Supplemental Material, M109.082990

Plasmids and site-directed mutagenesis –

mVDAC1 (obtained from W.J. Craigen, University of Houston) was cloned into plasmid pEGFP-N1 (Clontech) for construction of mVDAC1-GFP or pcDNA4/TO for constructing tetracycline regulated expression. Plasmid pEGFP-N1, carrying the wild type mVDAC1 gene, served as the template for amplification of mutant mVDAC1 genes. Site-directed mutagenesis of mVDAC1 was carried out *in vitro* by overlapping PCR amplification using the following primers: for E65Q, 5'-TGGACTCAGTATGG**G**CTGACG-3' (forward) and GCC**C**ATACTGAGTCCATCTG (reverse), for E72Q, GACGTTTACA**C**AGAAGTGGAAC (forward) and GTTCCACTTCT**G**TGTAAACGTC (reverse), For E202Q, GAAGTTG**C**AGACTGCTGTCAATCTC (forward) and GCGAGATTGACAGCAGTCT**G**CAAC (reverse), where the exchanged nucleotide is in bold.

Native or mutated mVDAC1 coding sequences were cloned into the *Bam*H1 and *Eco*RV restriction sites of the pcDNA4/TO vector (Invitrogen) containing the zeocin resistance gene and two tetracycline operator sites within the human cytomegalovirus (CMV) immediate-early promoter to allow for tetracycline-regulated expression of mVDAC1 in transfected cells.

Plasmid pEGFP-Bcl2 was obtained from R.J. Youle (NIH) (NIH). DNA encoding Bcl2α (obtained from E. Gross, Weizmann Institute of Science) was cloned into the *NcoI* and *EcoRI* restriction sites of the pHisparallel vector for construction of plasmid Bcl2(Δ23) using following primers: (forward) CACCATGGCGCACG CTGGGAGAACG and (reverse) CGGAATTCTTACAGAGACAGCCAGGAGAAATC.