Mitochondrial Function-Associated *APOE* Locus and its Implication in Alzheimer’s Disease and Aging

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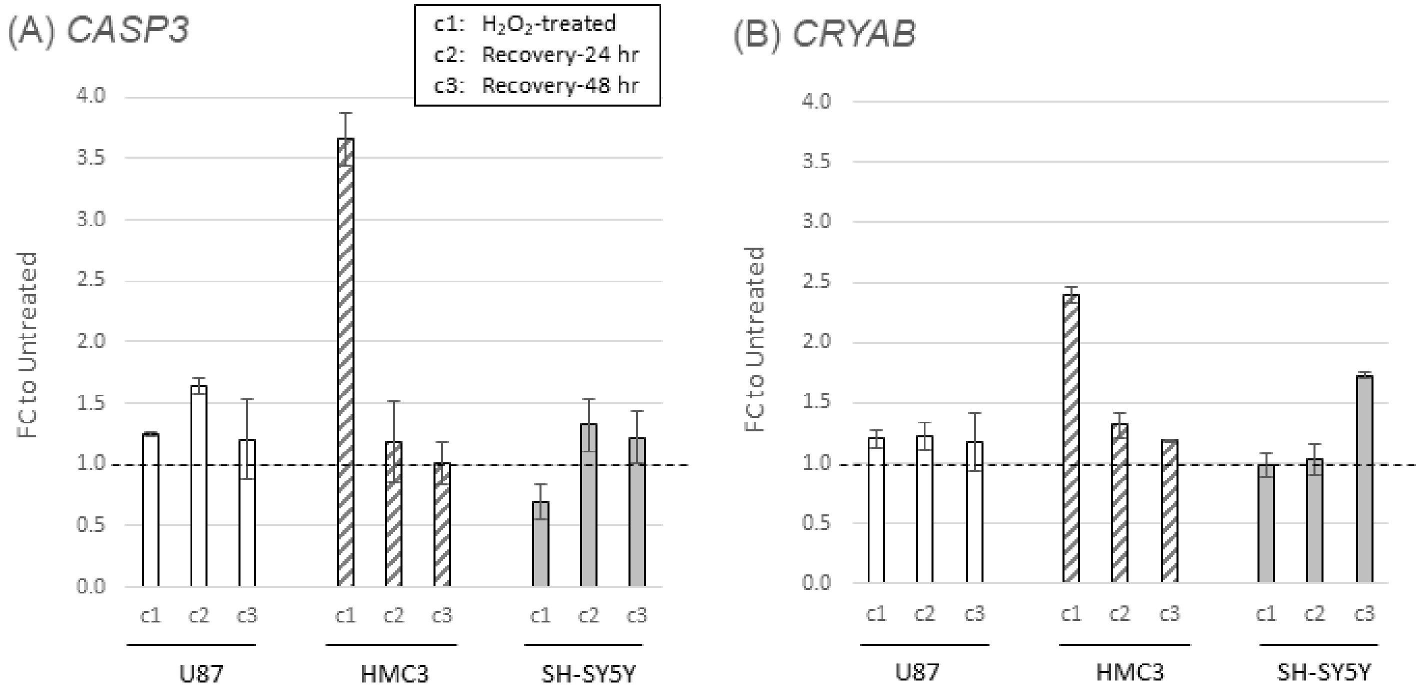
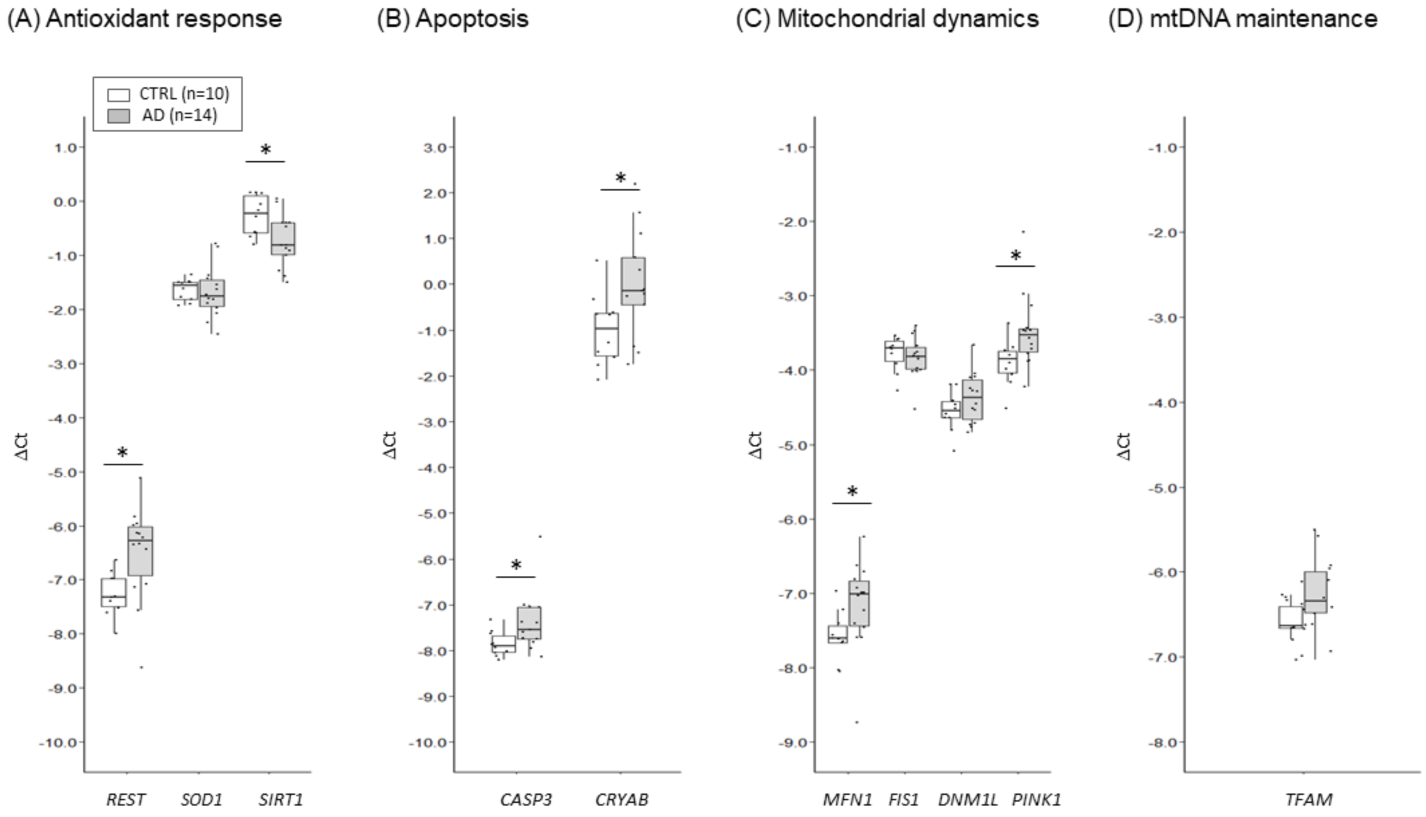
Table S1. Primers, probes, and TaqMan assays.

Method	Target	Primer	Sequence 5'-	Amplicon (bp)	Comment
SYBR-based PCR					
	MT-ND1	MT-ND1_F	CCTACTCCTCATTGTACCCATTC	93	Forward
		MT-ND1_R	GGGCCTTTGCGTAGTTGTA		Reverse
	NCOA3	NCOA3_F	GCCATACATTTAATTGCCGTATGT	94	Forward
		NCOA3_R	GTTTCATATCTCTGGCGCATTTC		Reverse
	REST	REST_F	GCGTACTCATTCAGGTGAGAAG	113	Forward
		REST_R	GAGGTTTAGGCCCATTGTGAA		Reverse
	SOD1	SOD1_F	GTGCAGGGCATCATCAATTTC	85	Forward
		SOD1_R	GGCCTTCAGTCAGTCCTTTAAT		Reverse
	SIRT1	SIRT1_F	GTAGGCGGCTTGATGGTAAT	101	Forward
		SIRT1_R	GGATAAGACGTCATCTTCAGAGTC		Reverse
	CASP3	CASP3_F	ATGGATTATCCTGAGATGGGTTTAT	89	Forward
		CASP3_R	TCTGTACCAGACCGAGATGT		Reverse
	CRYAB	CRYAB_F	TGGGAGATGTGATTGAGGTG	99	Forward
		CRYAB_R	CAGCTGGGATCCGGTATTT		Reverse
	MFN1	MFN1_F	AACCTGGCAGCTGAAGATAAA	83	Forward
		MFN1_R	TGGTTTCGAATAAAGTCCAGTCT		Reverse
	FIS1	FIS1_F	TGCGGAGCAAGTACAATGATG	97	Forward
		FIS1_R	AAGACGTAATCCCGCTGTTC		Reverse
	DNM1L	DNM1L_F	AGCTCCAGGACGTCTTCAAC	79	Forward
		DNM1L_R	CTGCTCTGCGTTCCCACTAC		Reverse
	PINK1	PINK1_F	TCGGCCTGTCAGGAGATC	78	Forward
		PINK1_R	CAAGCGTCTCGTGTCCAAC		Reverse
	TFAM	TFAM_F	AAAGACCTCGTTCAGCTTATAAC	94	Forward
		TFAM_R	TCCTTTACAGTCTTCAGCTTTTC		Reverse
	ACTB	ACTB_F	GGAATCCTTCTGACCCATGCC	70	Forward
		ACTB_R	CGTCTTCCCCTCCATCGTGG		Reverse
Digital PCR					
	TOMM40	TOMM40_F2	TCAGCACAATCGGGGAGTC	112	Forward
		TOMM40_R2	CCACTGTTGTCCATGTCACCC		Reverse
		TOMM40 probe	ACAAAGCAGCTGAGTCCCACAGAG		Internal probe
	ACTB	ACTB_F	TCGCCCACATAGGAATCCTTC	81	Forward
		ACTB_R	CGTCTTCCCCTCCATCGTGG		Reverse
		ACTB probe	TGACCCATGCCCACCATCAC		Internal probe
***TOMM40* Allelic Expression**					
	TOMM40	TOMM40_F1-Bio	Bio-AGATGGAGGGTGTCAAG	141	Forward
		TOMM40_R1	TGGGACTCAGCTGCTTTG		Reverse
TaqMan Assay					
	APOE RNA		ThermoFisher (Hs00171168_m1)		
	APOC1 RNA		ThermoFisher (Hs00155790_m1)		
	NECTIN2 RNA		ThermoFisher (Hs01071562_m1)		
	TOMM40 RNA		ThermoFisher (Hs01587378_mH)		
	ACTB RNA		ThermoFisher (Hs01060665_g1)		

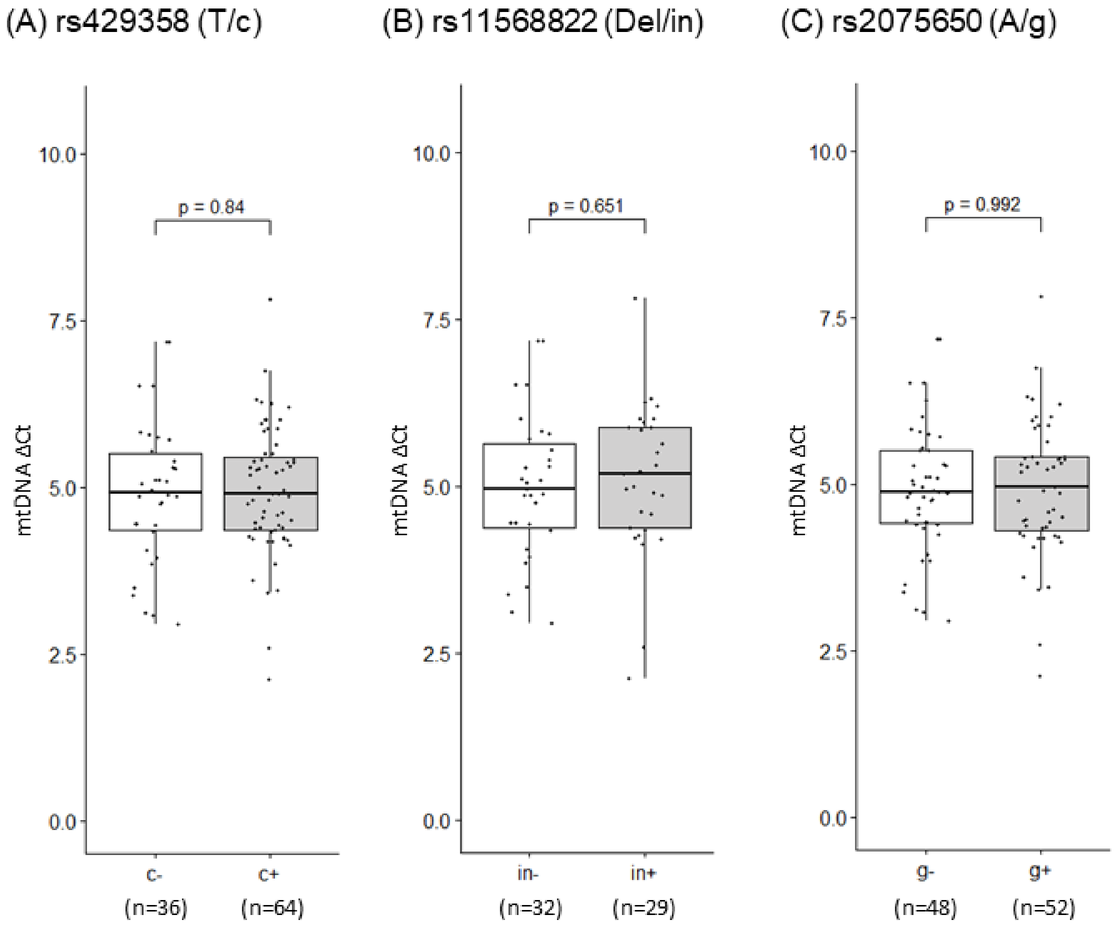
Supplementary Figure 1. Alterations of apoptosis-related gene expression during the H_2O_2 recovery phase. RNA levels of two genes, *CASP3* (A) and *CRYAB* (B) were quantified in experiment condition cells by RT-qPCR (SYBR-based). Fold changes (FC) to their untreated counterparts (set as baseline of 1.0) are plotted as average and standard deviation of three to four independent experiments. The first culture condition, c1, represents cells treated with H_2O_2 for 24 hr; c2, cells in H_2O_2 recovery phase after replenishing culture with fresh media and continuing to culture for additional 24 hr; c3, cells in H_2O_2 recovery phase for culturing additional 48 hr.



Supplementary Figure 2. Variations of mitochondrial structure and function-related gene expression in human PMB tissues. Using a subgroup of PMB samples [AD (n=14) and control (CTRL, n=10)] RNA levels were quantified by RT-qPCR (SYBR-based). (A) Antioxidant response genes, (B) Apoptosis genes, (C) Mitochondrial dynamics genes, and (D) a mtDNA maintenance gene. The ΔCt method was used, in which a larger ΔCt value indicates a higher RNA level. RNA levels of AD (grey filled) were compared with those of control (unfilled) by independent samples t-test using R-Program; *, $p<0.05$.



Supplementary Figure 3. Variations of mitochondrial DNA (mtDNA) copy numbers in human PMB tissues. Using PMB AD and control (CTRL) samples mtDNA copy numbers were quantified by RT-qPCR (SYBR-based). mtDNA copy numbers are stratified by alleles of SNPs: *APOE* rs429358 (A), *APOC1* rs11568822 (B) and *TOMM40* rs2075650 (C). The normalized ΔCt shows that a larger ΔCt value indicates a greater number of mtDNA. Independent samples t-test p-values are indicated. Numbers in parentheses denote sample sizes.



Supplementary Figure 4. Three-dimensional genome structure of the *APOE* locus. The UCSC Genome Browser's Hi-C/Micro-C tracks of the *APOE* locus. (A) a display of the entire region of *APOE* locus with a black triangle that is (B) a zoomed-in core region with blue triangles showing regions of strong interaction. This track provides heatmaps of chromatin folding data from in situ Hi-C and Micro-C XL experiments on the H1-hESC (embryonic stem cells) and HFFc6 (foreskin fibroblasts) cell lines. The data indicate how many interactions were detected between regions of the genome. A high score between two regions suggests that they are in close proximity in 3D space within the nucleus, which is shown by a more intense color in the heatmap. The color shade at any point within the triangle shows the proximity score for two genomic regions.