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Paternal Mitochondrial Transmission in Intra-Species *Caenorhabditis briggsae* Hybrids

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Abstract

To study mitochondrial–nuclear genetic interactions in the nematode *Caenorhabditis briggsae*, our three laboratories independently created 38 intra-species cytoplasmic–nuclear hybrid (cybrid) lines. Although the cross design combines maternal mitotypes with paternal nuclear genotypes, eight lines (21%) unexpectedly contained paternal mitotypes. All eight share in common ancestry of one of two genetically related strains. This unexpected parallel observation of paternal mitochondrial transmission, undesirable given our intent of creating cybrids, provides a serendipitous experimental model and framework to study the molecular and evolutionary basis of uniparental mitochondrial inheritance.

Key words: mitochondria, heteroplasmy, coevolution, transmission, fertilization.

The broad conservation of maternal mitochondrial transmission (MMT) (Birky 1995) suggests its evolutionary importance. However, forces promoting MMT are poorly understood (Carelli 2015). Presence of multiple mitochondrial genotypes (mitotypes) in a cell (heteroplasmy) causes dysfunction in mice (Sharpley et al. 2012) and humans (Schwartz and Vissing 2002). Thus, MMT might have evolved to prevent heteroplasmy (Mishra and Chan 2014). Studies in primates and in the nematode *Caenorhabditis elegans*, a relative of *C. briggsae*, suggest that paternally transmitted mitochondria are actively degraded following fertilization (Sutovsky et al. 1999; Al Rawi et al. 2011; Sato and Sato 2011, 2013), although the molecular signal distinguishing paternal mitochondria, and the oocyte receiver of this signal, remain unknown.

Fitness is impacted by mitochondrial genetic interactions not only between mitotypes (in heteroplasmy) but also between the mitochondrial and nuclear genomes (Rand et al. 2004; Gershoni et al. 2009). To identify dysfunctional mito-nuclear allele combinations for studying mitochondrial function, we conducted experimental crosses with *C. briggsae*, which exhibits substantial mitochondrial genetic variation among wild strains (Howe and Denver 2008). Our three laboratories independently generated replicate cytoplasmic–nuclear hybrids (cybrids) from nine wild isolate strains representing three phylogeographic clades (table 1) using the same cross design that demonstrated MMT in mice (Gyllenstein et al. 1985): we crossed two populations in the P0 generation, using males and self-sperm depleted hermaphrodites to ensure only cross progeny were produced. We then serially backcrossed sperm-depleted hybrid hermaphrodites

to P0 males for 10 generations (supplementary fig. S1, Supplementary Material online). This design produces cybrids: lines with the P0 maternal mitotype and P0 paternal nuclear genotype.

We next extracted genomic DNA from pools of each line and genotyped nuclear loci by polymerase chain reaction (PCR) of strain-specific or clade-specific amplified fragment length polymorphisms (AFLPs) (Koboldt et al. 2010; Hicks et al. 2012) and mitochondrial loci by PCR of a mitochondrial restriction fragment length polymorphism (RFLP) or by sequencing of the mitochondrial cytochrome oxidase II (COII) locus to identify strain-specific single-nucleotide polymorphisms (table 1). While we always observed the expected nuclear genotype in each line, we observed only P0 male mitotypes in 8 of 20 inter-population crosses that employed either HK104 or HK105, both isolated from Japan. Paternal mitotypes were not evident in the 18 lines initiated from other strains, and no lines appeared by PCR to be heteroplasmous. The eight lines containing paternal mitotypes are not cybrids; instead, they have mito-nuclear haplotypes identical to a P0 strain. The presence of a PCR-detectable paternal mitotype is evidence for paternal mitochondrial transmission (PMT): as no hermaphrodites from the P0 male population were used in producing cybrids, PMT is the only means by which P0 male mitotypes could exist in cybrids.

PMT was symmetrical, observed only when an “HK” strain served as the maternal or paternal P0 strain. While asymmetry with respect to cross direction is expected when hybrids suffer from Dobzhansky–Muller incompatibilities, symmetry is consistent with the interpretation that PMT is caused by

Table 1. Nuclear and Mitochondrial Genotypes of Cybrid Lines.

Line (Rep) ^a	P0 strains		Line nuclear genotype ^c		Line mitochondrial genotype ^e	
	Maternal	Paternal	Exp ^b	Obs	Exp ^d	Obs ^f
RC-PH (1)	HK105	PB800	Te	–	HK105	HK105
RC-PH (2)	HK105	PB800	Te	–	HK105	F
RC-PH (3)	HK105	PB800	Te	–	HK105	PB800
RC-HP (1)	PB800	HK105	Te	–	PB800	PB800
RC-HP (2)	PB800	HK105	Te	–	PB800	PB800
RC-HP (3)	PB800	HK105	Te	–	PB800	PB800
RC-EH (1)	HK104	EG4818	Te	–	HK104	EG4181
RC-EH (2)	HK104	EG4818	Te	–	HK104	HK104
RC-EH (3)	HK104	EG4818	Te	–	HK104	EG4181
RC-HE (1)	EG4818	HK104	Te	–	EG4818	HK104
RC-HE (2)	EG4818	HK104	Te	–	EG4818	HK104
RC-HE (3)	EG4818	HK104	Te	–	EG4818	HK104
MR-AH (1)	HK105	AF16	Tr	Tr	HK105	HK105
MR-AH (2)	HK105	AF16	Tr	Tr	HK105	AF16
MR-AH (3)	HK105	AF16	Tr	Tr	HK105	HK105
MR-AD (1)	DL232	AF16	Tr	Tr	DL232	DL232
MR-AD (2)	DL232	AF16	Tr	Tr	DL232	DL232
MR-AD (3)	DL232	AF16	Tr	Tr	DL232	DL232
MR-AE (1)	ED3101	AF16	Tr	Tr	ED3101	ED3101
MR-AE (2)	ED3101	AF16	Tr	Tr	ED3101	ED3101
MR-AE (3)	ED3101	AF16	Tr	Tr	ED3101	ED3101
MR-AJ4 (1)	JU403	AF16	Tr	Tr	JU403	JU403
MR-AJ4 (2)	JU403	AF16	Tr	Tr	JU403	JU403
MR-AJ4 (3)	JU403	AF16	Tr	Tr	JU403	JU403
MR-AJ1 (1)	JU1345	AF16	Tr	Tr	JU1345	JU1345
MR-AJ1 (2)	JU1345	AF16	Tr	Tr	JU1345	JU1345
MR-AJ1 (3)	JU1345	AF16	Tr	Tr	JU1345	JU1345
MR-AP (1)	PB800	AF16	Tr	Tr	PB800	PB800
MR-AP (2)	PB800	AF16	Tr	Tr	PB800	PB800
MR-AP (3)	PB800	AF16	Tr	Tr	PB800	PB800
MR-AV (1)	VT847	AF16	Tr	–	VT847	VT847
MR-AV (2)	VT847	AF16	Tr	–	VT847	VT847
MR-AV (3)	VT847	AF16	Tr	–	VT847	VT847
CP129 (1)	HK104	AF16	AF16	AF16	HK104	HK104
CP130 (2)	HK104	AF16	AF16	AF16	HK104	HK104
CP131 (1)	AF16	HK104	HK104	HK104	AF16	AF16
CP132 (2)	AF16	HK104	HK104	HK104	AF16	HK104
CP133 (3)	AF16	HK104	HK104	HK104	AF16	AF16

^aAll crosses were replicated (“Rep”) thrice, with the exception of AF16 male × HK104 hermaphrodite (producing CP129 and CP130), for which one of three lines initiated went extinct.

^bThe expected nuclear genotype is the P0 male wild isolate genotype.

^cIsolates belong to the tropical (“Tr”), temperate (“Te”) or equatorial Kenya phylogenetic clade (Cutter et al. 2010). Nuclear genotype was observed either by an AFLP that distinguishes temperate from tropical alleles or at five loci distinguishing AF16 and HK104 (CP129–CP133). Because the former assays only distinguish members of different clades, nuclear genotypes of within-clade (Te × Te or Tr × Tr) hybrids were unable to be obtained (–).

^dThe expected mitochondrial genotype is the P0 maternal wild isolate genotype.

^eMitochondrial genotype was observed either by sequencing the COII gene or by an RFLP distinguishing the AF16 and HK104 mitotypes (CP129–CP133).

^fThe observed and expected mitotypes occasionally did not match (gray shading), providing evidence for paternal mitochondrial transmission.

F, failed PCR reaction.

separation of co-evolved mitochondrial and nuclear loci (Turelli and Moyle 2007).

Our results suggest two possible mechanisms facilitating PMT. Co-evolved signal–receiver genes, as occur in species-specific sperm-egg protein recognition (Swanson and Vacquier 1998), could be separated in temperate–tropical cybrids. However, this explanation predicts more frequent inter-clade PMT, whereas our observations reveal more frequent intra-clade PMT. Thus, we favor the alternate explanation that natural genetic variation, perhaps passively accumulated through genetic drift, has reduced or eliminated the function of a paternal mitochondrial signal–receiver system in some members of the temperate clade.

PMT has been detected in a variety of taxa (Kondo et al. 1990; Gyllensten et al. 1991; Kaneda et al. 1995; Kvist et al. 2003; Aksyonova et al. 2005; Fontaine et al. 2007). However, such empirical observations rarely occur in tractable model systems and/or with sufficiently high frequency to encourage experimental pursuit of the mechanisms facilitating PMT. Recent discovery of mitochondrial–nuclear epistasis in AF16–HK104 hybrids (Chang et al. 2016) supports the possibility that mitonuclear interactions are important for the prevention of PMT. The role in *C. elegans* paternal mitochondrial elimination of a nuclear-encoded mitochondrial endonuclease, which translocates into mitochondria (Zhou et al. 2016), suggests that interaction of

the endonuclease with mitochondrial gene products is critical for preventing PMT. These possibilities promote the use of *C. briggsae* to elucidate the molecular and genetic mechanisms facilitating PMT.

Supplementary Material

Supplementary figure S1 is available at *Molecular Biology and Evolution* online (<http://www.mbe.oxfordjournals.org/>).

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References

- Aksyonova E, Sinyavskaya M, Danilenko N, Pershina L, Nakamura C, Davydenko O. 2005. Heteroplasmy and paternally oriented shift of the organellar DNA composition in barley-wheat hybrids during backcrosses with wheat parents. *Genome* 48:761–769.
- Al Rawi S, Louvet-Vallee S, Djeddi A, Sachse M, Culetto E, Hajjar C, Boyd L, Legouis R, Galy V. 2011. Postfertilization autophagy of sperm organelles prevents paternal mitochondrial DNA transmission. *Science* 334:1144–1147.
- Birky CW. Jr. 1995. Uniparental inheritance of mitochondrial and chloroplast genes: mechanisms and evolution. *Proc Natl Acad Sci U S A* 92:11331–11338.
- Carelli V. 2015. Keeping in shape the dogma of mitochondrial DNA maternal inheritance. *PLoS Genet*. 11:e1005179.
- Chang CC, Rodriguez J, Ross J. 2016. Mitochondrial-nuclear epistasis impacts fitness and mitochondrial physiology of interpopulation *Caenorhabditis briggsae* hybrids. *G3 (Bethesda)* 6:209–219.
- Cutter AD, Yan W, Tsvetkov N, Sunil S, Felix MA. 2010. Molecular population genetics and phenotypic sensitivity to ethanol for a globally diverse sample of the nematode *Caenorhabditis briggsae*. *Mol Ecol* 19:798–809.
- Fontaine KM, Cooley JR, Simon C. 2007. Evidence for paternal leakage in hybrid periodical cicadas (Hemiptera: Magicicada spp.). *PLoS One* 2:e892.
- Gershoni M, Templeton AR, Mishmar D. 2009. Mitochondrial bioenergetics as a major motive force of speciation. *Bioessays* 31:642–650.
- Gyllenstein U, Wharton D, Josefsson A, Wilson AC. 1991. Paternal inheritance of mitochondrial DNA in mice. *Nature* 352:255–257.
- Gyllenstein U, Wharton D, Wilson AC. 1985. Maternal inheritance of mitochondrial DNA during backcrossing of two species of mice. *J Hered*. 76:321–324.
- Hicks KA, Howe DK, Leung A, Denver DR, Estes S. 2012. *In vivo* quantification reveals extensive natural variation in mitochondrial form and function in *Caenorhabditis briggsae*. *PLoS One* 7:e43837.
- Howe DK, Denver DR. 2008. Muller's Ratchet and compensatory mutation in *Caenorhabditis briggsae* mitochondrial genome evolution. *BMC Evol Biol*. 8:62.
- Kaneda H, Hayashi J, Takahama S, Taya C, Lindahl KF, Yonekawa H. 1995. Elimination of paternal mitochondrial DNA in intraspecific crosses during early mouse embryogenesis. *Proc Natl Acad Sci U S A* 92:4542–4546.
- Koboldt DC, Staisch J, Thillainathan B, Haines K, Baird SE, Chamberlin HM, Haag ES, Miller RD, Gupta BP. 2010. A toolkit for rapid gene mapping in the nematode *Caenorhabditis briggsae*. *BMC Genomics* 11:236.
- Kondo R, Satta Y, Matsuura ET, Ishiwa H, Takahata N, Chigusa SI. 1990. Incomplete maternal transmission of mitochondrial DNA in *Drosophila*. *Genetics* 126:657–663.
- Kvist L, Martens J, Nazarenko AA, Orell M. 2003. Paternal leakage of mitochondrial DNA in the great tit (*Parus major*). *Mol Biol Evol* 20:243–247.
- Mishra P, Chan D. 2014. Mitochondrial dynamics and inheritance during cell division, development and disease. *Nat Rev Mol Cell Biol* 15:634–646.
- Rand DM, Haney RA, Fry AJ. 2004. Cytonuclear coevolution: the genomics of cooperation. *Trends Ecol Evol* 19:645–653.
- Sato M, Sato K. 2011. Degradation of paternal mitochondria by fertilization-triggered autophagy in *C. elegans* embryos. *Science* 334:1141–1144.
- Sato M, Sato K. 2013. Maternal inheritance of mitochondrial DNA by diverse mechanisms to eliminate paternal mitochondrial DNA. *Biochim Biophys Acta* 1833:1979–1984.
- Schwartz M, Vissing J. 2002. Paternal inheritance of mitochondrial DNA. *N Engl J Med*. 347:576–580.
- Sharpley MS, Marciniak C, Eckel-Mahan K, McManus M, Crimi M, Waymire K, Lin CS, Masubuchi S, Friend N, Koike M, et al. 2012. Heteroplasmy of mouse mtDNA is genetically unstable and results in altered behavior and cognition. *Cell* 151:333–343.
- Sutovsky P, Moreno RD, Ramalho-Santos J, Dominko T, Simerly C, Schatten G. 1999. Ubiquitin tag for sperm mitochondria. *Nature* 402:371–372.
- Swanson WJ, Vacquier VD. 1998. Concerted evolution in an egg receptor for a rapidly evolving abalone sperm protein. *Science* 281:710–712.
- Turelli M, Moyle LC. 2007. Asymmetric postmating isolation: Darwin's corollary to Haldane's rule. *Genetics* 176:1059–1088.
- Zhou Q, Li H, Li H, Nakagawa A, Lin JL, Lee ES, Harry BL, Skeen-Gaar RR, Suehiro Y, William D, et al. 2016. Mitochondrial endonuclease G mediates breakdown of paternal mitochondria upon fertilization. *Science* 353:394–399.