



Newsletter of the Freshwater Mollusk Conservation Society
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**Planning Well Underway for Our 2017
 Symposium in Cleveland, Ohio**

The 10th Biennial FMCS Symposium will be held on March 26 -30, 2017, at the Cleveland Downtown Marriott at Key Center, in Cleveland, Ohio. The Planning Committee is pleased to announce that our conference theme will be: *Ecosystems, Engineering, Valuation, and Practice - The Roles of Freshwater Mollusks in a Changing Environment*. The goal for this Symposium is to focus on how freshwater mollusks affect nature and society by: (1) providing ecosystem services, (2) being ecosystem engineers, (3) understanding their value relative to other biota and us, and (4) guiding the regulations and actions we use in

Contributed Articles

The following articles have been contributed by FMCS members and others interested in freshwater mollusks. These contributions are incorporated into Ellipsaria without peer review and with minimal editing. The opinions expressed are those of the authors.

Two Reproductively-Isolated Populations Cryptic Under *Pleurocera simplex* (Say, 1825) Inhabiting Pistol Creek in Maryville, Tennessee

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Although focused primarily upon genetic relationships among 11 populations of the *Pleurocera carinifera* group, the allozyme survey of Dillon (2011) also included four populations of *Pleurocera simplex* as controls. The results obtained for three of those four *simplex* populations were unremarkable. But certain observations made during the spring of 2008, at the initial sampling of *P. simplex* population S6 from Pistol Creek in Maryville Courthouse Park, Blount County, Tennessee (35.7535°N; 83.9711°W), prompted me to open a second line of inquiry described here.

In particular, my initial sample of nominal *P. simplex* from site S6 demonstrated striking deviations from Hardy-Weinberg expectations at three enzyme-encoding loci and strong evidence of linkage disequilibrium. Of the 30 individuals initially sampled from Pistol Creek in May of 2008, 17 appeared homozygous for allele 100 at the octanol dehydrogenase locus (*Oldh*) and allele 96 at the octopine dehydrogenase locus (*Opdh*), while 13 individuals appeared homozygous for allele 104 at *Oldh* and allele 98 at *Opdh*, with no putative heterozygotes in evidence at either locus. A striking difference was also noted between the two sets at the phosphoglucosmutase (*Pgm*) locus, the set of 17 showing *Pgm*⁹⁶ at a frequency of 0.882, the set of 13 showing *Pgm*¹⁰² at a frequency of 0.923, just six *Pgm* heterozygotes identified in total. No variation was uncovered at any of seven additional loci examined in the Pistol Creek sample.

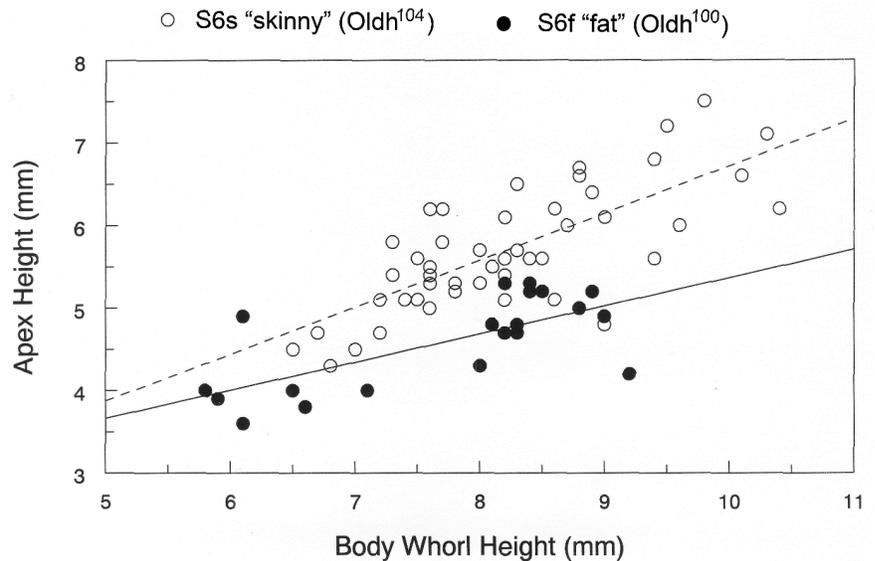
These observations suggested strongly that Pistol Creek might be inhabited by two reproductively isolated species indistinguishable [to my eyes] from *P. simplex*. So, In August of 2008, I returned to site S6 for a second sample. On this occasion, I examined the shell morphology of the animals collected more critically. It was my subjective impression that significant variation might exist in simple shell proportions within the sample, particularly with respect to the relative height of the body whorl. So, for the 71 snails sampled in August, I measured the maximum shell dimension (or "shell height"), and body whorl height (B), defined as the length of the final 360° of whorl, along the axis of coiling. I then defined apex height (A) as shell height minus body whorl height, and analyzed the relationship between body whorl height and apex height by analysis of covariance using the separate slopes model (JMP version 7).

Next, I classified my fresh sample of 71 individuals by their allozyme phenotype at 10 allozyme-encoding loci using the methods outlined in Dillon (1992, 2011). A total of 20 snails were homozygous for *Oldh*¹⁰⁰, while 51 were homozygous for *Oldh*¹⁰⁴, again with no putative heterozygotes. Differences were also very marked at the *Opdh* and *Pgm* loci, although a few heterozygotes were observed in both groups. The combined sample of 20 August snails plus 17 snails sampled in May showed *Opdh*⁹⁶ = 0.946 and *Pgm*⁹⁶ = 0.946, and the combined sample of 51 + 13 showed *Opdh*⁹⁸ = 0.953 and *Pgm*¹⁰² = 0.852. Again no variation was detected at the seven additional genetic loci examined, all individuals being fixed for the same putative alleles *Gpi*¹⁰⁰, *Mpi*¹⁰⁰, *Est*¹⁰⁰, *6pgd*¹⁰⁰, *Xdh*¹⁰⁰, *IsdhS*¹⁰⁰ and *IsdhF*¹⁰².

Figure 1 compares the regressions of (A) on (B) for the two subsamples, the N = 20 fixed for *Oldh*¹⁰⁰ and the N = 51 fixed for *Oldh*¹⁰⁴. The regressions of $A = 0.34B + 1.96$ ($r = 0.69$) for the former group and $A = 0.57B + 1.02$ ($r = 0.74$) for the latter group differed significantly in their slopes, although not in their intercept. Separate-slopes ANCOVA returned a value of $t = -7.14$ ($p < 0.0001$) testing for a difference between the two groups, the group of 20 showing a very significantly lower apex (holding body whorl constant) than the group of 51. The shells of this former group appeared stout or "fat," while the shells of the latter group appeared more slender or "skinny." I provisionally designated the (N = 20) snails

bearing fat shells and fixed for Oldh¹⁰⁰ as population S6f, and the (N = 51) snails bearing skinny shells and fixed for Oldh¹⁰⁴ as population S6s.

Figure 1. Shell apex height (A) as a function of body whorl height (B) in a sample of 71 putative "*Pleurocera simplex*" from Pistol Creek in Maryville, Tennessee.



Which of these two reproductively isolated populations might match bona fide *Pleurocera simplex*, and what might be the specific identity of the cryptic population underneath it is considered in the accompanying article by Dillon and Robinson (2016).

References:

- Dillon, R. T., Jr. 1992. Electrophoresis IV, Nuts and Bolts. *World Aquaculture* 23(2):48-51.
- Dillon, R. T., Jr. 2011. Robust shell phenotype is a local response to stream size in the genus *Pleurocera* (Rafinesque 1818). *Malacologia* 53:265-277.
- Dillon, R. T., Jr, and J. D. Robinson. 2016. The identity of the "fat *simplex*" population inhabiting Pistol Creek in Maryville, Tennessee. *Ellipsaria* 18(2):16-18.

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