

File S6. Potential off-target loci for gRNAs used in this study. Potential off-target sites were found using Cas-OFFinder (<http://www.rgenome.net/cas-offinder/>), with following selections: SpCas9 and 5'-NRG-4' for the PAM type, 2 for mismatch number, and Homo sapiens (hg19) for the target genome. Lowercase letters denote mismatched residues.

Target gene	gRNA-PAM sequence	Found sequence	Mismatches	Chromosome	Position (hg19)	Locus	On/Off-target
<i>GJB2</i>	CCTCCTTTGCAGCC ACAACGNRG	CCTCCTTTGCAGCC ACAACGAGG	0	chr13	20763593	<i>GJB2</i> , exon	On-target
<i>GJB2</i>	CCTCCTTTGCAGCC ACAACGNRG	CCTCCTTTGCtGCCA CAAgGTGG	2	chr6	41659304	<i>TFEB</i> , intron	Off-target
<i>GJB2</i>	CCTCCTTTGCAGCC ACAACGNRG	CCTCCTTTGCAGCC ACAAtCtTGG	2	chr5	149818830	Intergenic (between <i>CD74</i> and <i>RPS14</i>)	Off-target
<i>GJB5</i>	GGGCGCCTCTACCT GAACCCNNG	GGGCGCCTCTACCT GAACCCCGG	0	chr1	35223270	<i>GJB5</i> , exon	On-target
<i>GJB5</i>	GGGCGCCTCTACCT GAACCCNNG	GGaCGCCTCTtCCTG AACCCCTGG	2	chr6	106346458	Intergenic (between <i>PREP</i> and <i>PRDM1</i>)	Off-target
<i>GJA1</i>	TTTTCTCCGTGGGG CGAGAGNRG	TTTTCTCCGTGGGG CGAGAGAGG	0	chr6	121768589	<i>GJA1</i> , exon	On-target
<i>GJA1</i>	TTTTCTCCGTGGGG CGAGAGNRG	TTTTCTCCaTGGGG CaAGAGAGG	2	chr5	108387542	<i>FER</i> , intron	Off-target
<i>GJC1</i>	ATAGCCCAGGTACA TCACAGNRG	ATAGCCCAGGTACA TCACAGAGG	0	chr17	42882903	<i>GJC1</i> , exon	On-target