Older mothers are not at risk of having grandchildren with sporadic mtDNA deletions

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Purpose: Single large-scale mitochondrial DNA deletions account for a quarter of mitochondrial disease cases and occur sporadically with unknown risk factors. Mitochondrial DNA deletions accumulate with age in many tissues. Primordial germ cells, the precursors of oocytes are made by our grandmothers, therefore we wanted to determine whether age of maternal grandmother is a risk factor for sporadic mitochondrial DNA deletions. **Methods:** Twenty-nine patients with sporadic single mitochondrial DNA deletions from the Newcastle UK cohort provided dates of birth for mothers and maternal grandmothers plus father and paternal grandmother (healthy controls). **Results:** Mean age for grandmothers at birth of a mother of an affected patient was 28.5 years (SD ± 6.9) for single mitochondrial DNA deletions maternal grandmothers and 28.2 years (SD ± 6.1) for healthy control paternal grandmothers. **Conclusion:** Maternal grandmother age is not a risk factor for sporadic mitochondrial DNA deletions, an important observation in a population where many women are delaying reproduction. ***Genet Med* 2010:12(5):313–314.**

Key Words: *mitochondrial DNA, mitochondrial disease, MtDNA deletions, ageing, oocytes*

Mitochondrial disease affects 1/10,000 people in the UK[1] making it a common cause of genetic disease. Single large-scale deletions of the mitochondrial genome (mtDNA) account for ~25% of these cases.[2] The deletion varies between patients, but each patient possesses a single species of mtDNA deletion (ΔmtDNA) in affected tissues. The mechanism by which ΔmtDNA are created is unknown. When defining a ΔmtDNA, it worth noting that they can also exist within a wild-type molecule to become a partially duplicated molecule, which occur more frequently in single deletion patients with diabetes and deafness.[3] Other studies have shown partial duplications are phenotypically "silent" and can therefore be transmitted to offspring,[3] however, in single deletion patients where a duplication is not present, transmission of the deletion is rare and patients usually present as sporadic cases.[4]

In single deletion patients, the pivotal events occur before birth, but it is unknown at what point during development the initial ΔmtDNA is formed. We previously analyzed monozygotic twin brothers, where both harbored the same pathogenic ΔmtDNA in muscle although only one of which had developed a clinical phenotype.[5] The ΔmtDNA was heteroplasmic in both brothers indicating that the mutation was present in the oocyte before the formation of the embryo.[5]

The traditional view of how ΔmtDNA are formed involves a slipped-strand replication mechanism[6]; however, there are challenges to this, which are discussed in our recent article.[7,8] One of the main challenges is that if replication is the mechanism of mtDNA deletion formation then why do we not see them at high levels in replicating cells? We hypothesized that if ΔmtDNA were formed by repair this could explain their existence in oocytes where replication is rare,[7] but repair mechanisms exist to maintain the pool of mitochondria for as much as 50 years. In healthy women, ~50% of oocytes harbor low levels of ΔmtDNA (<0.1%).[9,10] In somatic cells, ΔmtDNA increases with age,[11,12] but the relationship of ΔmtDNA with age in oocytes is controversial.[9,10,13]

The familial origin of single large-scale ΔmtDNA has previously showed no association between maternal age and the risk of having an affected child.[14] However, it has not been investigated whether primordial germ cells which go on to form the oocytes for the second generation, are at greater risk of harboring ΔmtDNA (Fig. 1). Thus the aim of this study was to investigate whether increased grandmother age when the mother of an affected child is born is a risk factor for single ΔmtDNA patients.

MATERIALS AND METHODS

Fifty-one patients with sporadic single ΔmtDNA were identified from the Newcastle UK cohort of mtDNA disease patients. Twenty-nine patients gave consent and took part in the study by providing the date of births for mothers and maternal grandmothers (n = 29) as well as father and paternal grandmother (n = 21) (control). Information from the Office for National Statistics was used to ascertain national population data on ages of mothers at birth since 1938.[15] Mitochondrial disease controls (n = 17) were drawn randomly from our database of maternally inherited mtDNA point mutations. Statistical analysis was performed using a one-way analysis of variance.

RESULTS

The mean age for grandmothers at birth of a mother of an affected patient was 28.5 years (SD ± 6.9) for single ΔmtDNA maternal grandmothers and 28.2 years (SD ± 6.1) for healthy control paternal grandmothers (Fig. 2). For mitochondrial control maternal grandmothers, the mean age was 25.5 years (SD ± 6.1) (Fig. 2), and there was no significant difference between the mean ages of grandmothers in any of the groups ($P > 0.05$). Moreover, comparing our dataset to data from the Office for National Statistics showed no difference in the mean age of mothers at birth,[15] which showed the mean age of live births since 1938 to 2004 to be 27.8 years, with the mean ages varying from 26.1 to 29.4 years.

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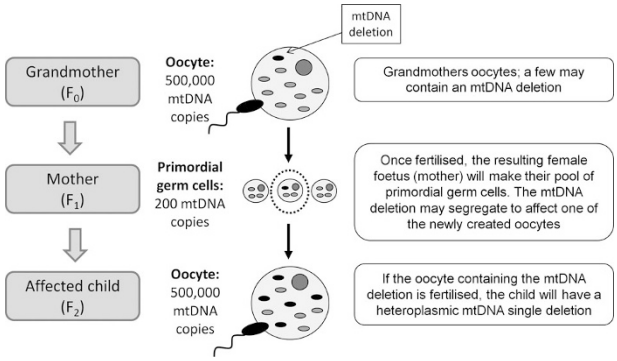

Fig. 1. Schematic diagram to show how grandmothers oocytes could lead to a single ΔmtDNA patient. Human oocytes contain ~500,000 mtDNA molecules. ΔmtDNA are reported to be present in ~50% of human oocytes. If an oocyte from the grandmother (F_0) containing a ΔmtDNA is fertilized and escapes the mitochondrial bottleneck, it could be segregated to form primordial germ cells for the developing female fetus (F_1). After rapid mtDNA amplification following the mitochondrial bottleneck this primordial germ cell will become a mature oocyte and have high levels of the ΔmtDNA. If this oocyte is fertilized, an affected individual (F_2) will be born.

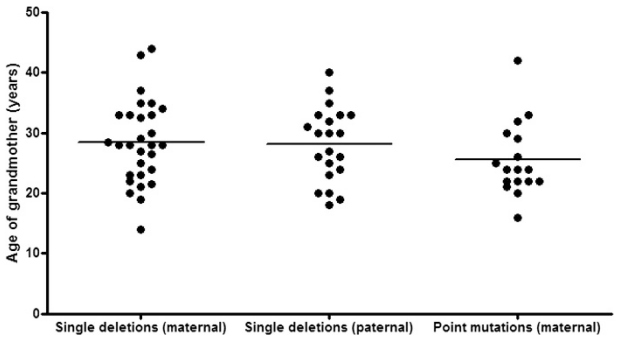

Fig. 2. Ages (years) of the grandmothers at birth of mother who has a child with sporadic ΔmtDNA. Data includes single deletion maternal grandmothers, healthy control paternal grandmothers, and mitochondrial point mutation control maternal grandmothers.

DISCUSSION

Our study shows that grandmother age is not a risk factor for sporadic, single ΔmtDNA patients. It is still uncertain whether there is an increase in ΔmtDNA in unfertilized oocytes with maternal age. Two studies reported no increase[9,10]; in contrast, a more recent study reports a significantly higher incidence of the common deletion in women ≥35 years.[13] Our study is well powered to show a difference in mean age between the two datasets of 5 years, which is similar to the mean age difference observed where ΔmtDNA occurred at a higher frequency in oocytes from older donors.[13] The results from our study are more supportive of no increase in ΔmtDNA with age in oocytes. However, further studies are needed to clarify the levels of ΔmtDNA in human oocytes, but in both species, there is good evidence to suggest that the mitochondrial bottleneck is efficient in preventing transmission of certain pathogenic mutations, in particular those seen in protein encoding genes[16] and in addition single deletion cases are almost always sporadic.[4] There has been some evidence to suggest that additional selection may act on oocytes throughout reproductive life as a study on transgenic mice harboring a large-scale ΔmtDNA showed that although the amount of deletion increased in most tissues with age, this was not the case in oocytes.[17] In conclusion, we have shown that age of maternal grandmother is not a risk factor for having a grandchild with a single mtDNA deletion disorder. The results from this study are important considering the increase in the number of women delaying reproduction.

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