# Mitochondrial DNA in human identification: a review

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## Introduction

Mitochondrial DNA (mtDNA) presents several characteristics useful for forensic studies:

- Lack of recombination
- High copy number
- Matrilineal inheritance

Human genetic identification for forensic purposes is achieved through the genetic profile or genetic fingerprint of an individual, which is the phenotypic description of a set of genomic loci that are specific to that individual.

### Mitochondrial DNA biology and genetics

Mitochondria are cellular organelles that contain an extrachromosomal genome, which is both different and separate from the nuclear genome, and consists of a five mm histone-free circular double-stranded DNA molecule, with around 16,569 base-pairs. mtDNA strands have different densities due to different base composition; the heavy (H) strand encodes more information than the light (L) strand. Another characteristics of mtDNA are the intronless genes and the limited intergenic sequences.

The apparent lack of mtDNA repair mechanisms and the low fidelity of the mtDNA polymerase lead to a significant higher mutation rate in the mitochondrial genome, compared to the nuclear genome (0.32x10^-6 /site/year vs 0.5x10^-9 /site/year).

Most of the sequence variation between individuals is found in two specific segments of the control region, namely in the hypervariable region 1 (HV1, pos 16,024-16,365) and in the hypervariable region 2 (HV2, pos 73-340), with additional polymorphic positions can be useful in the resolution of indistinguishable HV1/HV2 samples. The small size and relatively high inter-person variability of the HV regions are very ysefyl features for forensic testing purposes.

The mtDNA sequences defines the individual haplotype which is reported by the different base pairs relative to the rCRS mtDNA sequence (reference). The collection of similar haplotypes defined by the combination of SNPs in mtDNA inherited from a common ancestor defines an haplogroup which was formed as a result of the sequential accumulation of mutation through maternal lineage (mitchell et al., 2015).

A mitochondrion contains 2 to 10 copies of mtDNA and each somatic cell can have up to 1,000 mitochondria.

Contrarily to nuclear DNA, mtDNA is exclusively maternally inherited, which justifies the fact that, besides mutation, mtDNA sequence of siblings and all maternal relatives is identical.

**Heteroplasmy**. A person is considered as heteroplasmic if she/he carries more than one detectable mtDNA type. Heteroplasmy related to point substitutions is important for forensic human identification and it manifests itself in diverse ways (Stewart et al., 2001). An individual may show more than one mtDNA type in a single tissue. An individual may be heteroplasmic in one tissue sample and homoplasmic in another one. Finally, an individual may exhibit one mtDNA type in one tissue and a different type in another tissue. Of the 3 possible scenarios, the last one is the least likely to occur. When heteroplasmy is found in the mtDNA of an individual, it usually differs at a single base, in HV1 or HV2.

Attempting to improve the power of mtDNA in human identification, over the past decade some studies have been focused in the extension of the analysis to the whole mtDNA genome (Duan et al., 2019; Strobl et al., 2018; Woerner et al., 2018).

In the context of forensic analysis, both mtDNA sequences of a reference sample sand an evidence sample(s) are compared. When the sequences are unequivocally different, the conclusion is that they can be excluded as being originated from the same source. If the mtDNA sequences are identical, the samples must have the same origin or derive from the same maternal lineage. Similarly, samples can't be excluded when heteroplasmy is observed at the same nucleotide positions in both samples. When one sample is heteroplasmic and the other is honomplasmic but they both share at least one mtDNA species, the samples may have the same origin. Several authors have suggested that samples with mtDNA with one-base difference should be further evaluated, mainly regarding their rate of mutation.