Report on Molecular Systematics of Poorly-Known Freshwater Mollusks of Alabama

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Project Summary

This project used molecular sequence data to investigate several Priority 1 and 2 species of freshwater mollusks.

Main questions

- Is there significant genetic variation in *Elliptio arca* across the Mobile basin?
- Is *Elliptio purpurella* present in Alabama and genetically distinct from other *Elliptio* species?
- How different is *Elliptio mcmichaeli* from *E. crassidens*?
- What are the relationships of *Fusconaia escambia*?
- Are there significant differences between Coosa and Tennessee populations assigned to *Lasmigona holstonia*?
- Are there significant differences between middle and upper Coosa populations of *Strophitus connasaugaensis*? Does the species occur in the Black Warrior or Tombigbee?
- Is *Toxolasma corvunculus* different from *T. parvus?*
- Is *Toxolasma cylindrellus* different from *T. lividus*?
- Are there additional species of *Toxolasma* in the Escambia, Yellow, and Choctawhatchee systems? How are they related to the ACF and Mobile species?
- Are the priority species of *Elimia* in the Cahaba system truly distinct species or merely ecomorphs?
- Are the priority species of *Pleurocera* in the Tennessee system truly distinct species or merely ecomorphs?

Results

In addition to the specific target species (indicated in bold), some related priority 1 and 2 species were analyzed for comparison. Results for priority 1 and 2 species are as follows:

- ***Elliptio arca***: No significant genetic variation was detected between populations from different parts of the Mobile basin. Moderate genetic distance from other *Elliptio* species suggests its biological requirements might also be somewhat different, requiring corresponding design of conservation techniques.
- *Elliptio arctata*: Moderate genetic distance from other *Elliptio* species suggests its biological requirements might also be somewhat different, requiring corresponding design of conservation techniques.
- *Elliptio dilatata*: High genetic distance from other *Elliptio* species suggests its biological requirements are different, requiring corresponding design of conservation techniques.
- ***Elliptio mcmichaeli***: Slight genetic distance from *E. crassidens*. Biological requirements are probably very similar to that species.
- ***Elliptio purpurella***: Not clearly distinguished from the *icterina/complanata* group of species, due to both limited genetic divergence and the difficulty of confidently identifying reference specimens. This group is present in the Tallapoosa system, though previously it was not known in the Mobile basin.

- *Fusconaia barnesiana*: High genetic distance from other *Fusconaia* species suggests its biological requirements are different, requiring corresponding design of conservation techniques.
- *Fusconaia cor*: Closely related to *F. cuneolus* but distinct. They place within *Fusconaia* and probably have similar biological requirements to the more common *F. cerina*.
- *Fusconaia cuneolus*: Closely related to *F. cor* but distinct. They place within *Fusconaia* and probably have similar biological requirements to the more common *F. cerina*.
- ***Fusconaia escambia***: Closely related to *Quincuncina burkei* but distinct. They place within *Fusconaia* and probably have similar biological requirements to the more common *F. cerina*.
- *Fusconaia rotulata*: Closely related to the more common *F. ebena*. However, these two species are very distantly related to other *Fusconaia* species and probably have rather different biological requirements.
- *Fusconaia subrotunda*: Places within *Fusconaia* and probably has similar biological requirements to the more common *F. cerina*.
- *Lasmigona costata*: Very different from some of the other species currently assigned to *Lasmigona*.
- ***Lasmigona holstonia***: At least three different evolutionary units are currently placed under this name. In Alabama, the upper Coosa and upper Tennessee forms appear different; a third form only occurs in Tennessee. None are close relatives of *L. costata*.
- ***Strophitus connasaugaensis***: Closely related to but distinct from *S. subvexus*. The upper Coosa form (not currently known to live in Alabama) and the mid-Coosa form show a moderate level of genetic difference and probably require separate conservation. These species are not closely related to *S. undulatus* and probably have very different biological requirements. Sampled specimens from the Tombigbee and Black Warrior were *S. subvexus*, not *S. connasaugaensis*.
- *Strophitus undulatus*: Not very closely related to any of the other sampled species.
- ***Toxolasma corvunculus***: Is a valid species, but shells may be confused with the common *T. parvus* in the Black Warrior and Tombigbee systems.
- ***Toxolasma cylindrellus***: Is a valid species, most closely related to *T. lividus*.
- **Gulf drainage *Toxolasma*** species: Three overlooked evolutionary units exist: the Escambia and Pea form, the Choctawhatchee form, and a second species (along with *T. paulus*) in the ACF system.
- ***Elimia ampla***: Not clearly different from *E. variata*. The sampled *Elimia* species fell into several groups that were relatively distantly related to each other, suggesting that there may also be differences in their biological requirements.
- ***Elimia annettae***: Identification may be a problem, as two specimens appeared closely related to *E. bullula* from the Coosa but the third was quite different.
- ***Elimia bellacrenata***: Relatively close to two of the *E. annettae* and to *E. bullula*, but may be a distinct species. Similar specimens observed in Spring Creek upstream from Montevallo suggest that the species may be more widespread in the upper Little Cahaba system than currently thought.
- ***Elimia cochliaris***: Genetically very distinctive. A second population from Buck Creek may be sufficiently genetically different to need separate conservation management.

- ***Elimia varians***: Identification seems to be a problem, as three specimens all came out quite different from each other.
- ***Elimia variata***: Not clearly different from *E. ampla*.
- ***Pleurocera alveare***: Seems to fall genetically within the range of variation of *P. canaliculata*.
- ***Pleurocera pyrenella***: Seems to fall genetically within the range of variation of *P. canaliculata*.

Conservation implications

- Species with high levels of genetic difference between populations, especially from different drainages, should probably be treated as separate entities for conservation. This will probably raise the conservation priority for those species.
- Further study of genetically distinctive populations is needed to look for mrophological differences that can be used in the field in conservation work.
- Different species with low genetic difference between them may be simply ecomorphs, not requiring separate conservation.
- Current genus names are not always a reliable guide to the needs of rare species.

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Abstract: The conservation status of several species of freshwater mollusks in Alabama has been unclear due to uncertainties about their systematics and identification. This project investigated several problematic taxa using DNA sequencing to provide new evidence on their taxonomic status. In turn, these data provide information that suggests revisions in the conservation status of several taxa. In general, the unionids have more evolutionarily significant forms than currently recognized, whereas some of the pleurocerids may be oversplit. Many species reported to range across multiple river systems actually constitute multiple evolutionary units.

Introduction

Alabama has one of the most diverse freshwater mollusk faunas in the world, with over 350 species. Many species are highly sensitive to human-caused habitat modification such as impoundment and siltation. As a result, a large proportion of the priority 1 and 2 species in Alabama are freshwater mollusks. However, the classification and identification of species is problematic due to high individual variation, similarities between species, and lack of modern taxonomic review. Both unionids and pleurocerids are large and slow-moving, though the mussels have high dispersal potential as larvae parasitic on fish. Neither group can readily cross divides between river systems. This produces high potential for geographic isolation. As a result, similar forms in different river systems may actually be reproductively isolated species and deserve careful scrutiny. On the other hand, some freshwater mollusks show significant ecophenotypic variation within species. Because of this, some workers treat numerous named forms as merely variants of a single species. An extreme example is Hannibal (1912), who lumped almost the entire genus *Elimia* into a single species. The uncertainty regarding the pleurocerid snails is so high that no one has attempted to thoroughly revise them since Goodrich (1940, 1941a,b). Unfortunately, he did not explain most of his decisions, and some of his reasoning does not agree with modern practice or current understanding of systematics and evolution.

Also, molecular data suggest that the current genus-level classification of freshwater mollusks requires extensive revision (Lydeard *et al*., 2000; Minton *et al*., 2003; Campbell *et al*., 2005). This affects conservation because the biological requirements of poorly-known rare species often must be inferred from better-known related species. However, if we are seriously mistaken about the relationships of the species, then inferences about the biological requirements of the rare species are likely to be incorrect.

Because of the uncertainties in classification and identification of freshwater mollusks, current conservation priorities may be inaccurate. Molecular data provide a new source of evidence that can help address these problems. The project selected several species of unionid mussel and pleurocerid snail of high conservation concern in Alabama after consultation with several malacologists to identify some of the most problematic species and species groups in Alabama. The *Lasmigona holstonia* group occurs in small headwater streams in the New, Tennessee, Coosa, and Cumberland systems; historically, it also occurred in the Duck, Black Warrior, and Cahaba (Parmalee and Bogan, 1998; Pinder *et al*., 2002; Athearn, Museum of Freshwater Malacology collections). Such small streams are likely to undergo inter-drainage capture, so a species might be expected to range into multiple drainages. However, subtle morphological differences have prompted some workers to suspect that more than one species is present. *Lasmigona holstonia* is a priority 2 species in Alabama, as is *L. costata* (used for comparison). The *Strophitus subvexus* group ranges across Gulf drainages from easternmost Texas to the Apalachicola system (Vidrine, 1993; Brim Box and Williams, 2000). A distinct species, *S. connasaugaensis*, is recognized in the upper Coosa system. Morphological variation within the Black Warrior, Tombigbee, and Coosa systems has prompted speculation that multiple species may be present in these areas. Conversely, Johnson (1970) synonymized *S. connasaugaensis* with *S. subvexus*. *Strophitus connasaugaensis* is a priority 2 species, and *S. undulatus* (analyzed for comparison) is a priority 1 species.

Several species in the genus *Toxolasma* (lilliputs) are currently recognized by most authors (Turgeon *et al*., 1998), but some workers have synonymized several of these (Johnson, 1970). On the other hand, it is suspected that the Gulf drainages south and east of the Mobile system may harbor currently unrecognized species (Brim Box and Williams, 2000; Blalock-Herod *et al*., 2005). Thus, it is unclear whether supposedly rare species such as *T. cylindrellus* and *T. corvunculus* are truly distinct from more widespread forms such as *T. lividus* and *T. parvus*, but *T. parvus* and *T. paulus* might actually be species groups. Both *T. cylindrellus* and *T. corvunculus* are priority 1 species.

The genus *Fusconaia* contains many species of concern in Alabama, as well as three common species. However, Lydeard *et al*. (2000) found that species assigned to this genus are not all closely related to each other, a result substantiated by Campbell *et al*. (2005). The exact relationships of *F. escambia* were unclear due to disparate molecular results, so the present study reinvestigated it. *Fusconaia barnesiana* is priority 2 and *F. cor, F. cuneolus, F. escambia, F. rotulata,* and *F. subrotunda* are priority 1 species. The other species were analyzed for comparison with *F. escambia*.

The genus *Elliptio* is perhaps the most confusing of all mussels for identification. Two readily distinguished species are known from the Tennessee River system and three readily distinguished species are known from the Mobile basin. In contrast, the number of species and their distinguishing features in drainages from the Escambia east along the Gulf to the Atlantic drainages remains uncertain. Currently, nine species are recognized in the Alabama portions of the Escambia, Yellow, Choctawhatchee, and Chipola drainages (Garner *et al*., 2004). There is also some morphological variation within *E. arca* in the Mobile system and within *E. crassidens,* which occurs throughout Alabama. *Elliptio arca, E. arctata, E. dilatata, E. mcmichaeli,* and *E. purpurella* are priority 1 species, and three other species are considered extirpated from the state. We obtained specimens possibly assignable to *E. purpurella* but no definitively identified specimens; definite specimens of the other priority 1 species were analyzed.

Among the snails, *Elimia* is one of the most diverse genera in freshwater, with 55 species reported from Alabama. Of these, 13 are reported from the Cahaba system. One of these has been listed as extinct; *E. cochliaris* and *E. bellacrenata* are priority 1; and *E. ampla, E. annettae, E. variata,* and *E. varians* are priority 2. However, some of the "species" have been reported to intergrade, suggesting that they may be merely ecophenotypes. *Pleurocera* is also diverse, with 15 Alabama species, mostly in the Tennessee system. Although Garner *et al*. (2004) follow Burch and Tottenham (1980) and Goodrich (1940) in recognizing ten species in the Tennessee system, Hannibal (1912) and Rosewater (1960) recognize at most three. Some of the distinctive morphologies, treated by Goodrich as species, are associated with particular environments. For example, *P. alveare* occurs in high energy settings and *P. pyrenella* occurs in tupelo swamp areas of tributary streams. They could be ecologically specialized species or merely ecological forms of a more common and widespread species. In particular, the relatively stout shape of *P. alveare* would provide less surface for currents to push against and thus it is less likely to wash off of rocks in strong currents. In contrast, the elongate form of many of the other putative *Pleurocera* species provides greater surface area, preventing sinking into muddy substrates. *Pleurocera pyrenella* has been reported from the upper Black Warrior system as well as from the Tennessee system; however, species in the Mobile Basin generally seem quite distinct from the Tennessee River species. Both *P. alveare* and *P. pyrenella* are priority 2 species. *Pleurocera corpulenta* is a priority 1 species that was not found during the present study and so could not be analyzed genetically.

Materials and Methods

The present project examined DNA sequences of the selected species, as well as related, more common, species for comparison. The *cox1* gene was used for pleurocerids and unionids, and the ITS1-ITS2 region was also used for the unionids. Specimens were frozen, preserved in ethanol, or kept alive. For some specimens, a small clip of mantle tissue was preserved in ethanol, allowing release of the specimens after their identity was confirmed. DNA extraction, amplification, and sequencing used standard protocols (Campbell *et al*., 2005). ITS1 primers sequences followed King *et al*. (1999); *cox1* primers followed Campbell *et al.* (2005) for unionids and Folmer *et al*. (1994) and Minton and Lydeard (2003) for pleurocerids. Some amplifications for both groups used a new *cox1* primer instead of the Folmer *et al*. H primer (Giribet, pers. comm.). ITS2, 16S and *nadh1* genes were sequenced for some unionids, using primers from Krebs *et* al. (2003) for 16S, Campbell *et al*. (2004) for ITS2, and Campbell *et al.* (2005) for *nadh1*. The *cox1* fragments amplified by the Folmer *et al*. (1994) primers and the Minton and Lydeard (2003) primers overlap and were amplified and sequenced separately. ITS1 and ITS2 were generally amplified as an entire region and then the two primers annealing to the 5.8S region were used as internal sequencing primers. Sequences were aligned with BioEdit (Hall, 1999) and analyzed using PAUP* (Swofford, 1998). ITS1 and ITS2 had several indels, as is commonly the case in non-coding regions. These sequences were analyzed with gaps treated as a fifth base. PAUP* analyses used heuristic searches with 1000 random addition replicates, holding 10 trees at each step. Bootstrap analyses used 1000 bootstrap replicates of 10 random addition replicates each. An analysis of *cox1* data for over 100 amblemine species used 100 replicates for the parsimony search and 165 bootstrap replicates of 10 random replicates each. This places the sampled taxa into a broader phylogenetic framework. Previous work on the pleurocerids used either the Folmer *et al*. (1994) region or the Minton and Lydeard (2003) region of *cox1*. Therefore, three analyses were run, each region individually and one including those taxa with data for both regions. Some species had large autapomorphic insertions in ITS1 sequences. None of the primary target species had these, and the regions were excluded from the analyses, since they are not informative with regard to the species of interest. Percent differences were calculated in PAUP*, which requires that gaps be treated as unknown data. Percent differences for *Elliptio* were also calculated manually, treating gaps as a fifth base, since so many indels were present. Table 1 gives specimen locality information for the newly generated data. Although some taxa show considerably higher or lower levels of variation in the *cox1* gene, about 2% difference is frequently considered to suggest different species (Nielsen and Matz, 2006). Populations with higher differences should not automatically be considered separate species, nor should populations with lower differences be automatically synonymized, but it gives a reference point to say that the observed difference is high or low.

Much debate exists over how to recognize species, particularly in the context of needing to prioritize them for conservation. Although this research focused on molecular data, morphological data are also needed for species delineation. Phylogenetic analyses were used to identify diagnosable, mutually monophyletic populations as evolutionary units that seem to represent distinct species. However, morphological investigation is needed to confirm the distinctiveness of these units. Therefore, formal taxonomic changes are not proposed in this report.

Results

ITS1 amplified well for almost all specimens. However, most pleurocerids were polymorphic for ITS1, making the sequence unreadable. A few unionids amplified non-target sequences in addition to ITS1, but these differed enough in length to be readily separated by gel extraction. Several unionids had large insertions in the ITS region, sometimes duplicating part of the region. In these cases, both copies of the gene were aligned. *Strophitus undulatus* had both an allele with a nearly complete duplication of the ITS1 region and an allele of normal length, resulting in three copies of the region for that species. Several unionids had two or more alleles differing by one or two bases, usually within a string of several identical bases. No indels were found in the *cox1* gene, although other mollusks have lost or gained codons. *cox1* failed to amplify for some specimens that amplified well with ITS1. Percent differences for *cox1* were typically higher than for ITS1.

Figures 1-10 show the phylogenetic results. Tables 2 and 3 show percent differences for comparisons of specific interest. These include comparisons of species that had been treated as synonyms and variations within populations.

For the target species, mussels often showed differentiation between populations from different river systems. Morphological reinvestigation may support their recognition as distinct species. *Lasmigona holstonia* includes three clearly distinct evolutionary units. The New River and the Tennessee River populations are not genetically different, but the Coosa and Cumberland populations are both different. *Strophitus connasaugaensis* from the Coosa is distinct from *S. subvexus* and the sampled *S. "connasaugaensis"* from the Black Warrior, which was not different from *S. subvexus*. The upper and middle Coosa *S. connasaugaensis* were moderately distinct. *Toxolasma cylindrellus, T. corvunculus, T.* species from the Escambia, and *T.* species from the Choctawhatchee all appear to be valid species, and there seem to be two species of *Toxolasma* in the ACF system. Both occur in the Chattahoochee system, so they both potentially occur in Alabama, although the only locality confirmed genetically for one of the species is in Georgia. Preliminary data (not shown) indicate that the Escambia species also occurs in the Pea River. *T. parvus* is present in the western Mobile basin. *Fusconaia escambia* also appears valid and is most closely related to "*Quincuncina" burkei*. *Elliptio arca* did not show much variation across the Mobile basin. *Elliptio crassidens* showed some variation among samples from the Ohio, Tennessee, and Mobile systems, but at a relatively low level. *Elliptio dilatata* is very different from other *Elliptio* species. Four *Elliptio* species are present in the Mobile Basin. The molecular data failed to clearly sort out all of the Gulf Coast *Elliptio* forms, but there seem to be at least two species in the "*complanata/icterina"* group, and they seem different from true *E. complanata* and *E. icterina* from the Atlantic drainages.

In contrast to the mussels, some of the snails showed less variation than expected based on current classification. *Elimia cochliaris* was very different from all other Cahaba species and showed high differentiation between populations, suggesting that it may include more than one species. *Elimia bellacrenata* appeared genetically similar to, but distinct from, *E. annettae* and *E. bullula*. *Elimia ampla* and *E. variata* showed almost no genetic difference from each other and probably represent forms of one species. Specimens identified as *E. annettae* and *E. varians* did not all group together in the genetic analyses. Two of the three *E. annettae* were very similar to *E. bullula*, a Coosa River species. The third was closest, but not very close, to one of the "*E. varians"* specimens. All three samples specimens for *Elimia varians* came out very distinct from each other, with one appearing identical to *E. clara* and *E. showalteri* and the other two both seeming to be otherwise unsampled species. *Pleurocera alveare* and *P. pyrenella* showed less genetic differentiation from nearby populations of *P. canaliculata* in the mainstream of the Tennessee River than the difference between different populations of *P. canaliculata* along the Tennessee River.

Discussion

The results have numerous implications for the conservation of rare freshwater mollusks. Geographic isolation appears more important than previously thought in many cases. This suggests that populations from different drainages should be managed separately if possible until there is good evidence that they are the same. On the other hand, morphological variation within a drainage, especially in the snails, may be merely ecophenotypic and does not always reflect any genetic diversity. Another general result is that the currently recognized genera may not be a reliable guide to close relationships. Therefore, it is not safe to assume that a poorly-known species will have similar biological requirements to a better-known species currently assigned to the same genus.

Several target populations of mussels were confirmed as genetically distinct. The Coosa and Tennessee system "*Lasmigona holstonia"* and upper and middle Coosa "*Strophitus connasaugaensis"* deserve recognition as separate conservation entities (although the upper Coosa form of *S. connasaugaensis* seems extirpated from Alabama, if it ever occurred that far downstream). These forms have been described as distinct species (*L. etowaensis* (Conrad, 1849) for the Coosa "*holstonia*" and *S. alabamensis* (Lea, 1861) for the middle Coosa "*connasaugaensis*"), suggesting that morphological differences can be found. The present data support continued recognition of *Toxolasma corvunculus* and *T. cylindrellus* as distinct, imperiled species and new recognition of the Choctawhatchee and Escambia *Toxolasma* species and the two ACF basin *Toxolasma* species as distinct conservation units. They also confirm that the widespread and common *Toxolasma parvus* is established in the Mobile basin, primarily in the Black Warrior and Tombigbee systems. *Fusconaia escambia* is confirmed as a distinctive species. The paraphyly of *Q. burkei* to *F. escambia* in Figure 6 probably is an artifact of the low difference between the two *Quincuncina* specimens. . The status of *Elliptio* species in the drainages south and east of the Mobile system remains unclear. *Elliptio mcmichaeli* is very close to *E. crassidens.* Additional sampling with more genes and additional specimens, especially unambiguous specimens of *E. purpurella* and similar forms, may provide better resolution of this group. Likewise, further work is needed to fix the identity of the ACF *Toxolasma* species, identifying their ranges and distinguishing morphological characteristics. Although several workers have speculated that distinct species are present, the lack of any formal description suggests that the characters are more subtle. Paul Johnson's observation that the lure and display of *T. cylindrellus* and *T. lividus* are very different suggests that this feature should be examined in the other *Toxolasma* species.

Dividing currently recognized species also usually increases the conservation priority of the new, smaller groups. Thus, the Tennessee and Coosa populations of *L. holstonia* and the newly distinguished *Toxolasma* populations require reassessment of their conservation status. The Tennessee *L. holstonia* population in Alabama is confined to the upper Paint Rock system and probably deserves higher priority ranking in light of its difference from the Coosa population. If *L. holstonia*-like mussels are rediscovered in the upper Black Warrior or Cahaba, they deserve careful study to determine if they are also distinct from the Tennessee and Coosa forms. Because the entire known Alabama population of *Strophitus connasaugaensis* is assignable to the middle Coosa form, the present results do not alter its conservation status for the state; however, it does indicate a need for greater concern in Georgia and Tennessee. No changes in species identification and conservation status are indicated for *Fusconaia* or *Elliptio* species, apart from the recognition that an additional species of *Elliptio* reaches the Tallapoosa system.

For the snails, the present results suggest that conservation priorities for *Pleurocera alveare, P. pyrenella, Elimia ampla* and *E. variata* can be lowered because they do not appear genetically distinct. These results should be interpreted with caution, since some taxa that are good species on other grounds have very low genetic differentiation (Nielsen and Matz, 2006). On the other hand, high genetic variation within a single species has been reported for some pleurocerids (Dillon and Frankis, 2004). *Elimia bellacrenata* appears close to but distinct from some "*E. annettae"* specimens and *E. bullula*. Additional data for more populations will be needed to test whether the degree of differentiation observed in the present study indicates distinct but closely related species or merely high variation within a single species. The high priority of *E. bellacrenata* reflects its being found by Bogan and Pierson (1993) at only a single locality, Orr Park in Montevallo. However, in collecting for the present study, similar specimens were observed upstream of Montevallo in the Spring Creek system, so it may not be quite as rare as was feared. Similarly, *E. cochliaris* was found in Ebenezer Swamp, in addition to the single spring where Bogan and Pierson (1993) found it. An *E. cochliaris*-like form from Buck Creek was most closely related to the Little Cahaba *E. cochliaris* population, but had enough genetic difference to warrant separate conservation. As a species confined to springs and spring runs, *E. cochliaris* could easily become genetically isolated in small populations. This could easily produce speciation, as well as making each species in the group highly vulnerable to extinction due to localized range. *Elimia annettae* and *E. varians* require further study to determine if morphological features can be detected corresponding to the molecular variation found in the present study. Apparently their morphologies represent ecophenotypes or growth stages in more than one species. For comparison, other species of *Elimia* in the Cahaba system were also sequenced. *Elimia clara* and *E. showalteri*, along with one of the *E. "varians"*, were identical in sequence and appear synonymous. *E. cahawbensis* and *E. carinifera* were very similar (0.6% difference), though *E. carinifera* from elsewhere in the Mobile system may not be the same as the specimens from the Cahaba identified as *carinifera*. *Elimia carinocostata* was different from all other Cahaba species, but similar to *Elimia modesta* (also called *E. murrayensis*) from the upper Coosa.

Unexpectedly, the species currently assigned to *Elimia* in the Cahaba system appear to represent several distinct groups of pleurocerids, rather than a single invasion of the Cahaba.

Data for additional species, especially the types of genera, will help determine if genus-level changes are necessary.

The results of this study suggest some practical conservation measures. Because similar forms in different rivers are not always genetically compatible, species need protected in multiple locations throughout their range. This also helps protect against the risk of a catastrophe in one area wiping out the entire species. For restocking, species should not be transferred across major drainages unless genetic study confirms that they are the same. The many species that are very different genetically from others currently assigned to the same genus indicates that it's not safe to assume that a rare species will have similar biological needs (such as host fish, egg laying preferences, habitat requirements, *etc.*) to a better-known species in the same genus.

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Table 1. Collection data for specimens used. UAUC=University of Alabama collection number; NCSM=North Carolina State Museum number, B=Virginia DNR numbers for *Lasmigona holstonia* from southwestern Virginia, JDS *etc.*=Sides dissertation specimens. Other numbers are GenBank accession numbers for published data (*e.g.*, AY655029).

Species	Gene	Accession	Source
Actinonaias ligamentina	*cox1*	AF231730	Bogan and Hoeh, 2000
Actinonaias pectorosa	*cox1*	AY654990	Campbell *et al.*, 2005
Alasmidonta heterodon	*cox1*	AF093842	King *et al.*, 1999
Amblema elliottii	16S	AY655029	Mulvey *et al.*, 1997
Amblema elliottii	*cox1*	AY654991	Campbell *et al.*, 2005
Amblema elliottii	*nadh1*	AY655086	Campbell *et al.*, 2005
Amblema plicata 1	*cox1*	U56841	Hoeh *et al.*, 1998
Amuranodonta kijaensis	*cox1*, ITS1	UAUC3297	Mongolia
Anodonta anatina	*cox1*	DQ060168	Kallersjo *et al.*, 2005
Anodonta anatina	ITS1	AJ295287	Gerke and Tiedemann, 2001
Anodonta californiensis	*cox1*	AY493462	Mock *et al.*, 2004
Anodonta cygnea	*cox1*	DQ060170	Kallersjo *et al.*, 2005
Anodonta cygnea	ITS1	AJ295288	Gerke and Tiedemann, 2001
Anodonta oregonensis	*cox1*	AY493480	Mock *et al.*, 2004
Anodonta wahlamatensis	*cox1*	AY493471	Mock *et al.*, 2004
Cristaria plicata clessini	*cox1*	UAUC3361	Japan
Cyprogenia stegaria	*cox1*	AY654992	Campbell *et al.*, 2005
Cyrtonaias tampicoensis	16S	AY655032	Campbell *et al.*, 2005
Cyrtonaias tampicoensis	*cox1*	AF231749	Bogan and Hoeh, 2000
Cyrtonaias tampicoensis	ITS1	UAUC314	Lake Corpus Christi, Live Oak Co. TX
Cyrtonaias tampicoensis	*nadh1*	AY655090	Campbell *et al.*, 2005
Dromus dromas	*cox1*	AY654993	Campbell *et al.*, 2005
Ellipsaria lineolata	*cox1*	AY654994	Campbell *et al.*, 2005
Elliptio arca	*cox1*	AY654995	Campbell *et al.*, 2005
Elliptio arca BW	ITS1	UAUC501	Hurricane Ck. near FS Rd. 242, Winston Co. AL
Elliptio arca Coosa	ITS1	UAUC498	Oostanaula R. above Armuchee Ck., Floyd Co. GA
Elliptio arca Coosa 2	ITS1	UAUC503	Conasauga RM 43, Whitfield Co. GA
Elliptio arctata	*cox1*, ITS1	UAUC3496	Cahaba R., AL
Elliptio buckleyi	ITS1	UAUC3091	Wekiva R. Spring Run, Orange Co. FL

Elliptio complanata Chipola	ITS1	UAUC3489	Dry Creek at FL73, Jackson Co. FL
Elliptio complanata MD	ITS1	NCSM26964	NC State Museum
Elliptio complanata Savannah	ITS1	UAUC3448	Savannah R., HMc Sta. 854
Elliptio crassidens lower Coosa	*cox1*, ITS1	UAUC3150	between Wetumpka and Pipeline Shoals, Elmore Co. AL
Elliptio crassidens Cahaba	*cox1*	UAUC676	Cahaba R., CR 52 bridge, Shelby Co. AL
Elliptio crassidens Ohio	ITS1	UAUC3327	mouth of Sugar Creek, Braxton Co. WV
Elliptio crassidens Sipsey	ITS1	UAUC1169	Sipsey R., CR 2, Greene Co. AL
Elliptio crassidens Tenn	*cox1*, ITS1	UAUC3527	Diamond Island, Decatur/Hardin Co. TN
Elliptio dilatata	ITS1	UAUC2735	Alley Ford, Morgan Co. TN
Elliptio dilatata 1	*cox1*	AF231751	Bogan and Hoeh, 2000
Elliptio dilatata 2	*cox1*	AF156506	Graf and Ó Foighil, 2000
Elliptio dilatata 3	*cox1*	AF156507	Graf and Ó Foighil, 2000
Elliptio icterina Conecuh	ITS1	UAUC3438	Little Patsaliga Ck. at CR35, Crenshaw Co. AL
Elliptio icterina Conecuh 2	ITS1	UAUC3561	Gantt Lake at CR86, Covington Co. AL
Elliptio icterina Pea	*cox1*	UAUC3467	Pea R. CR 77, Pike/Barbour Co. AL
Elliptio icterina Pea	ITS1	UAUC3467	Pea R. CR 77, Pike/Barbour Co. AL
Elliptio icterina Pea 2	ITS1	UAUC1829	Pea R. CR 77, Barbour Co. AL
Elliptio icterina Pea 3	ITS1	UAUC3093	Pea R. CR 44, Pike/Barbour Co. AL
Elliptio icterina Savannah	ITS1	UAUC3494	Savannah R. RM150.9, Barnwell Co. SC
Elliptio mcmichaeli	*cox1*	UAUC3516	Pea R., AL87, Geneva Co. AL
Elliptio mcmichaeli	ITS1	UAUC3410	Pea R., AL87, Geneva Co. AL
Elliptio mcmichaeli 2	*cox1*, ITS1	UAUC3088	Choctawhatchee R., US Hwy 90, Holmes Co. FL
Elliptio purpurella? Loblockee	*cox1*, ITS1	A56Auburn	Loblockee Creek
Elliptio purpurella? Uchee	ITS1	A57Auburn	Uchee Creek tributary
Elliptio species rayed Chipola	ITS1	UAUC3571	Cowarts Creek @CR55, Houston Co. AL
Elliptio species unrayed Chipola	ITS1	UAUC3572	Cowarts Creek @CR55, Houston Co. AL
Elliptoideus sloatianus	*cox1*	AY613822	Campbell *et al.*, 2005
Epioblasma brevidens	*cox1*	AF156527	Graf and Ó Foighil, 2000
Epioblasma capsaeformis	*cox1*	AY654996	Campbell *et al.*, 2005
Epioblasma rangiana	*cox1*	EbVT	Virginia Tech
Epioblasma triquetra	*cox1*	AF156528	Graf and Ó Foighil, 2000
Fusconaia askewi	*cox1*	UAUC3395	Drakes Creek, Vernon Pa. LA
Fusconaia barnesiana	16S	AY655038	Campbell *et al.*, 2005
Fusconaia barnesiana	*cox1*	AY613822	Campbell *et al.*, 2005
Fusconaia barnesiana	*nadh1*	AY613791	Campbell *et al.*, 2005
Fusconaia cerina 1	16S	AY655039	Campbell *et al.*, 2005
Fusconaia cerina 1	*cox1*	AY613823	Campbell *et al.*, 2005
Fusconaia cerina 1	*nadh1*	AY655095	Campbell *et al.*, 2005
Fusconaia cerina 2	*cox1*	AF049522	Roe and Lydeard, 1998
Fusconaia cerina 2	*nadh1*	AY613792	Campbell *et al.*, 2005
Fusconaia cerina Louisiana	ITS1	UAUC3376	Twelvemile Creek at Rt1045, St. Helena Pa., LA
Fusconaia cor	16S	AY655040	Campbell *et al.*, 2005
Fusconaia cor	*cox1*	AY654997	Campbell *et al.*, 2005
Fusconaia cor	*nadh1*	AY655096	Campbell *et al.*, 2005
Fusconaia cuneolus	*cox1*	AY654998	Campbell *et al.*, 2005
Fusconaia cuneolus	*nadh1*	AY655097	Campbell *et al.*, 2005

Fusconaia ebena	16S	AF232790	Lydeard *et al.*, 2000
Fusconaia ebena	*cox1*	AY654999	Campbell *et al.*, 2005
Fusconaia ebena	ITS1	UAUC3149	between Wetumpka & Pipeline Shoals, Elmore Co. AL
Fusconaia ebena	*nadh1*	AY655098	Campbell *et al.*, 2005
Fusconaia escambia	ITS1	UAUC3403	Little Patsaliga Ck. at CR35, Crenshaw Co. AL
Fusconaia escambia 1	16S	AF232791	Lydeard *et al.*, 2000
Fusconaia escambia 1	*cox1*	AF232816	Lydeard *et al.*, 2000
Fusconaia escambia 1	*nadh1*	UAUC3403	Little Patsaliga Ck. at CR35, Crenshaw Co. AL
Fusconaia escambia 2	16S	AY655041	Campbell *et al.*, 2005
Fusconaia escambia 2	*cox1*	AF232817	Lydeard *et al.*, 2000
Fusconaia flava	*cox1*	AF156510	Graf and Ó Foighil, 2000
Fusconaia flava	ITS1	UAUC146	Ohio R., RM625, Jefferson/Harrison Co. KY/IN
Fusconaia flava 1	16S	AY238481	Krebs *et al.*, 2003
Fusconaia flava 1	*cox1*	AF231733	Bogan and Hoeh, 2000
Fusconaia flava 1	*nadh1*	AY613793	Campbell *et al.*, 2005
Fusconaia flava 2	16S	AY655042	Campbell *et al.*, 2005
Fusconaia flava 2	*cox1*	AF232822	Lydeard *et al.*, 2000
Fusconaia flava 2	*nadh1*	AY158781	Serb *et al.*, 2003
Fusconaia flava Missouri	*cox1*	UAUC2648	Mississippi R., Marion Co. MO
Fusconaia masoni	*cox1*	masoniNCSMH	NC State Museum
Fusconaia ozarkensis	*cox1*	UAUC3500	Bull Creek, pool 1/4 mi E Hwy 160, Taney Co. MO
Fusconaia subrotunda	16S	AY655043	Campbell *et al.*, 2005
Fusconaia subrotunda	*cox1*	AY613824	Campbell *et al.*, 2005
Fusconaia subrotunda	*cox1*	AY613824	Campbell *et al.*, 2005
Fusconaia subrotunda	*nadh1*	AY613794	Campbell *et al.*, 2005
Fusconaia succissa 1	16S	AF232794	Lydeard *et al.*, 2000
Fusconaia succissa 1	*cox1*	AF232819	Lydeard *et al.*, 2000
Fusconaia succissa 1	*nadh1*	AY158792	Serb *et al.*, 2003
Fusconaia succissa 2	16S	AF232795	Lydeard *et al.*, 2000
Fusconaia succissa 2	*cox1*	AF232820	Lydeard *et al.*, 2000
Fusconaia succissa 2	*nadh1*	AY158809	Serb *et al.*, 2003
Glebula rotundata	*cox1*	AF231729	Bogan and Hoeh, 2000
Gonidea angulata	*cox1*	AF231755	Bogan and Hoeh, 2000
Hemistena lata	16S	AY655046	Campbell *et al.*, 2005
Hemistena lata	*cox1*	AY613825	Campbell *et al.*, 2005
Hemistena lata	ITS1	UAUC2797	Frost Ford, RM 181.2, Hancock Co. TN
Hemistena lata	*nadh1*	AY613796	Campbell *et al.*, 2005
Hyriopsis cumingii	*cox1*	AY655000	Campbell *et al.*, 2005
Inversidens japanensis	*cox1*	AB055625	Okazaki and Ueshima, unpub
Lampsilis abrupta	*cox1*	UAUC3531	Diamond Island, Decatur/Hardin Co. TN
Lampsilis altilis 1	*cox1*	AF385105	Roe *et al.*, 2001
Lampsilis australis 1	*cox1*	AF385101	Roe *et al.*, 2001
Lampsilis cardium	*cox1*	AF120653	Giribet and Wheeler, 2002
Lampsilis hydiana	*cox1*	UAUC3508	Neches R., Rte 96 bridge, Hardin Co. TX
Lampsilis ornata 1	*cox1*	AY365193	Serb and Lydeard, 2003
Lampsilis ovata	*cox1*	AY613826	Campbell *et al.*, 2005
Lampsilis perovalis 1	*cox1*	AF385094	Roe *et al.*, 2001
Lampsilis siliquoidea 1	*cox1*	AF156521	Graf and Ó Foighil, 2000
Lampsilis straminea	*cox1*	UAUC3543	Sipsey R. at Benevola Island, Greene Co. AL
Lampsilis subangulata 1	*cox1*	AF385104	Roe *et al.*, 2001

Lampsilis teres 1	*cox1*	AF385113	Roe *et al.*, 2001
Lasmigona complanata	*cox1*	AF093845	King *et al.*, 1999
Lasmigona compressa MN	*cox1*	UAUC3519	3 mi W Milaca, CR140 bridge, Mille Lacs Co. MN
Lasmigona compressa1	*cox1*	AF093846	King *et al.*, 1999
Lasmigona compressa2	*cox1*	AF093847	King *et al.*, 1999
Lasmigona compressa3	*cox1*	AF156503	Graf and Ó Foighil, 2000
Lasmigona costata	*cox1*	AF093848	King *et al.*, 1999
Lasmigona costata	ITS1	UAUC3245	Venable Spring, Marshall Co., TN
Lasmigona decorata	*cox1*	AF093849	King *et al.*, 1999
Lasmigona etowahensis	ITS1	UAUC3433	Poplar Spring Creek, Whitfield Co., GA
Lasmigona etowahensis 1 2	*cox1*	UAUC3159	Campbell *et al.*, 2005
Lasmigona etowahensis3	*cox1*	UAUC3425	W. Fork Armuchee Creek, Walker Co., GA
Lasmigona holstonia 1	*cox1*, ITS1	B347	Bluestone R., Tazewell Co., VA
Lasmigona holstonia 2	*cox1*, ITS1	B348	Bluestone R., Tazewell Co., VA
Lasmigona holstonia 3	ITS1	B349	Bluestone R., Tazewell Co., VA
Lasmigona holstonia 4	*cox1*, ITS1	B350	Wolf Creek, Bland Co., VA
Lasmigona holstonia 5	*cox1*, ITS1	B351	Wolf Creek, Bland Co., VA
Lasmigona holstonia 6	ITS1	B352	Wolf Creek, Bland Co., VA
Lasmigona holstonia 7	ITS1	B353	Wolf Creek, Bland Co., VA
Lasmigona holstonia 8	ITS1	B354	Wolf Creek, Bland Co., VA
Lasmigona holstonia 9	*cox1*, ITS1	B355	Station Spring Creek, Tazewell Co., VA
Lasmigona holstonia 10	*cox1*, ITS1	B356	Station Spring Creek, Tazewell Co., VA
Lasmigona holstonia 11	ITS1	B357	Station Spring Creek, Tazewell Co., VA
Lasmigona holstonia 12	ITS1	B358	Station Spring Creek, Tazewell Co., VA
Lasmigona holstonia 13	ITS1	B359	Station Spring Creek, Tazewell Co., VA
Lasmigona holstonia 14	*cox1*, ITS1	B360	North Fork Clinch, Tazewell Co., VA
Lasmigona holstonia 15	*cox1*, ITS1	B361	North Fork Clinch, Tazewell Co., VA
Lasmigona holstonia 16	ITS1	B362	North Fork Clinch, Tazewell Co., VA
Lasmigona holstonia 17	ITS1	B363	North Fork Clinch, Tazewell Co., VA
Lasmigona holstonia 18	ITS1	B364	North Fork Clinch, Tazewell Co., VA
Lasmigona holstonia 19	*cox1*, ITS1	B365	North Fork Clinch, Tazewell Co., VA
Lasmigona holstonia 20	ITS1	B366	Maiden Spring Creek, Tazewell Co., VA
Lasmigona holstonia 21	ITS1	B367	Maiden Spring Creek, Tazewell Co., VA
Lasmigona holstonia 22	ITS1	B368	Maiden Spring Creek, Tazewell Co., VA
Lasmigona holstonia 23	*cox1*, ITS1	B369	Maiden Spring Creek, Tazewell Co., VA
Lasmigona holstonia Caney Fork	ITS1	UAUC3165	Collins R. at Hwy 56, Grundy Co., TN
Lasmigona holstonia Holston	ITS1	UAUC3189	Beech Creek near Light Mill, Hawkins Co., TN
Lasmigona subviridis	ITS1	AF093838	King *et al.*, 1999
Lasmigona subviridis 2	ITS1	AF091331	King *et al.*, 1999
Lasmigona subviridis1	*cox1*	AF091330	King *et al.*, 1999
Lasmigona subviridis2	*cox1*	AF093850	King *et al.*, 1999
Lasmigona subviridis3	*cox1*	AF093851	King *et al.*, 1999
Lemiox rimosus	*cox1*	AY655002	Campbell *et al.*, 2005
Leptodea fragilis 1	*cox1*	AF049518	Roe and Lydeard, 1998
Leptodea leptodon	*cox1*	AY655003	Campbell *et al.*, 2005
Lexingtonia dolabelloides	16S	AY655051	Campbell *et al.*, 2005
Lexingtonia dolabelloides	*cox1*	AY655004	Campbell *et al.*, 2005
Lexingtonia dolabelloides	ITS1	AY772175	Grobler *et al.*, 2006
Lexingtonia dolabelloides	*nadh1*	AY613798	Campbell *et al.*, 2005

Ligumia nasuta	*cox1*	AF156515	Graf and Ó Foighil, 2000
Ligumia recta	*cox1*	AF385110	Roe *et al.*, 2001
Megalonaias nervosa	*cox1*	AY655007	Campbell *et al.*, 2005
Nodularia douglasiae nipponensis	*cox1*	UAUC3363	Japan
Obliquaria reflexa	*cox1*	AY655008	Campbell *et al.*, 2005
Obovaria jacksoniana	*cox1*	AY655009	Campbell *et al.*, 2005
Obovaria olivaria	*cox1*	AF232812	Lydeard *et al.*, 2000
Obovaria rotulata	*cox1*	AF232814	Lydeard *et al.*, 2000
Obovaria subrotunda	*cox1*	AY655010	Campbell *et al.*, 2005
Obovaria unicolor	*cox1*	AF232811	Lydeard *et al.*, 2000
Plectomerus dombeyanus	16S	AY655057	Campbell *et al.*, 2005
Plectomerus dombeyanus	*cox1*	AY655011	Campbell *et al.*, 2005
Plectomerus dombeyanus	ITS1	UAUC2536	2.7 mi downstream of Jordan Dam, Elmore Co. AL
Plectomerus dombeyanus	*nadh1*	AY655110	Campbell *et al.*, 2005
Plethobasus cyphus	16S	AY655058	Campbell *et al.*, 2005
Plethobasus cyphus	*cox1*	AY613828	Campbell *et al.*, 2005
Plethobasus cyphus	ITS1	UAUC3157	Frost Ford, RM 181.2, Hancock Co. TN
Plethobasus cyphus	*nadh1*	AY613799	Campbell *et al.*, 2005
Pleurobema chattanoogaense	*cox1*	AY655012	Campbell *et al.*, 2005
Pleurobema clava	16S	AY655060	Campbell *et al.*, 2005
Pleurobema clava	*cox1*	AY655013	Campbell *et al.*, 2005
Pleurobema clava	ITS1	UAUC1477	Kennerdell and Clear Ck. SP, Venango Co. PA
Pleurobema clava	*nadh1*	AY613802	Campbell *et al.*, 2005
Pleurobema collina	16S	AY655061	Campbell *et al.*, 2005
Pleurobema collina	*cox1*	AY613830	Campbell *et al.*, 2005
Pleurobema collina	ITS1	UAUC1074	Wards Creek, CR 665, Albemarle Co. VA
Pleurobema collina	*nadh1*	AY613803	Campbell *et al.*, 2005
Pleurobema cordatum	*cox1*	AY613831	Campbell *et al.*, 2005
Pleurobema cordatum	ITS1	UAUC3530	Diamond Island, Decatur/Hardin Co. TN
Pleurobema cordatum	*nadh1*	AY613804	Campbell *et al.*, 2005
Pleurobema decisum 1	*cox1*	AF232801	Lydeard *et al.*, 2000
Pleurobema furvum	*cox1*	AY613833	Campbell *et al.*, 2005
Pleurobema georgianum	*cox1*	AY613834	Campbell *et al.*, 2005
Pleurobema gibberum	16S	AY655064	Campbell *et al.*, 2005
Pleurobema gibberum	*cox1*	AY613835	Campbell *et al.*, 2005
Pleurobema gibberum	*nadh1*	AY613808	Campbell *et al.*, 2005
Pleurobema hanleyianum 1	*cox1*	AY655016	Campbell *et al.*, 2005
Pleurobema oviforme 1	*cox1*	AY655017	Campbell *et al.*, 2005
Pleurobema perovatum	*cox1*	AY613838	Campbell *et al.*, 2005
Pleurobema pyriforme	*cox1*	AY613839	Campbell *et al.*, 2005
Pleurobema rubellum	*cox1*	AY613840	Campbell *et al.*, 2005
Pleurobema sintoxia	*cox1*	AY655019	Campbell *et al.*, 2005
Pleurobema stabile	*cox1*	UAUC3197	Conasauga R. below US 76, Whitfield/Murray Co. GA
Pleurobema taitianum	*cox1*	AY613844	Campbell *et al.*, 2005
Pleurobema troschelianum	*cox1*	AY613845	Campbell *et al.*, 2005
Popenaias popeii	*cox1*	AY655020	Campbell *et al.*, 2005
Popenaias popeii	ITS1	A48	Rio Grande, Laredo, Webb Co. TX
Potamilus alatus	*cox1*	AF049510	Roe and Lydeard, 1998
Potamilus purpuratus	*cox1*	AF049507	Roe and Lydeard, 1998

Pseudanodonta complanata	*cox1*	DQ060173	Kallersjo *et al.*, 2005
Pseudanodonta complanata	ITS1	AJ295289	Gerke and Tiedemann, 2001
Psilunio littoralis	*cox1*	AF303348	Machordom *et al.*, 2003
Psilunio littoralis	*cox1*	AF303348	Machordom *et al.*, 2003
Ptychobranchus fasciolaris	*cox1*	AF156514	Graf and Ó Foighil, 2000
Pyganodon grandis	*cox1*	AF156504	Graf and Ó Foighil, 2000
Pyganodon grandis	ITS1	AY319385	Campbell *et al.*, unpublished
Quadrula quadrula 1	16S	AY238485	Krebs *et al.*, 2003
Quadrula quadrula 1	*cox1*	AF231757	Bogan and Hoeh, 2000
Quadrula quadrula 1	*nadh1*	AY158790	Serb *et al.*, 2003
Quincuncina burkei 1	16S	AF2327779	Lydeard *et al.*, 2000
Quincuncina burkei 1	*cox1*	AF232804	Lydeard *et al.*, 2000
Quincuncina burkei 2	16S	AF2327779	Lydeard *et al.*, 2000
Quincuncina burkei 2	*cox1*	AF232803	Lydeard *et al.*, 2000
Quincuncina burkei 2	*nadh1*	AY158793	Serb *et al.*, 2003
Quincuncina infucata 1	16S	AF232782	Lydeard *et al.*, 2000
Quincuncina infucata 1	*cox1*	AF232807	Lydeard *et al.*, 2000
Quincuncina infucata 1	*nadh1*	AY655121	Campbell *et al.*, 2005
Quincuncina infucata 2	16S	AF232781	Lydeard *et al.*, 2000
Quincuncina infucata 2	*cox1*	AF232806	Lydeard *et al.*, 2000
Quincuncina kleiniana 1	16S	AF232783	Lydeard *et al.*, 2000
Quincuncina kleiniana 1	*cox1*	AF232808	Lydeard *et al.*, 2000
Quincuncina kleiniana 1	*nadh1*	AY158795	Serb *et al.*, 2003
Quincuncina kleiniana 2	16S	AF232784	Lydeard *et al.*, 2000
Quincuncina kleiniana 2	*cox1*	AF232809	Lydeard *et al.*, 2000
Sinanodonta calipygos	*cox1*	UAUC3360	Japan
Strophitus connasaugaensis	*cox1*, ITS1	UAUC3434	above Lower Kings/Norton Bridge, Murray/Whitfield Co. GA
Strophitus connasaugaensis alabamensis	ITS1	UAUC3177	Shoal Ck., near FS 500 bridge, Cleburne Co. AL
Strophitus connasaugaensis BW	ITS1	UAUC1683	Brushy Ck, FS Rd 254, Winston Co. AL
Strophitus subvexus	*cox1*	AY655021	Campbell *et al.*, 2005
Strophitus subvexus	ITS1	UAUC2715	Sucarnoochie Creek, Old Scooba Crossing, Kemper Co. MS
Strophitus undulatus	ITS1	UAUC2756	Big South Fork at Station Camp Creek, Scott Co. TN
Strophitus undulatus long	ITS1	UAUC2756	Big South Fork at Station Camp Creek, Scott Co. TN
Strophitus undulatus long part2	ITS1	UAUC2756	Big South Fork at Station Camp Creek, Scott Co. TN
Strophitus undulatus1	*cox1*	AF231740	Bogan and Hoeh, 2000
Strophitus undulatus2	*cox1*	AF093839	King *et al.*, 1999
Strophitus undulatus3	*cox1*	AF156505	Graf and Ó Foighil, 2000
Toxolasma corvunculus	*cox1*, ITS1	UAUC3465	Choctafaula Creek near CR54, Macon Co. AL
Toxolasma corvunculus 2	ITS1	UAUC3466	Opintlocco Creek near CR43 crossing, Macon Co. AL
Toxolasma cylindrellus	*cox1*, ITS1	UAUC3341	Estill Fork at end CR 175, Jackson Co. AL
Toxolasma lividus	*cox1*	AF231756	Bogan and Hoeh, 2000
Toxolasma lividus	ITS1	UAUC3340	Estill Fork at end CR 175, Jackson Co. AL
Toxolasma mearnsi	*cox1*	UAUC81	Lake Corpus Christi, Live Oak Co. TX
Toxolasma parvus BW	*cox1*	UAUC3575	South Needham Creek, Greene Co. AL
Toxolasma parvus Coosa	ITS1	UAUC3449	ponds at TARI, Whitfield Co. GA
Toxolasma parvus Pearl	ITS1	UAUC1274	Bogue Chitto, below sill, St. Tammany Pa. LA

Toxolasma parvus Tennessee	ITS1	UAUC3331	Mallard Point, Morgan Co. AL
Toxolasma parvus TN	*cox1*	AY655022	Campbell *et al.*, 2005
Toxolasma paulus	ITS1	UAUC261	Sawhatchee Ck.,SR 371, Early Co. GA
Toxolasma paulus Chatt	ITS1	UAUC3554	Chattahoochee RM118, Russell/Stewart Co. AL/GA
Toxolasma pullus	ITS1	UAUC571	Flat Tub Landing, Coffee Co. GA
Toxolasma species Choctawhatchee	ITS1	UAUC3556	Wrights Creek, FL 179, Holmes Co. FL
Toxolasma species Escambia	*cox1*	UAUC878	Panther Ck., Rt. 106, Butler Co. AL
Toxolasma species Escambia 2	*cox1*	UAUC3550	Point A lake, just below US 29, Covington Co. AL
Toxolasma species Escambia 3550	ITS1	UAUC3550	Point A lake, just below US 29, Covington Co. AL
Toxolasma species Escambia 3550 allele2	ITS1	UAUC3550	Point A lake, just below US 29, Covington Co. AL
Toxolasma texasensis	ITS1	UAUC80	Giddings State School Lake, Lee Co. TX
Toxolasma texasiensis	*cox1*	AY655023	Campbell *et al.*, 2005
Tritogonia verrucosa	*cox1*	AY655024	Campbell *et al.*, 2005
Truncilla truncata	*cox1*	AF156513	Graf and Ó Foighil, 2000
Unio crassus	*cox1*	DQ060174	Kallersjo *et al.*, 2005
Unio pictorum	*cox1*	DQ060175	Kallersjo *et al.*, 2005
Unio tumidus	*cox1*	DQ060176	Kallersjo *et al.*, 2005
Uniomerus declivus	*cox1*	AY613846	Campbell *et al.*, 2005
Venustaconcha ellipsiformis	*cox1*	AY655026	Campbell *et al.*, 2005
Venustaconcha pleasii	*cox1*	UAUC3520, 3525	Beaver Ck., T25N R17W S12, Douglas Co. MO
Villosa iris	*cox1*	AF156524	Graf and Ó Foighil, 2000
Villosa sima	*cox1*	UAUC3213	Witty Cr., South Prong Barren Fork, Bridge off Herman Lange Rd, Warren Co. TN
Villosa taeniata	*cox1*	UAUC2757	Big South Fork at Station Camp Creek, Scott Co. TN
Villosa trabalis	*cox1*	UAUC2723	Big South Fork, Parch Corn Creek, Scott Co. TN
Villosa villosa	*cox1*	AF385109	Roe *et al.*, 2001
Elimia ampla	*cox1*	Elimia1	Cahaba R. at Shelby CR 52 in Helena (CAH13)
Elimia ampla 2	*cox1*	Elimia1B	Cahaba R. above Marvel slab - 2nd shoal (CAH7)
Elimia annettae	*cox1*	Elimia2	Cahaba R., site above Marvel (CAH10)
Elimia annettae 2	*cox1*	Elimia2B	Cahaba R. at U.S. Hwy 280 dam (CAH18)
Elimia annettae 3	*cox1*	Elimia2C	Cahaba R. behind Hoover High School, off AL Hwy. 150 (CAH14)
Elimia bellacrenata	*cox1*	Elimia11	Orr Park, Montevallo
Elimia bullula	*cox1*	a	Sides, 2005
Elimia bullula 2	*cox1*	a	Yellowleaf Creek
Elimia cahawbensis	*cox1*	Elimia5	Cahaba R. at Shelby CR 52 in Helena (CAH13)
Elimia carinifera	*cox1*	Elimia6	Mud Creek, Tannehill, Tuscaloosa County
Elimia carinocostata	*cox1*	Elimia7	Cahaba R. at Grants Mill Road, ~300 yds. upstream of bridge (CAH16)
Elimia catenaria dislocata	*cox1*	AY063469	Dillon and Frankis, 2004
Elimia clara	*cox1*	Elimia8	Cahaba R. at Pratt's Ferry - Bibb CR 26 (CAH5)
Elimia cochliaris	*cox1*	Elimia12	spring west of CR10 crossing of Little Cahaba R.
Elimia cochliaris Buck	*cox1*	Elimia12B	Buck Creek
Elimia dickinsoni	*cox1*	Pleurocera5	Cowarts Creek, Houston Co. AL
Elimia hydei	*cox1*	AF435775	Minton and Lydeard, 2003
Elimia laqueata laqueata	*cox1*	JB827a	Sides, 2005

Elimia modesta "murrayensis"	*cox1*	E211	Swamp Creek, Redwine Rd., Whitfield Co. GA
Elimia olivula	*cox1*	Elimia9	Cahaba R., 1st large shoal below Booth's Ford (CAH9)
Elimia proxima race A	*cox1*	AY063464	Dillon and Frankis, 2004
Elimia proxima race B	*cox1*	AY063465	Dillon and Frankis, 2004
Elimia proxima race C variant 1	*cox1*	AY063466	Dillon and Frankis, 2004
Elimia proxima race C variant 2	*cox1*	AY063467	Dillon and Frankis, 2004
Elimia semicarinata	*cox1*	AY063468	Dillon and Frankis, 2004
Elimia showalteri	*cox1*	Elimia10	Cahaba R., 1st large shoal below Booth's Ford (CAH9)
Elimia showalteri2	*cox1*	Pleurocera1	Bibb County Glades
Elimia striatula	*cox1*	E111	Ponds at TARI
Elimia striatula JS	*cox1*	JDS02 1b	Sides, 2005
Elimia taitiana	*cox1*	JDS01 10c	Sides, 2005
Elimia varians	*cox1*	Elimia3	Cahaba R., site above Marvel (CAH10)
Elimia varians 2	*cox1*	Elimia3B	Cahaba R. at Centreville (CAH4)
Elimia varians 3	*cox1*	Elimia3C	Cahaba R., 1st large shoal below Booth's Ford (CAH9)
Elimia variata	*cox1*	Elimia4	Cahaba R. at Shelby CR 52 in Helena (CAH13)
Elimia variata 2	*cox1*	Elimia4B	Cahaba R. at U.S. Hwy 280 dam (CAH18)
Elimia virginica	*cox1*	Ev	Susquehanna R. above Harrisburg, PA
Io fluvialis	*cox1*	Io1	Nolichucky R.
Io fluvialis	*cox1*	AF435777	Minton and Lydeard, 2003
Juga acutifilosa	*cox1*	5834	Shoat Spring, California
Leptoxis ampla	*cox1*	AF469644	Minton *et al*., 2005
Leptoxis praerosa	*cox1*	AF435779	Minton and Lydeard, 2003
Lithasia armigera	*cox1*	AF435743	Minton and Lydeard, 2003
Lithasia armigera	*cox1*	AF469638	Minton *et al*., 2005
Pleurocera alveare	*cox1*	Pleurocera2	Tennessee R., Wheeler Dam tailwaters
Pleurocera annulifera	*cox1*	JDS00 24a	Sides, 2005
Pleurocera brumbyi	*cox1*	Pleurocera6	Spring Creek at Hook Road, Tuscumbia
Pleurocera canaliculata	*cox1*	Pleurocera4	Tennessee R., Wheeler Dam tailwaters
Pleurocera canaliculata 2	*cox1*	Pleurocera7	Tennessee R., Mussel Camp Road east of Decatur
Pleurocera canaliculata fila	*cox1*	JDS02 11b	Sides, 2005
Pleurocera canaliculata undulata	*cox1*	JB827b	Sides, 2005
Pleurocera chickasahaense	*cox1*	JDS00 8b	Sides, 2005
Pleurocera curta roanense	*cox1*	JDS02 7c	Sides, 2005
Pleurocera foremani	*cox1*	JDS02 12a	Sides, 2005
Pleurocera prasinata	*cox1*	JDS00 18a	Sides, 2005
Pleurocera pyrenella	*cox1*	Pleurocera3	Limestone Creek at US72, Limestone Co. AL
Pleurocera pyrenella	*cox1*	JDS01 19c	Sides, 2005
Pleurocera uncialis hastata	*cox1*	JDS02 10a	Sides, 2005
Pleurocera uncialis uncialis	*cox1*	JDS02 10d	Sides, 2005
Pleurocera vestita Cahaba	*cox1*	JDS00 5a	Sides, 2005
Pleurocera viridula	*cox1*	JDS02 5a	Sides, 2005
Pleurocera walkeri	*cox1*	JDS01 16a	Sides, 2005
Semisulcospira reticulata	*cox1*	Sret	Japan

Table 2. Range of percent differences for target unionid taxa. n.a.=not available. “ITS1, no indels” means that the percent was calculated by treating gaps as unknown. Two sequences that

differed only in one having more or fewer bases than the other would have a 0% difference. “ITS1, gaps as 5th base” means that the percent was calculated treating gaps as a fifth option, along with AGTC. Two sequences that differed only in one having more or fewer bases than the other would have a difference greater than zero.

Species/population	*cox1*	ITS1, no indels	Other
Elliptio arca Coosa-*E. arca* western Mobile	n.a.	0.00	0.00 ITS1, gaps as 5th base
Elliptio arca-*Elliptio arctata*	4.81	1.39	2.10 ITS1, gaps as 5th base
Elliptio arctata-*E. crassidens*	3.49-3.81	0.00	0.00-0.20 ITS1, gaps as 5th base
Elliptio complanata/icterina group Gulf-*E. complanata/icterina* Atlantic	n.a.	0.00-2.58	0.32-3.72 ITS1, gaps as 5th base
Elliptio complanata/icterina group Gulf-*E. complanata/icterina* group Gulf	0.75	0.00-1.81	0.00-2.58 ITS1, gaps as 5th base
Elliptio crassidens-*E. mcmichaeli*	0.95-1.43	0.00-0.60	0.00-0.97 ITS1, gaps as 5th base
Elliptio crassidens-*E. crassidens*	0.48-0.63	0.00-0.40	0.16-0.97 ITS1, gaps as 5th base
Elliptio dilatata-other *Elliptio*	6.53-10.26	1.78-3.78	2.11-4.86 ITS1, gaps as 5th base
Elliptio mcmichaeli-*E. mcmichaeli*	1.62	0.20	0.32 ITS1, gaps as 5th base
Fusconaia escambia-*Quincuncina burkei*	n.a.	n.a.	5.30-5.90 *cox1*, 16S, and *nadh1*
Lasmigona holstonia New/upper Tennessee-*L. holstonia* mid-Tennessee	n.a.	0.36	n.a.
Lasmigona holstonia New-*L. holstonia* upper Tennessee	0.00-0.31	0.00	n.a.
Lasmigona holstonia Tennessee/New-*L. “holstonia”* Coosa	13.44-13.96	0.91	n.a.
Strophitus “connasaugaensis” Black Warrior-*S. subvexus*	n.a.	0.00	n.a.
Strophitus connasaugaensis upper Coosa-middle Coosa	n.a.	0.36	n.a.
Strophitus connasaugaensis upper Coosa-*S. subvexus*	6.24	1.24	n.a.
Toxolasma corvunculus-*T. parvus*	8.75	0.97-0.98	n.a.
Toxolasma corvunculus-*T.* species Escambia	3.87-4.19	0.19	n.a.
Toxolasma cylindrellus-*T. lividus*	7.02	0.97	n.a.
Toxolasma parvus-*T. “paulus”*	n.a.	0.98-1.55	n.a.
Toxolasma parvus-*T.* species Choctawhatchee	n.a.	1.37-1.40	n.a.
Toxolasma parvus-*T.* species Escambia	9.73-9.90	1.17-1.19	n.a.
Toxolasma “paulus” 1-*T.* species Choctawhatchee	n.a.	0.40	n.a.
Toxolasma “paulus” 2-*T.* species Choctawhatchee	n.a.	1.61	n.a.
Toxolasma “paulus”-*T. “paulus”*	n.a.	1.00	n.a.
Toxolasma species Escambia-*T.* species Choctawhatchee	n.a.	1.31-1.32	n.a.

Table 3. Percent differences for priority *Elimia* species

	Elimia ampla	*Elimia ampla 2*	*Elimia annettae*	*Elimia annettae 2*	*Elimia annettae 3*	*Elimia bellacrenata*	*Elimia bullula JS*	*Elimia bullula*	*Elimia cahawbensis*	*Elimia carinifera*
Elimia ampla	0.00%	0.60%	7.07%	15.08%	7.02%	6.17%	5.72%	7.31%	7.21%	7.35%
Elimia ampla 2	0.60%	0.00%	6.84%	15.27%	6.84%	6.62%	4.34%	7.14%	6.99%	7.45%
Elimia annettae	7.07%	6.84%	0.00%	15.12%	0.91%	2.10%	1.08%	0.91%	3.19%	3.19%
Elimia annettae 2	15.08%	15.27%	15.12%	0.00%	14.82%	14.55%	12.51%	15.28%	14.97%	15.27%
Elimia annettae 3	7.02%	6.84%	0.91%	14.82%	0.00%	2.34%	0.00%	0.89%	3.50%	3.50%
Elimia bellacrenata	6.17%	6.62%	2.10%	14.55%	2.34%	0.00%	1.50%	2.34%	2.84%	2.82%
Elimia cochliaris	10.68%	10.90%	10.73%	15.26%	10.56%	10.32%	10.10%	11.15%	11.05%	11.17%
Elimia cochliaris Buck	10.36%	10.96%	11.27%	14.83%	11.10%	11.36%	11.42%	11.69%	11.42%	11.73%
Elimia varians	17.17%	17.48%	16.41%	17.26%	16.42%	16.08%	12.31%	16.42%	16.72%	16.57%
Elimia varians 2	16.71%	16.57%	16.87%	14.98%	17.03%	17.54%	12.27%	17.03%	17.33%	17.02%
Elimia varians 3	16.31%	16.55%	16.25%	16.49%	15.68%	16.12%	13.18%	15.83%	16.41%	16.71%
Elimia variata	0.00%	0.61%	7.14%	15.27%	7.14%	6.92%	5.34%	7.45%	7.30%	7.45%

	Elimia ampla	*Elimia ampla 2*	*Elimia annettae*	*Elimia annettae 2*	*Elimia annettae 3*	*Elimia bellacrenata*	*Elimia bullula JS*	*Elimia bullula*	*Elimia cahawbensis*	*Elimia carinifera*
Elimia variata 2	0.36%	0.74%	7.18%	15.34%	7.12%	6.51%	5.70%	7.41%	7.31%	7.44%
Pleurocera alveare	9.27%	8.96%	8.93%	14.35%	8.85%	8.40%	7.83%	9.43%	9.40%	9.23%
Pleurocera pyrenella	9.40%	9.69%	9.65%	15.23%	9.54%	8.53%	7.15%	10.12%	10.11%	9.94%

	Elimia carinocostata	*Elimia catenaria dislocata*	*Elimia clara*	*Elimia cochliaris*	*Elimia cochliaris* Buck	*Elimia dickinsoni*	*Elimia hydei*	*Elimia laqueata laqueata* JS	*Elimia modesta "murrayensis"*	*Elimia olivula*
Elimia ampla	4.65%	11.52%	17.17%	10.68%	10.36%	11.25%	11.48%	10.42%	4.67%	1.38%
Elimia ampla 2	5.02%	11.55%	17.48%	10.90%	10.96%	12.14%	13.13%	10.63%	5.45%	1.67%
Elimia annettae	5.78%	11.85%	16.41%	10.73%	11.27%	10.32%	10.60%	11.46%	5.89%	7.30%
Elimia annettae 2	15.27%	15.11%	17.26%	15.26%	14.83%	16.50%	12.32%	10.88%	15.06%	15.12%
Elimia annettae 3	6.08%	11.86%	16.42%	10.56%	11.10%	10.49%	9.43%	10.42%	6.16%	7.14%
Elimia bellacrenata	5.99%	11.89%	16.08%	10.32%	11.36%	9.57%	9.34%	9.17%	6.32%	7.24%
Elimia cochliaris	10.87%	11.52%	16.75%	0.00%	2.78%	11.89%	11.13%	9.88%	11.41%	10.61%
Elimia cochliaris Buck	11.57%	12.16%	17.19%	2.78%	0.00%	12.20%	9.38%	10.76%	12.11%	10.65%
Elimia varians	16.57%	17.78%	0.00%	16.75%	17.19%	16.19%	13.58%	11.65%	16.38%	17.48%
Elimia varians 2	16.57%	17.93%	19.61%	16.74%	16.11%	17.38%	15.14%	12.58%	16.97%	17.02%
Elimia varians 3	15.95%	15.79%	19.76%	15.29%	16.27%	18.00%	14.05%	11.53%	15.39%	16.70%
Elimia variata	4.71%	11.70%	17.33%	10.61%	10.65%	11.99%	13.13%	9.75%	5.14%	1.37%
Elimia variata 2	4.76%	11.61%	17.25%	10.79%	10.62%	11.28%	11.47%	10.29%	4.66%	1.52%
Pleurocera alveare	8.93%	8.50%	16.41%	9.46%	11.41%	9.92%	9.58%	7.60%	9.76%	9.12%
Pleurocera pyrenella	9.36%	8.94%	17.28%	9.81%	12.41%	9.85%	9.33%	7.93%	9.82%	9.85%

	Elimia proxima race A	*Elimia proxima* race B	*Elimia proxima* race C variant 1	*Elimia proxima* race C variant 2	*Elimia semicarinata*	*Elimia showalteri*	*Elimia showalteri 2*	*Elimia striatula* JS	*Elimia striatula*	*Elimia taitiana* JS
Elimia ampla	12.01%	12.11%	15.43%	15.15%	12.87%	17.17%	17.17%	10.21%	9.91%	14.12%
Elimia ampla 2	12.46%	12.31%	15.35%	15.65%	13.37%	17.48%	17.48%	9.33%	9.28%	17.08%
Elimia annettae	12.01%	11.40%	15.05%	14.59%	11.40%	16.41%	16.41%	9.54%	9.70%	15.49%
Elimia annettae 2	16.64%	15.87%	17.86%	17.24%	15.12%	17.26%	17.26%	8.77%	14.07%	9.88%
Elimia annettae 3	11.86%	11.86%	14.59%	14.74%	10.95%	16.42%	16.42%	8.33%	9.73%	14.14%
Elimia bellacrenata	12.10%	11.73%	14.92%	14.31%	11.89%	16.08%	16.08%	8.52%	9.34%	11.85%
Elimia cochliaris	9.01%	8.96%	14.74%	15.19%	11.07%	16.75%	16.75%	8.77%	8.98%	12.75%
Elimia cochliaris Buck	10.33%	10.65%	15.35%	15.62%	12.17%	17.19%	17.19%	11.67%	10.82%	13.54%
Elimia varians	18.39%	17.78%	17.93%	20.06%	18.24%	0.00%	0.00%	11.85%	15.69%	15.23%
Elimia varians 2	17.48%	16.87%	18.69%	18.54%	16.87%	19.61%	19.61%	16.72%	17.04%	13.80%
Elimia varians 3	16.39%	16.39%	18.67%	18.36%	16.24%	19.76%	19.76%	10.28%	14.71%	13.20%
Elimia variata	12.16%	12.31%	15.35%	15.20%	13.07%	17.33%	17.33%	9.33%	9.28%	17.07%
Elimia variata 2	12.27%	12.35%	15.55%	15.13%	13.11%	17.25%	17.25%	10.16%	9.88%	14.12%
Pleurocera alveare	11.37%	11.19%	15.29%	14.18%	11.07%	16.41%	16.41%	7.43%	7.19%	12.57%
Pleurocera pyrenella	11.52%	11.63%	15.62%	14.37%	11.52%	17.28%	17.28%	7.64%	7.82%	12.40%

	Elimia varians	*Elimia varians 2*	*Elimia varians 3*	*Elimia variata*	*Elimia variata 2*	*Elimia virginica*	*Pleurocera alveare*	*Pleurocera annulifera JS*	*Pleurocera brumbyi*	*Pleurocera canaliculata 2*
Elimia ampla	17.17%	16.71%	16.31%	0.00%	0.36%	14.66%	9.27%	9.51%	17.21%	9.83%
Elimia ampla 2	17.48%	16.57%	16.55%	0.61%	0.74%	14.43%	8.96%	8.46%	18.07%	9.26%
Elimia annettae	16.41%	16.87%	16.25%	7.14%	7.18%	14.13%	8.93%	8.43%	17.59%	9.23%
Elimia annettae 2	17.26%	14.98%	16.49%	15.27%	15.34%	15.42%	14.35%	9.53%	19.42%	14.36%
Elimia annettae 3	16.42%	17.03%	15.68%	7.14%	7.12%	13.91%	8.85%	8.74%	17.82%	9.14%
Elimia bellacrenata	16.08%	17.54%	16.12%	6.92%	6.51%	14.33%	8.40%	7.87%	16.05%	8.75%
Elimia cochliaris	16.75%	16.74%	15.29%	10.61%	10.79%	12.95%	9.46%	9.25%	16.76%	9.81%
Elimia cochliaris Buck	17.19%	16.11%	16.27%	10.65%	10.62%	13.46%	11.41%	11.12%	18.20%	11.42%
Elimia varians	0.00%	19.61%	19.76%	17.33%	17.25%	19.76%	16.41%	12.33%	20.55%	16.41%
Elimia varians 2	19.61%	0.00%	19.44%	16.72%	16.82%	17.77%	17.31%	16.81%	21.26%	17.33%
Elimia varians 3	19.76%	19.44%	0.00%	16.55%	16.40%	16.42%	15.58%	15.09%	20.90%	15.74%
Elimia variata	17.33%	16.72%	16.55%	0.00%	0.14%	14.43%	9.26%	8.46%	18.36%	9.56%
Elimia variata 2	17.25%	16.82%	16.40%	0.14%	0.00%	14.47%	9.52%	9.52%	17.62%	9.87%
Pleurocera alveare	16.41%	17.31%	15.58%	9.26%	9.52%	11.16%	0.00%	1.53%	16.71%	0.57%
Pleurocera pyrenella	17.28%	17.15%	16.01%	9.99%	9.72%	11.01%	1.79%	2.33%	16.33%	2.36%

	Pleurocera canaliculata	*Pleurocera canaliculata fila JS*	*Pleurocera canaliculata undulata JS*	*Pleurocera chickasahaense JS*	*Pleurocera curta roanense JS*	*Pleurocera foremani JS*	*Pleurocera prasinata JS*	*Pleurocera pyrenella*	*Pleurocera pyrenella JS*	*Pleurocera uncialis hastata JS*
Elimia ampla	9.67%	10.05%	9.40%	9.74%	11.88%	8.28%	10.16%	9.40%	11.99%	11.39%
Elimia ampla 2	9.53%	10.56%	7.22%	7.73%	14.89%	7.44%	12.42%	9.69%	10.64%	14.28%
Elimia annettae	9.35%	11.54%	6.79%	7.71%	15.13%	9.68%	11.65%	9.65%	10.60%	12.93%
Elimia annettae 2	15.23%	11.37%	8.19%	10.30%	18.22%	13.65%	10.78%	15.23%	13.74%	11.35%
Elimia annettae 3	9.25%	9.94%	8.06%	8.13%	14.53%	8.22%	9.99%	9.54%	11.99%	11.20%
Elimia bellacrenata	8.74%	8.12%	8.10%	8.11%	10.03%	6.24%	7.75%	8.53%	9.82%	9.06%
Elimia cochliaris	9.72%	10.38%	9.71%	9.84%	12.35%	10.78%	9.87%	9.81%	12.47%	10.19%
Elimia cochliaris Buck	11.95%	15.71%	10.42%	11.74%	18.51%	8.49%	14.61%	12.41%	15.04%	11.12%
Elimia varians	16.68%	14.33%	13.34%	13.11%	14.72%	14.91%	14.61%	17.28%	15.75%	12.86%
Elimia varians 2	17.14%	17.80%	15.32%	16.10%	16.33%	16.11%	16.62%	17.15%	15.63%	17.96%
Elimia varians 3	15.86%	14.53%	13.78%	14.43%	16.45%	12.64%	14.01%	16.01%	10.90%	12.91%
Elimia variata	9.83%	10.54%	7.22%	7.72%	15.67%	8.89%	12.42%	9.99%	11.44%	14.27%
Elimia variata 2	9.78%	10.14%	9.41%	9.75%	11.84%	8.26%	10.15%	9.72%	11.86%	11.35%
Pleurocera alveare	1.51%	4.75%	1.99%	1.88%	9.43%	9.20%	4.15%	1.79%	9.59%	7.24%
Pleurocera pyrenella	1.57%	4.95%	2.55%	2.56%	9.53%	8.57%	5.19%	0.00%	10.12%	7.47%

	Pleurocera uncialis uncialis JS	*Pleurocera vestita Cahaba JS*	*Pleurocera viridula JS*	*Pleurocera walkeri JS*
Elimia ampla	13.44%	9.90%	10.01%	12.03%
Elimia ampla 2	17.03%	7.09%	10.44%	11.80%
Elimia annettae	13.05%	7.11%	10.51%	13.55%
Elimia annettae 2	10.60%	8.20%	12.08%	11.35%
Elimia annettae 3	11.99%	6.84%	8.98%	15.12%

	Pleurocera uncialis uncialis JS	*Pleurocera vestita* Cahaba JS	*Pleurocera viridula* JS	*Pleurocera walkeri* JS
Elimia bellacrenata	11.50%	8.40%	8.01%	10.53%
Elimia cochliaris	12.53%	9.79%	9.75%	12.05%
Elimia cochliaris Buck	14.87%	10.39%	14.85%	16.25%
Elimia varians	14.42%	11.85%	15.06%	14.98%
Elimia varians 2	13.76%	17.13%	18.71%	15.97%
Elimia varians 3	14.69%	13.27%	15.09%	15.75%
Elimia variata	17.02%	7.09%	10.43%	12.61%
Elimia variata 2	13.44%	9.87%	9.98%	11.78%
Pleurocera alveare	12.23%	1.51%	4.48%	9.72%
Pleurocera pyrenella	11.94%	2.91%	4.46%	9.80%

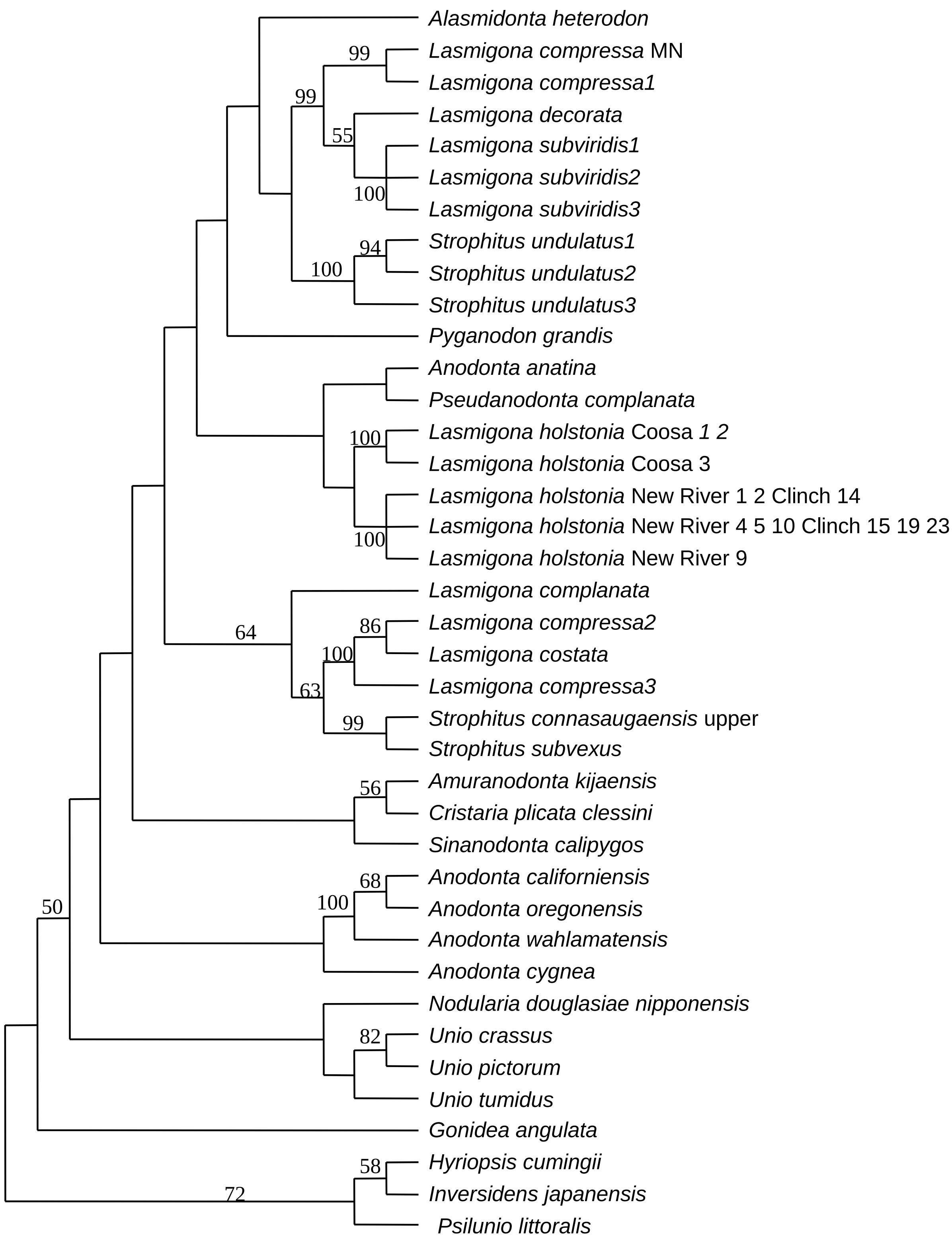


Figure 1. *cox1* data for *Lasmigona* and *Strophitus*. Strict consensus cladogram of 4 maximum parsimony trees, length 1205. Numbers are bootstrap percentages. Taxon names with multiple numbers after them (*e.g., Lasmigona holstonia* Coosa 1 2) indicate multiple specimens that yielded the same sequence.

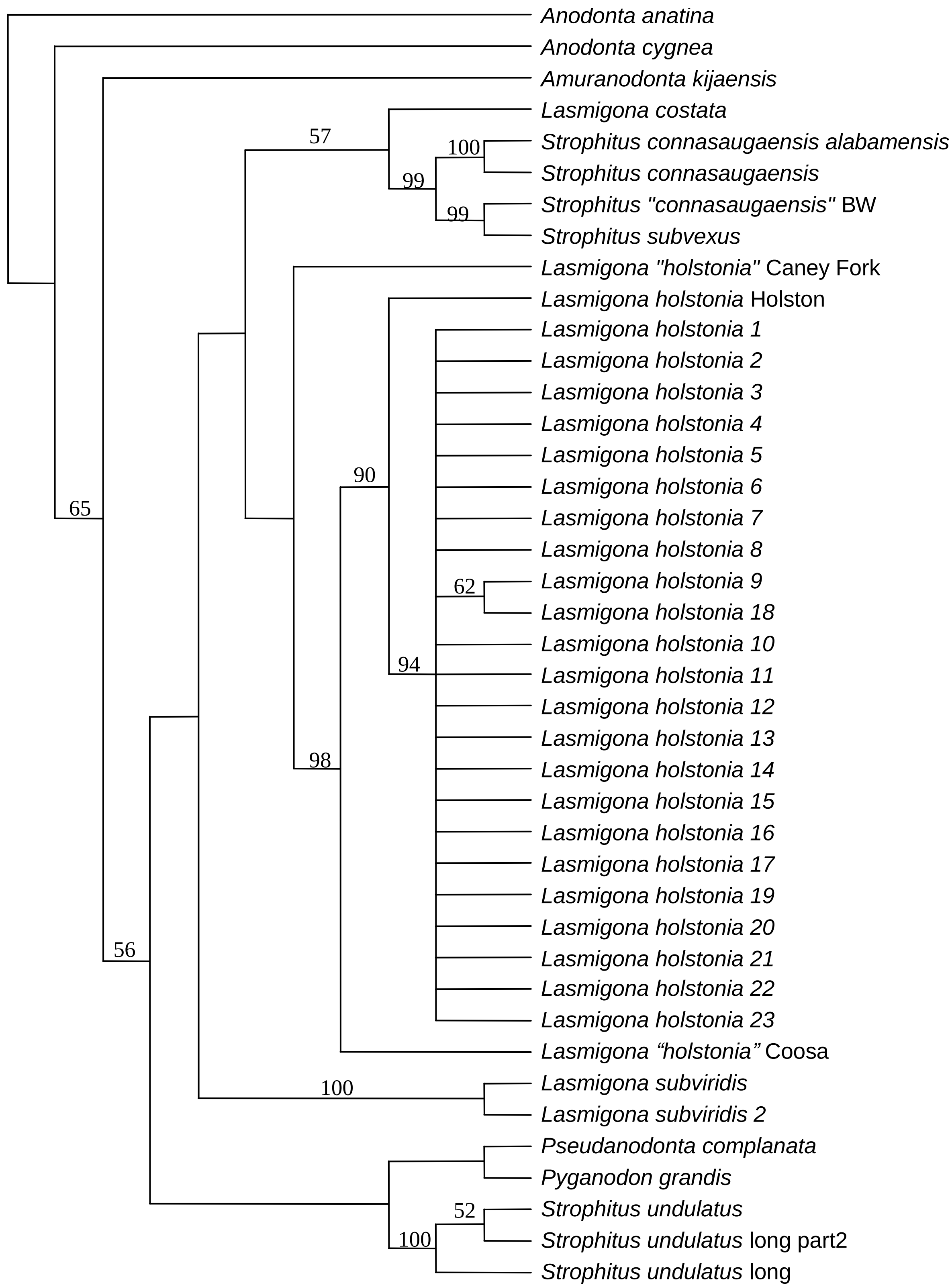
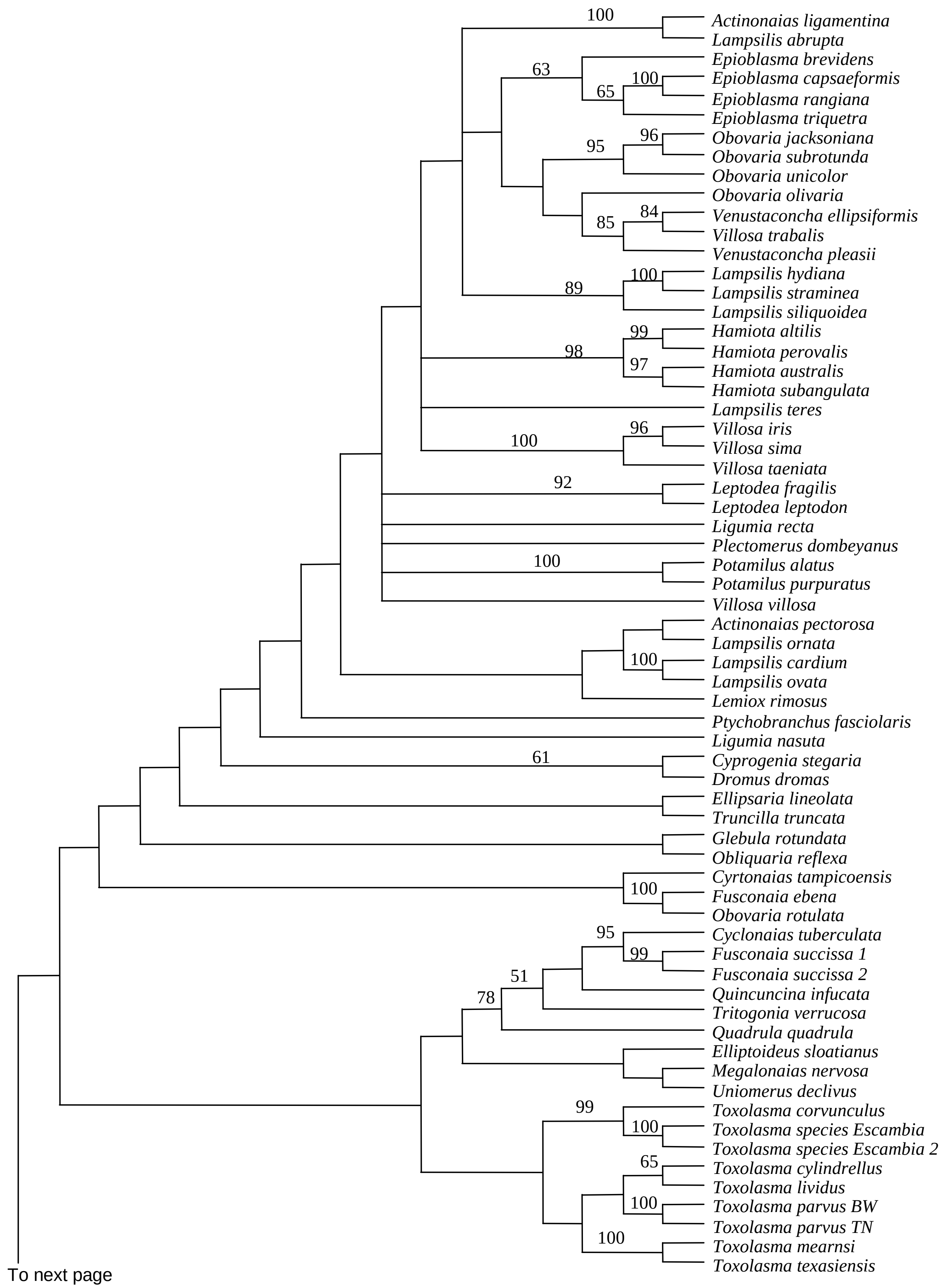


Figure 2. ITS1 data for *Lasmigona* and *Strophitus*. Single most parsimonious tree, length 503. Numbers are bootstrap percentages.


Actinonaias ligamentina
Lampsilis abrupta
Epioblasma brevidens
Epioblasma capsaeformis
Epioblasma rangiana
Epioblasma triquetra
Obovaria jacksoniana
Obovaria subrotunda
Obovaria unicolor
Obovaria olivaria
Venustaconcha ellipsiformis
Villosa trabalis
Venustaconcha pleasii
Lampsilis hydiana
Lampsilis straminea
Lampsilis siliquoidea
Hamiota altilis
Hamiota perovalis
Hamiota australis
Hamiota subangulata
Lampsilis teres
Villosa iris
Villosa sima
Villosa taeniata
Leptodea fragilis
Leptodea leptodon
Ligumia recta
Plectomerus dombeyanus
Potamilus alatus
Potamilus purpuratus
Villosa villosa
Actinonaias pectorosa
Lampsilis ornata
Lampsilis cardium
Lampsilis ovata
Lemiox rimosus
Ptychobranchus fasciolaris
Ligumia nasuta
Cyprogenia stegaria
Dromus dromas
Ellipsaria lineolata
Truncilla truncata
Glebula rotundata
Obliquaria reflexa
Cyrtonaias tampicoensis
Fusconaia ebena
Obovaria rotulata
Cyclonaias tuberculata
Fusconaia succissa 1
Fusconaia succissa 2
Quincuncina infucata
Tritogonia verrucosa
Quadrula quadrula
Elliptoideus sloatianus
Megalonaias nervosa
Uniomerus declivus
Toxolasma corvunculus
Toxolasma species Escambia
Toxolasma species Escambia 2
Toxolasma cylindrellus
Toxolasma lividus
Toxolasma parvus BW
Toxolasma parvus TN
Toxolasma mearnsi
Toxolasma texasiensis
To next page

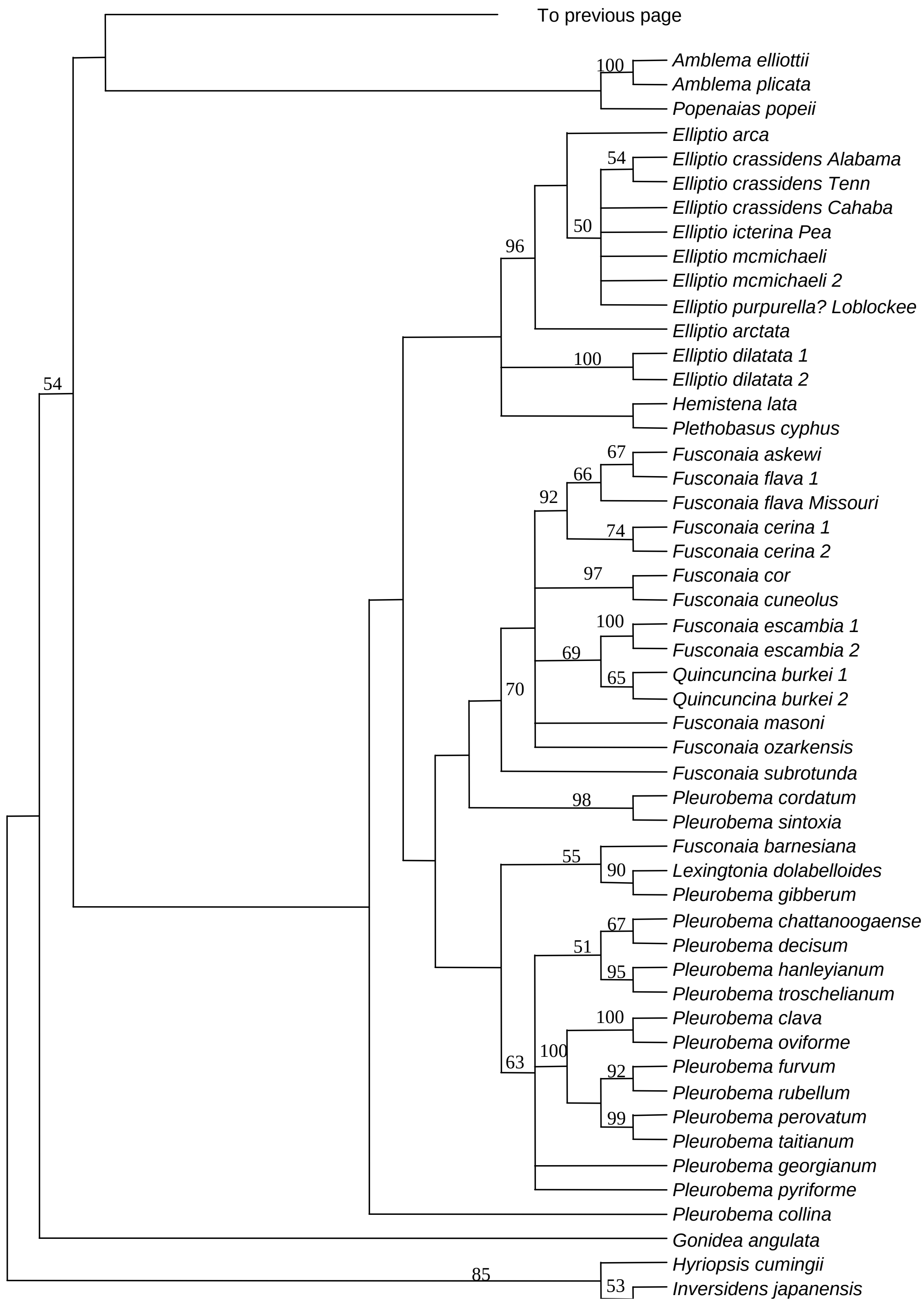


Figure 3. *cox1* data for Ambleminae, including *Toxolasma*, *Elliptio*, and *Fusconaia*. Strict consensus cladogram of 1523 maximum parsimony trees, length 2516. Numbers are bootstrap percentages.

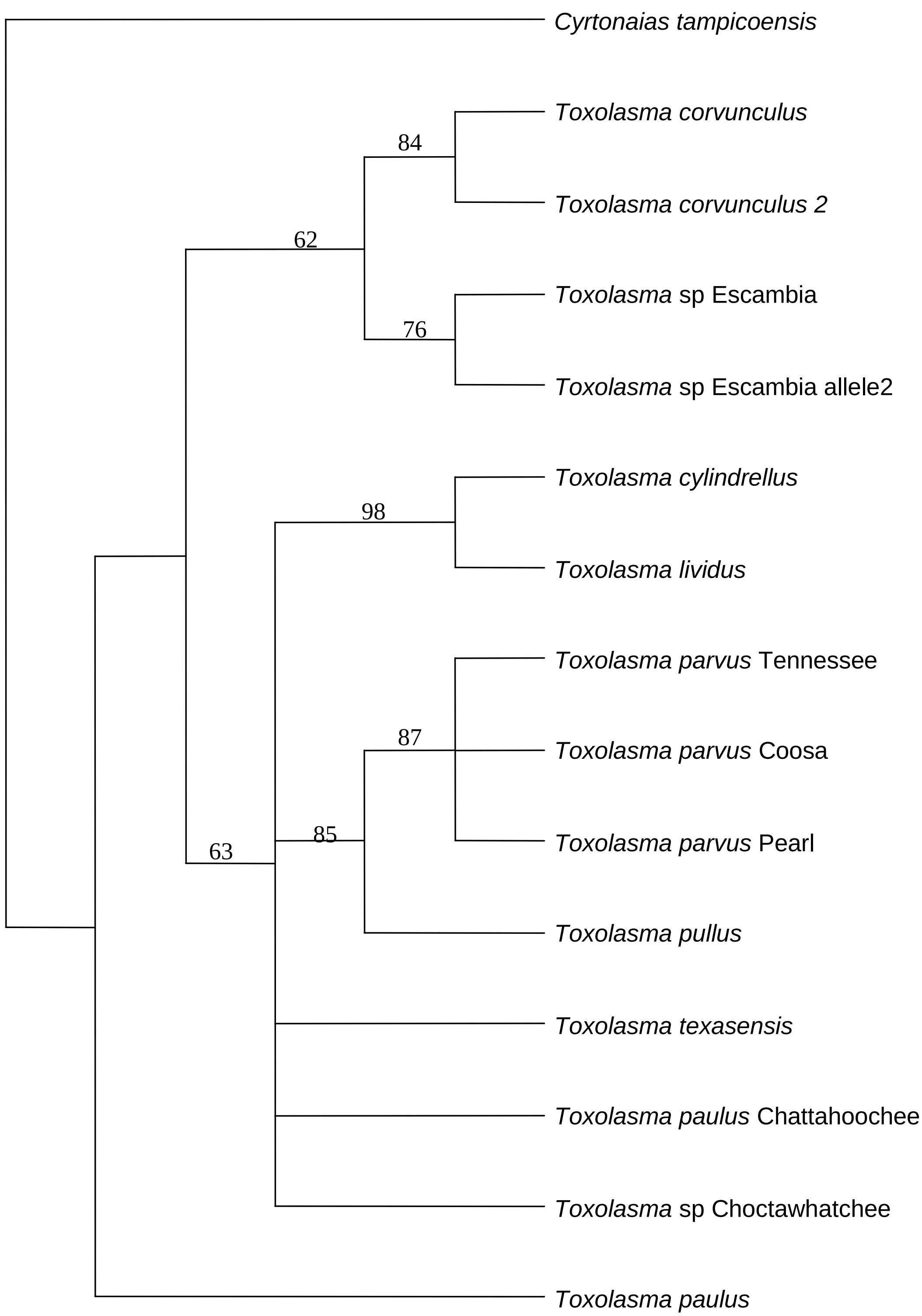


Figure 4. ITS1 data for *Toxolasma*. Strict consensus cladogram of 5 maximum parsimony trees, length 120. Numbers are bootstrap percentages.

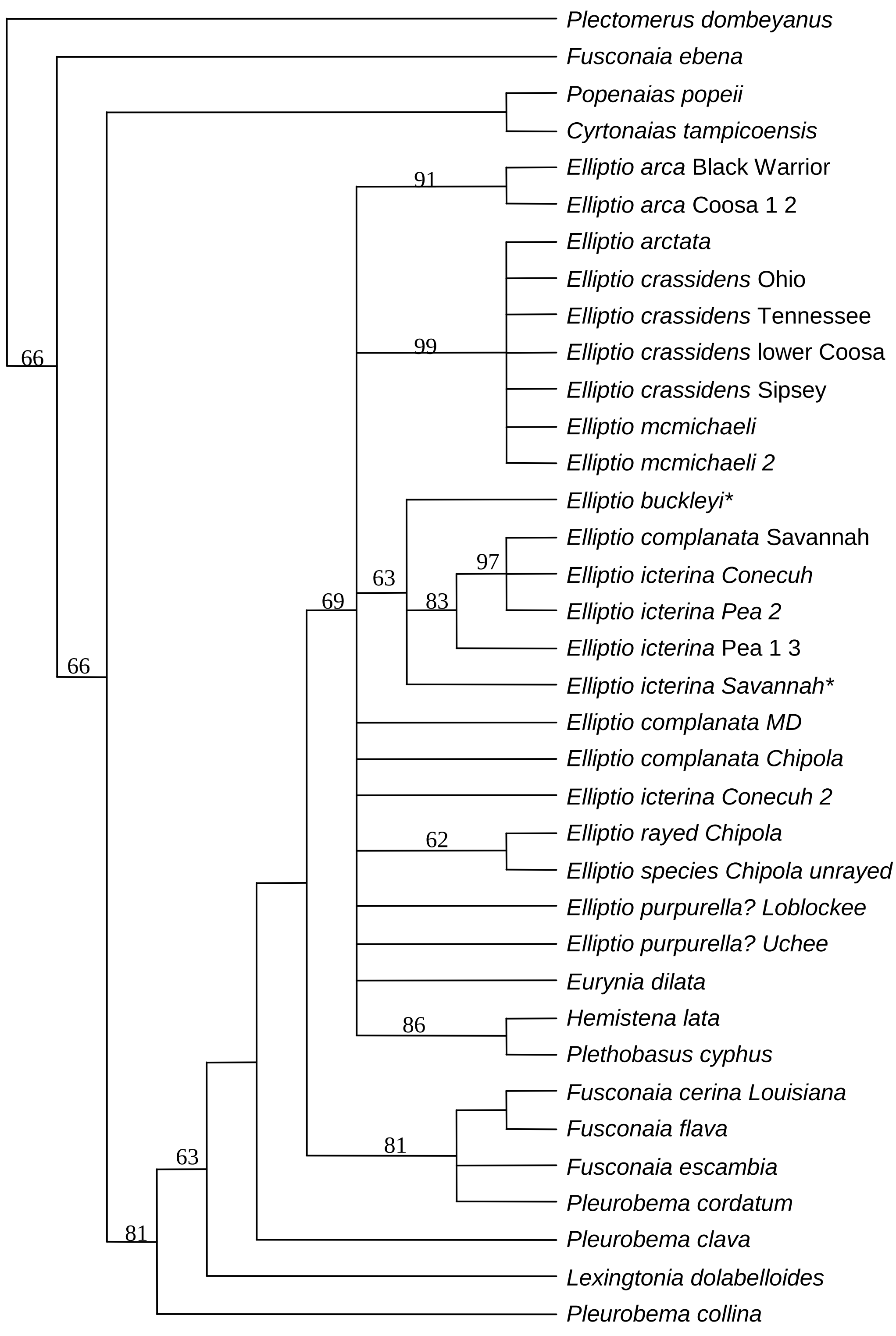


Figure 5. ITS1 data for *Elliptio* and *Fusconaia*. Strict consensus cladogram of 9 maximum parsimony trees, length 476. There was 57% bootstrap support for grouping *E. buckleyi* and *E. icterina* Savannah, marked with *. Taxon names with multiple numbers after them (*e.g.*, *Elliptio icterina* Pea 1 3) indicate multiple specimens yielded the same sequence. Numbers are bootstrap percentages.

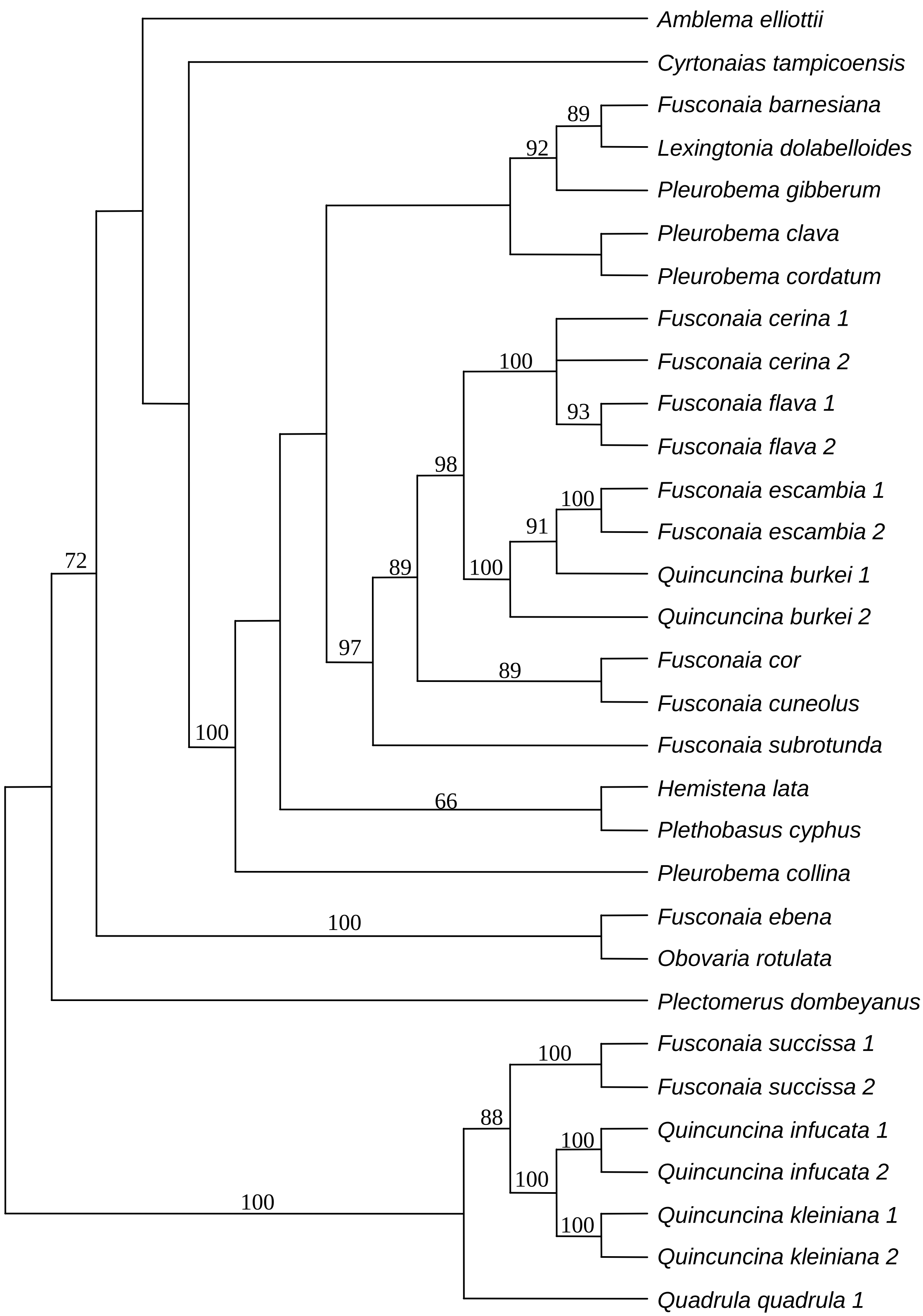


Figure 6. *cox1*, 16S, and *nadh1* data for *Fusconaia*. Strict consensus cladogram of 4 maximum parsimony trees, length 1819. Numbers are bootstrap percentages.

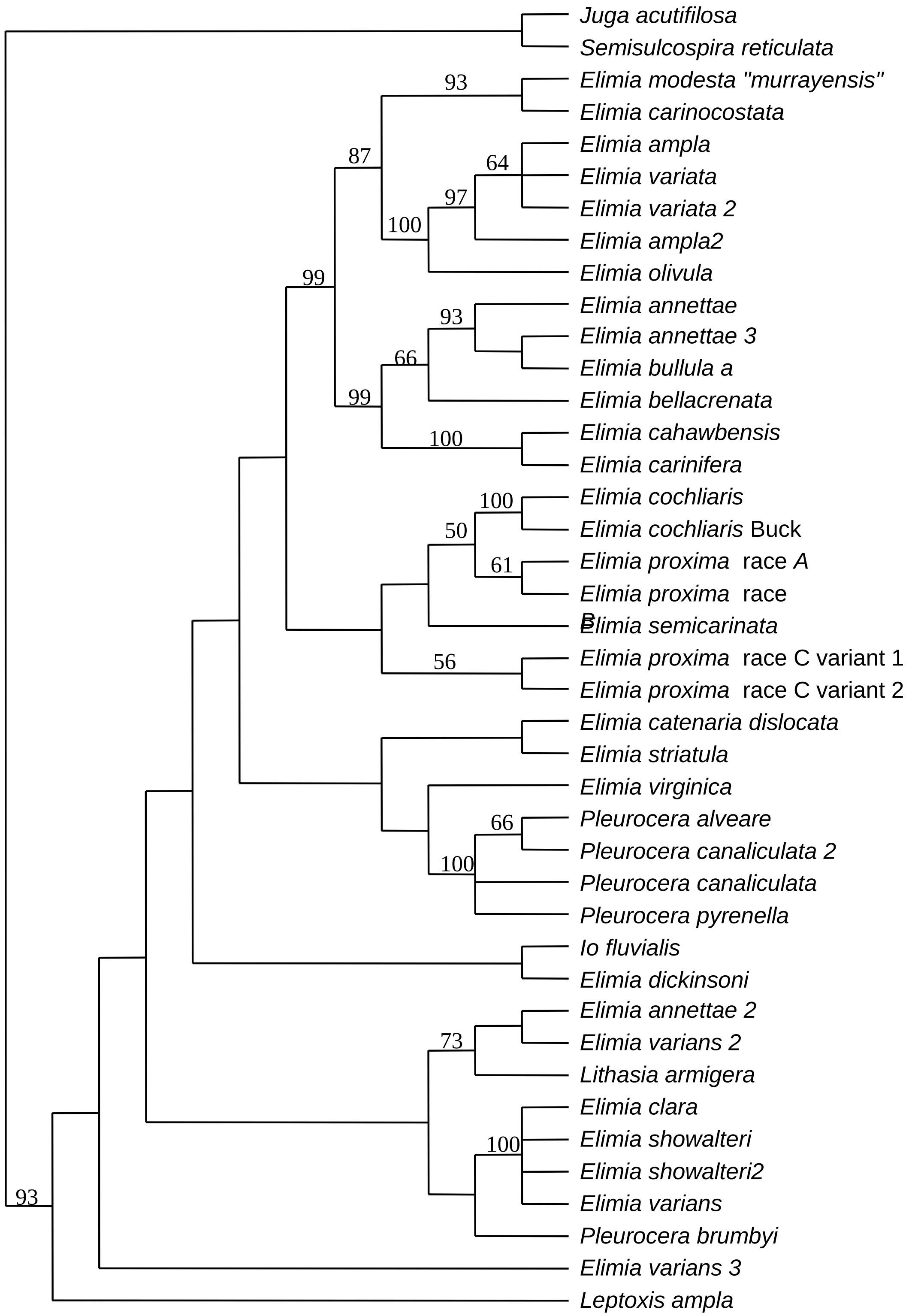


Figure 7. *cox1* data for pleurocerids, Folmer *et al*. (1994) region. Strict consensus cladogram of 2 maximum parsimony trees, length 1294. Numbers are bootstrap percentages. The bootstrap analysis also gave 67% support for a clade of *P. canaliculata* and *P. pyrenella*, which was not resolved in the strict consensus.

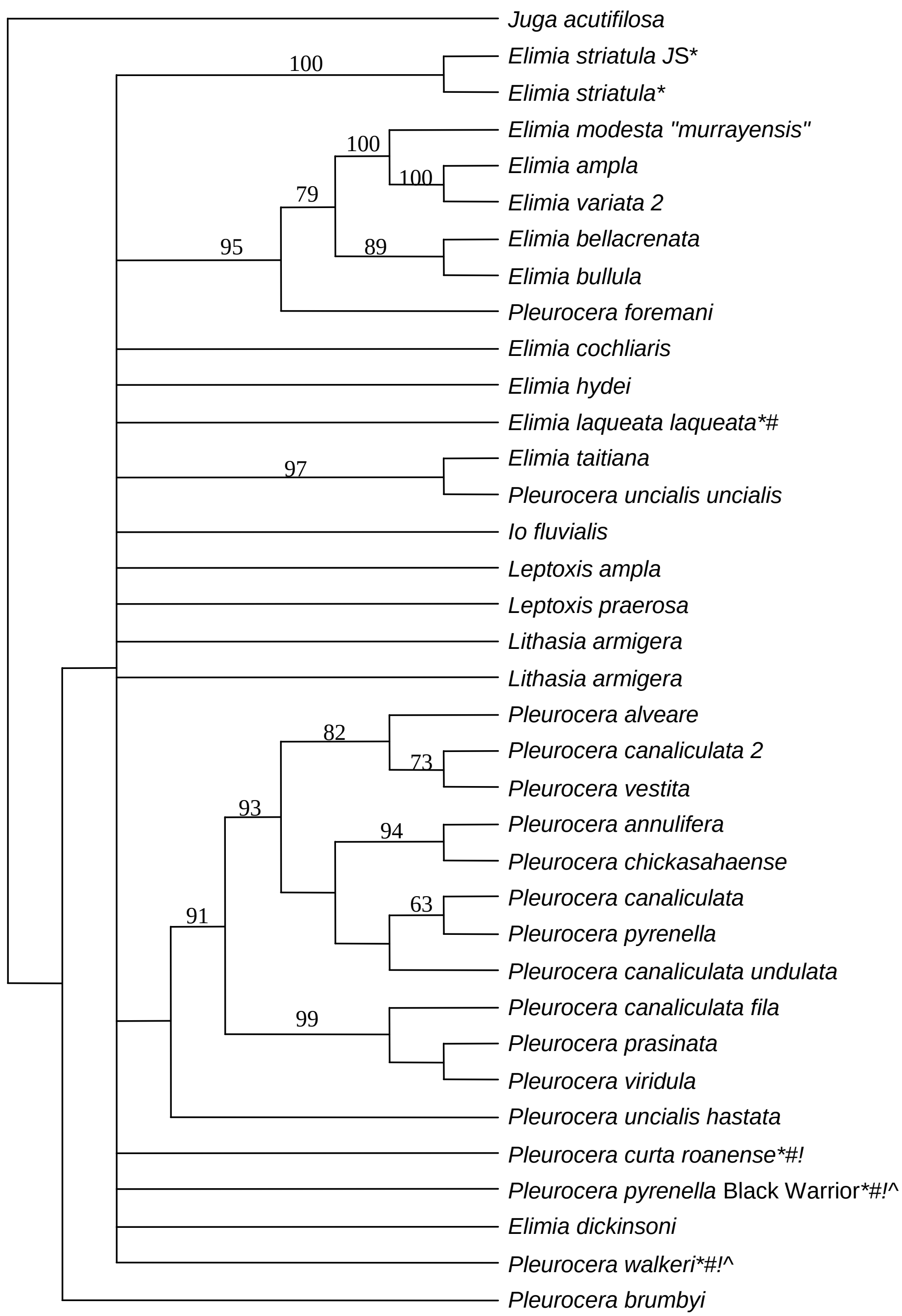


Figure 8. *cox1* data for pleurocerids, Minton and Lydeard (2003) region. Strict consensus cladogram of 63 maximum parsimony trees, length 1124. The bootstrap analysis supported several clades not resolved in the strict consensus (indicated by *, #, !, or ^ after the species name): 57% support for *E. striatula* (both), *E. laqueata laqueata, P. curta roanense, P. pyrenella* Black Warrior, and *P. walkeri* (*); 61% support for *E. laqueata laqueata, P. curta roanense , P. pyrenella* Black Warrior, and *P. walkeri* (#); 72% support for *P. curta roanense, P. pyrenella* Black Warrior, and *P. walkeri* (!), and 64% support for *P. pyrenella* Black Warrior and *P. walkeri* (^). Numbers are bootstrap percentages.

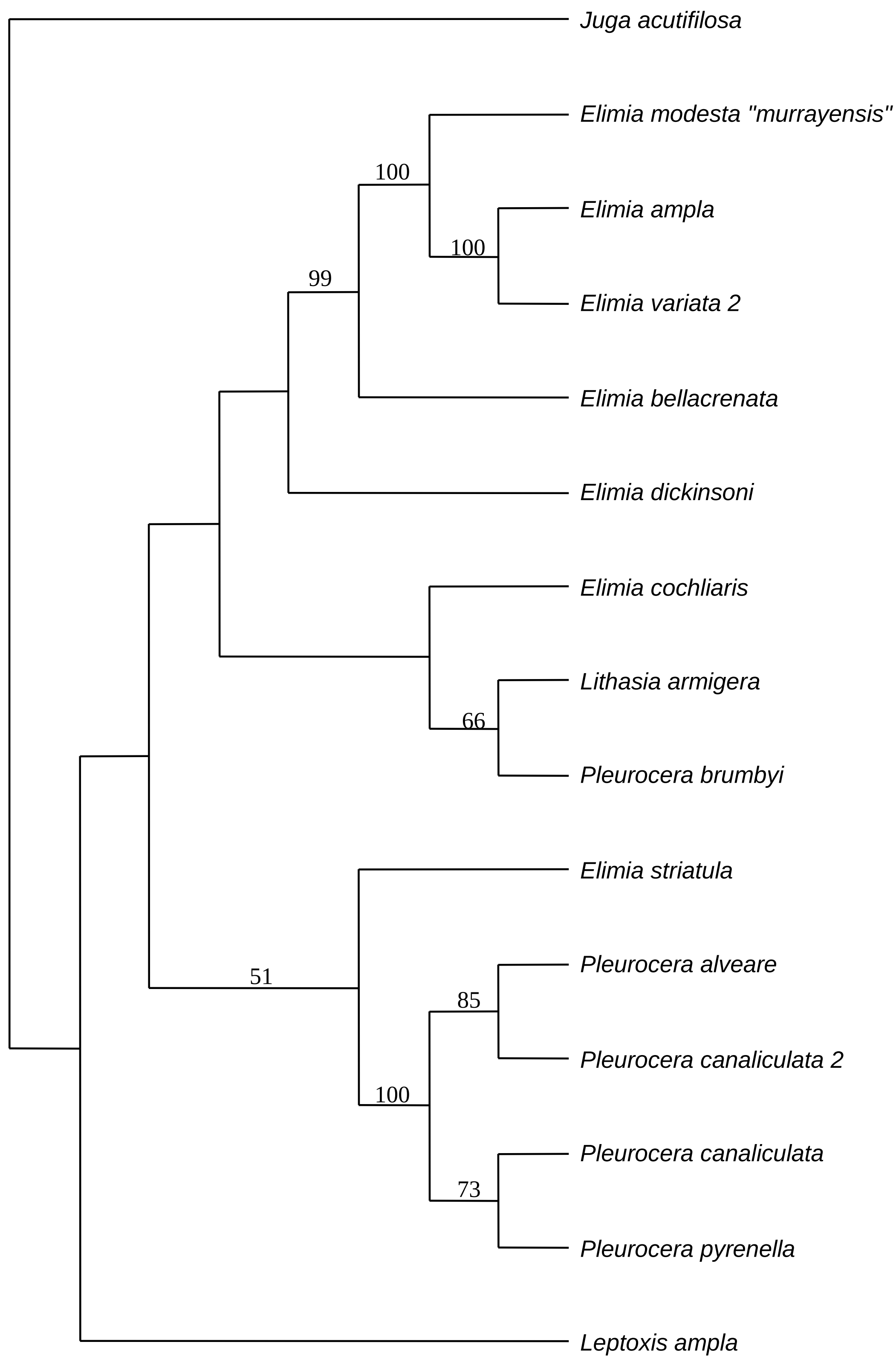


Figure 9. *cox1* data for pleurocerids, both regions. Single most parsimonious tree, length 929. Numbers are bootstrap percentages.