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Mitochondrial genetic variations in leukemia: a comprehensive overview

Ao Zhang[a,b], Wenbing Liu[a,b], Shaowei Qiu[a,b,*]

[a]State Key Laboratory of Experimental Hematology, National Clinical Research Center for Blood Diseases, Haihe Laboratory of Cell Ecosystem, Institute of Hematology & Blood Diseases Hospital, Chinese Academy of Medical Sciences & Peking Union Medical College, Tianjin, China; [b]Tianjin Institutes of Health Science, Tianjin, China

Abstract

Leukemias are a group of heterogeneous hematological malignancies driven by diverse genetic variations, and the advent of genomic sequencing technologies facilitates the investigation of genetic abnormalities in leukemia. However, these sequencing-based studies mainly focus on nuclear DNAs. Increasing evidence indicates that mitochondrial dysfunction is an important mechanism of leukemia pathogenesis, which is closely related to the mitochondrial genome variations. Here, we provide an overview of current research progress concerning mitochondrial genetic variations in leukemia, encompassing gene mutations and copy number variations. We also summarize currently accessible mitochondrial DNA (mtDNA) sequencing methods. Notably, somatic mtDNA mutations may serve as natural genetic barcodes for lineage tracing and longitudinal assessment of clonal dynamics. Collectively, these findings enhance our understanding of leukemia pathogenesis and foster the identification of novel therapeutic targets and interventions.

Key Words: Leukemia; mtDNA copy number variations; mtDNA mutations; mtDNA sequencing; Natural genetic barcodes; Prognostic marker

1. INTRODUCTION

Leukemias are a group of heterogeneous hematological malignancies resulting from uncontrolled neoplastic proliferation of undifferentiated or partially differentiated hematopoietic cells.[1] The occurrence of leukemia cells is closely associated with genetic variations. In addition, the genetic heterogeneity and clonal dynamics of leukemia have been extensively described based on somatic nuclear mutations during disease progression.[2–4] However, in addition to these nuclear genetic changes, non-chromosomal mitochondrial genetic variations also play a crucial role in the progression of leukemia.[5–8]

Mitochondria are the main energy-producing organelles of cells and are also involved in numerous cellular processes including calcium homeostasis, apoptosis, fatty acid oxidation, and the generation of metabolic intermediates.[9] Mitochondrial DNA (mtDNA) is a 16 kb double-stranded circular genome containing 37 genes related to the respiratory chain, including 13 messenger ribonucleic acids (mRNAs), 2 ribosomal RNAs (rRNAs), and 22 transfer RNAs (tRNAs) necessary for mitochondrial protein synthesis.[7] The mitochondrial genome also includes a noncoding displacement region (D-loop), which has a very significant transcriptional regulation function.[10] Unlike the nuclear genome, the mitochondrial genome has no intron (only composed of encoding genes and regulatory sequences), thus one point mutation may cause alterations of important structural genes, leading to changes in expression or property of the expressed proteins.[11] Damages easily occur to mtDNA due to its vulnerability without histone protection and a DNA repair system.[11] Moreover, mtDNA has a 10-fold higher mutation rate compared to the nuclear genome. Hence, mtDNA incrementally accumulates unique, irreversible genetic mutations and passes on to daughter cells even in healthy humans, which could be used for lineage tracing.[12–14] Importantly, the number of mitochondria (and therefore mtDNA) ranges from several hundred to >10,000 per cell in different cell types, facilitating robust mtDNA analysis even from a single cell[14] (Table 1).

The mtDNA variations encompass mtDNA copy number variations and mutations. Mutations in mtDNA are primarily responsible for mitochondrial abnormalities.[15] De novo mutations act as "inducers" of carcinogenesis, while functional variants act as "adaptors," and enable cancer cells to thrive in diverse environments. The mechanisms through which mtDNA mutations contribute to tumorigenesis involve the modulation of mitochondrial reactive oxygen species production, redox state, and mitochondrial intermediates that act as substrates for chromatin-modifying enzymes. In addition, alterations in the mtDNA can impact mitochondrial metabolites, subsequently influencing the epigenome and nuclear DNA gene expression.[16] Some mtDNA mutations exist heteroplasmically, meaning that individual cells contain a mixture of mitochondria with mutant or wild-type (WT) mtDNA. In heteroplasmic cells, the phenotype of a pathogenic mtDNA mutation is

Address correspondence: Shaowei Qiu, State Key Laboratory of Experimental Hematology, National Clinical Research Center for Blood Diseases, Haihe Laboratory of Cell Ecosystem, Institute of Hematology & Blood Diseases Hospital, Chinese Academy of Medical Sciences & Peking Union Medical College, China. E-mail address: qiushaowei@ihcams.ac.cn (S. Qiu).

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Table 1

Comparison between the nuclear and mitochondrial genomes.

Characteristic	Nuclear genome	Mitochondrial genome
Size	~3.3 × 10^9 bp	16,569 bp
Number of genes encoded	~20,000–30,000	37
Number of DNA molecules per cell	46	~100–>10,000
Gene density	~1 per 40,000 bp	1 per 450 bp
Introns	Frequently found	Absent
Percentage of the coding DNA	~3%	~93%
Histone proteins and DNA repair system	With	Without
Mode of inheritance	Mendelian inheritance and paternal inheritance	Matrilineal inheritance

determined by the ratio of mutant and WT genomes.[17] mtDNA copy number refers to the number of mitochondrial genome copies per nucleated cell. The most common approach to measuring mtDNA copy number is to calculate the ratio of the number of mtDNA copies and nDNA reads (2 copies per cell).[18] mtDNA copy number alterations can impact oxidative phosphorylation and energy metabolism, thereby contributing to mitochondrial dysfunction.[19,20] It is noteworthy that alterations in mitochondria number per cell are associated with the development of leukemia through the deregulation of respiratory profiles.[21]

In this review, we aimed to provide a comprehensive overview of mitochondrial genetic variations in leukemia, focusing on mtDNA mutations, copy number variations, mtDNA sequencing methods, and their significance as natural genetic barcodes and prognostic markers. It aims to facilitate the identification of potential therapeutic targets for the treatment of this hematological malignancy (**Fig. 1**).

2. mtDNA MUTATIONS IN LEUKEMIA

Mitochondria are under a double genetic control carried out by mtDNA and nuclear DNA. mtDNA mutations play critical roles in the reprogramming of mitochondrial metabolism. Studies have revealed that disruptive mutations of mtDNA are typically rare in normal cells but undergo a positive selection in cancer tissues, highlighting the critical role of mtDNA in tumorigenesis.[22] This observation has also been confirmed in leukemia since several mutations in mtDNA are associated with leukemia. For instance, Penter et al[4] discovered that chronic lymphocytic leukemia (CLL) cells carry at least 1 mtDNA mutation in over 50% of cells and can accumulate numerous additional mutations. He et al[23] demonstrated that somatic mtDNA mutations were present in approximately 40% of patients, with the A15296G mutation serving as a leukemia-specific marker. Furthermore, a study identified 2 mitochondrial protein-coding genes (*ND6* and *Cytb*), with mutation frequencies exceeding 5% in platelets from leukemia patients. In particular, the *ND6* gene exhibited the highest mutation frequency at 32.65%.[11] Recurrent variations at position T489C within the mtDNA D-Loop have been reported in various malignancies.[24–26] Tyagi et al[10] investigating pediatric acute myeloid leukemia (AML) cases, suggested that the T489C position might be a hotspot for recurrent variations associated with relapse, although these variations occur within the noncoding regions and appear functionally silent, they could potentially impact respiratory chain polypeptide levels and alter electron transport chain (ETC) activity and cellular energy capacity. Some somatic mutations were detected in both diagnosis and remission samples, probably due to the persistence of residual leukemia cells.[27]

In AML cases specifically, the most observed mtDNA sequence variants were single nucleotide substitutions which might have affected the binding of mtDNA to transcription factors, resulting in abnormal formation of protein complexes, thereby altering normal expression patterns.[28] Studies

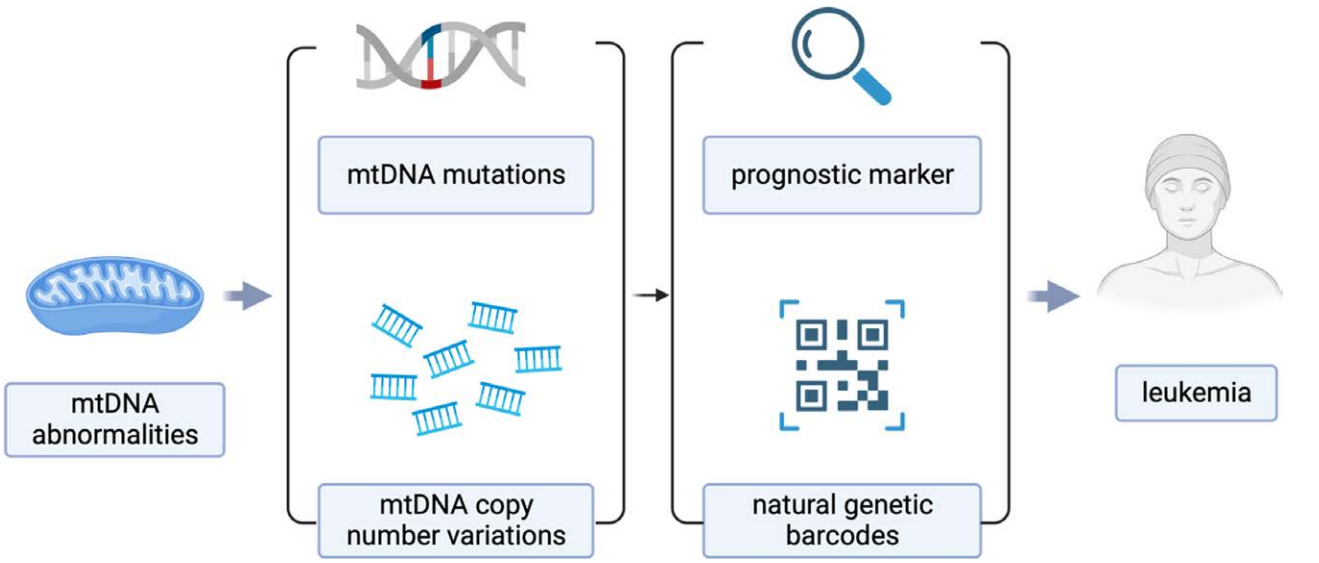

Figure 1. The mtDNA abnormalities encompass mtDNA copy number variations and mutations. The mtDNA abnormalities can serve as natural genetic barcodes and prognostic markers in leukemia. Increasing evidence indicates that mitochondrial genome abnormalities are important mechanisms of leukemia pathogenesis. mtDNA = mitochondrial DNA.

have demonstrated that patients with AML having high expression of mitochondrial transcription machinery (MTM) genes exhibit a significantly short median overall survival (OS; HR = 1.82, P = .003),[29] which might be explained by the deregulation of the MTM genes which may also influence the expression of mitochondrial genes and contribute to the pathogenesis of AML. In addition, some studies revealed a mutually exclusive relationship between somatic mitochondrial mutations and the *CBFB::MYH11* fusion gene in the beat AML cohort,[30] indicating potential substitutable functional mutations[31] and deducing that the *CBFB::MYH11* fusion gene was probably involved in mitochondrial metabolism reprogramming in AML. Additionally, alterations in copy number variations could co-occur with changes in mtDNA mutations and chromatin accessibility.[4] Moreover, some results indicate the congruence of mutations between mtDNA and RNA in AML.[30]

Mitochondrial mutation rates and the occurrence of leukemia are positively correlated.[32] However, the specific mechanism by which mtDNA mutations promote the pathogenesis of leukemia remains unknown. Whether accumulated mutated mtDNA is a consequence of rapid cell division, or if these mtDNA mutations confer selective advantages to transformed cells that contribute to leukemia progression and metastasis, such as affecting mitochondrial metabolism in that cell lineage remains controversial.

3. mtDNA COPY NUMBER VARIATIONS IN LEUKEMIA

Tumor cells display metabolic adaptations by modulating mitochondrial metabolism, ETC complex activities, and mtDNA copy numbers.[33] Transcriptional profile analysis has shown close correlations between transcript levels of enzymes of the tricarboxylic acid cycle and ETC, fatty acid β-oxidation, and branched-chain amino acid catabolism pathways with mtDNA copy numbers. Variations in mitochondria copy number reflect the cumulative effects of gene–environmental interactions, and the copy number has been implicated as a potential biomarker for various cancers.[7,34,35] Alterations in mtDNA copy number differ between certain tumor types. Studies have shown that the mean mtDNA copy number increased approximately 9-fold in patients with AML compared to that in controls (P < .0001). Pereira-Martins et al[36] demonstrated that patients with acute promyelocytic leukemia (APL) treated with chemotherapy and all-transretinoic acid whose leukemia cells had increased mtDNA content had a significantly low cumulative incidence of relapse. Another study suggested that mtDNA copy number was associated with anemia in hematologic malignancies, while red blood cell-bound mtDNA was negatively correlated with hemoglobin levels in patients.[37] mtDNA copy number is regulated by various mitochondrial biogenesis genes like *TFAM*, *POLG*, and *POLRMT*. Enhanced mitochondrial biogenesis is an important cellular adaptation in pediatric AML blasts, with a possible impact on disease biology and outcomes.[38] A recent study on adults with AML also reported increased expression of *TFAM* and *POLRMT* and other mitochondrial transcriptional factor genes like *TFB1M* and *TFB2M*, with poor prognostic significance.[29]

However, further investigation is needed to explore the relationship between mtDNA copy number variations and the pathophysiology and prognosis of leukemia. In some tumors, changes in mtDNA copy number might be an adaptive response secondary to mutations conferring growth advantage for certain tumor types. Furthermore, even within a class of tumors, differences in mtDNA content can correlate with cancer severity,[16] although these conclusions have not yet been confirmed in leukemia.

4. TECHNIQUES FOR mtDNA SEQUENCING

Currently, accessible techniques for sequencing mtDNA include the assay for transposase-accessible chromatin sequencing (ATAC-seq), RNA sequencing (RNA-seq), whole genome sequencing, quantitative real-time polymerase chain reaction (PCR), digital PCR, and others. However, since bulk sequencing entangles information from various cell types and obscures cellular heterogeneity, single-cell multi-omics has rapidly progressed.[39]

Longitudinal tracking of distinct clones within heterogeneous cell populations and linking clonal lineages to specific functional states at the single-cell level has been the focus of numerous endeavors aimed at comprehending disease evolution and mechanisms leading to therapeutic resistance in a wide range of cancers. The advent of genomic techniques has provided the means to concomitantly assess cell types and states with mtDNA genotypes.[40] It is worth mentioning that the combination of single-cell transcriptomics with mtDNA mutation detection could also establish lineage relationships.[41] Mitochondrial single-cell ATAC-seq (mtscATAC-seq) has emerged as one of the most successful single-cell sequencing methods in its ability to concomitantly profile accessible chromatin profiles alongside mitochondrial genotypes in a scalable manner.[42] However, characterizing and interpreting heterogeneous mixtures at the cellular level (also known as cell-type deconvolution) is also of great interest. Single-cell assays offer an opportunity for assigning single cells to the subpopulations (ie, clustering) and de-convolving the bulk data into subpopulation-specific data.[43] Other common deconvolution models are listed in Table 2.

5. mtDNA MUTATIONS AS NATURAL GENETIC BARCODES FOR LONGITUDINAL ASSESSMENT

Researchers have demonstrated the utility of somatic mtDNA mutations as natural genetic barcodes that may be stably propagated across cell divisions.[14,50] Cells sharing the same mitochondrial mutations are inferred to have descended from the same ancestral cell.[14] This offers opportunities for lineage tracing with enhanced resolution within cell populations defined by the same set of somatic mutations or even in the absence of such genetic events.[51] Compared to somatic nuclear mutations, mtDNA mutations possess several advantages. Mitochondria can replicate independently of the cell cycle, and mtDNA presents in hundreds of copies per cell. In combination with the small size, mtDNA can therefore be sequenced with high coverage at single-cell resolution without requiring additional targeted amplification steps while simultaneously providing information on cell states.[4,9,52] Somatic mtDNA mutations with levels as low as 5% heteroplasmy can be stably propagated, suggesting that even low-frequency heteroplasmic mtDNA mutations can be exploited for lineage tracing. Lineage tracing by mtDNA mutations is highly scalable and combined with assays to profile a cell's state at the chromatin or transcriptome level. In contrast, detecting nuclear somatic mutations by whole genome sequencing in individual cells remains costly, challenging to implement

Table 2

Summary of the current deconvolution models in ATAC-Seq data.

Deconvolution models	Key references
DC3	Zeng et al[43]
scAVENGERS	Han et al[44]
scDeconv	Liu[45]
DeconPeaker	Li et al[46]
Cellformer	Berson et al[47]
DECODER	Peng et al[48]
ConDecon	Aubin et al[49]

ATAC-Seq = assay for transposase-accessible chromatin sequencing.

on a large scale, and prone to substantial error rates.[50] In addition, nuclear mutations are often sparse events and frequently subject to selective pressures which could confound the inferences.

Stable propagation of mtDNA mutations over years in the absence of strong selective pressure indicates clonal persistence, but dramatic changes happen following tight bottlenecks including disease transformation and relapse post-therapy, paralleled by acquisition of copy number variants, alterations in chromatin accessibility, and gene expression. mtDNA mutations thus mirror disease history and provide natural genetic barcodes to enable patient-specific study of cancer subclonal dynamics.[4] Additionally, some scholars suggest that mtDNA mutations might be markers of a diverging subclone that emerged within the relapse population. They also observed distinct patterns over time for each mutation type, linking subclonal structures to different functional states.[4] One study utilizing mtsc ATAC-seq revealed that all the mtDNA mutation subclones were detected in different cell lineage clusters, suggesting multiple cell clones might contribute to the AML hierarchy, and different cells might fall into a similar type of "attractor" in AML.[53,54] Additionally, it was found the same phenotype contained multiple clones, implying there were certain key attractors responsible for determining the switching between different states.[54,55]

Overall, in one validation experiment, mitochondrial genotypes accurately inferred clonal lineage with >95% accuracy, achieving similar accuracy as widely applied genetic labeling methods. However, potential limitations include the horizontal transfer of mitochondria between cells under specific contexts and our incapacity to account for the phenotypic effects of the mtDNA mutations used for clonal tracing.[50] Lastly, the bulk nature and relative rarity of mtDNA transcriptome/genome coverage limit confident detection of low-frequency variants, which appeals to complementary bulk and single-cell genotyping assays optimized for mtDNA sequence capture.[40] Enhanced throughout and expanded coverage of single cells in the future will augment the level of detection of mtDNA mutations, thereby facilitating the tracking of clones using mtDNA mutations in settings such as premalignant states or minimal residual disease.[4] Applications of these methodologies also include the study of acquired therapeutic resistance and efficient tracking of donor and recipient chimerism during hematopoietic stem cell transplant.[50] Moreover, it is crucial to emphasize the integration of capturing mtDNA mutations with different types of nuclear mutations, which would further enhance the elucidation of both genomic and non-genomic evolution of malignant cells.[4]

6. mtDNA VARIATIONS AS A PROGNOSTIC MARKER IN LEUKEMIA

Mitochondria have a functional role in leukemia, numerous studies have reported that mtDNA variations may serve as a prognostic marker. mtDNA mutations in patients with AML were associated with inferior disease-free survival.[56] mtDNA copy number was often increased with a negative impact on disease aggressiveness and inferior survival in pediatric acute leukemia.[7,33] Mitochondrial heteroplasmic single nucleotide variants were also linked with prognosis in leukemia.[57] Mutations in mitochondrial genes that encode complexes I, III, and IV of the ETC have been linked to worst outcomes in patients with AML, potentially providing targets for therapeutic intervention.[58,59] Nicotinamide adenine dinucleotide hydrogen dehydrogenase subunit 4 (*ND4*) is a mitochondrial encoded transmembrane component of the ETC respiratory complex I, patients with somatically acquired *ND4* mutations had significantly longer relapse-free survival (P = .017) and OS (P = .021) than those with *ND4* (WT),[60] which implied the favorable prognostic influence of acquired *ND4* mutations in AML. Variations in one of the hypervariable regions in the D-Loop region (HV-1), namely 16126T-->C (P = .05), 16224T-->C (P < .01), and 16311T-->C (P < .001), were significantly associated with the inferior encrypting file system in pediatric AML.[61] Studies also found a strong association between deleterious heteroplasmic mitochondrial mutations and mortality.[57] However, some studies indicated that the impact of mtDNA copy number on outcomes in adult AML showed no significant association with survival,[7] further investigations should be conducted to examine this question.

7. CONCLUSION

mtDNA encodes critical components for mitochondrial function. Mitochondrial dysfunction caused by mtDNA mutations and copy number variations is closely implicated in tumorigenesis and progression.[62] Mitochondrial genetic analysis can enhance the stratification of malignant cells and improve our understanding of clonal dynamics in response to therapies, including putative treatment-sensitive and resistant clones.[40,50] However, how we can effectively and longitudinally study clonal dynamics in humans remains an important question. A systematic understanding of somatic mitochondrial variations and their interplay with the nuclear genome in leukemia remains poorly understood.

REFERENCES

[1] Nabavizadeh SA, Stein J, Mohan S. Neuroimaging in leukemia. *Hematol Oncol Clin North Am* 2016;30(4):823–842.

[2] Landau DA, Sun C, Rosebrock D, et al. The evolutionary landscape of chronic lymphocytic leukemia treated with ibrutinib targeted therapy. *Nat Commun* 2017;8(1):2185.

[3] Landau DA, Tausch E, Taylor-Weiner AN, et al. Mutations driving CLL and their evolution in progression and relapse. *Nature* 2015;526(7574):525–530.

[4] Penter L, Gohil SH, Lareau C, et al. Longitudinal single-cell dynamics of chromatin accessibility and mitochondrial mutations in chronic lymphocytic leukemia mirror disease history. *Cancer Discov* 2021;11(12):3048–3063.

[5] Carew JS, Zhou Y, Albitar M, Carew JD, Keating MJ, Huang P. Mitochondrial DNA mutations in primary leukemia cells after chemotherapy: clinical significance and therapeutic implications. *Leukemia* 2003;17(8):1437–1447.

[6] Grist SA, Lu XJ, Morley AA. Mitochondrial mutations in acute leukaemia. *Leukemia* 2004;18(7):1313–1316.

[7] Kim HR, Kang M-G, Lee YE, et al. Spectrum of mitochondrial genome instability and implication of mitochondrial haplogroups in Korean patients with acute myeloid leukemia. *Blood Res* 2018;53(3):240–249.

[8] Linnartz B, Anglmayer R, Zanssen S. Comprehensive scanning of somatic mitochondrial DNA alterations in acute leukemia developing from myelodysplastic syndromes. *Cancer Res* 2004;64(6):1966–1971.

[9] Stewart JB, Chinnery PF. Extreme heterogeneity of human mitochondrial DNA from organelles to populations. *Nat Rev Genet* 2021;22(2):106–118.

[10] Tyagi A, Pramanik R, Bakhshi R, Vishnubhatla S, Bakhshi S. Genetic landscape of mitochondrial regulatory region in pediatric acute myeloid leukemia: changes from diagnosis to relapse. *J Pediatr Genet* 2019;8(4):193–197.

[11] Wang D, Yang X, Cai D, et al. Genomic analysis of mutations in platelet mitochondria in a case of benzene-induced leukaemia: a case report. *Medicine (Baltim)* 2021;100(1):e24014.

[12] Fellous TG, McDonald SAC, Burkert J, et al. A methodological approach to tracing cell lineage in human epithelial tissues. *Stem Cells* 2009;27(6):1410–1420.

[13] Morris J, Na Y-J, Zhu H, et al. Pervasive within-mitochondrion single-nucleotide variant heteroplasmy as revealed by single-mitochondrion sequencing. *Cell Rep* 2017;21(10):2706–2713.

[14] Xu J, Nuno K, Litzenburger UM, et al. Single-cell lineage tracing by endogenous mutations enriched in transposase accessible mitochondrial DNA. *Elife* 2019;8:e45105.

[15] Zhang J, Zhang Z-X, Du P-C, et al. Analyses of the mitochondrial mutations in the Chinese patients with sporadic Creutzfeldt-Jakob disease. *Eur J Hum Genet* 2015;23(1):86–91.

[16] Kopinski PK, Singh LN, Zhang S, Lott MT, Wallace DC. Mitochondrial DNA variation and cancer. *Nat Rev Cancer* 2021;21(7):431–445.
[17] Yan C, Duanmu X, Zeng L, Liu B, Song Z. Mitochondrial DNA: distribution, mutations, and elimination. *Cells* 2019;8(4):379.
[18] Picard M. Blood mitochondrial DNA copy number: what are we counting? *Mitochondrion* 2021;60:1–11.
[19] Druzhyna NM, Wilson GL, LeDoux SP. Mitochondrial DNA repair in aging and disease. *Mech Ageing Dev* 2008;129(7-8):383–390.
[20] Grzybowska-Szatkowska L, Slaska B. Mitochondrial DNA and carcinogenesis (review). *Mol Med Rep* 2012;6(5):923–930.
[21] Mondet J, Lo Presti C, Chevalier S, et al. Mitochondria in human acute myeloid leukemia cell lines have ultrastructural alterations linked to deregulation of their respiratory profiles. *Exp Hematol* 2021;98:53–62.e3.
[22] Grandhi S, Bosworth C, Maddox W, et al. Heteroplasmic shifts in tumor mitochondrial genomes reveal tissue-specific signals of relaxed and positive selection. *Hum Mol Genet* 2017;26(15):2912–2922.
[23] He L, Luo L, Proctor SJ, et al. Somatic mitochondrial DNA mutations in adult-onset leukaemia. *Leukemia* 2003;17(12):2487–2491.
[24] Lièvre A, Blons H, Houllier AM, et al. Clinicopathological significance of mitochondrial D-Loop mutations in head and neck carcinoma. *Br J Cancer* 2006;94(5):692–697.
[25] Tan DJ, Chang J, Chen W-L, et al. Novel heteroplasmic frameshift and missense somatic mitochondrial DNA mutations in oral cancer of betel quid chewers. *Genes Chromosomes Cancer* 2003;37(2):186–194.
[26] Yu M, Shi Y, Zhang F, et al. Sequence variations of mitochondrial DNA D-loop region are highly frequent events in familial breast cancer. *J Biomed Sci* 2008;15(4):535–543.
[27] Cumbo C, Anelli L, Specchia G, Albano F. Monitoring of minimal residual disease (MRD) in chronic myeloid leukemia: recent advances. *Cancer Manag Res* 2020;12:3175–3189.
[28] Manfredi G, Fu J, Ojaimi J, et al. Rescue of a deficiency in ATP synthesis by transfer of MTATP6, a mitochondrial DNA-encoded gene, to the nucleus. *Nat Genet* 2002;30(4):394–399.
[29] Wu S, Fahmy N, Alachkar H. The mitochondrial transcription machinery genes are upregulated in acute myeloid leukemia and associated with poor clinical outcome. *Metabol Open* 2019;2:100009.
[30] Zhao C, Wang S, Wang K. Mutual exclusivity between the fusion gene CBFB::MYH11 and somatic mitochondrial mutations in acute myeloid leukaemia. *Br J Haematol* 2022;199(4):e25–e29.
[31] El Tekle G, Bernasocchi T, Unni AM, et al. Co-occurrence and mutual exclusivity: what cross-cancer mutation patterns can tell us. *Trends Cancer* 2021;7(9):823–836.
[32] Arnold RS, Fedewa SA, Goodman M, et al. Bone metastasis in prostate cancer: Recurring mitochondrial DNA mutation reveals selective pressure exerted by the bone microenvironment. *Bone* 2015;78:81–86.
[33] Chaudhary S, Ganguly S, Palanichamy JK, et al. PGC1A driven enhanced mitochondrial DNA copy number predicts outcome in pediatric acute myeloid leukemia. *Mitochondrion* 2021;58:246–254.
[34] Mondal R, Ghosh SK, Choudhury JH, et al. Mitochondrial DNA copy number and risk of oral cancer: a report from Northeast India. *PLoS One* 2013;8(3):e57771.
[35] Verma M, Naviaux RK, Tanaka M, Kumar D, Franceschi C, Singh KK. Meeting report: mitochondrial DNA and cancer epidemiology. *Cancer Res* 2007;67(2):437–439.
[36] Pereira-Martins DA, Coelho-Silva JL, Weinhäuser I, et al. Clinical significance of mitochondrial DNA content in acute promyelocytic leukaemia. *Br J Haematol* 2023;200(2):170–174.
[37] Yue L, Li Y-H, Ma R-L, et al. Circulating mitochondrial DNA is associated with anemia in newly diagnosed hematologic malignancies. *Leuk Lymphoma* 2023;64(1):178–187.
[38] Sriskanthadevan S, Jeyaraju DV, Chung TE, et al. AML cells have low spare reserve capacity in their respiratory chain that renders them susceptible to oxidative metabolic stress. *Blood* 2015;125(13):2120–2130.
[39] Guo X, Xu W, Zhang W, et al. High-frequency and functional mitochondrial DNA mutations at the single-cell level. *Proc Natl Acad Sci U S A* 2023;120(1):e2201518120.
[40] Lareau CA, Ludwig LS, Sankaran VG. Longitudinal assessment of clonal mosaicism in human hematopoiesis via mitochondrial mutation tracking. *Blood Adv* 2019;3(24):4161–4165.
[41] Miller TE, Lareau CA, Verga JA, et al. Mitochondrial variant enrichment from high-throughput single-cell RNA sequencing resolves clonal populations. *Nat Biotechnol* 2022;40(7):1030–1034.
[42] Lareau CA, Liu V, Muus C, et al. Mitochondrial single-cell ATAC-seq for high-throughput multi-omic detection of mitochondrial genotypes and chromatin accessibility. *Nat Protoc* 2023;18(5):1416–1440.
[43] Zeng W, Chen X, Duren Z, Wang Y, Jiang R, Wong WH. DC3 is a method for deconvolution and coupled clustering from bulk and single-cell genomics data. *Nat Commun* 2019;10(1):4613.
[44] Han S, Kim K, Park S, Lee AJ, Chun H, Jung I. scAVENGERS: a genotype-based deconvolution of individuals in multiplexed single-cell ATAC-seq data without reference genotypes. *NAR Genom Bioinform* 2022;4(4):lqac095.
[45] Liu Y. scDeconv: an R package to deconvolve bulk DNA methylation data with scRNA-seq data and paired bulk RNA-DNA methylation data. *Brief Bioinform* 2022;23(3):bbac150.
[46] Li H, Sharma A, Luo K, Qin ZS, Sun X, Liu H. DeconPeaker, a deconvolution model to identify cell types based on chromatin accessibility in ATAC-Seq data of mixture samples. *Front Genet* 2020;11:392.
[47] Berson E, Sreenivas A, Phongpreecha T, et al. Whole genome deconvolution unveils Alzheimer's resilient epigenetic signature. *Nat Commun* 2023;14(1):4947.
[48] Peng XL, Moffitt RA, Torphy RJ, Volmar KE, Yeh JJ. De novo compartment deconvolution and weight estimation of tumor samples using DECODER. *Nat Commun* 2019;10(1):4729.
[49] Aubin RG, Montelongo J, Hu R, Camara PG. Clustering-independent estimation of cell abundances in bulk tissues using single-cell RNA-seq data. *bioRxiv* 2023:2023.02.06.527318.
[50] Ludwig LS, Lareau CA, Ulirsch JC, et al. Lineage tracing in humans enabled by mitochondrial mutations and single-cell genomics. *Cell* 2019;176(6):1325–1339.e22.
[51] Penter L, Gohil SH, Wu CJ. Natural barcodes for longitudinal single cell tracking of leukemic and immune cell dynamics. *Front Immunol* 2021;12:788891.
[52] Lawless C, Greaves L, Reeve AK, Turnbull DM, Vincent AE. The rise and rise of mitochondrial DNA mutations. *Open Biol* 2020;10(5):200061.
[53] Mojtahedi M, Skupin A, Zhou J, et al. Cell fate decision as high-dimensional critical state transition. *PLoS Biol* 2016;14(12):e2000640.
[54] Wu J, Xiao Y, Sun J, et al. A single-cell survey of cellular hierarchy in acute myeloid leukemia. *J Hematol Oncol* 2020;13(1):128.
[55] Li C, Wang J. Quantifying the landscape for development and cancer from a core cancer stem cell circuit. *Cancer Res* 2015;75(13):2607–2618.
[56] Silkjaer T, Nørgaard JM, Aggerholm A, et al. Characterization and prognostic significance of mitochondrial DNA variations in acute myeloid leukemia. *Eur J Haematol* 2013;90(5):385–396.
[57] Hong YS, Battle SL, Shi W, et al. Deleterious heteroplasmic mitochondrial mutations are associated with an increased risk of overall and cancer-specific mortality. *Nat Commun* 2023;14(1):6113.
[58] Panina SB, Pei J, Kirienko NV. Mitochondrial metabolism as a target for acute myeloid leukemia treatment. *Cancer Metab* 2021;9(1):17.
[59] Wu S, Akhtari M, Alachkar H. Characterization of mutations in the mitochondrial encoded electron transport chain complexes in acute myeloid leukemia. *Sci Rep* 2018;8(1):13301.
[60] Damm F, Bunke T, Thol F, et al. Prognostic implications and molecular associations of NADH dehydrogenase subunit 4 (ND4) mutations in acute myeloid leukemia. *Leukemia* 2012;26(2):289–295.
[61] Sharawat SK, Bakhshi R, Vishnubhatla S, Bakhshi S. Mitochondrial D-loop variations in paediatric acute myeloid leukaemia: a potential prognostic marker. *Br J Haematol* 2010;149(3):391–398.
[62] Guo W, Liu Y, Ji X, et al. Mutational signature of mtDNA confers mechanistic insight into oxidative metabolism remodeling in colorectal cancer. *Theranostics* 2023;13(1):324–338.