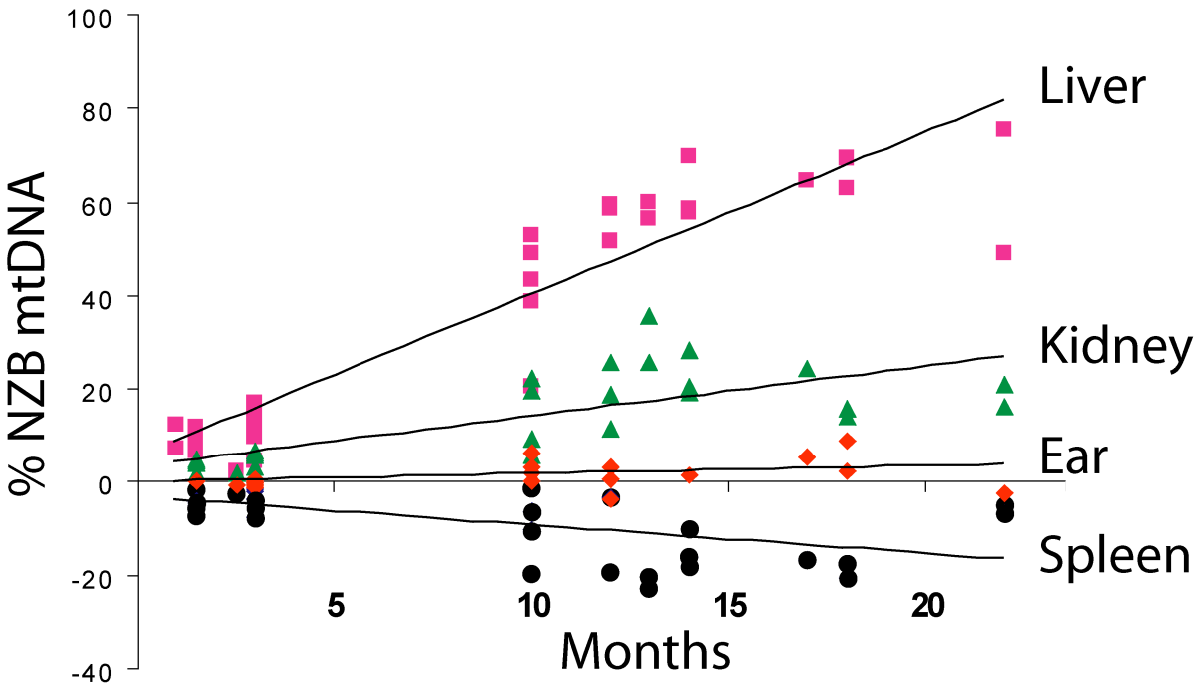

Figure S1. NZB mitochondrial DNA segregates over time in certain tissues. Ear DNA samples were obtained from heteroplasmic mice [BALB/cByJ (BALB) and NZB/BINJ (NZB) at the time of genotyping (3-4 weeks). DNA samples from liver, kidney and spleen were also obtained from different animals sacrificed at different time points over a 22 months period and tested for the levels of NZB by RFLP-PCR as described in methods. NZB levels in different tissues were normalized by subtracting the NZB mtDNA levels measured in the ear DNA.

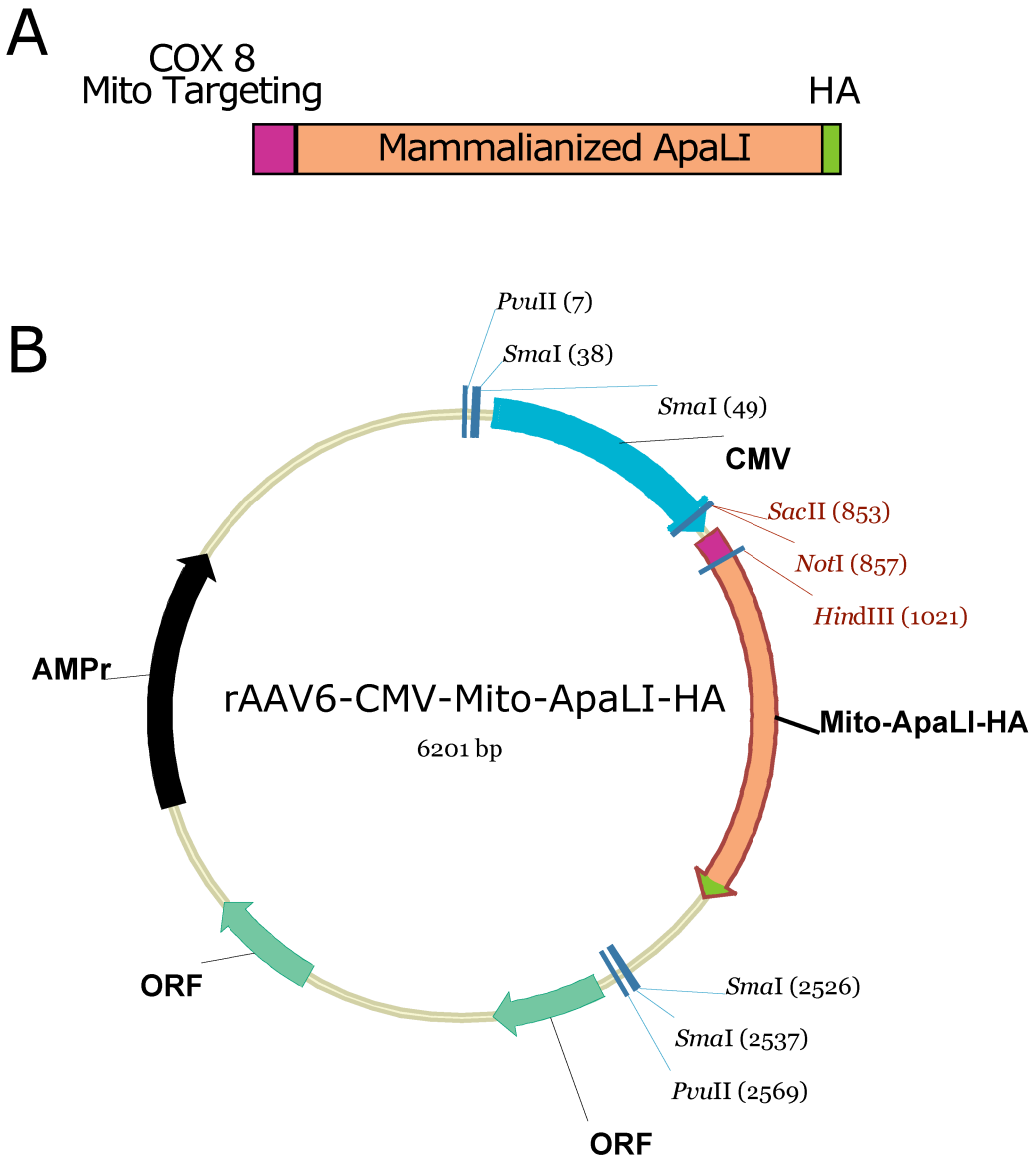

Figure S2. Construction and characterization of a rAAV6[Mito-ApaLI-HA]. The genetic code of a synthetic ApaLI gene was optimized for mammalian translation and a HA-tag coding sequence added to the 3'-end. The gene was cloned downstream of the cytochrome c oxidase subunit 8 (COX 8) mitochondrial targeting sequence. The recombinant rAAV6[mito-ApaLI-HA] contained a mitochondrial targeting sequence (cytochrome c oxidase subunit 8 (Cox8)), a gene coding for a mammalianized form of ApaLI restriction endonuclease synthesized in vitro and an HA tag for immunological detection. The construct was cloned in a recombinant adeno-associated serotype 6 backbone, under the control of cytomegalovirus promoter (CMV).

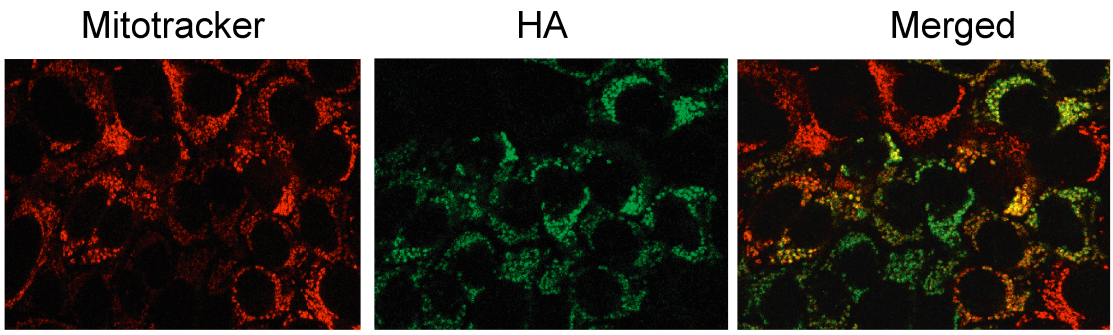

Figure S3. Expression of the recombinant [mito-ApaLI-HA] in cells. To test for the capacity of the rAAV6[mito-ApaLI-HA] to infect cells and localize to mitochondria with Mito Tracker Red CMXRos, hepatocytes derived from heteroplasmic mice were transduced with rAAV6[mito-ApaLI-HA] (5x10e4 vg/cell) for 7 days. Using an anti-HA antibody, we detect colocalization of the transgene in mitochondria of the cells that express the recombinant restriction endonuclease, but not the ones infected with the control (not shown).

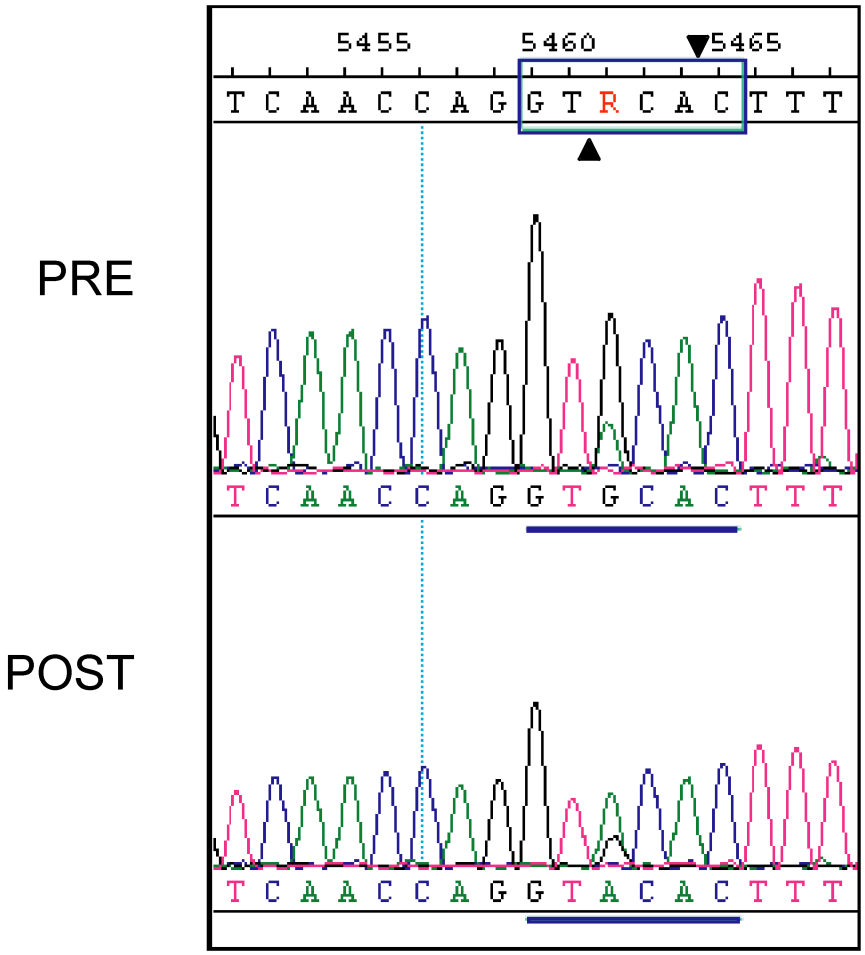

Figure S4. Electropherograms of the ApaLI restriction site region. Heteroplasmic mtDNA sequence is shown and pre- and post-injection reads with consensus base-calling below. The ApaLI restriction site [GTGCA`C] on BALB mtDNA (Pre-injection) is marked. The only altered peaks visible post-injection correspond to NZB mtDNA and no novel peaks corresponding to ApaLI site mutations are present.