

**REPORT FOR:**

Preliminary Analysis for Identification, Distribution, and Conservation Status of Species of  
*Fusconaia* and *Pleurobema* in Arkansas

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## EXECUTIVE SUMMARY

There are currently 13 species of *Fusconaia* and 32 species of *Pleurobema* recognized in the United States and Canada. Twelve species of *Pleurobema* and two species of *Fusconaia* are listed as Threatened or Endangered. There are 75 recognized species of Unionidae in Arkansas; however this number may be much higher due to the presence of cryptic species, many which may reside within the *Fusconaia* /*Pleurobema* complex. Currently, three species of *Fusconaia* and three species of *Pleurobema* are recognized from Arkansas. The true conservation status of species within these genera cannot be determined until the taxonomic identity of populations is confirmed.

The purpose of this study was to begin preliminary analysis of the species composition of *Fusconaia* and *Pleurobema* in Arkansas and to determine the phylogeographic relationships within these genera through mitochondrial DNA sequencing and conchological analysis. In order to do so, multiple populations from within Arkansas and from out-of-state, geographically disjunct populations, including topotypic or near-topotypic localities per taxon, were used to examine conchological and genetic affinities and divergence. The first objective was to conduct phylogenetic analysis on these specimens using the mitochondrial COI gene. The second objective was to quantify shell morphology and topographic anatomical characteristics. The third objective was to compare the conchological results to phylogenetic results to establish morphological limits of taxa and to construct a practical identification key to taxa.

Museum holdings from the following institutions have been examined: United States National Museum (USNM), Ohio State University Museum of Zoology (OSU), Mayborn Museum Complex-Baylor University (MMC-BU), Oklahoma Biological Survey (OBS), and Sam Noble Oklahoma Museum of Natural History, University of Oklahoma (SNOMNH). External view and internal view digital images were acquired for 16 taxonomic entities. Geometric morphometric analysis was conducted for external landmarks of five recognized *Fusconaia* species, four *Pleurobema* species, and five “unknown” clades from Arkansas to compare for congruence with the phylogenetic analysis.

DNA sequence data from 56 specimens *Fusconaia* or *Pleurobema* taxa have been aligned to date in a data set yielding 552 base pairs. Within this alignment 207 sites were polymorphic, 123 of which were phylogenetically informative under maximum parsimony. To date, only about half (n=56) of the individuals that have been sequenced (n = ~100) have undergone phylogenetic analysis. Sequence alignment continues on the unanalyzed individuals, and we are actively seeking additional specimens from both Arkansas populations and museum holdings to continue refining concepts of phylogenetic relationships that have been identified.

Analysis to date indicates there are two species of *Pleurobema* in Arkansas that have been previously overlooked. In addition, there are potentially one additional *Fusconaia* species

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and one *Pleurobema* species that may be present. Analyses are continuing to resolve these relationships, however, this project has provided a broad base from which to continue efforts to elucidate the composition and distribution of Arkansas *Fusconaia* and *Pleurobema*.

## INTRODUCTION

There are currently 13 species of *Fusconaia* and 32 species of *Pleurobema* recognized in the United States and Canada (Turgeon et al. 1998.) Twelve species of *Pleurobema* and two species of *Fusconaia* are listed as Threatened or Endangered. Harris et al. (1997) recognized 75 species of Unionidae in Arkansas; however this number may be much higher due to the presence of cryptic species, many which may reside within the *Fusconaia/Pleurobema* complex (Kandl et al. 2001). Currently, three species of *Fusconaia* and three species of *Pleurobema* are recognized from Arkansas. The true conservation status of species within these genera cannot be determined until the taxonomic identity of populations is confirmed.

Females of the genera are easily differentiated when gravid as the *Fusconaia* use all four gills as marsupia and the *Pleurobema* utilize only the outer pair (Oesch 1984). However, the *Fusconaia* and *Pleurobema* species generally lack diagnostic shell characteristics that make field identification reliable, especially on living specimens where internal shell characters are not visible without sacrificing the animal. Shell-based species identifications may be complicated by geographic variation due to ecophenotypically induced character expressions (e.g. clinal differences in shell length, inflation, and thickness relative to stream size) or genetic divergence with or without obvious variation to shell morphology (Ortmann 1920, Hoeh et al. 1995, Lydeard and Roe 1998).

## PURPOSE / OBJECTIVES

The purpose of this study was to begin preliminary analysis of the species composition of *Fusconaia* and *Pleurobema* in Arkansas and to determine the phylogeographic relationships within these genera through mitochondrial DNA sequencing. In order to do so, multiple populations from within Arkansas and from out-of-state, geographically disjunct populations, including topotypic or near-topotypic localities per taxon, were used to examine conchological and genetic affinities and divergence. Authorities and type localities for the taxa included in this study are:

*Fusconaia askewi* (Marsh 1896) – Texas pigtoe. Village Creek, Hardin Co. TX and Sabine R., TX (from Vidrine 1993)

*Fusconaia cerina* (Conrad 1838) – southern pigtoe. New Orleans, LA (from Vidrine 1993).

*Fusconaia ebena* (I. Lea 1831) – ebonyshell. Ohio River (from Parmalee and Bogan 1998).

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*Fusconaia flava* (Rafinesque 1820) – Wabash pigtoe. Small tributaries to Kentucky, Salt, and Green rivers (from Parmalee and Bogan 1998). Wisconsin and Fox rivers (from Vidrine 1993).

*Fusconaia lananensis* (Frierson 1901) – triangle pigtoe. Lanana and Bonita creeks near Nacodoches, Nacodoches County, TX (from Howells et al. 1996).

*Fusconaia ozarkensis* (Call 1887) – Ozark pigtoe. Current River, Shannon Co., MO and in Jacks Fork and Big Creek. Lectotype (selected by Johnson 1975) and paralectotype both from Jacks Fork, Shannon Co., MO (from Johnson 1980).

*Fusconaia subrotunda* (I. Lea 1831) – longsolid. Ohio (from Parmalee and Bogan 1998).

*Pleurobema beadleianum* (Lea 1861) – Mississippi pigtoe. Pearl River, Jackson, MS (from Vidrine 1993).

*Pleurobema cordatum* (Rafinesque 1820) – Ohio pigtoe. Ohio River (from Parmalee and Bogan 1998).

*Pleurobema riddellii* (Lea 1861) – Louisiana pigtoe. Dallas, TX (from Vidrine 1993).

*Pleurobema rubrum* (Rafinesque 1820) – pyramid pigtoe. Ohio (from Parmalee and Bogan 1998).

*Pleurobema sintoxia* (Rafinesque 1820) – round pigtoe. Mahoning R. near Pittsburg (from Parmalee and Bogan 1998).

The first objective was to conduct a phylogenetic analysis on these specimens using the COI gene. The second objective was to quantify shell morphology and topographic anatomical characteristics using geometric morphometric analyses. The third objective was to compare the geometric morphometric results to the phylogenetic results to establish morphological limits of individual taxa and to construct a practical identification key to taxa.

## MATERIALS AND METHODS

### DNA Extraction, Amplification, and Sequencing

Whole genomic DNA was extracted from 95% EtOH preserved mantle tissue using a standard CTAB/Cholorfom protocol (Sagahi-Marroof et al., 1984). Extracted DNA was visualized on an agarose gel and quantified on a Nanodrop ND-1000 spectrophotometer. DNA was then diluted to a standard concentration of 10ng/μl. A 600 bp portion of the cytochrome *c* oxidase gene (COI) was amplified using the universal primer LCO1490 (Folmer et al., 1994) and a modified version of HCO2198 (Graf and O Foighil, 2000). Amplicons were generated using Qiagen's Taq PCR Core Kit and the following thermal cycling conditions: an initial denaturation cycle of 95°C for 3min, 40 cycles of 95°C for 35s, 44°C for 45s, 72°C for 45s, followed by a 5min extension period at 72°C. Amplicons were purified using a shrimp alkaline phosphatase method, cycle sequenced using the Beckman DTCS Quick Start Kit, and visualized on a Beckman CEQ 8000 automated sequencer.

Sequences were visually aligned in the sequence editor BioEdit (Hall 1999) and the complete data set was converted into amino acid sequence to check the accuracy of the nucleotide sequence (Maddison and Maddison, 2000). Aligned sequences accessioned from Genbank (Table 1) and from specimens processed in this study (Table 2) were analyzed in Mr. Bayes version 3.0 (Huelsenbeck and Ronquist, 2001) criteria to infer phylogenetic relationships. Four Markov Chain Monte Carlo simulations were run simultaneously using a random starting tree for  $3 \times 10^6$  generations to estimate the topology and posterior probabilities for node support. Trees were sampled every 100 generations and burnin was determined by plotting the log-likelihood scores of all saved trees versus generation time. Trees with likelihood scores after stationarity was achieved were retained for inclusion in the majority-rule tree. Posterior probability support values were determined by the frequency at which a particular clade occurred within all trees retained after burnin.

### Morphometric analysis

More than 250 specimens were used in the morphological analysis. Effort was made to acquire digital data for the 6 nominal *Fusconaia* (*F. ebena*, *F. flava*, and *F. ozarkensis*) and *Pleurobema* (*P. cordatum*, *P. rubrum*, and *P. sintoxia*) taxa that occur in Arkansas, as well as from additional recognized or hypothesized taxonomic units that occur peripheral to the state, including *Fusconaia askewi*, *F.*

*cerina*, *F. lananensis*, *F. subrotunda*, *Pleurobema beadleianum*, and *P. riddellii* (Cummings and Mayer 1992, Jones et al. 2005, Howells et al. 1996, Oesch 1995, Parmalee and Bogan 1998, and Vidrine 1993).

Photographs of external and internal views of individual shells were taken with digital camera (Sony DSC-F717 Cyber-shot, KODAK EasyShare CX6330 or KODAK EasyShare Z712 IS). Shell valves were leveled using modeling clay to make the longitudinal axis parallel to the table surface to assure accuracy in length measurements. Length was quantified by measuring with Helios dial calipers to the nearest 0.05 mm or by including a ruler in the photograph to provide scale. Input files of the digital images were generated using tpsUtil Version 1.38 (Rohlf 2006), and tpsDIG2 Version 2.10 (Rohlf 2006) was used for landmark digitization. Two homologous (Type I) and 26 non-homologous (Type III) landmarks were generated from external views for a total of 28 external landmarks (Figure 1).

Geometric morphometric analysis was utilized to examine shape variation through principal components analysis (PCA) and canonical variates analysis (CVA). PCA is a technique for simplifying descriptions of variation among individuals while CVA simplifies descriptions of variation between groups (Zelditch et al. 2004). In traditional morphometric analysis, PCA usually suffers from the influence of size across the newly generated axes. However, in geometric morphometric analysis, all specimens are rescaled to be the same size thus eliminating size as a factor with the result that shape can be more thoroughly examined. In PCA, no *a priori* assumptions are needed to group individuals. CVA determines the set of axes which best discriminates between groups; therefore an *a priori* assumption of group membership is required. For CVA analysis, specimens were examined by species and by drainage. *Fusconaia* and *Pleurobema* species are not sexually dimorphic with regards to shell morphology (Bogan and Parmalee 2001). Therefore, males and females of all species and clades were combined for analysis.

All data analyses were performed using the Integrated Morphometrics Package (IMP) suite of programs (Sheets 2003). The scaled landmark coordinates from tpsDIG2 were imported to CoordGen6f. CoordGen6f converted landmark coordinates to Procrustes distances using least squares Procrustes superimposition methods. Procrustes superimposition methods are generally favored over other superimposition methods such as Bookstein coordinates or sliding baseline registration because landmarks are not constrained to a baseline that may transfer variance from the baseline to other landmarks (Zelditch et al. 2004, Hayes et al. 2007) and because generalized Procrustes methods have been shown to produce estimates with the least error and no pattern of bias (Rohlf 2006). For external views, the two homologous landmarks (landmarks 8 and 1) were utilized as baseline coordinates. Initial

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comparisons between all population combinations were performed in TwoGroup6h (Sheets 2003) to determine if there were significant shape differences assessed by Goodall's F. TwoGroup6h was also utilized to generate figures of the landmark mean Procrustes superimpositions of all species pairings to allow comparison of the mean shell outlines (shape). The significance level was adjusted using a Bonferroni correction for multiple pairwise comparisons. A PCA was performed with PCAGen6n on the data with a posteriori groups assigned by taxonomic unit. A CVA was also generated using CVAGen6k with groups assigned by drainage or taxon.

CVA was utilized to explore shape relationships of the "known" taxonomic units from type drainages with "unknown" taxonomic units collected in Arkansas for this study. The "assign unknown specimens" subroutine of CVA was used to compare specimens from 5 hypothesized Arkansas taxonomic units defined by the genetic analysis or by tentative taxonomic unit assignment that occurred at initial specimen collection and sorting. The assignment test determines the probability that a Mahalanobis' distance between an individual and the mean of the group is larger than expected under a null model of random variation around the mean of each group (Zelditch et al. 2004).

Recently acquired specimens utilized for genetic and conchological analyses in this study are summarized in Table 2. Museum specimens utilized strictly for the morphometric analyses are summarized as follows:

### *Fusconaia askewi*

**LA:** Vernon Parish: OSU 12993.1-4 (4), Sabine River at LA Highway 8 (Burr Ferry). **TX :** Panola County: OSU 49275.1-12 (12), Sabine River at U. S. Highway 79.

### *Fusconaia ebena*

**IL:** Pulaski County: OSU 30926.1-17 (17), Ohio River at Mound City.

### *Fusconaia flava*

**KY:** Hart County: OSU 11807.1- 8 (8) and OSU 16576.1-11 (11), Green River at Munfordville. Warren County: OSU 27163.1-9 (9) and OSU 39175.1-5 (5), Green River at KY Highway 185.

### *Fusconaia ozarkensis*

**MO:** Shannon County: OSU 34360.1-8 (8), Jacks Fork Current River at MO Highway 106. OSU 75488.1-7 (7), Jacks Fork Current River at MO Highway 106. Texas County: OSU 33628.1-10 (10), Jacks Fork Current River at MO Highway 17.



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### *Fusconaia subrotunda*

**OH:** Morgan County: OSU 17029.1-7 (7), Muskingum River at Lake Chute. Washington County: OSU 6635.1-3 (3), Muskingum River at Beverly. OSU 11027.1-4 (4) and OSU 17048.1-4 (4), Muskingum River at Lowell.

### *Pleurobema cordatum*

**TN:** Meigs County: ANSP 129491.1-4 (4), Tennessee River at Decatur.

### *Pleurobema riddellii*

**TX:** Angelina County: OSU 32887.1-15 (15), Angelina River at U. S. Highway 59. Hardin County: OSU 25059.1-22 (22), Village Creek at TX Highway 92.

### *Pleurobema rubrum*

**IL:** Pope County: OSU 68515.1-2 (2), Ohio River at Golconda. **OH:** Hamilton County: OSU 58511.1-8 (8), OSU 67910.1-3 (3), OSU 69474.1-2 (2), and OSU 69479.1-2 (2), Ohio River at Cincinnati.

### *Pleurobema sintoxia*

**AR:** Lawrence County: OSU 47681.1-8 (8), Black River at U. S. Highway 63. **OH:** Coshocton County: OSU 12879.1-9 (9), Walhonding River at OH State Highway 715. Mahoning County: OSU 58347, Mahoning River, site unknown. OSU 58350.1-5 (5), Mahoning River 3.0 miles northeast of Poland. Morgan County: OSU 49609.1-6 (6), Muskingum River north of Lake Chute. **PA:** Erie County: ASU 3595-3598 (4), French Creek at Dutch Road. Indiana County: ASU 3590-3594 (5), Little Mahoning River.

## RESULTS AND DISCUSSION

### Phylogenetic Analysis of Genetic Data

DNA sequence data from 56 specimens of *Fusconaia* or *Pleurobema* taxa have been aligned to date in a data set yielding 552 base pairs. Within this alignment 207 sites were polymorphic, 123 of which were phylogenetically informative. Figure 2 illustrates the results of the Bayesian analysis of the the COI dataset. Representative specimens included in the genetic and the morphometric analyses can be found in Plates 1-9.

With the exclusion of '*Fusconaia*' *ebena*, our analysis produced reciprocally monophyletic *Pleurobema* and *Fusconaia* clades. Within each *Fusconaia* and *Pleurobema* clade,

a subclade consisting of Ouachita region individuals was recovered, although high support was found for the *Pleurobema* Ouachita clade and only moderate support was found for the *Fusconaia* Ouachita clade. This result is consistent with that of Serb (2006), which indicates that the Ouachita region is biogeographically unique for some unionid species.

Our analysis of the COI gene region produced a largely unresolved *F. flava* clade with little geographic structuring. Our results indicate that *F. cerina* may not be a valid taxon, but rather synonymous with *F. flava*. Our results also indicate that *F. ozarkensis*, including topotypic material, is a valid taxon and is distinct from *F. flava*.

The *Pleurobema* clade consists of a largely unresolved polytomy consisting of *P. rubrum*, *P. sintoxia*, and *P. cordatum*, none of which were found to be monophyletic. This result is consistent with previous studies which also found these species to be paraphyletic (Burdick and White, 2007; Campbell et al., 2005). This could be a result of incomplete lineage sorting (Nei, 1987), or it could be that these species have poorly defined conchological characters, making species assignment problematic. Two highly divergent haplotypes were found in *Pleurobema* specimens from the Middle Fork of the Little Red River and Spring rivers which were basal to all non-Mobile River *Pleurobema*.

Within the *Pleurobema* clade is a well supported St. Francis River clade, although one St. Francis individual was placed within the unresolved *Pleurobema* polytomy. The St. Francis River has been shown to be an evolutionarily unique area for *Epioblasma* (Zanatta and Murphy, 2008), but not for *Cyprogenia* (Serb, 2006). Our molecular results suggest that the St. Francis River *Pleurobema* may share a unique evolutionary history, and closer evaluation of this fauna is warranted.

The placement of GR2 is puzzling. The specimen was identified as *P. rubrum*, however it falls out basal to the remaining *Fusconaia* species (excluding '*F.*' *ebena*). Highly divergent haplotypes of specimens identified as *F. flava* have been found in a previous study (Burdick and White 2007) which allied with *P. cordatum*. The placement of GR2 as basal to the remaining *Fusconaia* may indicate that it is a unique evolutionary lineage (i.e. a new species). Additional species of *Fusconaia* as well as additional Green River specimens are needed to determine the identity/uniqueness of our GR2 specimen.

Our samples of *Fusconaia ebena* form a well supported clade outside of the remaining *Fusconaia* and *Pleurobema* samples. This result is consistent with previous studies (Lydeard et

al. 2000, Campbell et al. 2005, Burdick and White 2007), and it is likely that '*F.*' *ebena* along with '*Obovaria*' *rotulata*, belong to a new genus. A more thorough examination of *Obovaria*, including the type species, *O. retusa* will be needed before any taxonomic recommendation can be made for this clade.

### **Geometric Morphometric Analysis of Conchology**

Table 3 summarizes the results of Goodall's F tests, and all pairwise comparisons resulted in values of  $p=0$  with the exceptions of *Fusconaia flava* versus *Pleurobema riddellii* and *Fusconaia subrotunda* versus *Pleurobema sintoxia*. Figure 3 illustrates comparisons in mean Procrustes superimpositions of the landmark data for selected pairings representing "species" pairs that are vastly different in shape and some that are very similar in shape. These illustrations provide a representation of the mean shell shape (2-dimensional, shell outline) of the species. Specimens representing the various "species" can be seen in Plates 1-9.

The PCA of the *Fusconaia* taxa yielded only one distinct eigenvalue. The PC1 axis described 67.5% of the total variation, and the PC2 axis described approximately 14% of the total variation. Figure 4.1 illustrates the separation achieved within the PCA, and *Fusconaia ozarkensis* was the only taxon that did not overlap with the other congeners. The PCA of the *Pleurobema* taxa yielded 3 distinct eigenvalues with the PC1 axis describing 67.5%, PC2 axis 16.2%, and PC3 axis 9% of the total variation. Figure 4.2 shows the scatter plot resulting from the PCA, and there is a high degree of segregation of the taxa with only minor overlaps.

Results of the CVA analyses are illustrated in Figures 5.1 and 5.2. The *Fusconaia* CVA (Fig. 5.1) resulted in 4 distinct canonical variates, complete segregation among taxa, and 100% correct *a posteriori* assignment to corresponding groups based on Mahalanobis' distance between the individual and the group mean. The *Pleurobema* group CVA (Figure 5.2) resulted in 3 distinct canonical variates, complete separation among taxa, and 100% correct *a posteriori* assignment to corresponding groups.

The CVA subroutine "assign unknown taxa" was utilized to explore shape relationships of specimens collected during this study with shapes of the "known" taxa type drainage specimens. The 5 groups explored were 1) Ouachita *Fusconaia flava* clade (see Fig. 2), 2) *Pleurobema* sp. Group 1 Ouachita River drainage (initial determination *P. rubrum*), 3) *Pleurobema* sp. Group 2 Ouachita River

drainage (initial determination *Fusconaia* sp.), 4) St. Francis *Pleurobema* clade (see Figure 1), and 5) White River drainage *Pleurobema* sp.

Results of the assignments for Ouachita *Fusconaia flava* clade specimens (n=15) are illustrated in Figure 6.1. Based on external shape, the following assignments resulted: *F. askewi* (1, ASU 2766, see Fig. 2), *F. ebena* (2, including ASU 2804, see Figs. 2, 3.2, 4.2), *F. flava* (11, including ASU 2779, see Figs. 2, 3.1, 4.1 and ASU 2780, see Fig. 2), *F. subrotunda* (1), and *F. ozarkensis* (0).

CVA based assignments of *Pleurobema* sp. Group 1 Ouachita River drainage specimens that were initially identified as *Pleurobema rubrum* are illustrated in Figure 6.2. Assignments based on external shell landmarks for the 14 specimens included in this group are: *P. cordatum* (0), *P. riddellii* (0), *P. rubrum* (11, including ASU 2789, see Figs. 2, 5.5, 6.5 and ASU 2797, see Fig. 2), and *P. sintoxia* (3). Assignments for *Pleurobema* sp. Group 2 Ouachita River drainage (initially identified as *Fusconaia*) are shown in Figure 6.3. The assignments for these 11 specimens are: *P. cordatum* (1), *P. riddellii* (8, including ASU 2771, see Fig. 2), *P. rubrum* (0), and *P. sintoxia* (2, including ASU 2765, see Figs. 2, 8.3, 9.3).

The St. Francis *Pleurobema* clade CVA assignment results are shown in Figure 6.4. Assignments for the 11 specimens included in this clade analysis are: *P. cordatum* (2, including ASU 2727, see Figs. 2, 7.3, 7.4 and ASU 2728, see Fig. 2, ), *P. riddellii* (2, including ASU 2721, see Figs. 2, 7.1, 7.2), *P. rubrum* (1), and *P. sintoxia* (6, including ASU 2722, see Figs. 2, 5.4, 6.4 and ASU 2724, see Figs. 2, 7.5, 7.6).

Finally, the CVA-based assignments test for White River drainage *Pleurobema* sp. is illustrated in Figure 6.5. Both specimens of this clade (including ASU 2733, see Figs. 2, 8.5, 9.5) most closely resembled *P. sintoxia* in external shape comparison. However, this slab-sided and compressed “species” does not closely resemble any specimens we have previously encountered in Arkansas.

## CONCLUSION

Both genetic data and morphological data indicate there are two taxonomic units that have not been recognized in Arkansas, at least in the recent state literature. The Ouachita *Pleurobema* clade most likely represents *Pleurobema riddellii*, although final resolution of its exact phylogenetic relationship is pending acquisition of COI sequences from topotypic *P. beadleianum* and *P. riddellii*. Regardless of its final placement, this “species” has previously been identified as *Fusconaia flava*, *Pleurobema rubrum* or *P. sintoxia* by recent workers. The White River drainage *Pleurobema* sp. is a unique taxon both genetically and morphologically. Its compressed, slab-like conchology is unique and does not compare favorably with any other species previously examined. Likewise, resolution of its phylogenetic relationship is pending acquisition of additional specimens and COI sequences for both White River drainage *Pleurobema* sp. and topotypic *P. cordatum*.

There is potential for recognition an additional two “species” within the *Fusconaia/Pleurobema* group. The relationship between *Fusconaia flava*, *F. cerina*, and the *Fusconaia* Ouachita clade of this study is unclear, and recent studies have shown there may be two distinct taxonomic units residing within current concepts of *Pleurobema rubrum* (Campbell et al. 2005, Burdick and White 2007). Additional COI data and acquisition of more comparative taxonomic units for geometric morphological analyses are needed to resolve placement of the *Fusconaia* Ouachita clade and the St. Francis *Pleurobema* clade.

Resolution of the phylogeny for *Fusconaia* and *Pleurobema* species will likely require a large cooperative effort between multiple researchers with sampling from more individuals, drainages and gene regions, including the incorporation of nuclear markers, as well as comparisons between male and female mitotypes. However, this study provides the basis from which to develop these research questions and continue to build an understanding of the phylogenetic relationships for Arkansas *Fusconaia* and *Pleurobema* taxa.

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*Fusconaia* and *Pleurobema* Preliminary Analysis

Table 1. List of “known” *Pleurobema*, *Fusconaia* and outgroup taxa COI sequences acquired from Genbank and used for the phylogenetic analysis in this study.

<i>Taxa</i>	Genbank Accession #
<i>Fusconaia cerina</i>	AY613823
<i>Fusconaia cuneolus</i>	AY654998
<i>Fusconaia ebena</i>	AY654999
<i>Fusconaia flava</i> (Ohio River, IN - KY)	AF232822
<i>Pleurobema furvum</i>	AY613833
<i>Pleurobema oviforme</i>	AY613837
<i>Pleurobema perovatum</i>	AY654998
<i>Pleurobema rubrum</i>	AY613841
<i>Quadrula quadrula</i>	AF156511
<i>Amblema plicata</i>	DQ648083

*Fusconaia* and *Pleurobema* Preliminary Analysis

Table 2. Collection data [Arkansas State University Museum of Zoology Catalog Number (ASUMZ#), Tentative ID, Locality, Date, and Collector] for the strict consensus phylogenetic analysis (Fig. 1) completed to date.

ASUMZ#	Preliminary ID	Locality	Date	Collector
2762	<i>F. ebena</i>	AR, Ouachita Co. Ouachita River ca. 4.0 river miles upstream of Camden. Coll. 20050724:ORB14M. N33.62172 W092.83901.	24-Jul-05	J. L. Harris, A. J. Harris
OR3Lc	<i>F. ebena</i>	Ohio River	unknown	Monte McGregor lab
113	<i>Pleurobema</i> sp.	AR, Cross Co. St. Francis R. at Ash Bend ca. 6.5 rkm downstream of U.S. Hwy. 64 at Parkin. 20050211:01 Lat. 35.25360 Long. - 90.58167.	11-Feb-05	J. L. Harris, W. R. Posey
2736	<i>F. flava</i>	AR, Monroe Co. White River at River Mile 91 (Aberdeen) N34.60611; W91.33011.	14-Sep-99	J. L. Harris, W. R. Posey
2746	<i>F. flava</i>	AR, Yell Co. Petit Jean River ca. 4.0 km downstream of Co. Rd. 27. (Site PJR 3). N35.07767; W93.53241	29-July-01	J. L. Harris, et al.
2738	<i>F. flava</i>	AR, Monroe Co. White River at River Mile 91 (Aberdeen) N34.60611; W91.33011.	14-Sept-99	J. L. Harris, W. R. Posey
2744	<i>F. flava</i>	AR, Yell Co. Petit Jean River ca. 4.0 km downstream of Co. Rd. 27. (Site PJR 3). N35.07767; W93.53241	29-Jul-01	J. L. Harris, et al.
2751	<i>F. flava</i>	AR, Conway Co. West Fork Point Remove Creek ca. 1.6 km downstream of Hacker's Creek. N35.31942; W92.86933 (Site 3).	29-Aug-99	J. L. Harris
2755	<i>F. flava</i>	AR, Yell Co. Petit Jean River ca. 4.0 km upstream of confluence with Big Piney Creek. (Site PJR 4) N35.07680; W93.54244 .	29-Jul-01	J. L. Harris, M. Scott, M. Barnett
2766	<i>Fusconaia</i> sp.	AR, Ouachita Co. Ouachita River ca. 4.0 river miles upstream of Camden. Coll. 20050724:ORB14M. N33.62172 W092.83901.	24-Jul-05	J. L. Harris, A. J. Harris
2779	<i>F. flava</i>	AR, Ouachita Co. Ouachita River ca. 4.0 river miles upstream of Camden. Coll. 20050724:ORB14M. N33.62172 W092.83901.	24-Jul-05	J. L. Harris, A. J. Harris
2780	<i>F. flava</i>	AR, Ouachita Co. Ouachita River ca. 4.0 river miles upstream of Camden. Coll. 20050724:ORB14M. N33.62172 W092.83901.	25-Jul-05	J. L. Harris, A. J. Harris
2884	<i>F. flava</i>	AR, Clark-Ouachita Co. Little Missouri R. at Site 121 ca. 1.0 river km upstream of confluence with White Oak Cr. N33°78.660' W093°06.261'.	5-Sep-04	J. L. Harris, A. D. Christian
2804	<i>P. sintoxia</i>	AR, Ouachita Co. Ouachita River ca. 4.0 river miles upstream of Camden. Coll. 20050724:ORB14M. N33.62172 W092.83901.	25-Jul-05	J. L. Harris, A. J. Harris

*Fusconaia* and *Pleurobema* Preliminary Analysis

ASUMZ#	Tentative ID	Locality	Date	Collector
OSU7	<i>F. flava</i>	OH, Muskingum Co. Muskingum River at Dresden. N40.12076; W81.99998.	unknown	OSUMZ mussel facility - tissue sample by J. L. Harris; D. M. Hayes
110	<i>Pleurobema sp.</i>	AR, Cross Co. St. Francis R. at Ash Bend ca. 6.5 rkm downstream of U.S. Hwy. 64 at Parkin. 20050211:01 Lat. 35.25360 Long. - 90.58167.	11-Feb-05	J. L. Harris, W. R. Posey
104	<i>Pleurobema sp.</i>	AR, Cross Co. St. Francis R. at Ash Bend ca. 6.5 rkm downstream of U.S. Hwy. 64 at Parkin. 20050211:01 Lat. 35.25360 Long. - 90.58167.	11-Feb-05	J. L. Harris, W. R. Posey
107	<i>Pleurobema sp.</i>	AR, Cross Co. St. Francis R. at Ash Bend ca. 6.5 rkm downstream of U.S. Hwy. 64 at Parkin. 20050211:01 Lat. 35.25360 Long. - 90.58167.	11-Feb-05	J. L. Harris, W. R. Posey
111	<i>Pleurobema sp.</i>	AR, Cross Co. St. Francis R. at Ash Bend ca. 6.5 rkm downstream of U.S. Hwy. 64 at Parkin. 20050211:01 Lat. 35.25360 Long. - 90.58167.	11-Feb-05	J. L. Harris, W. R. Posey
105	<i>Pleurobema sp.</i>	AR, Cross Co. St. Francis R. at Ash Bend ca. 6.5 rkm downstream of U.S. Hwy. 64 at Parkin. 20050211:01 Lat. 35.25360 Long. - 90.58167.	11-Feb-05	J. L. Harris, W. R. Posey
FR1	<i>Pleurobema sintoxia</i>	PA, Crawford/Erie Co., French Cr. at Dutch Rd xing N41.8520048 W79.9778362	11-May-07	A. Peck
3252	<i>Pleurobema sp.</i>	AR, Dallas/Grant Co. Saline River at U.S. Hwy 167 xing ca. 11 rdmi. South of Sheridan. Lat. 34.11545° Long. 92.40587°	May 18, 19, 23, 24 and June 6 2006	J. L. Harris, J. Seagraves, A. Peck, K. Inoue
2753	<i>P. sintoxia</i>	AR, Yell Co. Petit Jean River ca. 3.0 km upstream of Co. Rd. 27 (Site PJR 11). N35.08999; W93.58037	19-Aug-2001	J. L. Harris, J. Seagraves
2801	<i>P. rubrum</i>	AR, Ouachita Co. Ouachita River ca. 4.0 river miles upstream of Camden. Coll. 20050724:ORB14M. N33.62172 W092.83901.	24-Jul-05	J. L. Harris, A. J. Harris
2856	<i>F. flava</i>	AR, Clark-Ouachita Co. Little Missouri R. at Site 121 ca. 1.0 river km upstream of confluence with White Oak Cr. N33°78.660' W093°06.261'.	5-Sept-04	J. L. Harris, A. D. Christian
2789	<i>P. rubrum</i>	AR, Ouachita Co. Ouachita River ca. 4.0 river miles upstream of Camden. Coll. 20050724:ORB14M. N33.62172 W092.83901.	24-Jul-05	J. L. Harris, A. J. Harris
2797	<i>P. rubrum</i>	AR, Ouachita Co. Ouachita River ca. 4.0 river miles upstream of Camden. Coll. 20050724:ORB14M. N33.62172 W092.83901.	24-July-05	J. L. Harris, A. J. Harris

*Fusconaia* and *Pleurobema* Preliminary Analysis

ASUMZ#	Tentative ID	Locality	Date	Collector
2803	<i>Pleurobema sintoxia</i>	AR, Ouachita Co. Ouachita River ca. 4.0 river miles upstream of Camden. Coll. 20050724:ORB14M. N33.62172 W092.83901.	24-Jul-05	J. L. Harris, A. J. Harris
2911	<i>F. ozarkensis</i>	AR, Washington Co. Illinois River ca. 800 m upstream of Robinson Rd. crossing. Lat. 36.13266 Long. -94.34980.	30-Jun-05	W. R. Posey
3279	<i>F. flava</i>	AR, Dallas/Grant Co. Saline River at U.S. Hwy 167 xing ca. 11 rdmi. South of Sheridan. Lat. 34.11545° Long. 92.40587°	May 18, 19, 23, 24 and June 6 2006	J. L. Harris, J. Seagraves, A. Peck, K. Inoue
BR211		AR, Lawrence-Randolph Co. Black R. ca. 1.0 km upstream of Spring River. N36.11904; W91.06327. (RM 72.8)	28-Jul-07	J. L. Harris, D. M. Hayes
LM1	<i>P. sintoxia</i>	PA, Little Mahoning River	unknown	T. Smith
2879	<i>P. rubrum</i>	AR, Clark-Ouachita Co. Little Missouri R. at Site 121 ca. 1.0 river km upstream of confluence with White Oak Cr. N33°78.660' W093°06.261'	5-Sept-04	J. L. Harris, A. D. Christian
2733	<i>Fusconaia sp.</i>	AR, Cleburne Co. Middle Fork Little Red River at Winterringer thesis Site 4. Lat. N38°48.844 W092°33.111.	18-Nov-01	J. L. Harris, A. D. Christian, R. Winterringer
2704	<i>F. askewi</i>	Texas, Panola-Harrison Co. Sabine River upstream of U. S. Hwy. 59. N32°20.210 W94°21.643.	14-May-03	J. L. Harris, A. D. Christian
2765	<i>Fusconaia sp.</i>	AR, Ouachita Co. Ouachita River ca. 4.0 river miles upstream of Camden. Coll. 20050724:ORB14M. N33.62172 W092.83901.	24-Jul-05	J. L. Harris, A. J. Harris
2788	<i>F. flava</i>	AR, Ouachita Co. Ouachita River ca. 4.0 river miles upstream of Camden. Coll. 20050724:ORB14M. N33.62172 W092.83901.	24-Jul-05	J. L. Harris, A. J. Harris
3274	<i>P. sintoxia</i>	AR, Dallas/Grant Co. Saline River at U.S. Hwy 167 xing ca. 11 rdmi. South of Sheridan. Lat. 34.11545° Long. 92.40587°	May 18, 19, 23, 24 and June 6 2006	J. L. Harris, J. Seagraves, A. Peck, K. Inoue
3253	<i>Pleurobema sp.</i>	AR, Dallas/Grant Co. Saline River at U.S. Hwy 167 xing ca. 11 rdmi. South of Sheridan. Lat. 34.11545° Long. 92.40587°	May 18, 19, 23, 24 and June 6 2006	J. L. Harris, J. Seagraves, A. Peck, K. Inoue
3264	<i>Pleurobema sp.</i>	AR, Dallas/Grant Co. Saline River at U.S. Hwy 167 xing ca. 11 rdmi. South of Sheridan. Lat. 34.11545° Long. 92.40587°	May 18, 19, 23, 24 and June 6 2006	J. L. Harris, J. Seagraves, A. Peck, K. Inoue
3257	<i>Pleurobema sp.</i>	AR, Dallas/Grant Co. Saline River at U.S. Hwy 167 xing ca. 11 rdmi. South of Sheridan. Lat. 34.11545° Long. 92.40587°	May 18, 19, 23, 24 and June 6 2006	J. L. Harris, J. Seagraves, A. Peck, K. Inoue
2771	<i>Fusconaia sp.</i>	AR, Ouachita Co. Ouachita River ca. 4.0 river miles upstream of Camden. Coll. 20050724:ORB14M. N33.62172 W092.83901.	24-Jul-05	J. L. Harris, A. J. Harris

*Fusconaia* and *Pleurobema* Preliminary Analysis

Table 3. Results of Goodall's F (Procrustes) analyses for *Fusconaia* species. D = minimized partial Procrustes distance between means.

	<i>Fusconaia askewi</i> (n=15)	<i>Fusconaia ebena</i> (n=17)	<i>Fusconaia flava</i> (n=31)	<i>Fusconaia ozarkensis</i> (n=24)	<i>Fusconaia subrotunda</i> (n=18)
<i>Fusconaia askewi</i> (n=15)	NA	F=24.06; p=0; df 52, 1560; D=0.0887	F=6.05; p=0; df 52, 2288; D=0.0458	F=92.14; p=0; df 52, 1924; D=0.1808	F=19.71; p=0; df 52, 1612; D=0.0842
<i>Fusconaia ebena</i> (n=17)	NA	NA	F=17.88; p=0; df 52, 2392; D=0.0733	F=115.65; p=0; df 52, 2028; D=0.1883	F=9.33; p=0; df 52, 1716; D=0.0537
<i>Fusconaia flava</i> (n=31)	NA	NA	NA	F=110.30; p=0; df 52, 2756; D=0.1732	F=17.86; p=0; df 52, 2444; D=0.072
<i>Fusconaia ozarkensis</i> (n=24)	NA	NA	NA	NA	F=61.27; p=0; df 52, 2080; D=0.1401
<i>Fusconaia subrotunda</i> (n=18)	NA	NA	NA	NA	NA

Table 4. Results of Goodall's F (Procrustes) analyses for *Pleurobema* species. D = minimized partial Procrustes distance between means.

	<i>Pleurobema cordatum</i> (n=4)	<i>Pleurobema riddellii</i> (n=19)	<i>Pleurobema rubrum</i> (n=17)	<i>Pleurobema sintoxia</i> (n=22)
<i>Pleurobema cordatum</i> (n=4)	NA	F=24.97; p=0 df 52, 1092; D=0.1568	F=4.07; p=0; df 52, 988; D=0.0861	F=7.62; p=0; df 52, 1248; D=0.1003
<i>Pleurobema riddellii</i> (n=19)	NA	NA	F=35.37; p=0; df 52, 1768; D=0.1254	F=14.28; p=0; df 52, 2028; D=0.0684
<i>Pleurobema rubrum</i> (n=17)	NA	NA	NA	F=13.81; p=0; df 52, 1924; D=0.0824
<i>Pleurobema sintoxia</i> (n=22)	NA	NA	NA	NA

Table 5. Results of Goodall's F (Procrustes) analyses for *Fusconaia* versus *Pleurobema* species. D = minimized partial Procrustes distance between means.

	<i>Fusconaia askewi</i> (n=15)	<i>Fusconaia ebena</i> (n=17)	<i>Fusconaia flava</i> (n=31)	<i>Fusconaia ozarkensis</i> (n=24)	<i>Fusconaia subrotunda</i> (n=18)
<i>Pleurobema cordatum</i> (n=4)	F=22.34; p=0; df 52, 884; D=0.1618	F=17.25; p=0; df 52, 988; D=0.1303	F=21.68; p=0; df 52, 1716; D=0.1592	F=16.79; p=0; df 52, 1352; D=0.1393	F=9.75; p=0; df 52, 1040; D=0.1047
<i>Pleurobema riddellii</i> (n=19)	F=5.85; p=0; df 52, 1664; D=0.0433	F=12.55; p=0; df 52, 1768; D=0.0586	F=2.78; p=2.75e-010; df 52, 2496; D=0.0280	F=121.94; p=0; df 52, 2132; D=0.1885	F=17.49; p=0; df 52, 1820; D=0.0723
<i>Pleurobema rubrum</i> (n=17)	F=38.58; p=0; df 52, 1560; D=0.1450	F=13.95; p=0; df 52, 1664; D=0.0809	F=41.71; p=0; df 52, 2392; D=0.1298	F=64.62; p=0; df 52, 2028; D=0.1686	F=13.74; p=0; df 52, 1716; D=0.0819
<i>Pleurobema sintoxia</i> (n=22)	F=14.32; p=0; df 52, 1820; D=0.0757	F=11.57; p=0; df 52, 1924; D=0.0632	F=13.99; p=0; df 52, 2652; D=0.0650	F=52.58; p=0; df 52, 2288; D=0.1312	F=2.82; p=1.741e-010; df 52, 1976; D=0.0319

Figure 1. External landmarks used in geometric morphometric analysis.



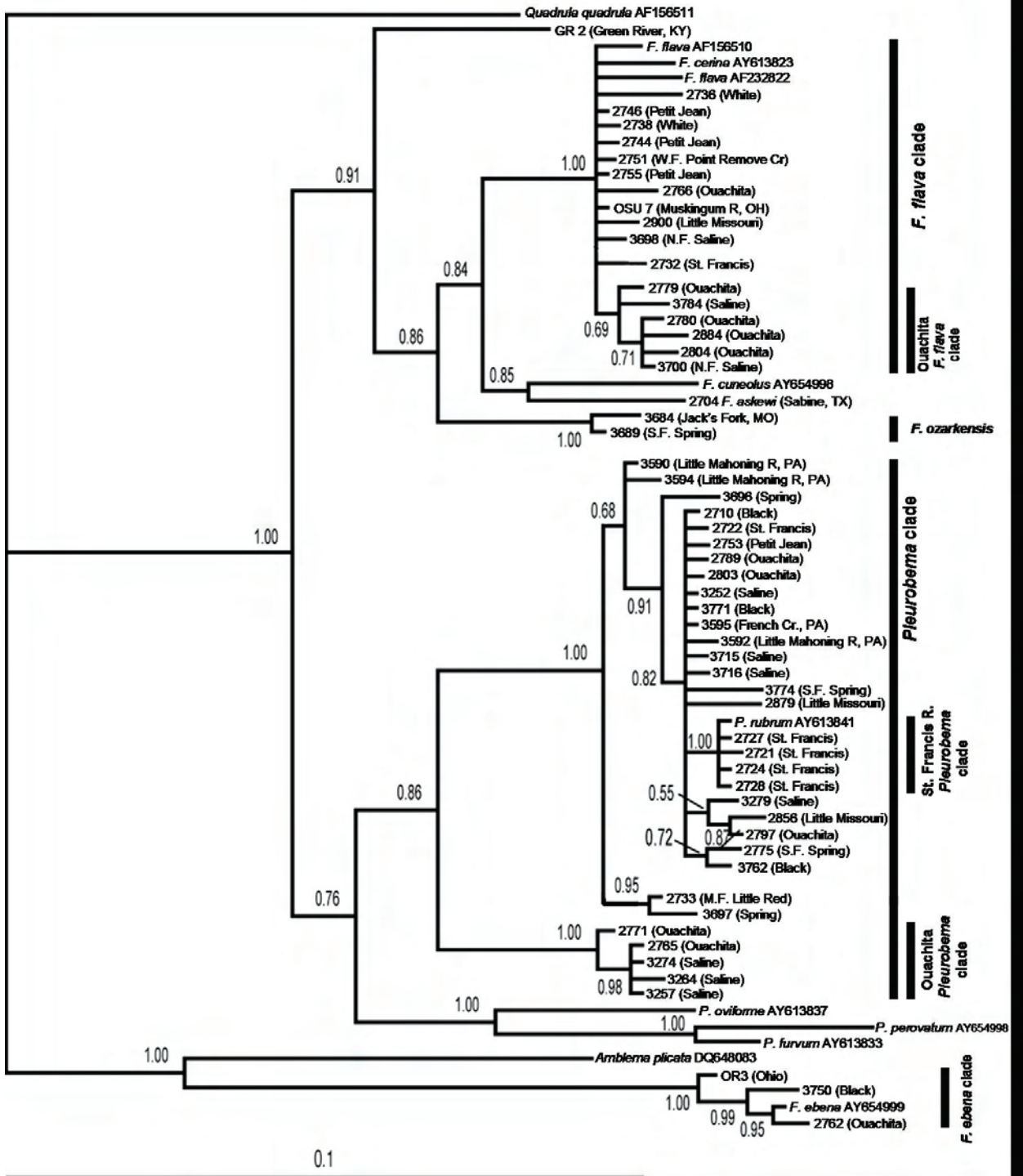


Figure 2. Majority-rule tree from Bayesian analysis of the COI data set. Numbers above the branches are the posterior probability support for those nodes. Individuals are numbered with ASUMZ number and drainage locality and correspond to Table 2. Previously published sequences are labeled with taxon and GenBank Accession number (see Table 1).

*Fusconaia* and *Pleurobema* Preliminary Analysis

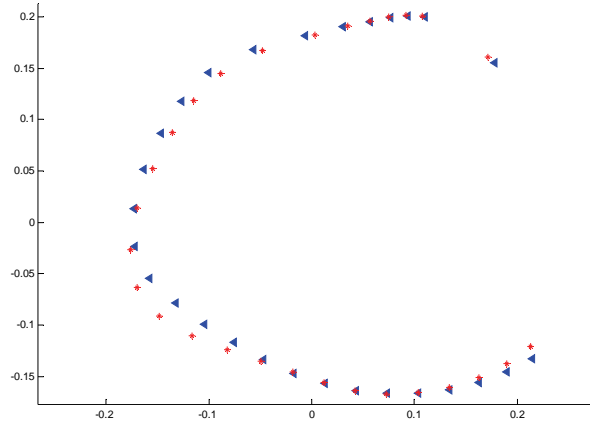


Figure 3.1. *Fusconaia askewi* (blue triangles) versus *F. flava* (red stars) landmarks mean Procrustes superimposition.

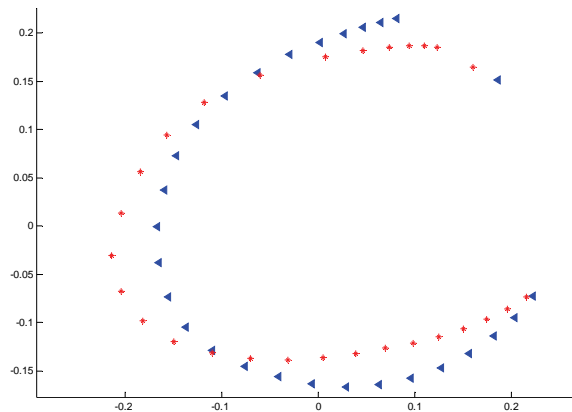


Figure 3.2. *Fusconaia ebena* (blue triangles) versus *F. ozarkensis* (red stars) landmarks mean Procrustes superimposition.

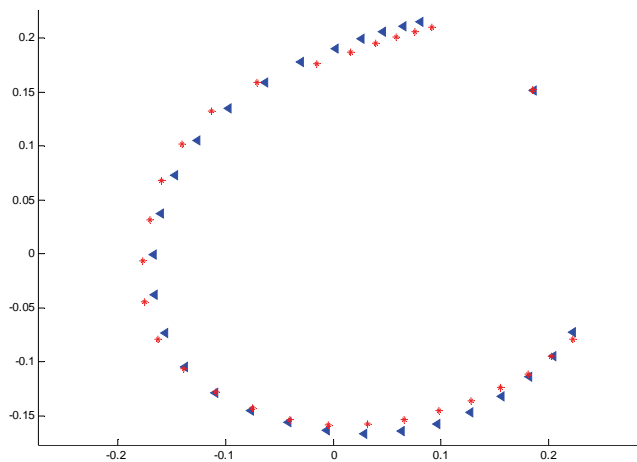


Figure 3.3. *Fusconaia ebena* (blue triangles) versus *F. subrotunda* (red stars) landmarks mean Procrustes superimposition.



*Fusconaia* and *Pleurobema* Preliminary Analysis

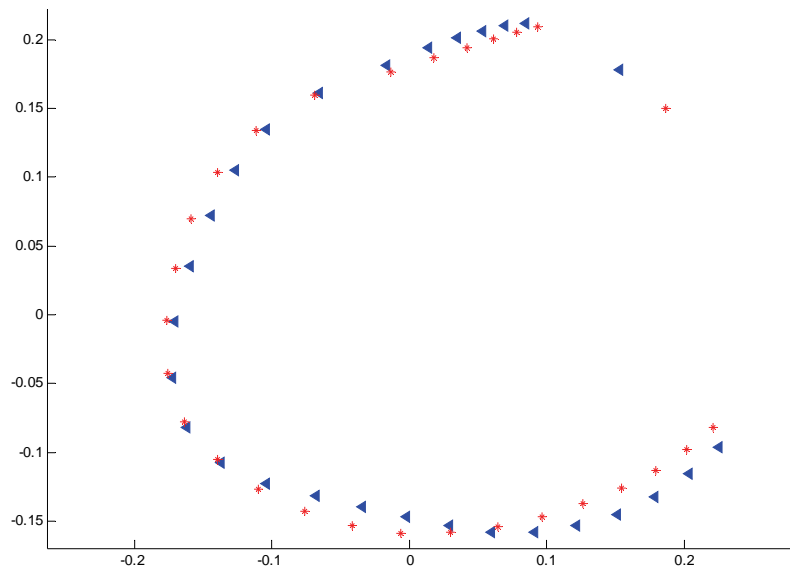


Figure 3.4. *Fusconaia flava* (blue triangles) versus *F. subrotunda* (red stars) landmarks mean Procrustes superimposition.

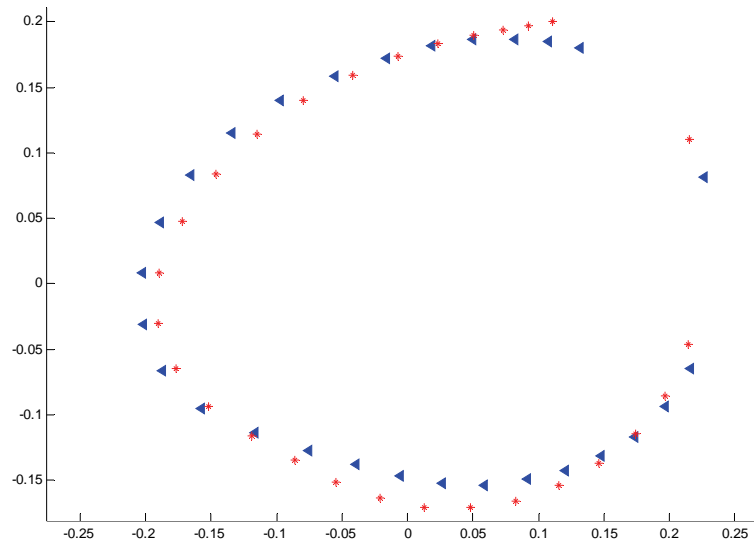


Figure 3.5. *Pleurobema cordatum* (blue triangles) versus *P. rubrum* (red stars) landmarks mean Procrustes superimposition.

*Fusconaia* and *Pleurobema* Preliminary Analysis

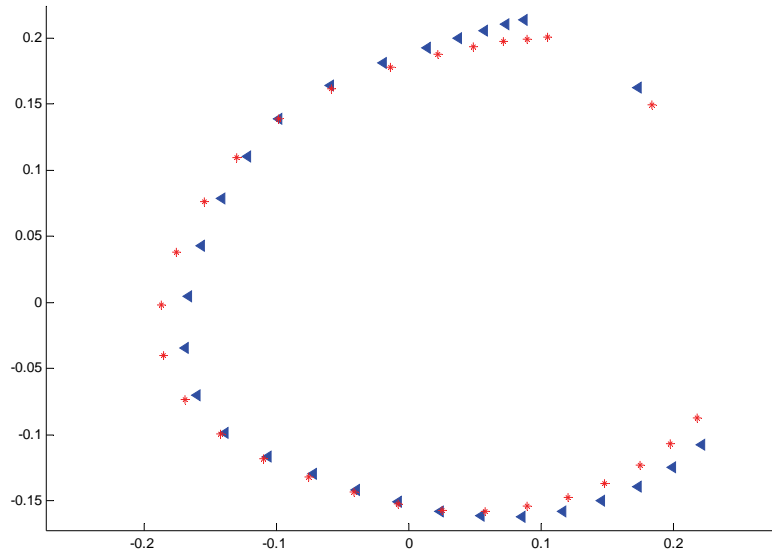


Figure 3.6. *Pleurobema riddellii* (blue triangles) versus *P. sintoxia* (red stars) landmarks mean Procrustes superimposition.

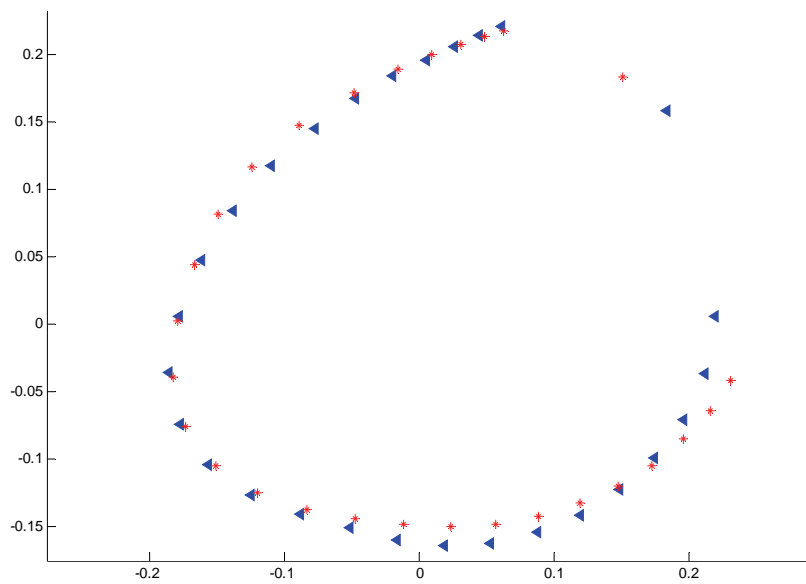


Figure 3.7. *Pleurobema rubrum* (blue triangles) versus *P. sintoxia* (red stars) landmarks mean Procrustes superimposition.

*Fusconaia* and *Pleurobema* Preliminary Analysis

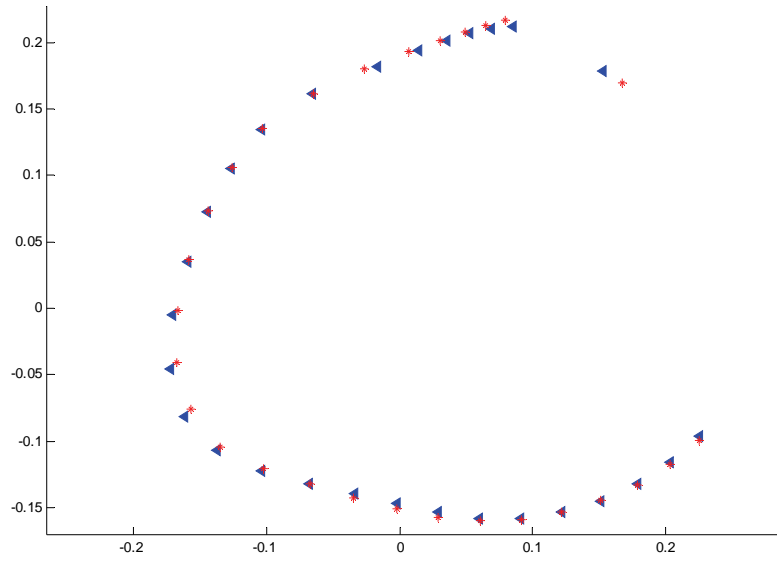


Figure 3.8. *Fusconaia flava* (blue triangles) versus *Pleurobema riddellii* (red stars) landmarks mean Procrustes superimposition.

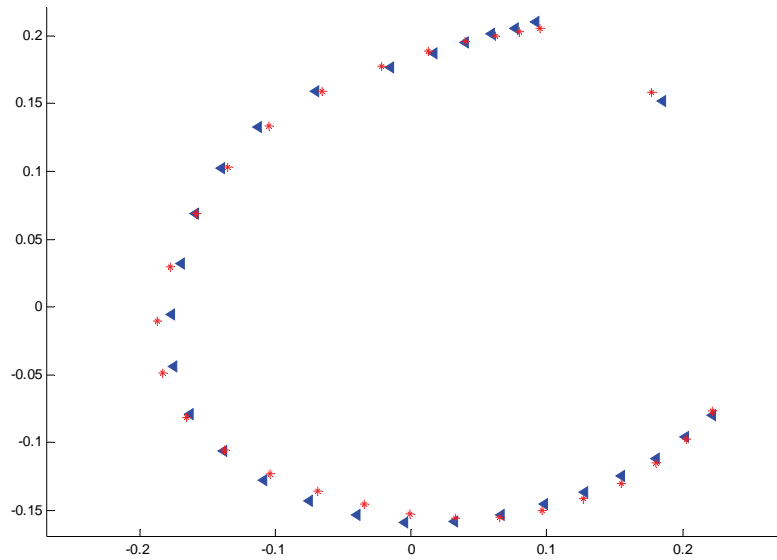


Figure 3.9. *Fusconaia subrotunda* (blue triangles) versus *Pleurobema sintoxia* (red stars) landmarks mean Procrustes superimposition.

*Fusconaia* and *Pleurobema* Preliminary Analysis

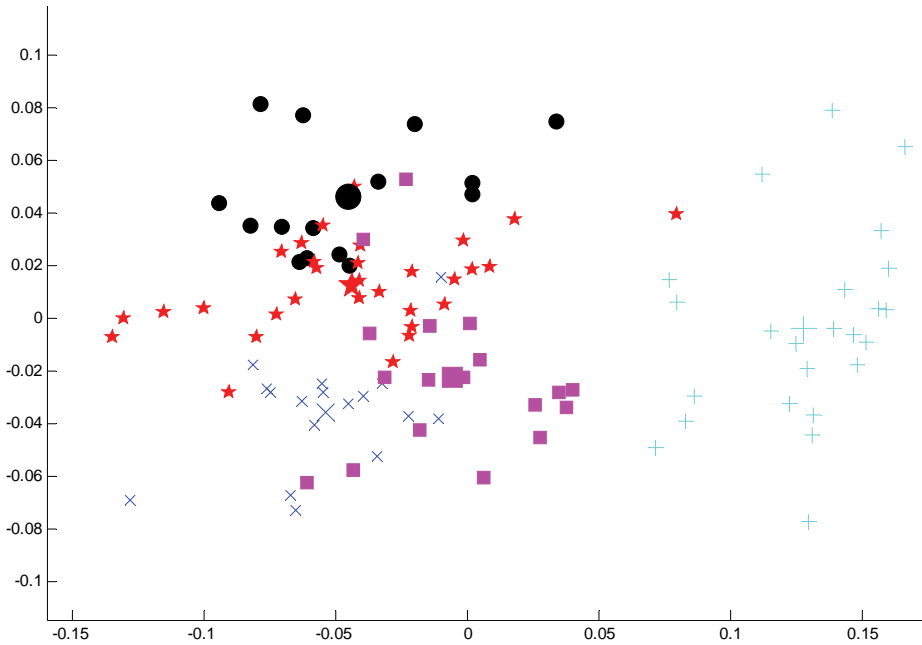


Figure 4.1. Scatter plot from PCA of *Fusconaia* external landmarks from type drainage specimens. *F. askewi* (black circle), *F. ebena* (blue x), *F. flava* (red star), *F. subrotunda* (magenta square), and *F. ozarkensis* (aquamarine +). Enlarged symbols represent group mean. (n specimens=105, n distinct eigenvalues=1).

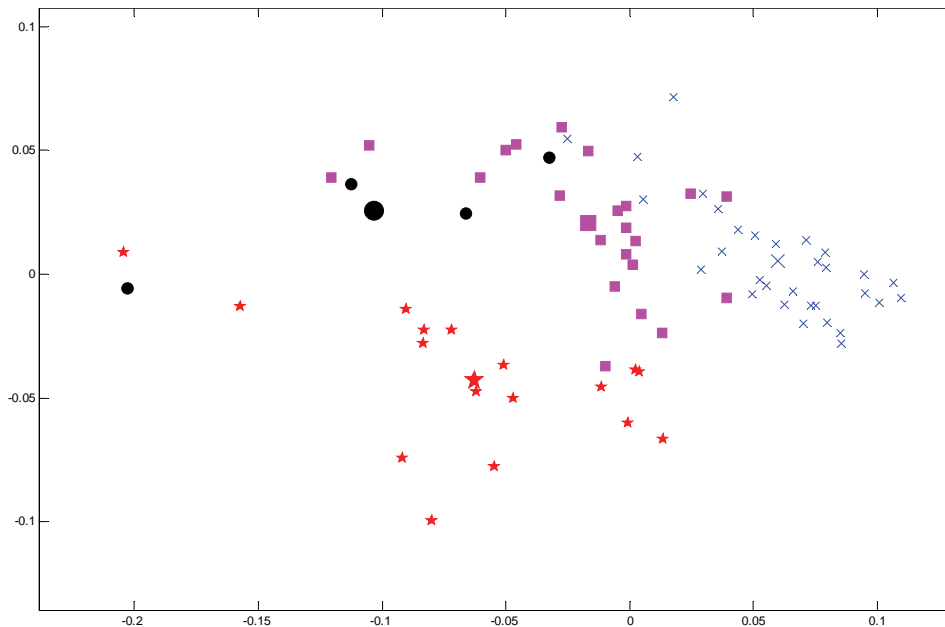


Figure 4.2. Scatter plot from PCA of external landmarks from *Pleurobema* type drainage specimens. *P. cordatum* (black circle), *P. riddellii* (blue x), *P. rubrum* (red star), and *P. sintoxia* (magenta square). Enlarged symbols represent group mean. (n specimens=74, n distinct eigenvalues=3).

*Fusconaia* and *Pleurobema* Preliminary Analysis

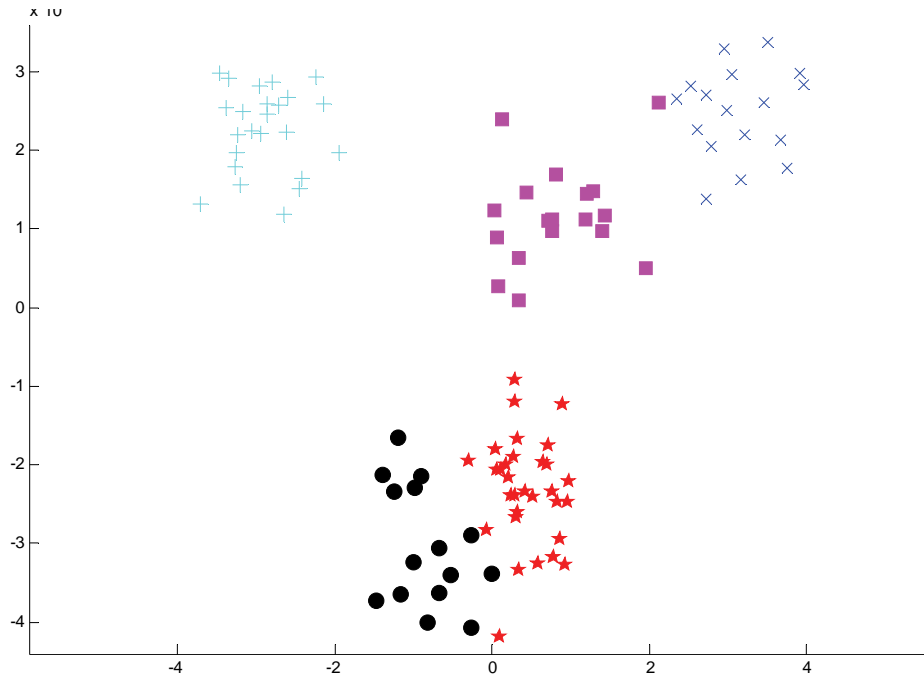


Figure 5.1. Scatter plot from CVA of external landmarks from *Fusconaia* type drainage specimens. *F. askewi* (black circle), *F. ebena* (blue x), *F. flava* (red star), *F. subrotunda* (magenta square), and *F. ozarkensis* (aquamarine +). (n specimens=105, n distinct canonical variates=4). All 105 *a posteriori* specimen assignments to correct groups.

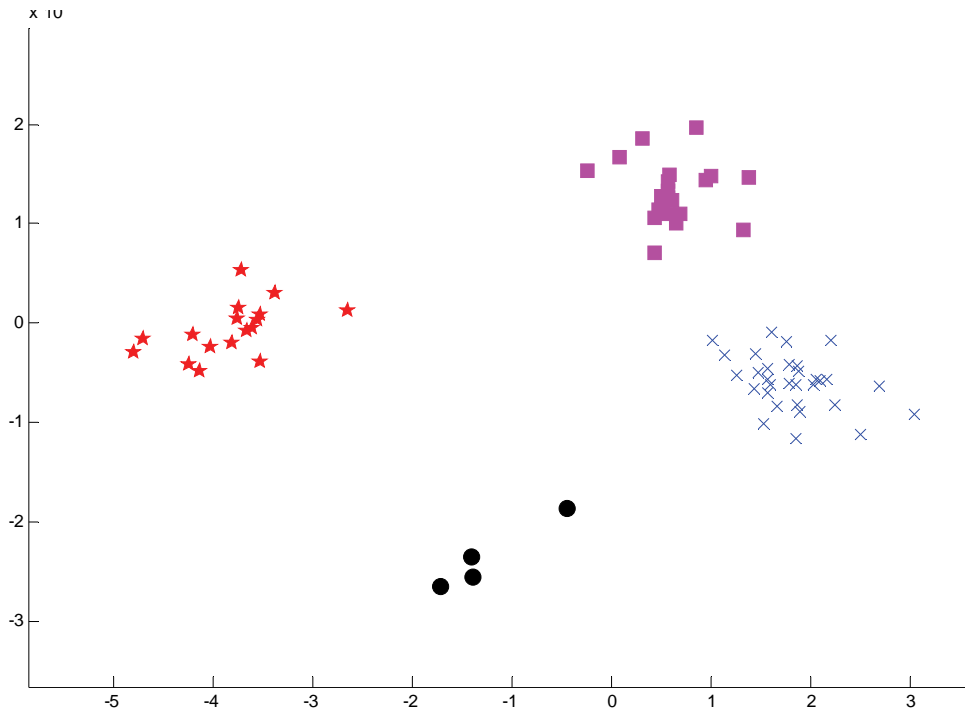


Figure 5.2. Scatter plot from CVA of external landmarks from *Pleurobema* type drainage specimens. *P. cordatum* (black circle), *P. riddellii* (blue x), *P. rubrum* (red star), and *P. sintoxia* (magenta square). (n specimens=74, n distinct canonical variates=3). All 74 *a posteriori* specimen assignments to correct groups.

*Fusconaia* and *Pleurobema* Preliminary Analysis

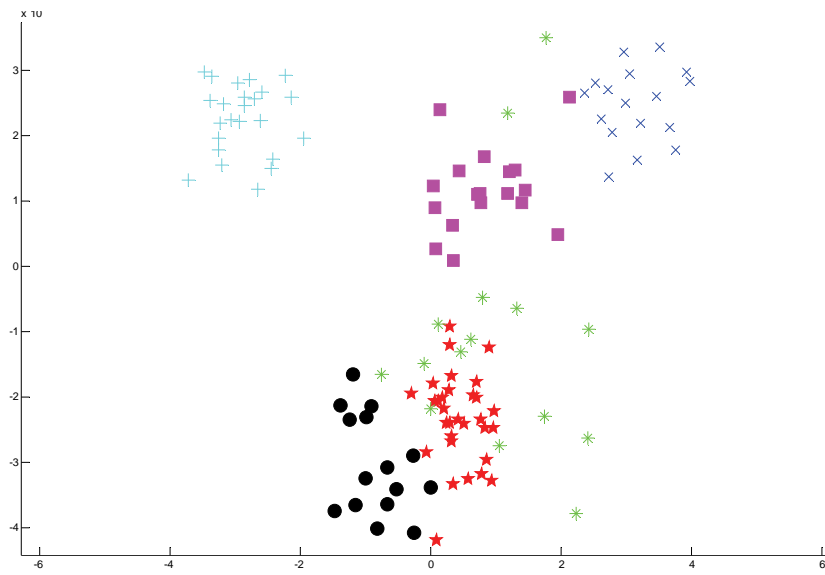


Figure 6.1. Scatter plot from CVA of external landmarks from *Fusconaia* type drainage specimens and assignment of *Fusconaia* sp. unknowns from Ouachita River drainage. *F. askewi* (black circle), *F. ebena* (blue x), *F. flava* (red star), *F. subrotunda* (magenta square), *F. ozarkensis* (aquamarine +), and *Fusconaia* sp. Ouachita River (green \*, n=15). All 105 *a posteriori* specimen assignments to correct groups. *Fusconaia* sp. assignments: *F. askewi* (1), *F. ebena* (2), *F. flava* (11), *F. subrotunda* (1), *F. ozarkensis* (0).

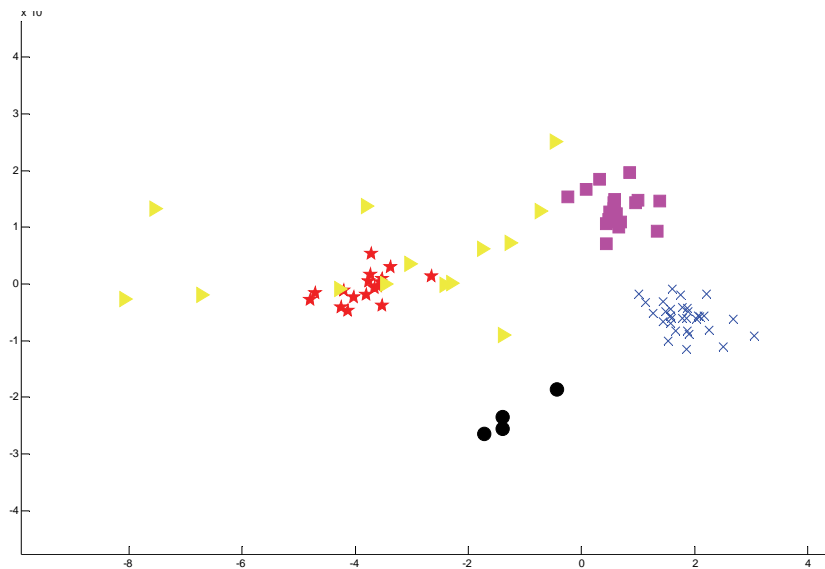


Figure 6.2. Scatter plot from CVA of external landmarks from *Pleurobema* type drainage specimens and assignment of *Pleurobema* sp. Group 1 (tentative *P. rubrum*) unknowns from Ouachita River drainage. All 74 *a posteriori* specimen assignments to correct groups. *P. cordatum* (black circle), *P. riddellii* (blue x), *P. rubrum* (red star), *P. sintoxia* (magenta square), and *Pleurobema* sp. Group 1 Ouachita River (green \*, n=14). 3 distinct canonical variates. *Pleurobema* sp. assignments: *P. cordatum* (0), *P. riddellii* (0), *P. rubrum* (11), *P. sintoxia* (3).

*Fusconaia* and *Pleurobema* Preliminary Analysis

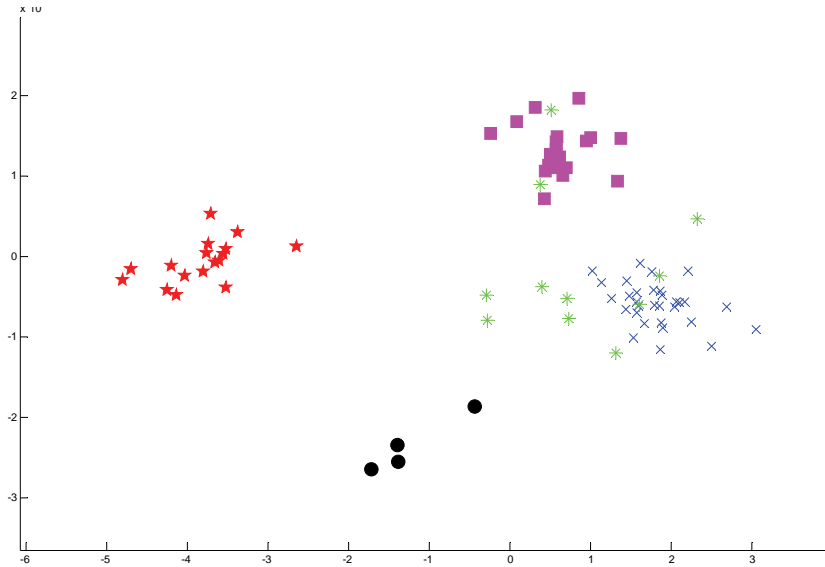


Figure 6.3. Scatter plot from CVA of external landmarks from *Pleurobema* type drainage specimens and assignment of *Pleurobema* sp. Group 2 unknowns (tentative *Fusconaia*) from Ouachita River drainage. All 74 *a posteriori* specimen assignments to correct groups. *P. cordatum* (black circle), *P. riddellii* (blue x), *P. rubrum* (red star), *P. sintoxia* (magenta square), and *Pleurobema* sp. Ouachita River (green \*, n=11). 3 distinct canonical variates. *Pleurobema* sp. assignments: *P. cordatum* (1), *P. riddellii* (8), *P. rubrum* (0), *P. sintoxia* (2).

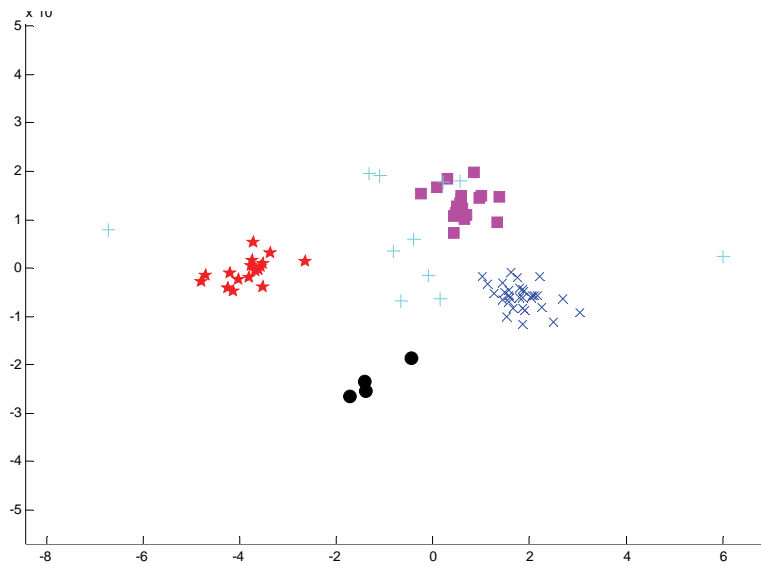


Figure 6.4. Scatter plot from CVA of external landmarks from *Pleurobema* type drainage specimens and assignment of *Pleurobema* sp. unknowns from St. Francis River drainage. All 74 *a posteriori* specimen assignments to correct groups. *P. cordatum* (black circle), *P. riddellii* (blue x), *P. rubrum* (red star), *P. sintoxia* (magenta square), and *Pleurobema* sp. St. Francis River (aquamarine +, n=11). 3 distinct canonical variates. *Pleurobema* sp. assignments: *P. cordatum* (2), *P. riddellii* (2), *P. rubrum* (1), *P. sintoxia* (6).

*Fusconaia* and *Pleurobema* Preliminary Analysis

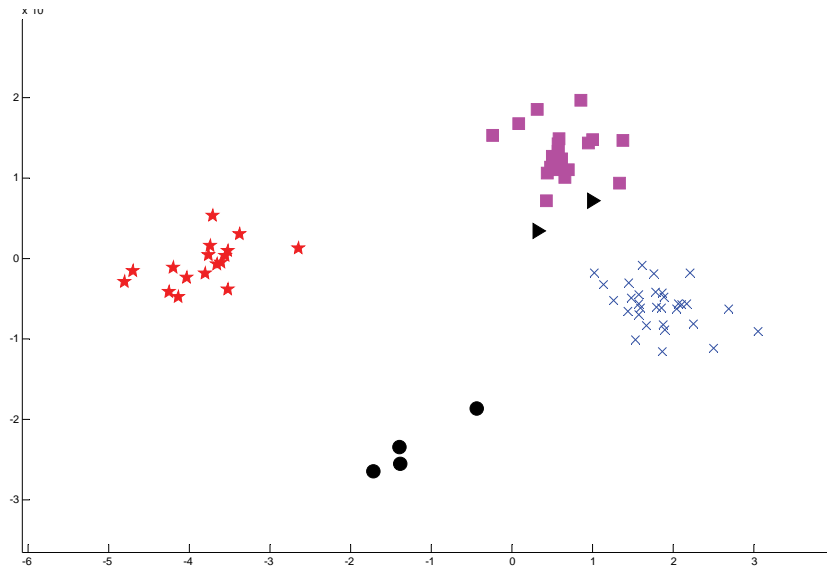


Figure 6.5. Scatter plot from CVA of external landmarks from *Pleurobema* type drainage specimens and assignment of *Pleurobema* sp. unknowns from White River drainage. All 74 *a posteriori* specimen assignments to correct groups. *P. cordatum* (black circle), *P. riddellii* (blue x), *P. rubrum* (red star), *P. sintoxia* (magenta square), and *Pleurobema* sp. White River (black triangle, n=2). 3 distinct canonical variates. *Pleurobema* sp. assignments: *P. cordatum* (0), *P. riddellii* (0), *P. rubrum* (0), *P. sintoxia* (2).



*Fusconaia* and *Pleurobema* Preliminary Analysis



1.1



1.2



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Plate 1. *Fusconaia flava* Arkansas specimens (external view).

1.1) AR, Craighead County. Cache River, USNM 272249, *F. selecta* syntype. L 55.9, H 44.9.

1.2) AR, Cross County. St. Francis River. ASU 2732. L 80.4, H 68.7

1.3) AR, Monroe County. White River. ASU 2738. L 32.3, H 29.0

1.4) AR, Yell County. Petit Jean River. ASU 2746. L 84.4, H 62.2

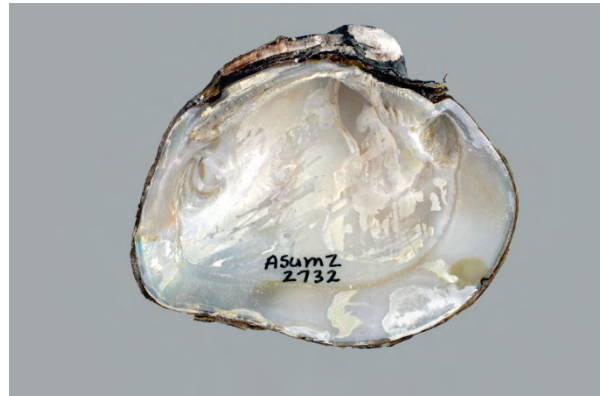
1.5) AR, Yell County. Petit Jean River. ASU 2755. L 63.6, H 54.0

1.6) AR, Conway County. West Fork Point Remove Creek. ASU 2751. L 67.2, H 49.1

*Fusconaia* and *Pleurobema* Preliminary Analysis



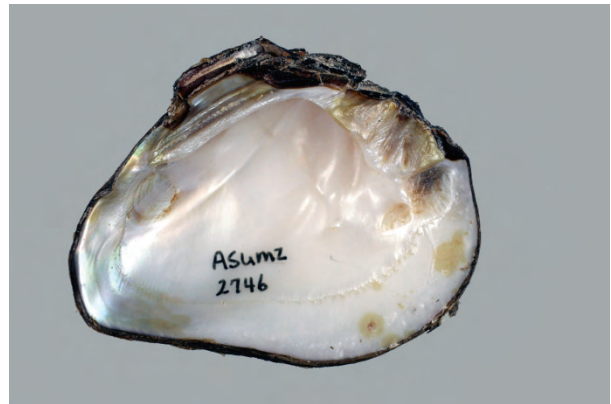
2.1



2.2



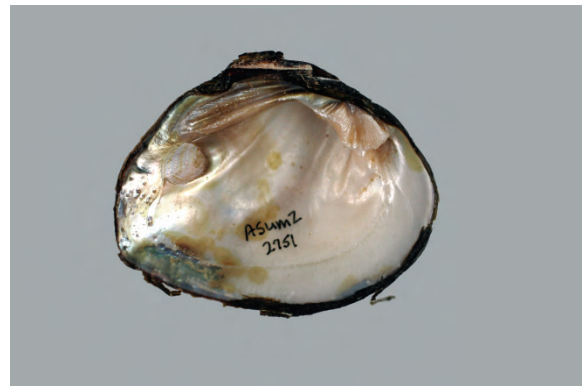
2.3



2.4



2.5



2.6

Plate 2. *Fusconaia selecta* and *F. flava* Arkansas specimens (internal view).

2.1) AR, Craighead County. Cache River, USNM 272249, *F. selecta* syntype. L 55.9, H 44.9, W 36.7.

2.2) AR, Cross County. St. Francis River. ASU 2732. L 80.4, H 68.7

2.3) AR, Monroe County. White River. ASU 2738. L 32.3, H 29.0

2.4) AR, Yell County. Petit Jean River. ASU 2746. L 84.4, H 62.2

2.5) AR, Yell County. Petit Jean River. ASU 2755. L 63.6, H 54.0

2.6) AR, Conway County. West Fork Point Remove Creek. ASU 2751. L 67.2, H 49.1

*Fusconaia* and *Pleurobema* Preliminary Analysis



3.1



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3.3



3.4



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3.6

Plate 3. *Fusconaia flava* Ouachita clade, *F. askewi*, *F. ozarkensis*, and *F. ebena* (external view).

3.1) AR, Ouachita County. Ouachita River. ASU 2779. L 53.8, H 45.8

3.2) AR, Ouachita County. Ouachita River. ASU 2804. L 61.7, H 51.0

3.3) *F. askewi*. TX, Harrison-Panola Co. Sabine River. ASU 2704.

3.4) *F. ozarkensis*. MO, Shannon Co. Jacks Fork Current River. ASU 3684

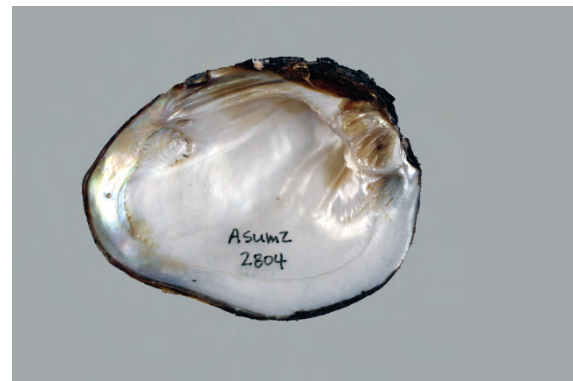
3.5) *F. ebena*. OH, Cincinnati. Ohio River. USNM 85792. L 43.8, H 36.2

3.6) *F. ebena*. AR, Ouachita Co. Ouachita River. ASU 2762. L 52.4, H 45.3

*Fusconaia* and *Pleurobema* Preliminary Analysis



4.1



4.2



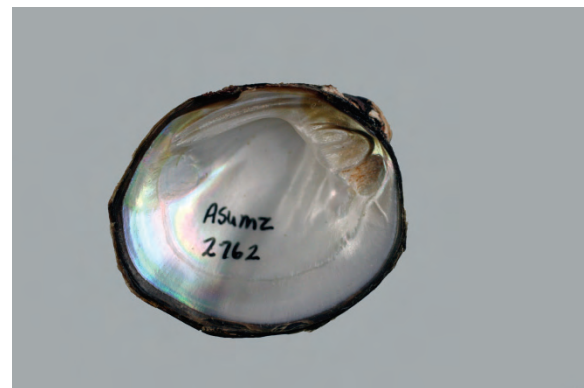
4.3



4.4



4.5



4.6

Plate 4. *Fusconaia flava* Ouachita clade, *F. askewi*, *F. ozarkensis*, and *F. ebena* (internal view).

4.1) AR, Ouachita County. Ouachita River. ASU 2779. L 53.8, H 45.8

4.2) AR, Ouachita County. Ouachita River. ASU 2804. L 61.7, H 51.0

4.3) *F. askewi*. TX, Harrison-Panola Co. Sabine River. ASU 2704. L

4.4) *F. ozarkensis*. MO, Shannon Co. Jacks Fork Current River. ASU 3684

4.5) *F. ebena*. OH, Cincinnati. Ohio River. USNM 85792. L43.8, H 36.2

4.6) *F. ebena*. AR, Ouachita Co. Ouachita River. ASU 2762. L 52.4, H 45.3

*Fusconaia* and *Pleurobema* Preliminary Analysis



5.1



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5.4



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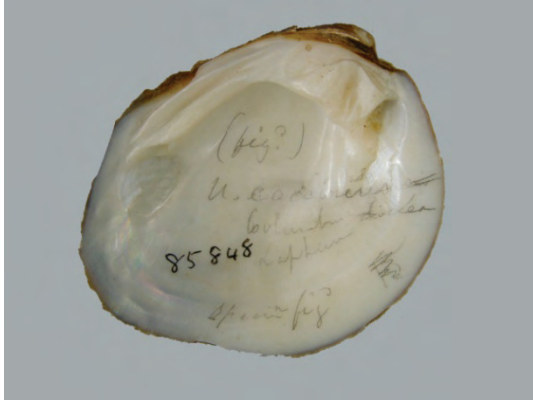


5.6

Plate 5. *Pleurobema* clade specimens (external view).

- 5.1) *P. sintoxia*. OH, Cincinnati. Ohio River. USNM 85848 type. L 56.2, H 51.8  
5.2) *P. sintoxia*. PA, unknown County. Little Mahoning River. ASU 3590. L 65.2, H 52.4  
5.3) *P. sintoxia*. PA, unknown County. Little Mahoning River. ASU 3594. L 76.1, H 58.0  
5.4) *Pleurobema* sp. AR, Cross County. St. Francis River. ASU 2722. L 61.8, H 50.1  
5.5) *Pleurobema* sp. AR, Ouachita County. Ouachita River. ASU 2789. L 64.6, H 44.7  
5.6) *Pleurobema* sp. AR, Ouachita County. Ouachita River. ASU 2803. L 68.7, H 53.7

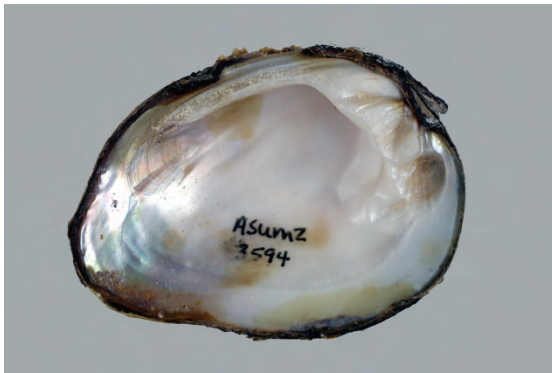
*Fusconaia* and *Pleurobema* Preliminary Analysis



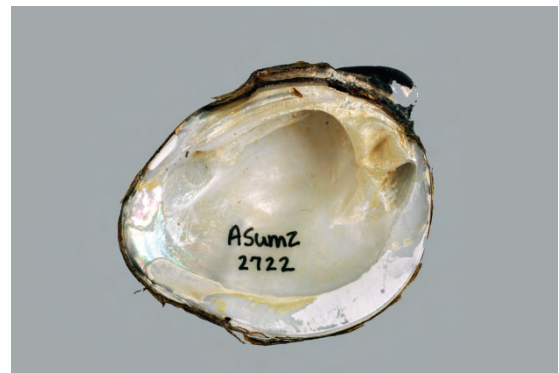
6.1



6.2



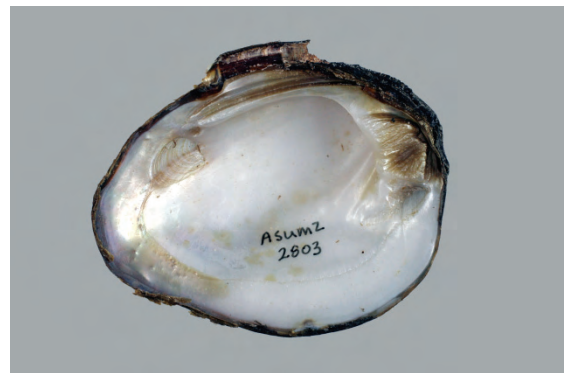
6.3



6.4



6.5



6.6

Plate 6. *Pleurobema* clade specimens (internal view).

- 6.1) *P. sintoxia*. OH, Cincinnati. Ohio River. USNM 85848 type. L 56.2, H 51.8
- 6.2) *P. sintoxia*. PA, unknown County. Little Mahoning River. ASU 3590. L 65.2, H 52.4
- 6.3) *P. sintoxia*. PA, unknown County. Little Mahoning River. ASU 3594. L 76.1, H 58.0
- 6.4) *Pleurobema* sp. AR, Cross County. St. Francis River. ASU 2722. L 61.8, H 50.1
- 6.5) *Pleurobema* sp. AR, Ouachita County. Ouachita River. ASU 2789. L 64.6, H 44.7
- 6.6) *Pleurobema* sp. AR, Ouachita County. Ouachita River. ASU 2803. L 68.7, H 53.7

*Fusconaia* and *Pleurobema* Preliminary Analysis



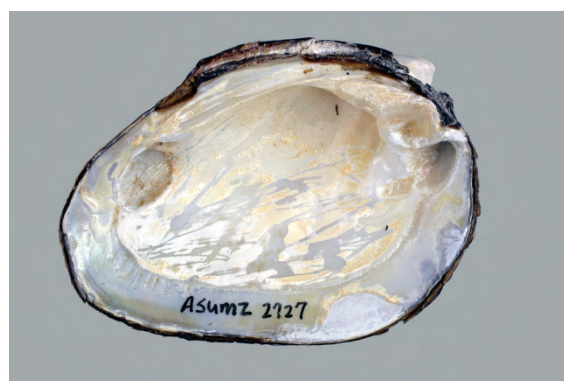
7.1



7.2



7.3



7.4



7.5



7.6

Plate 7. *Pleurobema* St. Francis River clade (external and internal views)

7.1 and 7.2) *Pleurobema* sp. AR, Cross County. St. Francis River. ASU 2721 L 55.1, H 46.3

7.3 and 7.4) *Pleurobema* sp. AR, Cross County. St. Francis River. ASU 2727 L 92.2, H 65.5

7.5 and 7.6) *Pleurobema* sp. AR, Cross County. St. Francis River. ASU 2724 L 73.3, H 59.7

*Fusconaia* and *Pleurobema* Preliminary Analysis



8.1



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Plate 8. *Pleurobema beadleianum*, *P. riddellii*, *Pleurobema* Ouachita River clade, and *Pleurobema* sp. White River drainage (external view).

8.1) *P. beadleianum*. MS, unknown County. Pearl River. L 44.0, H 38.4

8.2) *P. riddellii*. TX, Dallas County. Trinity River. L 35.7, H 32.4

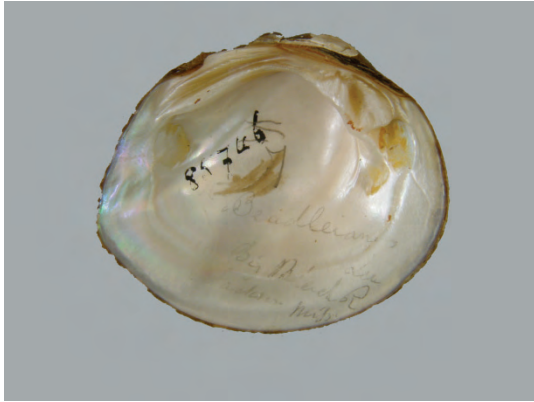
8.3) *Pleurobema* sp. AR, Ouachita County. Ouachita River. ASU 2765. L 42.9, H 38.8

8.4) *Pleurobema* sp. AR, Ouachita County. Ouachita River. ASU 2778. L 35.0, H 33.0

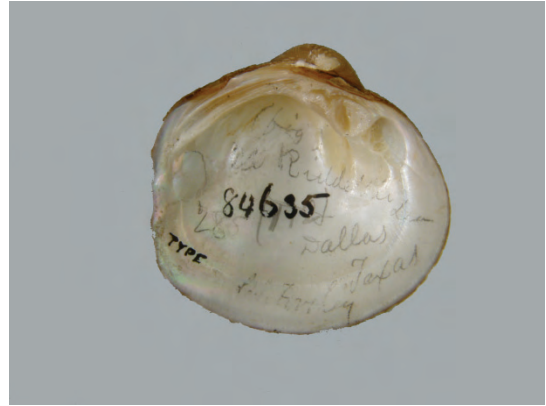
8.5) *Pleurobema* sp. AR, Cleburne County. Middle Fork Little Red River. ASU 2733. L 128.6, H 86.7



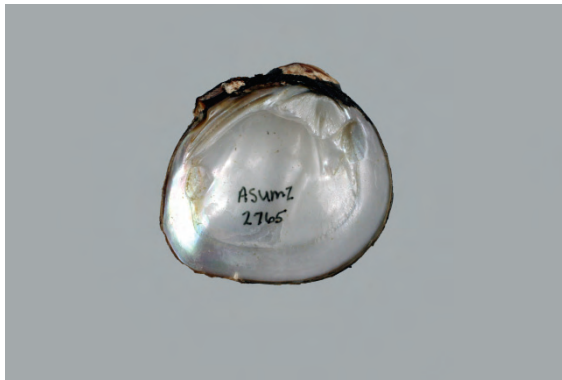
*Fusconaia* and *Pleurobema* Preliminary Analysis



9.1



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9.3



9.4



9.5

Plate 9. *Pleurobema beadleianum*, *P. riddellii*, *Pleurobema* Ouachita River clade, and *Pleurobema* sp. White River drainage (internal view).

9.1) *P. beadleianum*. MS, unknown County. Pearl River. L 44.0, H 38.4

9.2) *P. riddellii*. TX, Dallas County. Trinity River. L 35.7, H 32.4

9.3) AR, Ouachita County. Ouachita River. ASU 2765. L 42.9, H 38.8

9.4) AR, Ouachita County. Ouachita River. ASU 2771 L 35.0, H 33.0

9.5) AR, Cleburne County. Middle Fork Little Red River. ASU 2733. L 128.6, H 86.7