae, левая створка; *E* — Sphaeriinae с прямыми кардинальными зубами (левая створка); *F* — Euperinae (левая створка).

ration of siphons and peculiar mode of their contraction — with apical parts turning inside (Mansur, Meier-Brook, 2000) are also interpreted here as synapomorphies of Euperinae. Upper retractors of the inhalant siphon suffer some reduction in *Sphaerinova* (tabl. 2). Their complete reduction in *Pisidium* s. l. might be a consequence of the inhalant siphon reduction (species of this group may have only a simple branchial opening). Noteworthy, lower retractors of the inhalant siphon or their rudiments are distinguishable also in the species, wich have no branchial opening (fig. 5, *E*).

Sphaeriids are also characterized by relatively short outer radial mantle muscles. Furthermore, arrangement of the inner radial muscles in bundles is shown to be a synapomorphy of Sphaeriinae (fig. 6). Orientation of these bundles is an important character in *Euglesa*, since its advanced states support the clade *Cyclocalyx* including at least five species (tabl. 2).

Elongation of the mantle fusion is shown to be an advanced feature which developed independently in Euperinae and several clades within *Pisidum* s. l.

Gills. The main evolutionary trend in the studied group is the gradual reduction of the outer demibranch, indicated by its reduced size, shift in posterior direction, transition from two-lamellar to one-lamellar structure and delay in the ontogenetic development. According to our analyses, one-lamellar outer demibranch is an apomorphy supporting the clade which includes all genera of *Pisidium* s. l. group except *Lacustrina*, and complete reduction of the outer demibranch took place once, defining a clade of “neotenic pisidia” (*Neopisidium, Odhneripisidium* and *Afropisidium* in the present understanding). In comparison with the outgroups, Sphaeriidae are characterized by relatively short ascending lamella of the inner demibranch.

Concerning development of the interlamellar septae, results of our analyses are controversial. Depending on the applied constraints, development of septae on each filament is shown as an initial state for all Sphaeriinae, or only for the *Sphaerium* s. l. clade. Consequently, peculiar arrangement of septae (on each second filament) characterizing some species of *Pisidium* s. l. can be iterpreted as an apomorphy supporting some its subgroupings (in the former case) or an ancestral state for the whole group.

Alimentary system. In comparison with the outgroups, labial palps of sphaeriids are characterized by somewhat reduced ridged area. Sphaeriinae are distinguished by the angulate anterior edge of these palps (in the outgroups and Euperinae it is straight).

Concluding from the earlier ivestigations (Mansur, Meier-Brook, 2000) and results of this study, we assume that sphaeriid stomach is a reduced venerid one. The

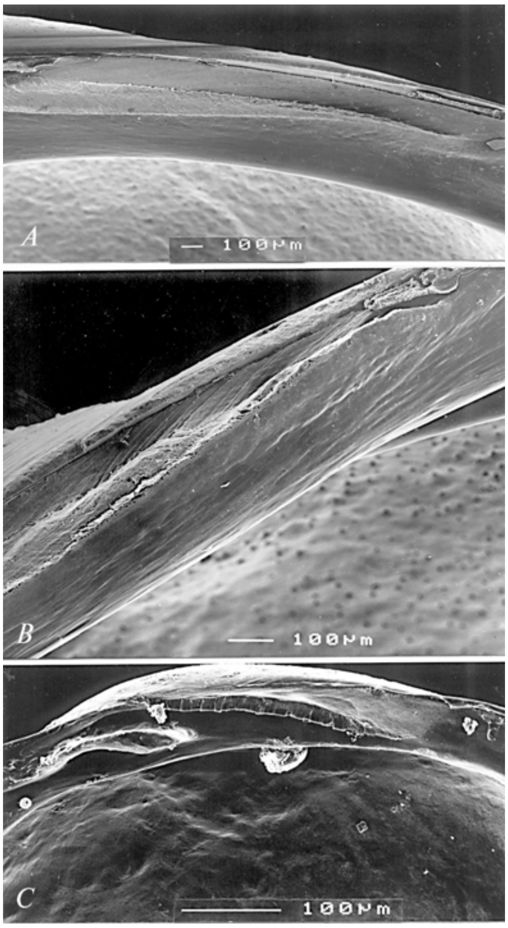

Fig. 4. Ligament: *A* — *Amesoda rivicola* (ligament with nympha); *B* — *Musculium? hartmanni* (ligament with nympha); *C* — *Odhneripisidium tenuilineatum* (introverted ligament).

Рис. 4. Лигамент: *A* — *Amesoda rivicola* (лигамент с нимфой); *B* — *Musculium? hartmanni* (лигамент с нимфой); *C* — *Odhneripisidium tenuilineatum* (интровертированный лигамент).

most remarkable advanced feature is absence of caeca containing winds of the major typhlosole, which are well seen in the outgroups. Two loops of the major typhlosole seen in larger sphaeriids can be treated as the rudiments of these caeca (fig. 7). Sphaeriinae are distinguished by the posterior extension of the stomach (its length exceeds height).

Distribution of the other stomach characters (stomach separation, elevation of anterior fold, and course of the minor typhlosole) is controversial: each scenario include cases of reversion from advanced to primitive character states (tabl. 1). It concerns especially the group *Amesoda*, which is most similar in the stomach characters to Euperinae and the outgroups; these similarities can be interpreted either as symplesiomorphies or reversions.

Simplification of the midgut coil is shown in all our analyses as a synapomorphy for *Pisidium* s. l.

Nephridia. In contrast to the organs observed above, sphaeriid nephridia do not shw any trend for reduction. Moreover, they demonstrate progressive development within the family. Presence of such structures as long funnel, pericardial tube, dorsal lobe and excretory sac are apparently synapomorphies for the family. These complications may intensify osmoregulatory function which is very important in the freshwater environment.

Anterior extension of the excretory sac is shown here as a synapomorphy of *Sphaerium* and narrow funnel — as a tentative synapomorphy for the *Musculium* + *Herringtonium* + *Pisidium* s. l. clade (in the analysis without constraints). Transformations of the type of nephridium (open/closed) apparently occured many times within the group. In all probability, the common ancestor of *Pisidium* s. l. had closed nephridia — with pericardial portion not visible between the branches of dorsal lobe (tabl. 2), but the ancestral state in the subfamily Sphaeriinae cannot be defined with certainty.

Reproductive system. All sphaeriids are brooders, but Euperinae produce large eggs with much yolk developing directly between gill lamellae (ovoviviparity), while Sphaeriinae develop small eggs nourished from the mother in brood pouches formed by inner demibranch filaments (transition to the true viviparity). These two modes evidently demonstrate different reproductive strategies of the two major sphaeriid subgroupings, which may be identified as ovoviviparity and transition to the true viviparity.

Furthermore, the latter subfamily is characterized by a small gonad placed at the base of the foot and not extended dorsally.

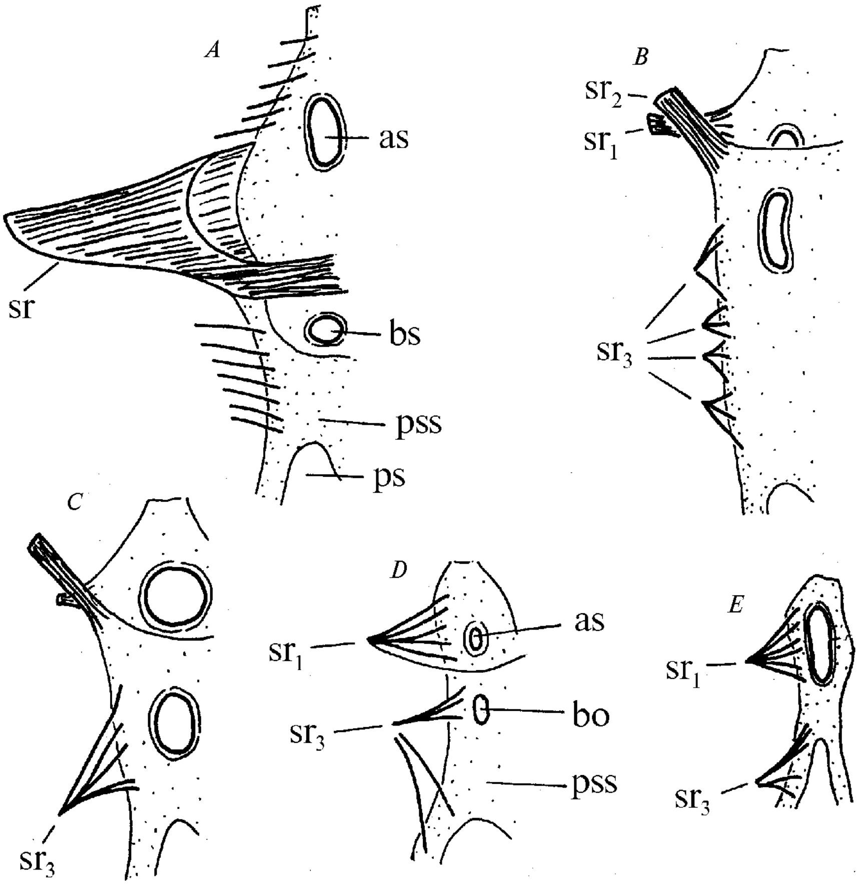

Fig. 5. Siphons and siphonal musculature (diagrammatic): *A — Neocorbicula*; *B — Eupera*; *C — Sphaerium*, *Musculium* and *Herringtonium*; *D — Lacustrina*, *Pisidium* s. str. and *Euglesa*; *E — Odhneripisidium* and *Afropisidium*.

Рис. 5. Сифоны и сифональная мускулатура (схематизировано): *A — Neocorbicula*; *B — Eupera*; *C — Sphaerium*, *Musculium* и *Herringtonium*; *D — Lacustrina*, *Pisidium* s. str. и *Euglesa*; *E — Odhneripisidium* и *Afropisidium*.

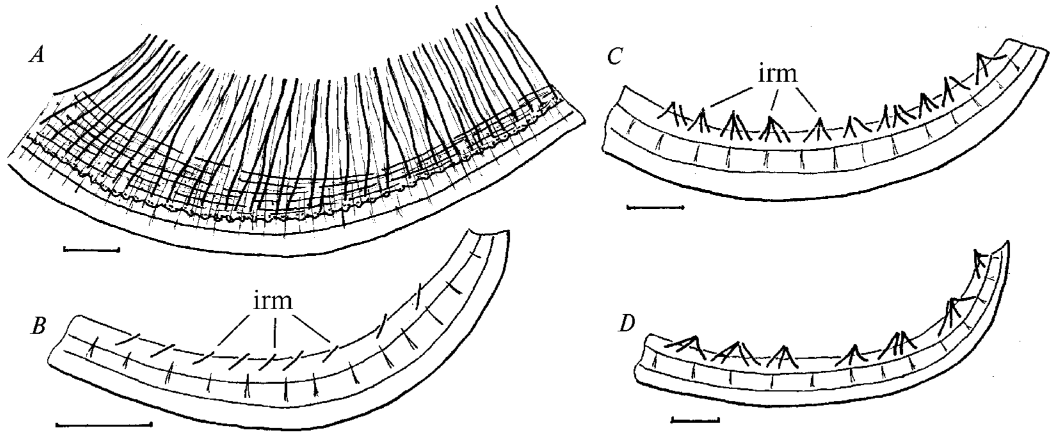

Fig. 6. Musculature of mantle edge (diagrammatic): *A — Corbicula*; *B — Eupera*; *C — Amesoda*; *D —* majority of Sphaeriinae. Scale bar 1 mm.

Рис. 6. Мускулатура мантийного края (схематизировано): *A — Corbicula*; *B — Eupera*; *C — Amesoda*; *D —* большинство Sphaeriinae. Масштабная линейка 1 мм.

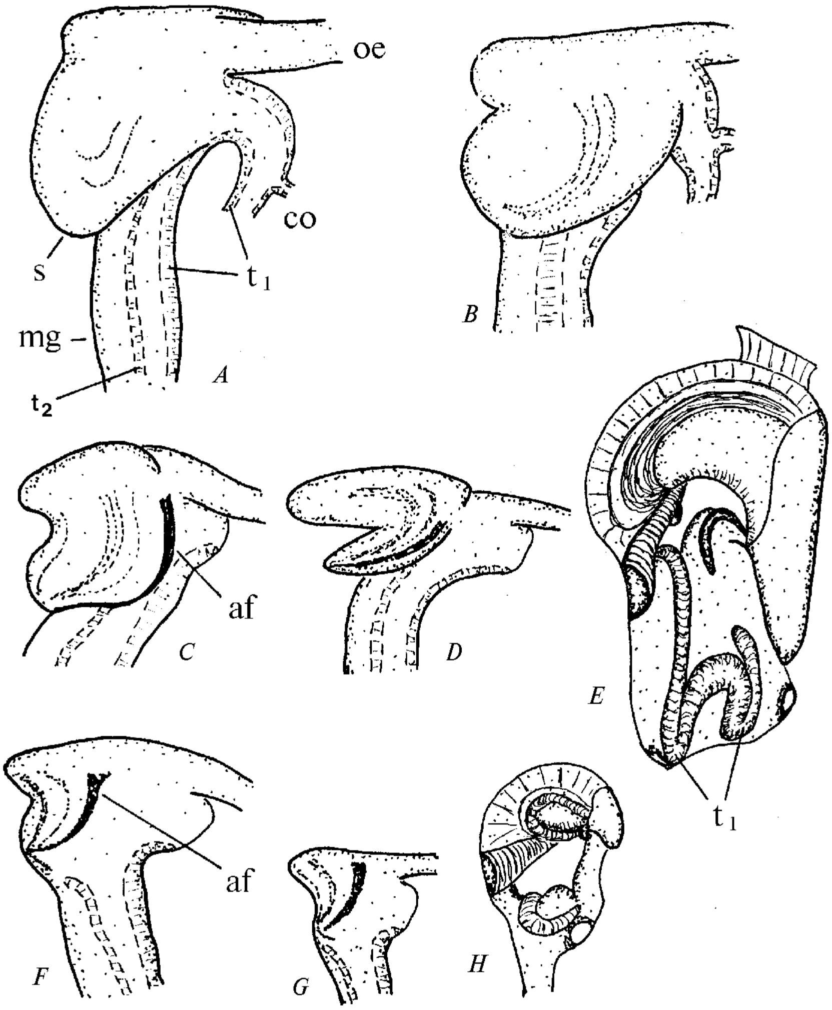

Fig. 7. Stomach characters (diagrammatic): *A* — *Chamelea gallina*; *B* — *Corbicula fluminea*; *C* — *Eupera platensis*; *D* — *Amesoda striatina*; *E* — *Amesoda similis*; *F* — *Sphaerium corneum*; *G* — *Pisidium amnicum*; *H* — *Euglesa supina* (*A*—*D*, *F*—*G* — view from right side; *E*, *H* — internal view of dissectted stomach).

Рис. 7. Строение жедудка (схематизировано): *A* — *Chamelea gallina*; *B* — *Corbicula fluminea*; *C* — *Eupera platensis*; *D* — *Amesoda striatina*; *E* — *Amesoda similis*; *F* — *Sphaerium corneum*; *G* — *Pisidium amnicum*; *H* — *Euglesa supina* (*A*—*D*, *F*—*G* — вид справа; *E*, *H* — вскрытый желудок изнутри).

From this study we still cannot decide, whether sequential or asynchronous brooding (when many broods on different stages of development are found in one animal) is a synapomorphy for *Sphaerium* s. l. clade, or a plesiomorphic feature in Sphaeriinae, since the evidence for the latter scenario is still very tentative (difference in one step only). Sequential brooding in *Neocorbicula* reported by Ituarte (1994) is apparently a result of parallel evolution.

Position of the brood pouch proved to be an informative character within *Pisidium* s. l.: peculiar states of this character found in *Pisidium* s. str. (brood pouch occupies major part of the inner demibranch even on initial stages of its development), and *Odneripisidium* + *Afropisidium* clade (brood pouch placed near the dorsal edge of the gill) are interpreted here as apomorphies. The whole group is also characterized by enlarged number of filaments involved in formation of the brood pouch.

Larval development. Different taxa of sphaeriids may release their larvae on different stages of development defined by the structure of their gills. The longest incubation takes place in *Sphaerium* s. str. and *Amesoda*, which newborn are released on the most advanced stages characterized by presence of the 2nd lamella in outer demibranchs (fig. 8). This condition may be interpreted as an ititial state in Sphaeriinae, or advanced character supporting its terminal clade, depending on our assumptions concerning relationships of *Sphaerium, Musculium* and *Pisidium*. Released larvae of *Musculium* and *Herringtonium* have only one outer demibranch lamella. The young of *Eupera* is released before appearance of the outer demibranch as such, repetition of this feature in *Pisidium* s. l. may be iterpreted as a symplesiomorphy or a reversion.

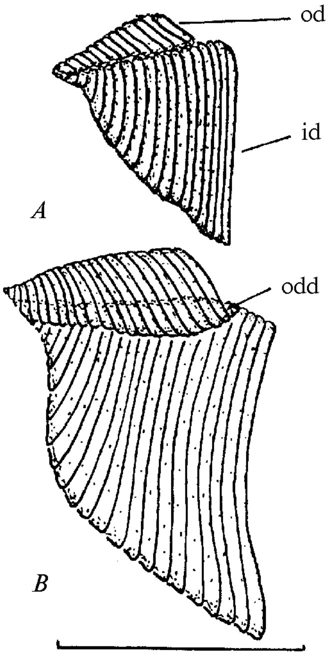

Fig. 8. Gills in released larvae: *A* — *Musculium*; *B* — *Sphaerium* s. str. Scale bar 1 mm.

Рис. 8. Жабры у молоди: *A* — *Musculium*; *B* — *Sphaerium* s. str. Масштабная линейка 1 мм.

This review of characters transformations in the family Sphaeriidae shows, that the data of different analyses are controversial and many aspects of evolution within this group are still not clear. However, evolutionary trends characterizing the whole family and its two basic partitions (subfamilies) could be defined with more certainty. Among these trends, reductions involving many organs and structures (hinge, ligament, siphons, gills and stomach) are the most remarkable. In general, such reductions are consistent with the diminution of size, but it could not be the only cause of reduction. For example, *Pisidium* s. str. is quite comparable in size to many species of *Sphaerium* and *Musculium*, but has many common features with much smaller species of *Euglesa*: reduced exhalant siphon with simplified musculature, one lamellar outer demibranch, shortened coil of midgut and pericardial tube of nephridium; it is even more advanced in its profound reduction of the outer demibranch indicated by its great posterior shift (tabl. 2).

On the other hand, such organs as siphonal and mantle edge musculature, nephridia and brood pouches were subject of progressive development and became more complicated and specialized in the course of evolution. Apparently, some of these transformations, namely development of the coiled nephridium and brood pouches for incubation of young were crucial for the group, determining its success in freshwater environments whith their hypoosmotic conditions and strong water currents. Factors which triggered transformation of the siphonal musculature are still not clear. The subfamily Euperinae is in many characters (e. g. ligament, labial palps and stomach) more primitive than Sphaeriinae. At the same time, two subfamilies show quite different adaptations in such structures as siphons, mantle edge and nephridium, as well as in the reproductive characters.

Taxonomic implications

The above analysis shows that the whole family Sphaeriidae and its two traditional subfamilies (Euperinae and Sphaeriinae) are well supported clades. Monophyly of some preliminarily defined taxa of Sphaeriinae, such as *Amesoda*, *Sphaerium* s. str., *Musculium* s. str., *Sphaerinova*, *Pisidium* s. str., *Henslowiana*, *Cingulipisidium*, *Pseudeupera* and *Odhneripisidium* (excluding *O. moitessierianum*) is also confirmed, but not all of these taxa have good bootstrap support. On the other hand, *Musculium* s. l., *Euglesa* and *Afropisidium* were shown to be paraphyletic or polyphyletic. Outstdanding position of *Herringtonium* is also shown, and its closer relationship to *Musculium* than to *Sphaerium* is noteworthy. *Sphaerinova* and *Paramusculium* proved to be separate line-

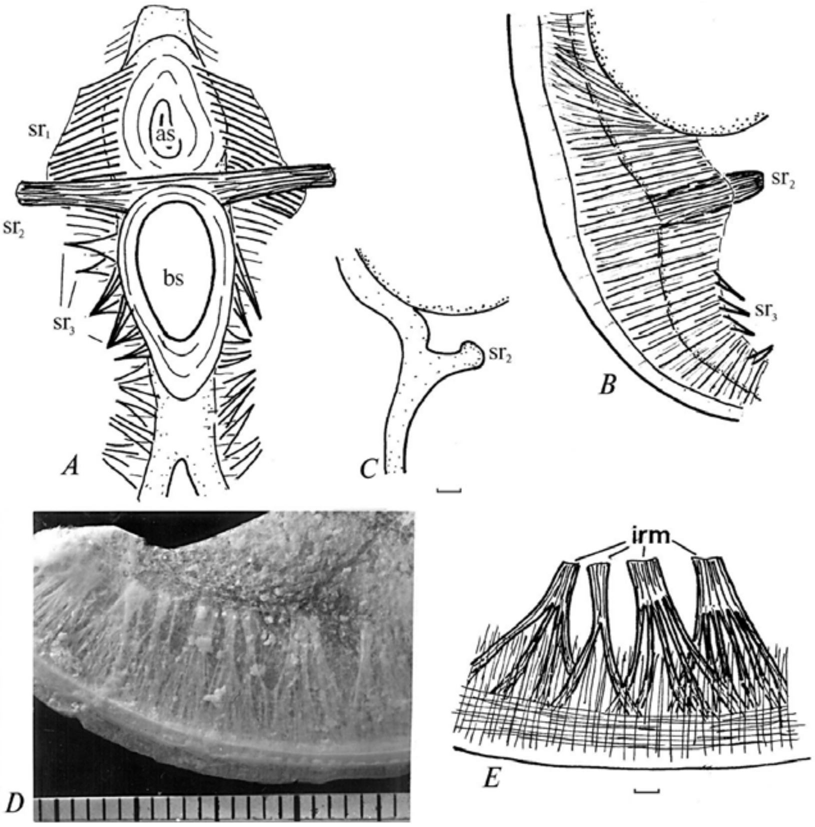

Fig. 9. Mantle musculature in *Polymesoda* (*Geloina*) *erosa*: *A* — siphonal muscles from inside; *B* — siphons, lateral view; *C* — mantle line near posterior adductor scar; *D*, *E* — mantle edge from outside (ruler in *D* 1 mm).

Рис. 9. Мантийная мускулатура *Polymesoda* (*Geloina*) *erosa*: *A* — сифональные мышцы изнутри; *B* — сифоны сбоку; *C* — мантийная линия вблизи отпечатка заднего аддуктора; *D*, *E* — мантийный край изнутри (*D:* цена деления 1 мм).

ages with uncertain relationships. Furthermore, close relationship of several *Musculium* species from tropical and southern temperate regions (*M. incomitatum*, *M. argentinum* and *M. indicum*) to *Sphaerinova* is shown by the search under constraint; if this relationship is confirmed, the mentioned species may be included in *Sphaerinova*.

Phylogenetic relationships of several species are tentatively defined by this study: *E. cara* is probably related to the group *Henslowiana* (shown only in the majority rule consensus), *E. hibernica* and *E. langleyana* — to *Cingulipisidium*, while *E. obtusalis*, *E. ovampicum* and *E. milium* — to *Pseudeupera* (the whole clade may bear the name *Cyclocalyx*). A group of species from different continents showed close affinity to *E. casertana* (tabl. 2; fig. 1, 2), the name *Casertiana* being available for this group. Relationships of *E. globularis* and *E. personata* are still not defined.

All “neotenic” pisidia belonged to a single terminal clade in both our reconstructions, with *Neopisidium conventus* in the basal position. Phylogenetic affinities and taxonomic status of *Odhneripisidium? moitessierianum* remain uncertain.

Possible sister groups of Sphaeriidae

Synapomorphies of the family Sphaeriidae, especially those which presume progressive development of organs, can be used as key characters in a search for possible outgroups of Sphaeriidae. Brackish-water bivalves living in conditions of transition between marine and freshwater habitats seem to be the most perspective in this aspect. As a first stage of this search, we checked some brackish water taxa traditionally assigned to Corbiculidae but not included in any previous phylogenetic analysis, namely *Polymesoda* (*Geloina*) and *Batissa*. Since characters of the mantle musculature important, as shown above, for understanding phylogeny of the Sphaeriidae were not described by previous morphological studies of corbiculids (Morton, 1976, 1989), we focused our examinations on these characters.

According to our observations, *P.* (*G.*) *erosa* is characterized by a very peculiar arrangement of siphonal muscles: the muscles of branchial siphons are separated from those of the anal siphon, forming a pair of strong retractors (fig. 9, *A—B*). This arrangement can be interpreted as a first step in formation of the retractors system, characteristic to Sphaeriidae. Mutiplied lower siphonal muscles can be seen as well, and they are very similar to those of *Eupera*. Unusual arrangement of siphonal muscles is reflected also in the mantle line, which has no mantle sinus, but a narrow projection in postrior part (fig. 9, *C*).

Musculature of the mantle edge of *Geloina* is also demonstrative, since the outer radial muscles are relatively short and the bundles of the inner radial muscles are prominent and attached well above the mantle line (fig. 9, *D—E*). This condition is rather similar to that observed in Sphaeriinae, but not in Euperinae.

Organization of the mantle musculature in *Batissa* is similar to that observed in *Corbicula*: siphonal muscles are rather short and musculature of the inhalant siphon are not clearly distinguished from that of the exhalant siphon, lower muscles of the inhalant siphon are not seen, outer radial mantle muscles are as long as the inner ones, the latter are organized in a band, not in separate bundles (fig. 10).

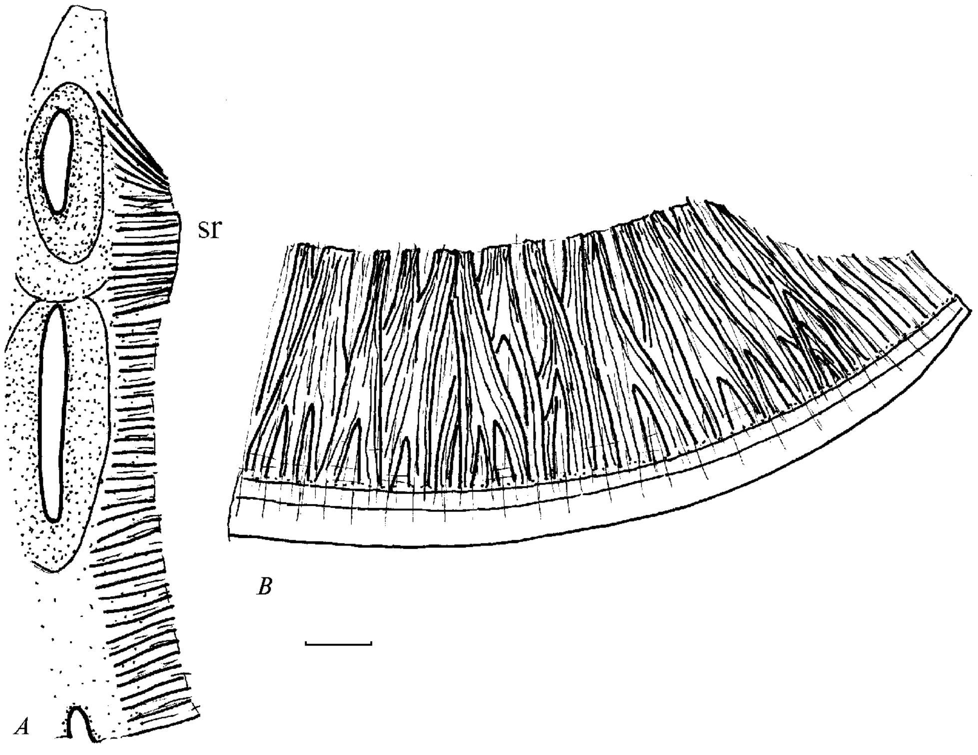

Fig. 10. Mantle musculature in *Batissa violacea*: *A* — siphons and their muscles from inside; *B* — mantle edge from outside.

Рис. 10. Мантийная мускулатура *Batissa violacea*: *A* — сифоны и их мускулатура изнутри; *B* — мантийный край снаружи.

Fixed material of the North American *Polymesoda* s. str. was not available for this study, however the course of the mantle line shows that its musculature is organised in the same way as in *Neocorbicula*, and differs from that of *Corbicula* and *Batissa* only in having deep mantle sinus.

Thus, the family Corbiculidae appears to be rather heterogeneic in respect of its mantle musculature, with *Polymesoda* (*Geloina*) falling apart from the other taxa and demonstrating some similarity to the family Sphaeriidae. Noteworthy, *Geloina* is not distinguished from other corbiculids or venerids in the principal characters of hinge, ligament siphons and stomach (Morton, 1976, this study).

Discussion

Monophyletic status of the family Sphaeriidae and its two major subgroupings is in a good agreement with the previous morphological (Mansur, Meier-Brook, 2000) and molecular (Park, Ó Foighil, 2000) studies. However, the rank of these subgroupings is still disputable. Taking into account that synapomorphies of Euperinae and Sphaeriinae concern many organs and their reproductive systems demonstrate different strategies of adaptation to the freshwater environments (e. g. ovoviviparity and viviparity), we cannot reject also those classifications, which recognize Euperinae and Sphaeriinae as separate families (Starobogatov, 1992).

Reconstructions of relationships between the major partitions in Sphaeriinae (or Sphaeriidae in the strict sense suggested above) are still controversial. Since *Pisidium* s. l. clade is well supported by each morphological analysis (fig. 1,2), and the clade *Sphaerium* s. l. (*Sphaerium* + *Musculium*) well supported by molecular data (Park, Ó Foighil, 2000; Cooley, Ó Foighil, 2000) requires only one additional step in our analysis (fig. 2), monophyly of both clades seems very probable. Noteworthy, all characters supporting *Pisidium* s. l. are reductions, but synchronous repetition of reductions in several organs (mantle, gills, alimentary system and nephridium) seems to be improbable. If the basic dichotomy of viviparous clams is confirmed by the further study (e. g. total evidence analysis), each of the partitions may obtain the rank of subfamily (Sphaeriinae and Pisidiinae respectively).

The other major discrepancy between morphological and molecular reconstructions need to be explained, namely position of *Afropisidium*, which is shown to be a sister clade to all remaining Sphaeriidae in the trees, based on 18S and 16S ribosomal DNA data (Park, Ó Foighil, 2000; Cooley, Ó Foighil, 2000). Probably, this taxon or the whole group *Neopisidium* s. l. (including *Neopisidium* s. str., *Afropisidium* and *Odhneripisidium*) is a basal clade for *Pisidium*, not a terminal one as shown by this study. Such a topology apparently needs fewer additional steps than assuming paraphyly of *Pisidium* s. l. (Korniushin, Glaubrecht, in press). Two circumstances should be mentioned in this respect. First of all, *Afropisidium* is distinguished from other *Pisidium* s. l. by the external type of ligament. This type could not be coded unequivocally because of numerous transitions to the other types, but may indicate plesiomorphic condition and thus ancient origin of the group. Detailed examination of the ligament pit for presence of rudimentary nympha seems necessary. Furthermore, *Afropisidium* (as well as *Odhneripisidium*) demonstrate similarity to *Eupera* in configuration of nephridium. According to Mansur and Meier-Brook (2000), the lateral section of nephridium in *Eupera* is a part of the dorsal lobe, while similarly looking structure in observed in the above mentioned taxa and, partly, in *Amesoda* and *Neopisidium conventus* — as the lateral loop. Consequently, these structures were coded as different characters. However, characters of nephridium apparently need further investigation. Probably, the coding accepted here is not adequate to the homologies between different parts of this organ. Noteworthy, *Euglesa* is monophyletic either in the published molecular trees (Cooley, Ó Foighil, 2000), or in the recostructions obtained under constraint, fixing *Neopisidium* as a basal lineage of *Pisidium* s. l.

In showing the trend for gill reduction, this study is in agreement with earlier reviews on gill development and evolution in sphaeriids (Korniushin, 1996 b). At the same time, monophyletic status of the group characterized by complete reduction of the outer demibranch (*Neopisidium* s. l.) contradicts to some earlier views (Kuiper, 1962; Korniushin, 1992, 1998 a). Spontaneous reduction of the outer demibranch was also reported for some species belonging to *Euglesa*, e. g. *E. keniana* (Kuiper, 1966 a; Piechocki, Korniushin, 1994), but later investigations have shown that some populations of these species may have small rudiments of the outer demibranch (Korniushin, 1998 d), which never happens in *Neopisidium* species.

Similar length of the trees with rather different topologies, obtained by our analyses can be explained by controversial distribution of character states. In particular, several taxa have peculiar combinations of primitive and advanced states, e. g. *Amesoda* is characterized by the primitive features of ligament (presense of nympha) and stomach (clear separation from the midgut), alongside specialized or advanced features of siphonal musculture (long and strong retractors) and brood pouches (absense of compartmentation). Since position of these groups on the phylogenetic tree is still not defined, they deserve a closer study. The cases of character reversions suggested by our analyses (e. g. concerning nympha, stomach and nephridia) also need a careful investigation. Probably, involving more characters and taxa may help to find more definite solutions of these puzzles. Resolving uncertainty about the nearest sister group of Sphaeriidae is also very important in a search for such solutions. Until more data for phylogenetic analyses are available, we consider premature any extensive taxonomic rearrangements.

Discrepancies in characters of the mantle muscles found between different taxa of Corbiculidae make phylogenetic analysis of this group (either morphological or molecular) rather urgent. *Polymesoda* (*Geloina*) should be included in such analyses and its subgeneric vs generic status, as well as monophyly of the whole family need to be tested.

Despite rather simplified composition of hinge and structure of stomach characterizing Sphaeriidae, we consider improbable their direct origin from a primitve heterodont group like Astartidae or Astartoidea, as suggested by Starobogatov (1992). Also molecular analysis shows, that Sphaeriidae belong to the clade of more advanced Veneroida including superfamilies Veneroidea, Mactroidea, and Tellinoidea, as well as freshwater families Corbiculidae and Dreissenidae (Park, Ó Foighil, 2000). However, *Geloina* may not be the only potential sister group of Sphaeriidae, therefore a broader phylogenetic analysis involving the characters defined here and including a selection of taxa from different heterodont superfamilies seem to meaningful.

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Attachment 1. List of characters and states. For detailed description of characters see A. Korniushin, M. Glaubrecht (in press).

Приложение 1. Список признаков и их состояний. Детальное описание признаков дано А. Корнюшиным, М. Глаубрехтом (в печати).

Shell

1. *Position of umbo*: 0 — anterior; 1 — central; 2 — posterior.
2. *Caps (calyculi)*: 0 — absent; 1 — present.
3. *Position of ligament*: 0 — not introverted; 1 — introverted.
4. *Nympha*: 0 — present; 1 — absent.
5. *Inner cardinal tooth of the right valve (c1)*: 0 — present; 1 — absent.
6. *Inner cardinal tooth of the left valve (c2)*: 0 — divided into 2 parts; 1 — bent (arched); 2 — straight; 3 — absent.

7. *Outer cardinal of the right valve (c3)*: 0 — divided into 2 parts; 1 — not divided; 2 — absent.
8. *Outer cardinal tooth of the left valve (c4)*: 0 — present; 1 — absent.
9. *Lateral teeth*: 0 — absent; 1 — present.
10. *Folds of periostracum*: 0 — present; 1 — absent.
11. *Size of periostracum folds*: 0 — not pronounced; 1 — pronounced.
12. *Periumbonal striae*: 0 — absent; 1 — present

Mantle

13. *Branchial mantle opening*: 0 — present; 1 — absent.
14. *Siphons*: 0 — two siphons; 1 — only anal siphon.
15. *Fusion of siphons*: 0 — fused; 1 — free.
16. *Contraction of siphons*: 0 — without apical part turning inside; 1 — with apical part turning inside.
17. *Retractors of the exhalant siphons*: 0 — integrated with muscles of inhalant siphon; 1 — separated.
18. *Upper muscles of the branchial siphon*: 0 — present; 1 — absent.
19. *Strength of the upper retractors of the branchial siphon*: 0 — strong; 1 — weak.
20. *Attachment of upper the muscles of branchial siphon*: 0 — along the mantle line; 1 — tightly adjoining adductor muscles (the scars are not separated); 2 — apart from posterior adductors (the scars are separated).
21. *Arrangement of lower muscles of the branchial siphon*: 0 — organised in broad muscle bands; 1 — form several paired bundles; 2 — form one pair of bundles.
22. *Perisiphonal mantle fusion (suture)*: 0 — present; 1 — absent.
23. *Length of perisiphonal suture*: 0 — short; 1 — slightly elongate (about 1/5 the length of the pedal slit); 2 — markedly elongate (1/4 to 1/2 the length of the pedal slit).
24. *Inner radial mantle muscles*: 0 — dispersed; 1 — organised in bundles.
25. *Bundles of inner radial muscles*: 0 — strong; 1 — weak.
26. *Differentiation of the mantle muscle bundles*: 0 — bundles uniform; 1 — anterior bundles markedly bigger.
27. *Orientation of mantle muscle bundles*: 0 — perpendicular to the mantle margin; 1 — converging anteriorly; 2 — converging medially.
28. *Number of muscle bundles*: 0 — ten to fourteen; 1 — seven to nine; 2 — four to six.
29. *Outer radial muscles*: 0 — long; 1 — short.
30. *Inner mantle fold*: 0 — normally developed; 1 — poorly developed.

Gills

31. *Outer demibranch*: 0 — present; 1 — absent.
32. *Position of outer demibranch*: 0 — before 6th filament; 1- at 7th to 10th filament; 2 — behind 11th filament.
33. *Outer demibranch descending lamella*: 0 — present; 1 — absent.
34. *Inner demibranch ascending lamella*: 0 — high; 1 — relatively low.
35. *Interlamellar septae in the inner demibranch*: 0 — developed on each 5th-7th filament; 1 — developed on each 2nd filament; 2 — developed on all filaments.

Alimentary system

36. *Anterior edge of the outer palps*: 0 — straight; 1 — with a projecting angle.
37. *Ridged area on palps*: 0 — broad (covers the whole inner surface); 1 — narrow (covers about ½ of the inner surface).
38. *Separation between the stomach and the midgut*: 0 — present; 1 — absent.
39. *Form of the stomach.* States: 0 — stretched dorso-ventrally; 1 — stretched in posterior direction.
40. *Sorting area on the stomach roof (SA3)*: 0 — broad; 1 — narrow.
41. *Anterior fold*: 0 — not elevated; 1 — elevated.
42. *Caeca*: 0 — present; 1 — absent.
43. *Anteriorly directed branch of the right digestive gland duct*: 0 — present; 1 — absent.
44. *Course of the major typhlosole*: 0 — with two loops; 1 — simple, without any loops.
45. *Course of the minor typhlosole in the stomach*: 0 — runs parallel to the major typhlosole; 1 — turns posteriorly.
46. *Coil of the intestine*: 0 — complicated, with several loops; 1 — simple, with one loop.
47. *Funnel*: 0 — short; 1 — long.

Nephridia

48. *Form of the funnel*: 0 — broad; 1 — narrow.
49. *Pericardial tube*: 0 — absent; 1 — present.
50. *Course of the pericardial tube*: 0 — with at least 3 loops; 1 — with only 2 loops.
51. *Dorsal lobe*: 0 — absent; 1 — present.
52. *Splitting of the dorsal lobe*: 0 — in three sections (branches); 1 — in two sections.
53. *Form of the dorsal lobe*: 0 — elongated (length more than width); 1 — square (length equal to width); 3 — broad (length less than width).
54. *Position of the pericardial tube in relation to the dorsal lobe*: 0 — covered by the dorsal lobe (closed nephridium); 1 — visible dorsally (open nephridium).
55. *Anterior extension of the lateral loop*: 0 — absent; 1 — present.

56. *Position of the anterior extension*: 0 — completely covered by the dorsal lobe; 1 — partly covered by the dorsal lobe; 2 — open from the dorsal side.
57. *Excretory sac*: 0 — absent; 1 — present.
58. *Form of excretory sac*: 0 — not extended; 1 — extended anteriorly

Reproductive system

59. *Gonad*: 0 — extending dorsally; 1 — not extending dorsally.
60. *Nutrition of embryos*: 0 — planktotrophic; 1 — lecithotrophic (ovo-viviparity); 2 — nutrition provided by the parental animal (eu-viviparity).
61. *Simultaneous development of several broods (asynchronous brooding)*: 0 — absent; 1 — present.
62. *Brood pouches*: 0 — absent; 1 — present.
63. *Number of filaments in the pouch*: 0 — two to three; 1 — five to nine; 2 — more than ten.
64. *Position of pouch*: 0 — upper position; 1 — lower position; 2 — not localised.
65. *Compartmentalisation of brood pouches*: 0 — absent; 1 — present.
66. *Byssus in adults*: 0 — present; 1 — absent.

Development

67. *Velum*: 0 — present, 1 — absent.
68. *Outer demibranch in the released young*: 0 — absent, 1 — present.
69. *Second lamella of outer demibranch in the incubated larvae*: 0 — absent, 1 — present.

Attachment 2. **Preliminary classification of the studied taxa, with characteristics of their distribution** (taxa with indefinite status marked with “?”, type species of genera and subgenera in bold).

Приложение 2. **Предварительная классификацияизученных таксонов и характеристика их распространения** (таксоны с неопределенным статусом обозначены “?”, типовые виды родов и подродов выделены шрифтом)

Family Veneridae
Chamelea gallina (Linne, 1758) — Atlantic and Mediterranean

Family Corbiculidae
Corbicula fluminea (Müller, 1774) — Oriental
Neocorbicula limosa (Maton, 1809) — South American

Family Sphaeriidae
Subfamily Euperinae
Genus *Eupera* Bourguignat, 1854
E. platensis Doello-Jurado, 1921 — South American
Genus *Byssanodonta* Orbigny, 1846
B. paranensis Orbigny, 1835 — South American
Subfamily Sphaeriinae
Genus *Amesoda* Rafinesque, 1820
Subgenus *Amesoda* s. str.
A. similis (Say, 1816) — North American
A. striatina (Lamarck, 1818) — North American
Subgenus *Rivicoliana* Servain, 1888
A. rivicola (Lamarck, 1818) — European
Genus *Sphaerium* Scopoly, 1777
S. corneum (Linnaeus, 1758) — Palaearctic
S. nucleus (Studer, 1820) — Palaearctic
S. rhomboideum (Say, 1822) — North American
S. nitidum Clessin in Westerlund, 1876 — Circum-boreal
S. solidum (Normand, 1844) — European
Genus? *Herringtonium* Clarke, 1973
H. occidentale (Prime, 1860) — North American
Genus *Musculium* Link, 1807
Subgenus *Musculium* s. str.
M. lacustre (Müller, 1774) — Holarctic
M. securis (Prime, 1851) — North American
Subgenus *Paramusculium* Alimov et Starobogatov, 1968
M. transversum (Say, 1829) — North American
Subgenus *Sphaerinova* Iredale, 1843
M. tasmanicum (Tenison Woods, 1870) — Australian
M. novaezelandiae (Deshayes, 1854 — New Zealand
Subgenus *Afromusculium* Korniushin, 1998
M. incomitatum (Kuiper, 1966) — South African

Species with uncertain subgeneric placement
M. hartmanni (Jickeli, 1874) — African
M. argentinum (Orbigny, 1835) — South American
M. indicum (Deshayes, 1854) — Oriental (Indian)
Genus *Pisidium* C. Pfeiffer, 1821
P. amnicum (Müller, 1774) — Palaearctic
P. dubium (Say, 1816) — North American
Genus *Lacustrina* Sterki, 1816
P. subtilestriatum Lindholm, 1909 — Euro-Siberian (Arctic)
Genus *Euglesa* Jenyns, 1832
Subgenus *Euglesa* s. str.
E. personata (Malm, 1855) — European
E. casertana (Poli, 1791) — Holarctic?
E. globularis Clessin in Westerlund, 1873 — Palearctic
Subgenus *Cyclocalyx* Dall, 1903
E. obtusalis (Lamarck, 1818) — Palaearctic
Subgenus *Hiberneuglesa* Starobogatov in Dolgin, 1983
E. hibernica (Westerlund, 1897) — European
Subgenus *Pseudeupera* Germain, 1913
E. subtruncata (Malm, 1855) — Holarctic
E. pulchella (Jenyns, 1832) — Palearctic
Subgenus *Henslowiana* Fagot, 1792
E. henslowana (Sheppard, 1823) — Palearctic
E. lilljeborgi (Clessin, 1886) — Holarctic (Boreal)
E. supina A. Schmidt, 1851 — Euro-Siberian
Subgenus *Cingulipisidium* Pirogov & Starobogatov, 1974
E. nitida (Jenyns, 1832) — Holarctic
E. pseudosphaerium (Favre, 1927) — European
Subgenus *Tetragonocyclas* Pirogov & Starobogatov, 1974
E. milium (Held, 1836) — Holarctic
Species with uncertain subgeneric placement
Euglesa langleyana (Melvill & Ponsonby, 1891) — South African
E. ovampicum (Ancey, 1890) — South African
E. viridaria (Kuiper, 1956) — African
E. etheridgei (Smith, 1883) — Australian
E. atkinsoniana (Theobald, 1876) — Oriental (Indian)
E. compressa (Prime, 1851) — North American
E. cara (Cotton, 1953) — Australian
E. keniana (Preston, 1911) — African
Genus? *Neopisidium* Odhner, 1821
N. conventus (Clessin, 1877) — Holarctic (Boreo-Alpine)
Genus? *Afropisidium* Kuiper, 1962
A. pirothi (Jickeli, 1880) — African
A. sterkianum (Pilsbry, 1897) — South American
A. clarckeanum (G. & H. Nevill, 1871) — Oriental
A. aslini (Kuiper, 1983) — Australian
A. hodgkini (Suter, 1905) — New Zeland
Genus? *Odhneripisidium* Kuiper, 1962
O. stewarti (Preston, 1909) — Central Asian
O. annandalei (Prashad, 1925) — Oriental
O. tenuilineatum (Stelfox, 1918) — European
Odnneripisidium? *moitessierianum* (Paladilhe, 1866) — Euro-Siberian

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