

### Tissue-specific maternal and paternal mitochondrial DNA in the freshwater mussel, *Anodonta grandis grandis*

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Until recently, inheritance of mitochondrial DNA (mtDNA) in animals was thought to be strictly maternal. Evidence for incidental paternal mtDNA leakage was obtained in hybrid crosses of mice<sup>1</sup> and *Drosophila*.<sup>2,3</sup> An unusual pattern of mtDNA inheritance, i.e. double uniparental inheritance, was described in the blue mussel, *Mytilus edulis*.<sup>4,5</sup> and in the giant floater, *Anodonta grandis grandis*.<sup>6</sup> Here we report the distribution of maternal and paternal mitochondrial DNA in *A. g. grandis*, which differs from that in *M. edulis*.

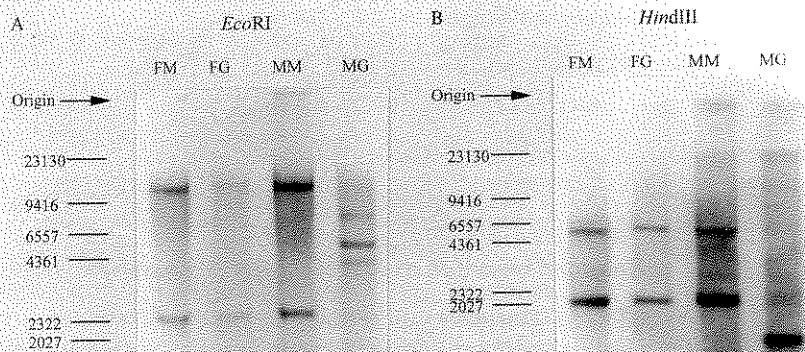
In the mode of double uniparental inheritance of *Mytilus edulis*, the transmission of mitochondrial types depends upon the sex. Female offspring receive predominantly maternal mtDNA and transmit the maternal type mtDNA into eggs, while males receive both maternal and paternal mtDNA and preferentially package the paternal type mtDNA into sperm. Heteroplasmy of the maternal and paternal mtDNA is commonly found in the somatic tissues of male *M. edulis*.<sup>5,7,8,9,10</sup> Liu and associates did

not examine somatic tissue<sup>6</sup> and it is not known whether heteroplasmy is also a common phenomenon in male *Anodonta grandis grandis*.

To determine the distribution of mitochondrial types, eight *A. g. grandis* (six males, two females) were collected from Flagler Reservoir (T9S, R50W, sections 4/5/8/9) at Kit Carson County, Colorado. Total DNA was extracted from gonadal tissue and from mantle tissue for each individual<sup>6</sup>. Five restriction enzymes, *AseI*, *EcoRI*, *HaeIII*, *HindIII*, and *HinII* were used to digest the total DNA. Mitochondrial DNA fragments were detected and scored as described in Liu *et al.*<sup>6</sup>

For all five mtDNA restriction fragment patterns, female mussels exhibited only the maternal type mtDNA in both gonadal and somatic tissues (Fig. 1). Male mussels showed predominantly the paternal type mtDNA in the gonadal tissue, but only the maternal type mtDNA in the somatic tissues (Fig. 1).

Males gave a weak signal of the maternal type mtDNA in the gonadal tissue. This phenomenon was



**Figure 1.** Sex- and tissue-specific mitochondrial DNA distribution in the giant floater, *Anodonta grandis grandis*. **A.** Southern blot of DNA extracted from somatic tissue (mantle) and gonadal tissue of each sex. DNA was restricted with *EcoRI*. **B.** Same as A except DNA was restricted with *HindIII*. Abbreviations: FM, female mantle tissue; FG, female gonadal tissue; MM, male mantle tissue; MG, male gonadal tissue.

also observed in *M. edulis*.<sup>4</sup> Skibinski *et al.* suggested that maternal type mtDNA might occasionally leak into the paternal mtDNA inheritance system. However, the weak maternal type mtDNA signal in the gonadal tissue of males might be from the surrounding somatic tissue of the gonad, and does not necessarily indicate that maternal type mtDNA are packaged into sperm.

In *M. edulis*, zygotes contain approximately 10<sup>4</sup> mitochondria contributed by the egg<sup>11</sup> and approximately five copies of mitochondria contributed by the sperm<sup>12</sup>. As males develop and grow, the paternal type mtDNA becomes codominant with the maternal type mtDNA in the somatic tissue. If the paternal type mtDNA is in females, it cannot be detected by the usual detection methods.<sup>4</sup> In contrast, the maternal type is the only type of mtDNA in the somatic tissues of both males and females in *A. g. grandis*. The contrasting results suggest that the regulation of replication of mitochondria differs between these two bivalve species.

The analysis of different tissues within individual animals is not usual in studies of mtDNA variation. Foot, mantle and adductor muscle are the tissues more commonly used for DNA analysis in bivalves.<sup>7,13</sup> If maternal type mtDNA is the predominant type of mtDNA in the somatic tissues, as in *A. g. grandis*, the detection of the double uniparental inheritance could easily be missed by using only somatic tissue in the DNA analysis. Thus double uniparental inheritance might occur widely in bivalves, but may have been missed.

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### About preferential ingestion of organic matter by bivalves

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It is well known that filter-feeding bivalves produce two sorts of biodeposits: the faeces (the food rejected after passage through the digestive tract) and, simultaneously when the sestonic concentration is high, the pseudofaeces (a part of the collected particles rejected before injection). Because analyses revealed that pseudofaeces usually have a lower organic fraction than the food, numerous authors have claimed that bivalves are able to preferentially ingest organic

matter.<sup>2,6,7,8,9</sup> For example, we have obtained similar results with the venerid *Ruditapes philippinarum* (Adams & Reeve) fed with a mixture of particles of SiO<sub>2</sub> (size range: 5-47 μm) and of *Dunaliella primolecta* (mean diameter: 5 ± 0.3 μm). While the organic fraction of this food ranged from 50-60%, the pseudofaeces produced had an organic fraction between 20% and 30%. While, doubtless, such a result is experimental evidence of the existence of