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Mitochondrial DNA Variations in Tumors: Drivers or Passengers?

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Abstract

Mitochondrial DNA alterations, including point mutations, deletions, inversions and copy number variations, have been widely reported in many age-related degenerative diseases and tumors. However, numerous studies investigating their pathogenic role in cancer have provided inconsistent evidence. Furthermore, biological impacts of mitochondrial DNA variants vary tremendously, depending on the proportion of mutant DNA molecules carried by the neoplastic cells (the so-called heteroplasmy). The recent discovery of inter-genomic crosstalk between nucleus and mitochondria has reinforced the role of mitochondrial DNA variants in perturbing this essential signaling pathway and thus indirectly targeting nuclear genes involved in tumorigenic and invasive phenotype. Therefore, mitochondrial dysfunction is currently considered a crucial hallmark of carcinogenesis as well as a promising target for anticancer therapy. This chapter describes the role of different types of mitochondrial DNA alterations by mainly considering the paradigmatic model of colorectal carcinogenesis and, in particular, it revisits the issue of whether mitochondrial mutations are causative cancer drivers or simply genuine passenger events. The advent of high-throughput next-generation sequencing techniques, as well as the development of genetic and pharmaceutical interventions for the treatment of mitochondrial dysfunction in cancer, are also discussed.

Keywords: mitochondrial DNA variants, heteroplasmy, nuclear-mitochondrial crosstalk, oxidative stress, mtDNA copy number alterations, D-loop, cancer therapy, mitogenomics

1. Introduction

Mitochondria are highly dynamic organelles whose biogenesis and functions are tightly regulated by the nucleus through a constant bidirectional crosstalk. Indeed, only about 1%

of mitochondrial proteins are encoded by mitochondrial DNA (mtDNA), with all the others encoded by the nuclear genome, including proteins involved in mtDNA replication and transcription [1].

The human mtDNA is a small circular double-stranded DNA molecule of approximately 16.6 kb that encodes for 2 ribosomal RNAs (12S and 16S), 22 transfer RNAs required for protein synthesis and 13 essential protein subunits of the oxidative phosphorylation system (OXPHOS) (**Figure 1**) [2]. The electron transport chain, the primary metabolic pathway which generates energy in the form of ATP, is composed of five protein complexes (I–V) localized in the inner membrane of mitochondria, including complex II that is exclusively coded by the nuclear genome. This system includes seven subunits of respiratory enzyme complex I, one subunit of complex III, three subunits of complex IV and two subunits of complex V. As mentioned before, all other mitochondrial proteins, including those involved in mtDNA replication, transcription and translation, are encoded by nuclear genes and are targeted to the mitochondrion by specific transport systems. The discovery of over 2000 mitochondrial small non-coding RNAs (mitosRNAs), playing a pivotal role in the control of normal mitochondrial gene expression, revealed an underestimated level of mitochondrial functional complexity [3]. Furthermore, studies on antisense anti-termination tRNAs and delRNAs shed new light on novel mechanisms expanding the coding potential of mitogenome [4, 5].

Byproducts of the electron transport chain (ETC) constantly generate reactive oxygen species (ROS) that may severely damage the mitochondrial DNA. If not efficiently repaired, the accumulation of oxidative lesions in the mtDNA molecules lead to gradual mitochondrial dysfunction, which is reflected in changes in the number, morphology and functioning of mitochondria, as observed in cancer cells [6].

mtDNA is more susceptible to mutations than nuclear DNA, due to the lack of histones and chromatin protective structures, paucity of introns, less efficient mtDNA repair mechanisms and a higher exposure to deleterious ROS generated during ATP synthesis within the mitochondrial compartment [7].

Although low levels of intracellular ROS normally regulate cellular signaling and are essential for normal cell survival and proliferation, aberrant ROS production is frequently observed in neoplastic cells. In the mitochondrial free radical theory of aging accumulation of damaging mtDNA mutations, impairment of oxidative phosphorylation as well as an imbalance in the expression of antioxidant enzymes results in exponential overproduction of ROS. This elicited condition forms a “vicious cycle” that is the basis of a wide range of pathologies, termed as “free radical diseases” such as cancer, neurodegeneration, atherosclerosis, diabetes mellitus and chronic inflammation [8]. Importantly, besides the obvious induction of oxidative nucleotide damage to mtDNA, ROS promotes tumorigenesis through several other mechanisms, including stabilization of hypoxia-inducible factor (HIF)- α , increased calcium flux, inactivation of key phosphatases, such as Pten and PP2A, and activation of both the NRF2 and NF- κ B transcription factors [9–11].

Since the Warburg theory of cancer postulated in 1956 [12], mitochondrial dysfunction has been regarded as a hallmark of cancer progression and as a promising target for anticancer therapies [13, 14]. For instance, enhancing complex I activity has been demonstrated to inhibit tumorigenicity and metastasis of breast cancer cells [15]. More recently, mitochondrial dysfunction

Figure 1. Map of the human mitochondrial DNA and distribution of somatic variants in colorectal cancer. mtDNA somatic mutations are mainly represented by homoplasmic alterations (black arrowheads), although rarer heteroplasmic substitutions (blue arrowheads) have been detected in the MT-RNR2 (16S) region or mixed homoplasmic/heteroplasmic variants (red arrowheads) in the MT-ND5 locus.

has also been associated with a crucial step for tumorigenesis, that is, epithelial-to-mesenchymal transition (EMT), enabling cancer dissemination and metastatic spread [16]. Importantly, mtDNA alterations may also disrupt the inter-genomic crosstalk between nucleus and mitochondrion and is associated with increased oxidative stress, ROS and cytosolic calcium accumulation, reduction of cell ATP levels and an imbalance in the NADH/NAD⁺ ratio. Moreover, ROS-induced oxidative stress may also affect the expression of nuclear genes involved in tumorigenic and invasive phenotypes, as it has been shown in colorectal cancer cells [17].

2. mtDNA alterations: a focus on colorectal carcinogenesis

2.1. Somatic mtDNA variants

Cancer is caused by the accumulation of multiple genetic alterations, such as point mutations, copy number variations (CNVs), inversions and epigenetic modifications [18]. This multi-step

process has been depicted in detail for colorectal cancer, which represents an ideal paradigm of tumorigenesis. In 1990, Fearon and Vogelstein [19] postulated a multi-step model of colorectal carcinogenesis, the long established “adenoma-carcinoma sequence”, in which the inactivation of the APC tumor-suppressor gene occurs first in normal colonic epithelial cells, followed by activating mutations in the KRAS gene and subsequent additional alterations in other tumor-suppressor genes, such as TP53 and TGF- β pathway genes.

Accumulating evidence emphasizes the functional role of mtDNA abnormalities in mitochondrial dysfunction and colorectal carcinogenesis. In a whole-genome comparative study of five different tumors, it has been demonstrated that the frequencies of deleterious non-synonymous somatic variants vary tremendously across tumor types, with the higher frequency (63%) in colorectal adenocarcinomas [20]. The vast majority of these mtDNA variants were represented by G > A and C > T transitions, the typical molecular fingerprint due to oxidative stress in mtDNA [21].

Thus far, mtDNA variants have been found to affect different regions with an essential role in mitochondrial protein synthesis machinery and oxidative phosphorylation (**Figure 1**) [22–24]. Importantly, it has been shown that mtDNA mutations may generate unprocessed transcripts by precluding RNA processing that impair mitochondrial biogenesis and energy maintenance [25, 26]. It is noteworthy to mention that mtDNA variants not only affect genes directly involved in the ETC, but also genes related to mitochondrial metabolism, such as tRNA genes, in which pathogenic mutations are 6.5 times more frequent than in other mitochondrial loci [27, 28].

MUTHY-associated polyposis (MAP) patients carry a significant increase of non-synonymous changes in conserved amino acid residues of the MT-CO₂ gene, particularly the hotspot m.7763G > A transition [29]. Nevertheless, there is no compelling evidence in the literature propending for a single common coding-region mtDNA variant or haplogroup that may strongly influence the risk of developing a colorectal adenocarcinoma. Alternatively, it is likely that mtDNA alterations influencing colorectal cancer risk may be in the form of heteroplasmic low frequency variants, possibly restricted to specific subsets of patients with colorectal cancer [30]. Curiously, it has been demonstrated that mutations disrupting the respiratory complex I in pituitary adenomas are somatic modifiers of tumorigenesis associated with less aggressive and genome-stable oncocytic lesions [31].

It is commonly believed that mtDNA variants arise due to positive selection of those “driver” variants conferring clonal growth advantage. Accordingly, we observed that likely non-pathogenic mtDNA variants (“passengers”) reverted to the wild-type homoplasmic status during tumor progression in colorectal cancer patients [29]. On the contrary, the mtDNA variants that are positively selected during tumor progression might be considered the most tolerable alterations for neoplastic cells. However, a deleterious impact of mtDNA passenger variants on cancer progression may not be completely excluded, as it has been previously evidenced in nuclear DNA passenger alterations [32].

2.2. Mitochondrial DNA heteroplasmy

Mitochondrial DNA heteroplasmy has been involved in a large spectrum of human diseases. Beside classical mitochondrial diseases, such as mitochondrial myopathy, myoclonic epilepsy with ragged red fibers, and mitochondrial encephalomyopathy, lactic acidosis, and stroke-like

episodes (MELAS), mitochondrial heteroplasmy also plays a pivotal role in complex disorders, including type 2 diabetes mellitus, late-onset neurodegenerative diseases and cancer [30].

mtDNA variants are maternally-inherited or arise as *de novo* somatic mutations in a fraction (heteroplasmic) or all (homoplasmic) mitochondrial genomes within each cell containing hundreds of copies of mtDNA molecules. Over time, the proportion of the mutant mtDNA within the cell may vary and drift toward predominantly mutant or wild-type form to achieve homoplasmy. Accordingly, the biological impact of a mtDNA variant may fluctuate, depending on the proportion of mutant mtDNA molecules carried by the neoplastic cell. Moreover, the level of heteroplasmy increases significantly with age and may vary between tissues and ethnic groups [33, 34]. By using high-throughput sequencing technology, Guo et al. [35] showed that very low heteroplasmy variants, down to almost 0.1%, are generally inherited from the mother, thus implying their likely neutral effect, and that this inheritance begins to decrease at about 0.5%. Accordingly, it has been demonstrated that high heteroplasmic mtDNA mutation loads, generally above 80%, are required to trigger substantial dysfunctions in the oxidative phosphorylation process. For instance, the m.3571insC mutation in the MTND1 gene of respiratory complex I is commonly detected in oncocytic tumors, in which it causes a severe mitochondrial dysfunction when mutant load is above 83% [36]. Importantly, this mitochondrial threshold effect strictly regulates the balance between tumor growth and suppression [37]. Interestingly, low-level mitochondrial heteroplasmies are commonly found in healthy individuals, and the advent of next-generation sequencing (NGS) technologies revealed that 25–65% of the general population harbor at least one heteroplasmic variant across the entire mitochondrial genome [38, 39]. By studying human colorectal cancer cell lines, Polyak et al. [40] showed that the vast majority of mutations were ROS-related homoplasmic transitions, indicating that mtDNA molecules could rapidly become homogeneous under high clonal selection conditions. Nevertheless, several other *in vivo* studies demonstrated that mtDNA heteroplasmy is far more common in colorectal neoplasms [41–43]. As occasionally observed in the case of revertant mosaicism, a naturally occurring phenomenon involving spontaneous correction of a pathogenic mutation in a somatic cell, heteroplasmic somatic variants may also naturally revert to wild-type homoplasmy [44, 45].

2.3. mtDNA copy number alterations

Epidemiological studies have indicated significant association of leukocyte mtDNA copy number with risk of several malignancies, including glioma, colorectal and breast tumors, and its use has been proposed as a potential biomarker to select patients who benefit from adjuvant chemotherapy [46–50]. A reduced mtDNA content has also been correlated with lymph node metastasis and lower survival rates in patients with colorectal cancer [51].

In the past years, it has been demonstrated that mtDNA depletion leads to tumorigenesis by inducing changes in the redox status, membrane potential, ATP levels, gene expression, nucleotide pools, and increased chromosomal instability (e.g. translocations) [52, 53]. However, other findings reported a gain of mtDNA copy number, thus suggesting that mtDNA replication could be increased to compensate for detrimental metabolic effects caused by mtDNA variations and/or oxidative stress [54]. These conflicting data may be partly explained by the non-homogeneous timing of blood DNA analyses for mtDNA copy number determination. Interestingly, depletion

of mtDNA results in significant changes in methylation patterns of a number of nuclear-encoded genes, and these epigenetic modifications are reversed by the restoration of mtDNA content [55].

The molecular mechanism altering mtDNA copy number is still under investigation. In a study of 65 colorectal cancers, it has been suggested that hypomethylation of specific sites on CpG islands of the D-loop promoter may be involved in the regulation of mtDNA copy numbers [56]. Moreover, it has been reported that polymorphisms within the nuclear-encoded polymerase gamma gene (POLG), which codifies for a key component of the mitochondrial genome maintenance machinery, may lead to a decrease in mtDNA content and mitochondrial dysfunction [57]. Curiously, a homozygous polymorphic insertion (AluYb8MUTYH) in the 15th intron of the MUTYH base excision repair gene has been associated with a significant reduction of the type 1 MUTYH protein that localizes to mitochondria as well as lowered mtDNA content in age-related diseases [58]. Since biallelic mutations of MUTYH are associated with the MAP syndrome, it might be speculated that homozygous or compound heterozygous MUTYH variants may correlate with the mtDNA content in colorectal cancer [30].

2.4. D-loop and mitochondrial instability

The non-coding D-loop region contains essential transcription and replication elements and is formed by two hypervariable regions, namely HV-I (nt. 16,024–16,383) and HV-II (nt. 57–333) [59]. The latter includes the D310 sequence, a polycytidine repeat (nt. 303–309), which is essential for mtDNA replication in virtue of the H-strand replication origin. Replication of the leading strand initiates at the origin of H-strand synthesis and proceeds unidirectionally, displacing the parental H-strand as single-stranded DNA [60]. The D-loop is a well-known hotspot for somatic mutations in many types of cancer, with a mutation rate 100- to 200-fold higher than nuclear DNA. This finding may be partly explained by considering the direct relationship between mutational frequency and single-strandedness during mitochondrial replication [61].

mtDNA variants in the D-loop region have been repeatedly associated with risk and survival rates in cancer patients and, thus, they have been proposed as valuable prognostic markers. However, it has been argued that most of these studies could be biased due to artifacts related to genotyping errors or inadequate experimental design [62]. Mitochondrial microsatellite instability (mtMSI), that is a change in length in the repetitive sequences of the D-loop segment between normal and tumor tissues, has been described as a frequent molecular event in different cancers, but its prognostic value is still debated [63]. The variation of the homopolymeric tract length mainly arises through replication slippage of mitochondrial DNA polymerase and, importantly, this process may affect mtDNA replication and transcription. Intriguingly, the oxidative damage to mitochondrial polymerase γ may also contribute to the alteration in the length of the polycytidine repeat by impacting on mtDNA replication [64].

Instability of the D-loop hypervariable region-II (HV-II) has been associated with variants specifically grouped inside the MT-CO₂ gene in MAP patients, thus suggesting that genome instability might contribute to drive non-random accumulation of MT-CO₂ variants in the early stages of MAP colorectal tumorigenesis [29]. Therefore, D-loop mutations probably do not directly drive carcinogenesis but are more likely an epiphenomenon, used as a universal clonal marker (“molecular clock”) to estimate the relative mitotic history of tumors [65, 66].

3. Mitochondrial-nuclear crosstalk

Tight coordination between the nucleus and mitochondria is required for proper mitochondrial functioning and includes both anterograde (nucleus to mitochondria) and retrograde (mitochondria to nucleus) signals. This crosstalk is critical for the maintenance of cellular homeostasis, and accumulated mtDNA variants may perturb this subtle pathway [67]. It has been demonstrated that somatically acquired mitochondrial-nuclear genome fusion sequences are present in human cancer cells [68]. Although most of the genes encoding proteins of the OXPHOS machinery are transcribed in the nucleus (anterograde signaling), mitochondria may also exert retrograde regulatory control over the nucleus in terms of nuclear gene expression modulation [69]. This phenomenon suggests a strong association between nuclear and mitochondrial DNA alterations in driving tumor development and progression. Variants in nuclear-encoded mitochondrial genes, such as fumarate hydratase, iso-citrate dehydrogenase and succinate dehydrogenase) have been associated with a wide variety of human cancers, such as paragangliomas, uterine leiomyomas, renal carcinomas, breast cancers, gastrointestinal stromal cancers, leukemia, prostate cancer, glioblastomas and colorectal carcinomas [70–78]. Furthermore, it has been demonstrated that mtDNA changes and MAPK pathway alterations synergize to drive colorectal malignant transformation [79].

In a study on colorectal adenoma and adenocarcinoma samples, an increased number of mutations in nuclear genes encoding proteins involved in critical mitochondrial processes, such as fusion, fission and localization were found [80]. It has also been suggested that mtDNA depletion may disrupt crucial nuclear processes, leading to centrosome amplification and mitotic spindle multipolarity, both participating in cancer cell transformation [81, 82]. mtDNA variants have the potential to induce molecular signals through the mitochondrial-nuclear crosstalk mechanism, thereby promoting nuclear compensation in response to mitochondrial malfunction [67]. Interestingly, some typical nuclear transcription factors, such as the tumor-suppressor p53 and estrogen receptor (ER), are localized within mitochondria, where they exert various transcription-independent functions [83]. By using transmitochondrial cybrid systems (“cybrids”), Kaiparettu et al. [69] elegantly demonstrated that mitochondria derived from the non-transformed breast epithelial cell line MCF10A reverse the tumorigenic properties of osteosarcoma metastatic cells (e.g. cell proliferation and viability under hypoxic conditions, anchorage-independent cell growth, resistance to anticancer drugs) by suppressing several oncogenic pathways involving HER2, SRC, RAS and TP53; on the other hand, some of the tumor-suppressor genes including VHL, PTEN and RB1 were overexpressed in cytoplasmic hybrids (cybrids) with non-cancerous mitochondria.

Other studies suggested that mitochondrial dysfunction may induce epigenetic modifications within the nuclear genome, such as aberrant methylation patterns in CpG-rich regions [84, 85]. These epigenetic alterations, including DNA and chromatin modifications and signaling through small RNAs, may contribute to the maintenance of mitochondria-mediated oncogenic transformation. However, the mitochondrial signals that potentially might trigger these epigenetic changes in the nucleus remain still largely unknown [30].

ROS-induced mitochondrial deregulation has been reported to trigger a survival response by inducing the nuclear factor NF- κ B pathway and stimulating the synthesis of anti-apoptotic molecules (such as Bcl-xL/Bcl-2), which in turn promote cell survival and proliferation [86].

Moreover, oxidative stress may also affect the expression of nuclear genes involved in tumorigenic and invasive phenotypes [87]. Altogether these findings suggest that targeting the retrograde signaling could be a successful therapeutic strategy for cancer.

4. Targeting mitochondria for cancer therapy

Numerous studies suggested that mtDNA alterations may contribute to chemotherapy resistance and affect radiotherapy outcome. For instance, Guerra et al. [88] showed that mutations in the NADH dehydrogenase subunit 4 (MT-ND4) lead to acquired chemoresistance during treatment with paclitaxel carboplatin.

In the last few years, spindle transfer, a promising emerging strategy aimed at generating clinical germline gene therapy against inherited mitochondrial disorders, has supported the idea of a possible gene therapy approach for the editing of somatic mtDNA alterations [89]. Ideally, repairing the mutated mtDNA sequence would also restore the normal mitochondrial function and likely induce tumor regression. Taylor et al. [90] proposed a strategy that aimed to specifically block the replication of the mutant mtDNA by peptide nucleic acid (PNA), thereby allowing the selective propagation of the wild-type DNA. Moreover, mitochondrial dysfunction might also be restored by stimulating the mitophagy process in order to eliminate the deleterious mtDNA variants [91]. Targeting DNA repair enzymes to mitochondria may be a suitable strategy to correct mtDNA mutations. For instance, cell transfection with an expression vector containing the gene coding the DNA repair enzyme human 8-oxoguanine DNA glycosylase/apurinic lyase (hOGG1) has been used to reduce free fatty acids (FFAs)-induced mtDNA damage [92]. Furthermore, overexpression of hOGG1 in mitochondria has been shown to attenuate breast cancer progression and metastasis in transgenic mice [93]. Although hOGG1 has been the most frequently employed enzyme to enhance mtDNA repair, alternative strategies targeting other proteins transferred to mitochondria, such as endonuclease III (EndoIII) and endonuclease VIII (EndoVIII), have been proposed in the last years [94–96]. Other therapeutic approaches for patients carrying mtDNA mutations are based on allotopic gene expression, as preliminary demonstrated in different mitochondrial disorders [97], and targeted restriction endonucleases. In this regard, SmaI and PstI have been used as a powerful tool for treatment of mitochondrial dysfunction, resulting in the elimination of the mutant mtDNA and restoration of normal mitochondrial functionality [98]. In the last decade, many other approaches and compounds targeting dysfunctional mitochondria have been experienced, such as signal peptides. Lipophilic cations, cell-penetrating peptides and nanoparticles. A promising approach is based on the reprogramming of energy metabolism in colorectal cancer cells, through specific mitochondria-targeting agents, such as the second-generation rosamine analogs that target complex II and ATP synthase activities of the mitochondrial oxidative phosphorylation pathway [99]. More recently, it has been argued that mitochondria of tumor-initiating cells (TICs), which play a prominent role in cancer initiation, metastasis and resistance to therapy, may be targeted by mitocan vitamin E succinate in a complex II-dependent manner [100]. Another original approach has been developed to trigger cell death signaling pathways in colorectal cancer cells [101], such as ROS-dependent apoptosis and autophagy [102]. The recent improvement of high-throughput drug-screening platforms allowed the identification of novel non-toxic mitochondrial inhibitors, as in the

case of diphenyleneiodonium chloride (DPI), a strong inhibitor of mitochondrial complex I and II flavin-containing enzymes, which effectively depletes cancer stem-like cells (CSCs), one of the main drivers of poor clinical outcome in a wide variety of tumor types and especially in advanced disease states [103]. Interestingly, mitochondrial inhibition with VLX600 has also been proposed in combination with imatinib in the treatment of drug-resistant gastrointestinal stromal tumors (GISTs) [104].

Recently, morphological and ultrastructural changes in the mitochondrial cristae structure (cristae remodeling), for example, through the optic atrophy 1 (OPA1) pathway, represent an important step in apoptosis and autophagy, and a potential target for future pharmacological modulation in cancer [105].

Chromosomal translocations generating in-frame oncogenic gene fusions also represent successful examples of targeted cancer therapies, and recently it has been shown that the FGFR3-TACC3 (F3-T3) gene fusion—initially discovered in human glioblastoma and then reported in many other cancers—promotes oxidative phosphorylation, mitochondrial biogenesis and tumor growth [106–108].

5. Ultra-sensitive next-generation sequencing techniques and mitogenomics

Whole mitochondrial genome analysis by high-throughput next-generation sequencing (NGS) techniques enables the detection of low-level heteroplasmic mtDNA variants and completely revolutionized mitogenomics in the last few years [109]. This approach has been extensively applied to different mitochondrial disorders to carefully investigate the transmission dynamics of low-level maternal germline mtDNA variants across generations [110–112]. In a comparative analysis, it has been demonstrated that Sanger sequencing is valid for quantification of heteroplasmies with more than 10% of cells/mitochondria carrying the mutation, whereas NGS is capable of reliably detecting and quantifying heteroplasmic variants down to the 1% level [113]. Recently, a massive parallel sequencing (MPS) protocol reliably quantified low frequency, large mtDNA deletions in single cells with a lower detection limit of 0.5% [114]. mtDNA NGS has been also suggested as a useful quality check of pluripotent stem cells for drug discovery and regenerative medicine purposes [115].

Conventionally, DNA variants detected in a tumor sample but not in the germline counterpart (such as peripheral blood, buccal swab or saliva) are scored as somatic (likely pathogenic) mtDNA variants, otherwise they are considered as germinal variants (likely polymorphic/benign). High-throughput NGS approaches may unveil low-level germinal heteroplasmies having a tumoral tissue counterpart with higher heteroplasmy simply because of increased cell replication rate or random genetic drift phenomena and, therefore, without any deleterious oncogenic effect. The ultra-sensitive detection rate of NGS methods may be used to monitor even subtle shifts in the heteroplasmy levels of the tumor during time and potentially correlate them with tumor evolution [116]. Moreover, the possibility to easily analyze the circulating cell-free mtDNA isolated from plasma/serum (“liquid biopsy”) or urine [117–119], may allow non-invasive serial sampling from the same patient.

6. Conclusions

In the last decades, evidence on the contribution of mtDNA variants to tumorigenesis has incredibly grown. Therefore, mitochondria are actually considered one of the most promising targets for novel anticancer therapies. Accordingly, mtDNA variants can be regarded as useful tumor biomarkers for clinical practice, whereas the tight communication between nuclear and mitochondrial genomes sheds new light on the molecular and functional mechanisms underlying the onset and progression of complex human diseases, such as cancer and neurodegenerative diseases.

Conflict of interest

The authors declare no conflicts of interest. This article does not contain any studies with human participants performed by the authors.

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