Mitochondrial DNA heteroplasmy in human health and disease (Review)

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Abstract. The biomedical literature has extensively documented the functional roles of genetic polymorphisms in concert with well-characterized somatic mutations in the etiology and progression of major metastatic diseases afflicting human populations. Mitochondrial heteroplasmy exists as a dynamically determined co-expression of inherited polymorphisms and somatic mutations in varying ratios within individual mitochondrial DNA genomes with repetitive patterns of tissue specificity. Mechanistically, carcinogenic cellular processes include profound alterations of normative mitochondrial function, notably dependence on aerobic and anaerobic glycolysis, and aberrant production and release of lactate, according to a classic theory. Within the translational context of human health and disease, the present review discusses the necessity of establishing critical foci designed to probe multiple biological roles of mitochondrial heteroplasmy in cancer biology.

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1. Introduction

Extensive biomedical literature has documented the functional roles of heritable genetic polymorphisms in concert with accumulated somatic mutations and epigenetic factors in the etiology and progression of major metastatic (1,2), autoimmune (3,4) and neurodegenerative (2,5,6) disorders. By contrast, a considerably smaller subset of heritable polymorphisms in the haploid mitochondrial DNA (mtDNA) genome is functionally linked to the phenotypic manifestation of severe genetic disorders originating from markedly compromised mitochondrial function (7-10). Notably, a relatively recent body of literature has demonstrated interactive effects of heritable mtDNA polymorphisms on the altered expression of select nuclear genes that are associated with several human disease states (11-13). Conversely, specific allele imbalances within the nuclear mitochondrial genome are associated with altered translocation of key mitochondrial proteins involved in normative bioenergetics and are negatively linked to cancer progression (14,15).

2. Biological significance of mitochondrial heteroplasmy

Mitochondrial heteroplasmy may be defined as a dynamically determined co-expression of wild-type (WT)-inherited polymorphisms and somatic mutations in varying ratios within individual mtDNA genomes distributed throughout the intraorganelle compartments of individual cells. Within the translational context of human health and disease, the necessity of establishing critical foci designed to probe multiple biological roles of mitochondrial heteroplasmy has become apparent. Human mtDNA exists as a 16.6-kilobase circular genome that contains 13 protein-encoding sequences corresponding to subunits ND1-6, including ND4 and ND4L, of respiratory complex I, catalytic subunits cytochrome c oxidase subunit I-III (CO1-3) of respiratory complex IV, subunits adenosine triphosphate 6 (ATP6) and ATP8 of F1F0 ATPase, and cytochrome B of respiratory complex III. The remaining genes encode 22 tRNAs and 12, and 16S rRNAs (16). As the number of mitochondrial genomes normalized against each diploid nuclear genome varies according to cell type and total mtDNA copy number, estimated values of 100-10,000 have been reported in the biomedical literature (8,16,17). The complex sequence heterogeneity within mixed intra-mitochondrial populations of several thousand individual heteroplasmic mtDNA genomes within individual cells dictates that empirical sorting of putative biological activities of distinct patterns of mtDNA heteroplasmy, according to molecular biological, biochemical, physiological and bioinformatic criteria will be technically difficult (18-20).

Notably, a previous study monitored the intra-mitochondrial organization of heterologous heteroplasmic mtDNA
genomes into DNA-protein complexes termed nucleoids (21). Heterologous mtDNAs were stably maintained in distinct nucleoid populations, whereas trans-complementation of heteroplasmic nucleoids was apparently achieved by the diffusion of mtDNA-derived transcripts within the mitochondrial matrix. Although the investigators speculated on a putative restorative mechanism of trans-complementation to operationally increase homoplasmic WT mtDNA and mitochondrial bioenergetics, other studies have demonstrated distinct and repeatable patterns of mtDNA heteroplasmy that varied across different cell types from the same individual (22,23) and were stably maintained in individual daughter cells over multiple cell divisions (19). An ostensibly straightforward interpretation of these observations indicates highly regulated normative expression of heteroplasmic mtDNA genomes within the intra-mitochondrial compartment in individual human cell types that complements normative mitochondrial function. Mechanistically, tissue- and cell-specific patterns of heteroplasmic mtDNA appear to be maintained via intra-mitochondrial trans-complementation of heteroplasmic nucleoids and mtDNA-derived transcripts (21), as well as the intercellular exchange of mtDNA (19). It also becomes apparent that the polycentric nature of heteroplasmic mtDNA-encoded transcripts introduces an additional level of complexity by which to evaluate putative facilitative roles of preserved patterns of mtDNA heteroplasmy on homeostatic metabolic processes (16).

Validation of the potential existential role of cell-specific patterns of mtDNA heteroplasmy on normative mitochondrial functions is also provided by a preclinical Drosophila genetic model employing a temperature-sensitive-lethal mtDNA mutation in functional linkage to the COI locus (24). Notably, the viability of homoplasmic flies at restrictive temperatures was fully maintained by expressing an alternative COI oxidase, which specifically conferred restorative mtDNA heteroplasmy that was associated with fully viable and tissue-specific phenotypes. Conversely, using a genetic replacement paradigm to induce tissue-specific mutant COI homoplasm in heteroplasmic flies, it was observed that restoration of mtDNA homoplasmy in the eye resulted in severe neurodegeneration at restrictive temperatures. Of note, utilizing the same temperature sensitive-lethal mtDNA model of COI dysfunction, the frequency of the mutant allele in heteroplasmic flies was significantly decreased in the germline and over multiple generations (25). A critical analysis of these two studies concludes that selection against potentially deleterious mtDNA heteroplasmic mutations during the process of oogenesis may be in marked contrast to developmentally determined, cell-specific patterns of mtDNA heteroplasmy generated during various stages of tissue differentiation.

3. Mitochondrial heteroplasmy and cancer

The mechanistic staging of carcinogenic cellular processes includes profound alterations of normative mitochondrial function (26-29), notably dependence on aerobic and anaerobic glycolysis, aberrant production and release of lactate, and metabolic downregulation of mitochondrial oxidative processes according to the classic theory promoted by Warburg et al (30). The primacy of cancer as a mitochondrial metabolic disease has been proposed (31-33), in marked contrast to widely espoused theories supporting the causative role of multiple somatic mutations in the etiology and persistence of numerous types of cancer. In light of the relatively high somatic mutational rates of mtDNA, the multi-modal coding mechanisms provided by cell-specific patterns of mtDNA heteroplasmy may reflect the evolutionary linkages of mitochondria to primordial protobacterial precursors and trillions of enteric bacteria contained within the human microbiome (20,34-36). As a corollary, cellular processes that regulate physiologically compatible patterns of heteroplasmic mtDNA may undergo state-dependent dysregulation resulting in an altered metabolically compromised phenotype characteristic of cancer cells and other pathophysiologically altered cell types (22,37). A relevant previous study has linked allele-specific expression of nuclear DNA with mtDNA heteroplasmy with functional impairment in bioenergetics and an apparent positive selection for reduced mitochondrial function (38). The overall results and conclusions of the study provide supportive evidence for positive selection processes driving higher order cellular pattern recognition of heteroplasmic mtDNA genomes in ordered stages of tumor progression (18,22,23,39).

Recently, a critical review has outlined a binary regulatory system responsible for selective expression of genes contained within mitochondrial and chloroplast genomes (40). The evolutionarily conserved mtDNA genome represents a self-contained genetic system that encodes existentially required catalytic and regulatory subunits of respiratory complexes I, III, and IV, and two subunits of F1F0 ATPase. The unifying principle responsible for reciprocal regulation of intraorganelle energy production is critically linked to maintenance of redox potential by electron transport through respiratory complexes. Accordingly, the functional transformation of cell-specific mitochondria into high-efficiency bio-engines appears to be dependent on the veracity of ongoing gene expression within restricted metabolic or physiological demands. The critical regulatory roles of cell-specific patterns of mtDNA heteroplasmy are fundamental to the maintenance of requisite metabolic capacity during the normal aging processes (18-20,39). These contentions are also supported by a recent study that has underlined the critical importance of functional mitochondria in the maintenance of differentiation and reprogramming of induced pluripotent stem cells (iPSCs) (41). Notably, a transition from somatic mitochondrial oxidative metabolism to glycolytic metabolism, highly reminiscent of cancer cells, was observed to be required for successful reprogramming of iPSCs. Accordingly, somatic mitochondria and associated oxidative bioenergetics are extensively remodeled with the induction of an iPSC-like phenotype, and the transition from oxidative to glycolytic metabolic processes appears to be strongly regulated by hypoxia, specifically by hypoxia-inducible factor 1α (HIF1α) signaling pathways (42,43). Early induction of HIF1α target genes may be required for iPSC derivation via the activation of a glycolytic program that is highly reminiscent of undifferentiated cancer cells.

4. Conclusions

We hypothesize that the biological significance of mtDNA heteroplasmy is reflected by the ability of cellular mitochondria to modulate effectively state-dependent changes in energy
requirements by concerted transcriptional and translational mechanisms (19,44,45). An apparent perturbation of homeo-
static regulation of intra-mitochondrial patterns of mtDNA heteroplasmy may be amplified during the initiation and
progression of pathophysiological processes associated with major human disease states (17,19,23,46,47). Furthermore, bi-
directional communication between cytosolic and mitochondrial signaling pathways provide co-ordinate regulation of nuclear DNA- and mtDNA-derived gene expression within a constantly changing physiological environment designed to promote
molecular switching of cellular metabolic machinery from meeting anabolic to catabolic demands (48). As technological
transplantation of functionally viable mitochondria comes with the anticipation of the significant restoration of normative
cellular function, understanding and mapping of cell-specific mosaic patterns of heteroplasmic mtDNA expression appears
to represent a prime prerequisite for future translational studies.

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