BRIEF COMMUNICATION | Genetics in Medicine

Biparental inheritance of mitochondrial DNA in humans is not a common phenomenon

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Purpose: A recent report has raised the possibility of biparental mitochondrial DNA (mtDNA) inheritance, which could lead to concerns by health-care professionals and patients regarding investigations and genetic counseling of families with pathogenic mitochondrial DNA variants. Our aim was to examine the frequency of this phenomenon by investigating a cohort of patients with suspected mitochondrial disease.

Methods: We studied genome sequencing (GS) data of DNA extracted from blood samples of 41 pediatric patients with suspected mitochondrial disease and their parents.

Results: All of the mtDNA variants in the probands segregated with their mother or were apparently de novo. There were no variants that segregated only with the father and none of these families showed evidence of biparental inheritance of their mtDNA.

Conclusion: Paternal mitochondrial transmission is unlikely to be a common occurrence and therefore at this point we would not recommend changes in clinical practice.

Genetics in Medicine (2019) 21:2823–2826; https://doi.org/10.1038/s41436-019-0568-0

Keywords: mitochondrial genome; genetic testing; genetic counseling; mitochondrial diseases; paternal inheritance

INTRODUCTION

The underlying molecular diagnosis of patients with mitochondrial diseases can be due to nuclear or mitochondrial DNA (mtDNA) pathogenic variants. In humans, mtDNA is generally inherited from the mother. Thus, the cascade diagnostic investigations, genetic counseling, and reproductive choices for patients with pathogenic mtDNA variants are currently based on that premise.[1]

In a recent study, Luo et al.[2] reported evidence of paternal transmission of mtDNA to offspring in three families. Some of the issues emerging from these findings relate specifically to mitochondrial disease diagnosis and genetic counseling and could prompt concerns about current practice.

The possibility of paternal inheritance of mtDNA has been reported previously in mice, including across multiple generations.[3,4] Tissue-specific inheritance of paternal mtDNA in humans has also been reported in a single individual with mitochondrial myopathy whose skeletal muscle had a de novo homoplasmic 2-bp deletion in the *MT-ND2* gene on the paternal mtDNA haplotype, while only maternal mtDNA was detected in other tissues tested.[5] The recent finding by Luo et al.[2] prompts reconsideration of whether paternal inheritance may be more common in humans than previously considered. To our knowledge, there have not been any other systematic investigations in human trios.

MATERIALS AND METHODS

All procedures followed were in accordance with the ethical standards of the responsible committee on human experimentation (institutional and national) and with the Helsinki Declaration of 1975, as revised in 2000. Informed consent was obtained from all patients included in the study. This project was approved by the Human Research Ethics Committee of the Sydney Children's Hospitals Network (ID number 10/CHW/114).

We studied genome sequencing (GS) data of DNA extracted from blood samples of 41 pediatric patients recruited in Australia with suspected mitochondrial disease and their parents, as previously reported.[6] Thirty-seven of the patients had trio GS, while in four patients DNA from the father was not available.

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Submitted 2 March 2019; accepted: 24 May 2019
Published online: 7 June 2019

Table 1 Mitochondrial DNA (mtDNA) variant segregation

				Proband			Mother			Father		
Patient	All inherited variants shared with the mother	Paternal specific variants	Number of de novo variants	Number of variants	Heteroplasmic variants	Haplotype[14]	Number of variants	Heteroplasmic variants	Haplotype[14]	Number of variants	Heteroplasmic variants	Haplotype[14]
1	Yes	No	0	31	0	M35b	31	0	M35b	16	0	HV
2	Yes	No	0	9	0	H1aq	9	0	H1aq	9	0	H1aq
3	Yes	NA	1	38	1 m.7925G>A (70%)	T2a1b1a	37	0	T2a1b1a	NA	NA	NA
4	Yes	No	0	9	0	H1	9	0	H1	32	0	U5b2b4a
5	Yes	No	0	10	0	H1	10	0	H1	33	0	T2b30
6	Yes	No	0	43	0	M1a1i	43	0	M1a1i	13	0	H1m1
7	Yes	No	0	17	0	V10a	17	0	V10a	10	0	H1c
8	Yes	No	0	33	1 m.8489A>C (3%)	K1a2a	33	1 m.8489A>C (28%)	K1a2a	36	0	T2b5a
9	Yes	No	0	29	0	A1a	29	0	A1a	35	0	T2b
10	Yes	No	0	12	0	H1a3a	12	0	H1a3a	35	0	J1c2c1
11	Yes	No	0	0	0	H2a2a1	0	0	H2a2a1	29	0	J1c2
12	Yes	No	0	39	0	J2a1a1	39	0	J2a1a1	12	0	H45
13	Yes	No	0	16	0	H4a1a1a	16	0	H4a1a1a	2	1 m.16183A>C (20%)	H2a2a1
14	Yes	No	0	33	0	J1c2b1	33	0	J1c2b1	35	0	J1c2b1
15	Yes	No	0	11	0	H1	12	1 m.16150C>T (25%)	H1	11	0	H5
16	Yes	No	1	34	1 m.3243A>G (40%)	U5b2b4a	33	0	U5b2b4a	31	0	U3a1a
17	Yes	No	0	36	0	U2d2	36	0	U2d2	32	0	X4
18	Yes	No	0	24	0	HV2a1	24	0	HV2a1	39	0	G2a2
19	Yes	No	0	31	0	J1c2	31	0	J1c2	NA	NA	NA
20	Yes	No	0	31	0	M3c2	31	0	M3c2	31	0	U7a2
21	Yes	No	0	35	0	T2g2	35	0	T2g2	14	0	H1bb
22	Yes	No	1	36	0	A12a	35	0	A12a	15	0	H13a1a1a
23	Yes	No	0	37	0	K1a4f1	37	0	K1a4f1	NA	NA	NA
24	Yes	No	0	31	0	P4b1	31	0	P4b1	42	0	M42a
25	Yes	No	0	28	0	J1c3g	28	0	J1c3g	16	0	V10a
26	Yes	No	0	31	0	J1c1b	31	0	J1c1b	11	0	H1c
27	Yes	No	0	41	0	I1a1	41	0	I1a1	15	0	H1e1a3
28	Yes	No	0	27	0	U5a2 + 16294	27	0	U5a2 + 16294	34	0	I2c
29	Yes	No	0	8	0	H2a3b	8	0	H2a3b	25	0	U5a2a2
30	Yes	No	0	14	1 m.16399A>G (8%)	H1a	14	1 m.16399A>G (83%)	H1as	38	1 m.16188CT>C (20%)	T2
31	Yes	No	0	12	0	V + @72	12	0	V + @72	24	1 m.16188CT>C (70%)	B4c2
32	Yes	No	2	35	2 m.15617G>A (12%) m.153A>G (96%)	T2b4a1	33	0	T2b4a1	39	0	I1a1b
33	Yes	No	0	12	0	H3 + 152	12	0	H3 + 152	11	0	H3z
34	Yes	No	0	29	0	B4b1a2	29	0	B4b1a2	32	0	W4a1
35	Yes	No	0	12	0	H11a	12	0	H11a	9	0	H1k
36	Yes	No	0	35	0	K1a3a1b	35	0	K1a3a1b	6	0	H2a2a2
37	Yes	No	0	13	0	H45b	13	0	H45b	13	0	H3a1a
38	Yes	No	0	40	0	U1a1a	40	0	U1a1a	24	0	U5a1g
39	Yes	No	0	15	0	H11a2a	15	0	H11a2a	34	0	U4c1
40	Yes	No	0	37	0	F1f	37	0	F1f	NA	NA	NA
41	Yes	No	0	41	0	U2e2a1a	41	0	U2e2a1a	14	0	H1c3

Variants are reported relative to the Revised Cambridge Reference Sequence NC_012920.1.

Variants were filtered using Seave[7] and reported relative to the Revised Cambridge Reference Sequence NC_012920.1. Variants with heteroplasmy levels >5% were included in the analysis and if discordant with the mother, lower levels of heteroplasmy were further investigated using the bioinformatic pipeline *mity* (Puttick et al., unpublished). Variants in positions where common sequencing errors occur due to a homopolymer stretch (MT:302–315 and MT:3105–3109) were excluded.

RESULTS

Using GS we observed a median of medians coverage of 4586× (range 1179–22,965×) in the mitochondrial genome. We identified a median of 31 mtDNA variants per proband (Table 1). In all studied families, homoplasmic variants detected in the mother were also found in the offspring.

Only five probands had any heteroplasmic variants. In two families, heteroplasmic variants detected in the mother presented lower heteroplasmy levels in the probands (families 8 and 30); in one family (family 15) the m.16150C>T heteroplasmic variant (mutant load 25%) detected in the mother was not present in the proband. These different heteroplasmy levels can be explained by the mitochondrial bottleneck during oogenesis.

In one family (family 32), two de novo heteroplasmic variants (m.15617G>A; mutant load 12% and m.153A>G; mutant load 96%) were detected in the proband that were not detected in the mother or father. In family 3, a de novo heteroplasmic variant (m.7925G>A; mutant load 70%) was present in the proband that was not identified in the mother's blood; no DNA was available from the father, however, all 37 variants present in the mother were also identified in the proband and they both shared the same haplotype (T2a1b1a).

We did not find any homoplasmic or heteroplasmic variants in the cohort that segregated only with the father. In addition, none of these families presented with high numbers of heteroplasmic variants or other suggestive patterns of biparental inheritance of their mtDNA. However, because only blood samples were analyzed, tissue-specific heteroplasmy cannot be ruled out.

DISCUSSION

Previous studies reporting paternal mtDNA transmission have been controversial, with most cases later regarded as sample cross-contamination.[8] The recent study by Luo et al.[2] has reactivated the concern about the possibility of paternal mtDNA transmission; they described three multigeneration families with a large number of mtDNA variants with high levels of heteroplasmy, corresponding to mixed haplogroups in a pattern apparently corresponding to autosomal dominant inheritance. They attempted to exclude sample contamination by conducting the mtDNA sequencing in different laboratories, however, their results have also been challenged. Lutz-Bonengel[9] and Vissing[10] speculated that the apparent biparental mtDNA inheritance pattern in the families could be attributed to amplification of nuclear mtDNA segments (NuMTs).[9,10] Luo et al.[11] contested that claim by arguing that it is unlikely that the NuMT would amplify with the different sequencing methods used. In addition, they argued that there would be a low probability of all offspring studied inheriting the NuMT variants. Nonetheless, they plan to conduct further single-cell sequencing studies with additional families to rule out that possibility. Even if paternal mtDNA transmission is confirmed, the mechanism of this event is still not clear, as well as the potential consequences of this phenomenon, given that no actual causal relationship with mitochondrial disease was established in the reported families.

Our results further support the prevailing view that paternal inheritance of mtDNA appears to be a rare event in humans, and suggest that the scenario described by Luo et al.[2] is uncommon. We suggest that genetic counseling and genetic investigations for families with suspected mitochondrial disease do not need to change. Nonetheless, it may be worth considering sequencing paternal mtDNA where a patient has an apparent de novo mtDNA variant.

We believe that this is the largest cohort of patients with suspected mitochondrial disease undergoing trio GS reported to date. As access to GS technology and genomic data sharing become more widespread in clinical practice,[12] the analysis of nuclear and mtDNA sequencing data in larger cohorts could aid in better understanding the frequency and consequences of biparental mtDNA inheritance events, and could lead to the identification of variants in nuclear genes involved in paternal mtDNA elimination that may be contributing to this mechanism.[2,13]

ACKNOWLEDGEMENTS

This research was supported by a New South Wales (NSW) Office of Health and Medical Research Council Sydney Genomics Collaborative grant (J.C.), National Health and Medical Research Council (NHMRC) project grant 1026891 (J.C.), NHMRC research fellowship 11022896 (D.R.T.), and a NSW Health Early-Mid Career Fellowship (M.J.C.). We are grateful to the Crane and Perkins families for their generous financial support. The research conducted at the Murdoch Children's Research Institute was supported by the Victorian Government's Operational Infrastructure Support Program.

DISCLOSURE

The authors declare no conflicts of interest.

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