Putative ancient asexual Darwinulidae (Crustacea, Ostracoda) go genomics

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The ostracod family Darwinulidae is one of the three remaining animal groups with a putative ancient asexual status besides bdelloid rotifers and oribatid mites (SCHÖN et al. 2009). The recent debate on its asexual status (SMITH et al. 2006; HAYDEN 2008; MARTENS & SCHÖN 2008) has remained inconclusive, because of the lack of suitable molecular markers. Previous genetic analyses revealed low levels of genetic variability in nuclear and mitochondrial regions of *Darwinula stevensoni* (see SCHÖN et al. 2009 for a recent summary) and other darwinulids (SCHÖN et al. 2010).

Retrotransposons can provide a genomic test for ancient asexuality. Evolutionary theory predicts that they should either be degenerated or absent in ancient asexuals, because sex seems to be required to spread these elements (HICKEY 1982). Fulfilling these predictions, ARKHIPOVA & MESELSON (2000) indeed found no retrotransposons in ancient asexual bdelloids. Further research on the bdelloids meanwhile revealed one functional retrotransposon, which probably became domesticated for other functions (ARKHIPOVA et al. 2003). With a PCR approach, we found one group of functional retrotransposons in *D. stevensoni* (SCHÖN & ARKHIPOVA 2006). This is not necessarily evidence against ancient asexuality, because these retrotransposons could also have been domesticated for other functions like in the bdelloids. But to test such predictions, genomic data are required.

An alternative genomic test for ancient asexuality is the functionality of genes being required for meiosis and sex. However, no sequence information on meiotic genes from ostracods is yet available.

To apply these novel approaches to darwinulids, we have constructed a genomic library of *D. stevensoni* from a monoclonal Belgian population. Fossil data indicate that this species has reproduced exclusively asexually for at least 25 million years (Straub 1952). From 1,000 living specimens of *D. stevensoni*, a cosmid library with 18,418 clones has been constructed. Since the haploid genome size of *D. stevensoni* is estimated at 420–450 Megabasepairs (Paczesniak, in prep.), the library covers the whole genome 1.5 to 2 times and there is a chance of 90–95% to find a particular gene. We will use this library to check for the functionality of retrotransposons and meiotic genes in *D. stevensoni* as genomic tests for ancient asexuality and to develop more variable nuclear markers and microsatellites.

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