Mitochondrial DNA Aberration of Hematopoietic Stem Cells: Implication and Clinical Application

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Somatically acquired mitochondrial DNA (mtDNA) mutations also have been linked to aging, degenerative diseases, cancer and autoimmunity. MtDNA mutations accumulate with age in postmitotic tissues such as muscle and brain but have been postulated to be diluted and lost in continually proliferating tissues such as bone marrow (BM). A large deletion of mtDNA is a hallmark of Pearson's syndrome, a constitutional disorder that includes sideroblastic anemia. MtDNA mutations recently were reported also in apparently acquired sideroblastic anemia and in myelodysplastic syndromes. While we were unable to confirm these results, we observed marked sequence variation in marrow mtDNA among different normal donors (Shin MG et al. Blood 2003;101:3118-25). We speculated that mtDNA mutations might arise and become homoplasmic in hematopoietic progenitor and stem cells during life, and mutations might then become fixed after clonal expansion of individual $CD34^+$ cells in certain hematologic diseases.

Unexpected and marked heterogeneity in the sequence of the mitochondrial DNA (mtDNA) control region of individual $CD34^+$ cells from normal adult bone marrow, not present in $CD34^+$ cells from umbilical cord blood, indicates that age-related homoplasmic resolution of mtDNA mutations occurs in an actively replicating tissue. This finding has important implications since the polymorphisms of this region are used for forensic identifications and anthropological research and are currently under investigation as tumor biomarkers. Furthermore mtDNA mutations may be useful in the study of hematopoietic stem cell biology and for the measurement of the mtDNA mutation rate in mammalian cells (Shin MG et al. Blood 2004;103:553-61). After then, we showed direct evidence of clonal expansion of cells containing mtDNA mutations and that the mtDNA sequence may be easily determined using peripheral blood (PB) as a CD34 cell source. Analysis of 594 circulating CD34 clones showed that 150 (25%) had mtDNA sequences different from the same donor's corresponding aggregate sequence. Examination of single granulocytes indicated that 103 (29%) from the same six individuals showed mtDNA heterogeneity, with sequences distinct from the corresponding aggregate tissue sequence and from the sequences of other single granulocytes. Circulating and BM CD34 cells showed virtually identical patterns of mtDNA heterogeneity, and the same changes were seen in progeny granulocytes as in their progenitors, indicating that blood sampling could be utilized in studies to determine whether mtDNA reflects an individual's cumulative or recent exposure to mutagens; as a marker of individual hematopoietic progenitors, stem cells and their expansion; and for the detection of minimal residual disease in hematological malignancies of CD34 cell origin (Shin MG et al. Blood 2004;103:4466-77).

The length heteroplasmies in the hypervariable regions (HV) of mitochondrial DNA (mtDNA) from blood cells were examined in 57 healthy Korean donors and 26 USA DNA samples. The Korean mtDNA HV exhibited 57 different haplotypes, originating from 77 distinct nucleotide differences. These length heteroplasmies were classified into 6 and 8 variant types, respectively. Interestingly, all the healthy Korean subjects displayed length heteroplasmies in both the HV1 and HV2 regions, a finding that directly contradicts the results of other studies on Korean subjects (*Electrophoresis* 2004;25:28-34). Closer examination of the HV2 length heteroplasmies indicated that most of these donors (84 %) exhibited a minimal 303-315 poly-C tract frame shift of one base pair (mixture of one major and minor mtDNA type). Sixteen percent of the donors, however, had poly-C tract frame shifts of two base pairs or more. The donor group with the greater numbers of length heteroplasmies had about a two-fold decrease in mtDNA copy number compared with the group exhibiting only a one bp frame shift. This result directly supports the possibility that a severe frame shift in the 303-315 poly-C tract may also cause the impairment of mtDNA replication in hematopoietic tissue.