SUPPLEMENTARY DATA

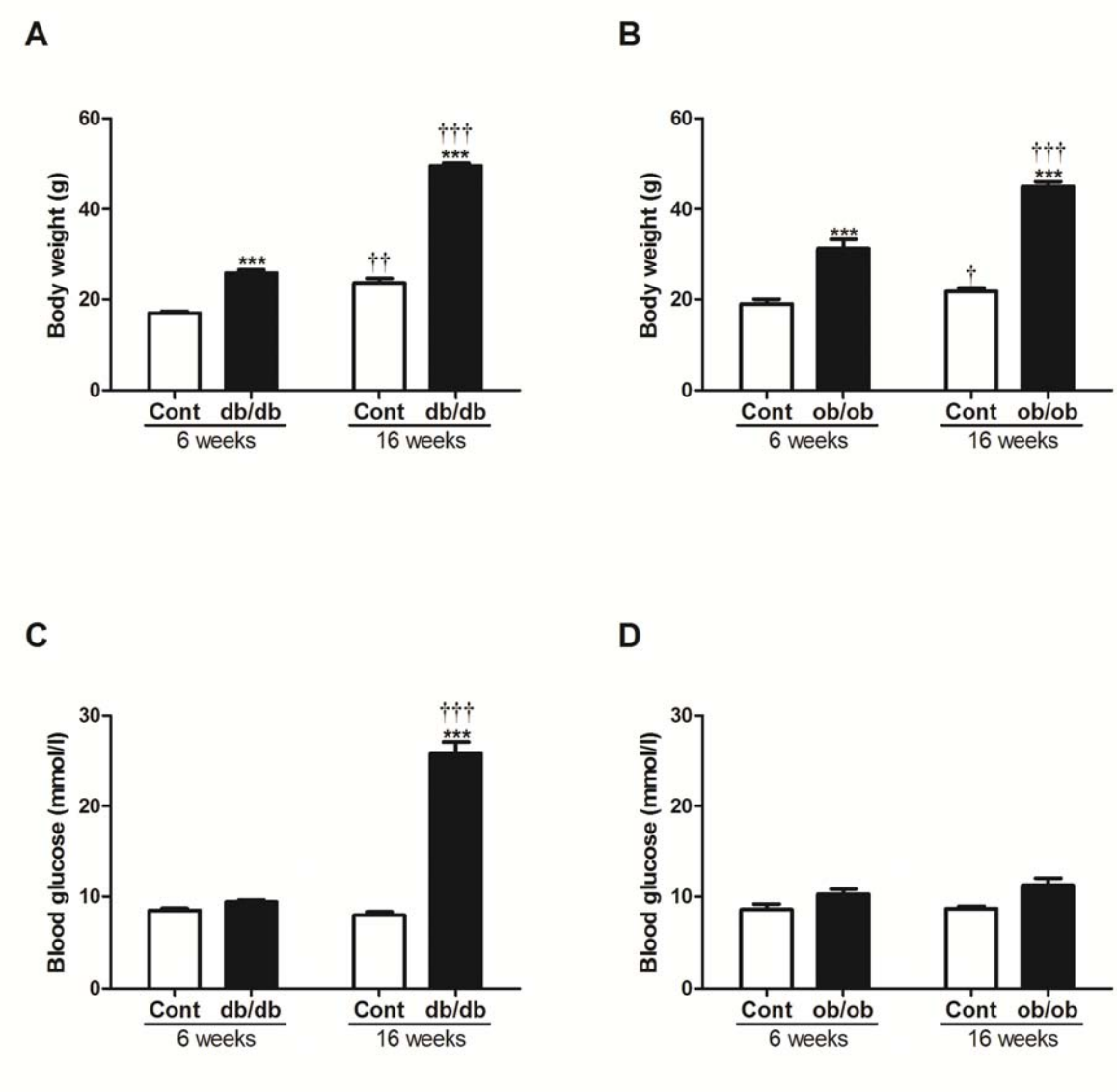
Supplementary Table 1. Sequences of oligonucleotide primers.

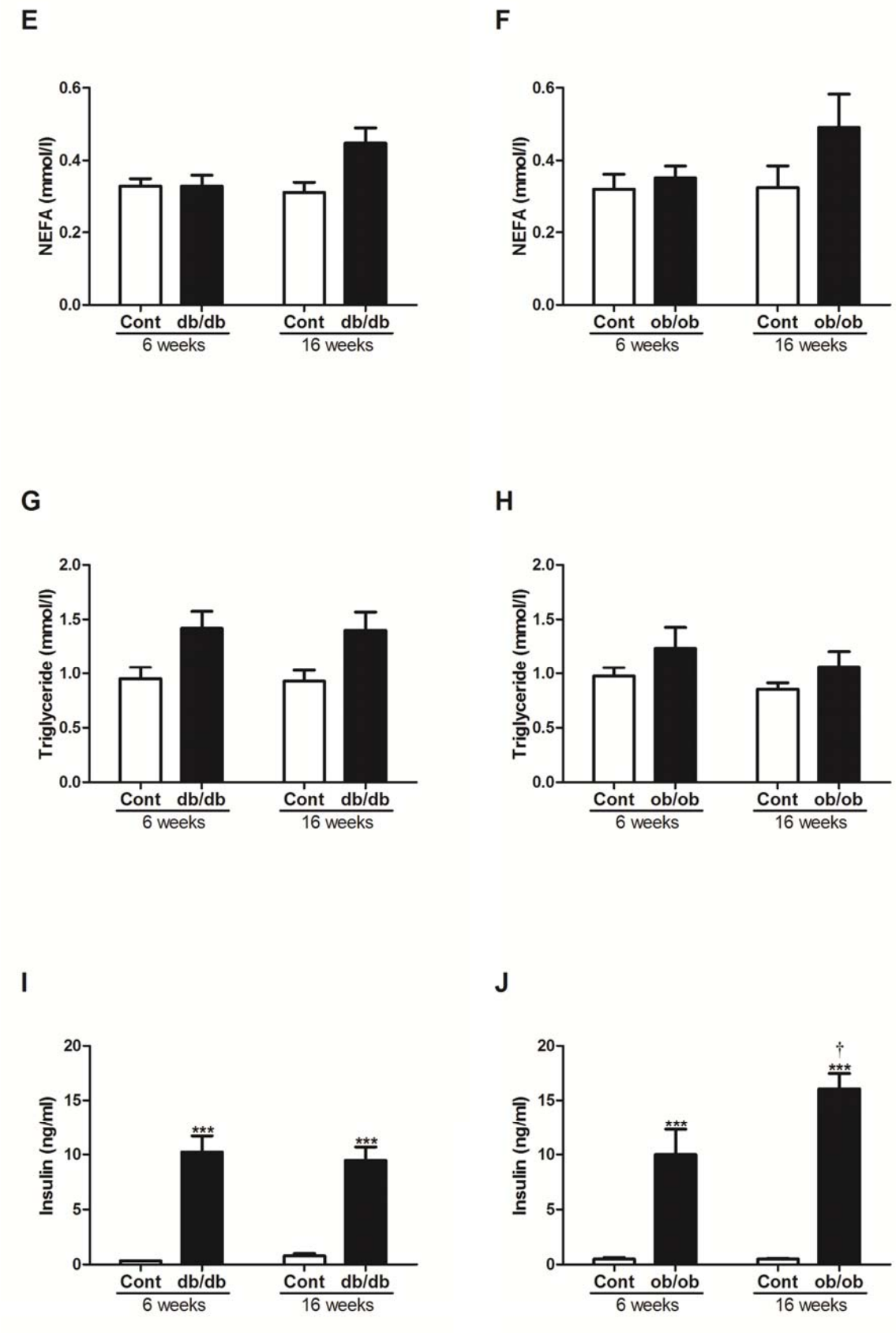
Gene symbol	5' Oligonucleotide	3' Oligonucleotide
Atf3	GCTGCCAAGTGTCGAAACAAG	CAGTTTTCCAATGGCTTCAGG
Atf4	ATCCAGCAAAGCCCCACAAC	CAAGCCATCATCCATAGCCG
Bak1	CGCTACGACACAGAGTTCCA	GGTAGACGTACAGGGCCAGA
Bax	TGCAGAGGATGATTGCTGAC	GATCAGCTCGGGCACTTTAG
Beta2 (Neurod1)	ACTCCAAGACCCAGAAACTGTC	ACTGGTAGGAGTAGGGATGCAC
Bip (Hspa5, Grp78)	AGGACAAGAAGGAGGATGTGGG	ACCGAAGGGTCATTCCAAGTG
Catalase	ATGAAGCAGTGGAAGGAGCAGC	CTGTCAAAGTGTGCCATCTCGTC
Ccl2 (Mcp-1)	CCACTCACCTGCTGCTACTCATTC	TCTGGACCCATTCCTTCTTGG
Chop (Ddit3; Gadd153)	TTCACTACTCTTGACCCTGCGTC	CACTGACCACTCTGTTTCCGTTTC
Cxcl1 (Gro-1)	CAAACCGAAGTCATAGCCACACTC	TTGTCAGAAGCCAGCGTTCAC
Cyclophilin A	TGTGCCAGGGTGGTGACTTTAC	TGGGAACCGTTTGTGTTTGG
Erp72 (Pdia4)	AGTCAAGGTGGTGGTGGGAAAG	TGGGAGCAAAATAGATGGTAGGG
Fkbp11	ACACGCTCCACATACACTACACGG	ATGACTGCTCTTCGCTTCTCTCCC
Gipr	GCGTGCTCTACTGCTTCATCAAC	AACTTTCCAAGACCTCATCCCC
Gk (Gck, MODY2)	CATTGAATCAGAGGAGGGCAGC	TAGTGGACTGGGAGCATTTGTGGG
Glp1r	GGGTCTCTGGCTACATAAGGACAAC	AAGGATGGCTGAAGCGATGAC
Glut2 (Slc2a2)	CATTCTTTGGTGGGTGGC	CCTGAGTGTGTTTGGAGCG
Gpr40 (Ffar1)	TATTCCTGGGGTGTGTGTGTGG	CCAAGGGCAGAAAGAAGAGCAG
GPx (GPx-1, CGPx)	ACAGTCCACCGTGTATGCCTTC	CTCTTCATTCTTGCCATTCTCCTG
Grp94 (Hsp90b1)	AAACGGCAACACTTCGGTCAG	GCATCCATCTCTTCTCCCTCATC
HO-1 (Hmox1, Hsp32)	CCACACAGCACTATGTAAAGCGTC	GTTCGGGAAGGTAAAAAAAGCC
Id1	TTGGTCTGTCGGAGCAAAGC	GCAGGTCCCTGATGTAGTCGATTAC
IL-1β	TGTTCTTTGAAGTTGACGGACCC	CCACAGCCACAATGAGTGATACTG
IL-6	CAAGAGACTTCCATCCAGTTGCC	CATTTCCACGATTTCCCAGAGAAC
Kir6.2 (Kcnj11)	TCGTGTCCAAGAAAGGCAACTG	GGAAGGCAGATGAAAAGGAGTGG
MafA	CGGGAACGGTGATTGCTTAG	GGAGGTTGGGACGCAGAA
mGPDH (Gpd2)	AAAGACTGGAGCCCCACACTCTAC	ATCCCGTATTTCACCTCTGCTTC
Nkx6.1	GGACCAGAGAGAGCACGC	TTCGGGTCCAGAGGTTTG
p58 (Dnajc3, Prkri)	AAGCCCGTGGAAGCCATTAG	GGTCATTTTCATTGTGCTCCTGAG
PC (Pcx)	GTTCCGTGTCCGAGGTGTAAAG	CGCAGAAGGATGTCCCTGAAAC
Pdx1 (Mody4)	CGGACATCTCCCCATACG	AAAGGGAGCTGGACGCGG
Sod1 (Cu/ZnSOD)	ATGGGGACAATACACAAGGCTG	CAATGATGGAATGCTCTCCTGAG
TNF-α	CCCCTTTACTCTGACCCCTTTATTG	AACCTGACCACTCTCCCTTTGC
Trib3	TCTTCAGCAACTGTGAGAGGACG	TCCAGACATCAGCCGCTTTG
Xbp1	AAACAGAGTAGCAGCGCAGACTGC	GGATCTCTAAAACTAGAGGCTTGGTG

Aliases of gene symbols given in parentheses

SUPPLEMENTARY DATA

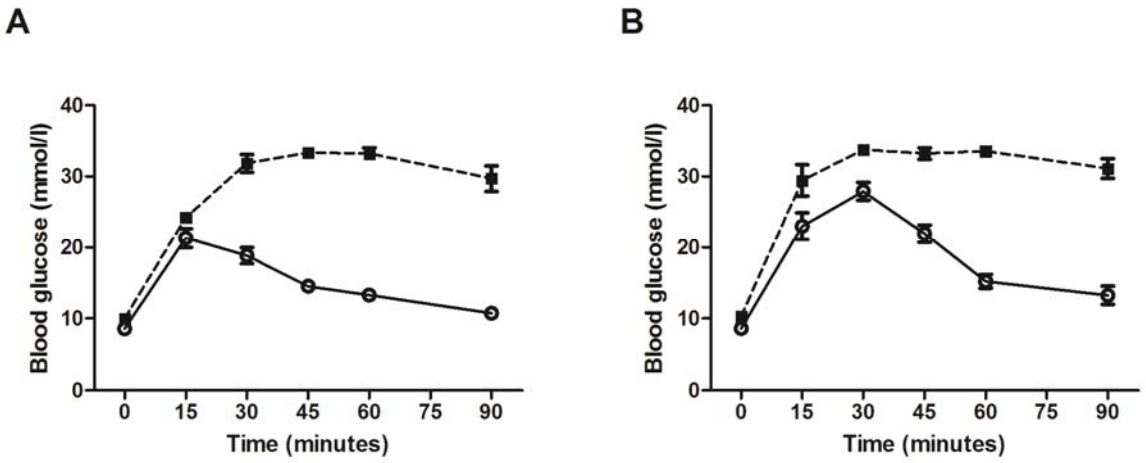
Supplementary Figure 1. Characteristics of *db/db* and *ob/ob* mice at 6 and 16 weeks of age. Body weight (A, B), blood glucose (C, D), and plasma NEFA (E, F), triglyceride (G, H) and insulin (I, J) levels for C57BL/KsJ control (n = 8-22) and *db/db* (n = 10-14) mice at 6 weeks of age, C57BL/KsJ control (n = 9-12) and *db/db* (n = 5-10) mice at 16 weeks of age, C57BL/6J control (n = 6) and *ob/ob* (n = 6) mice at 6 weeks of age, and C57BL/6J control (n = 6) and *ob/ob* (n = 7) mice at 16 weeks of age. All results shown are mean±SEM; ***p<0.001 genotype effect in each age group; †p<0.05, †††p<0.001 age effect in each genotype.





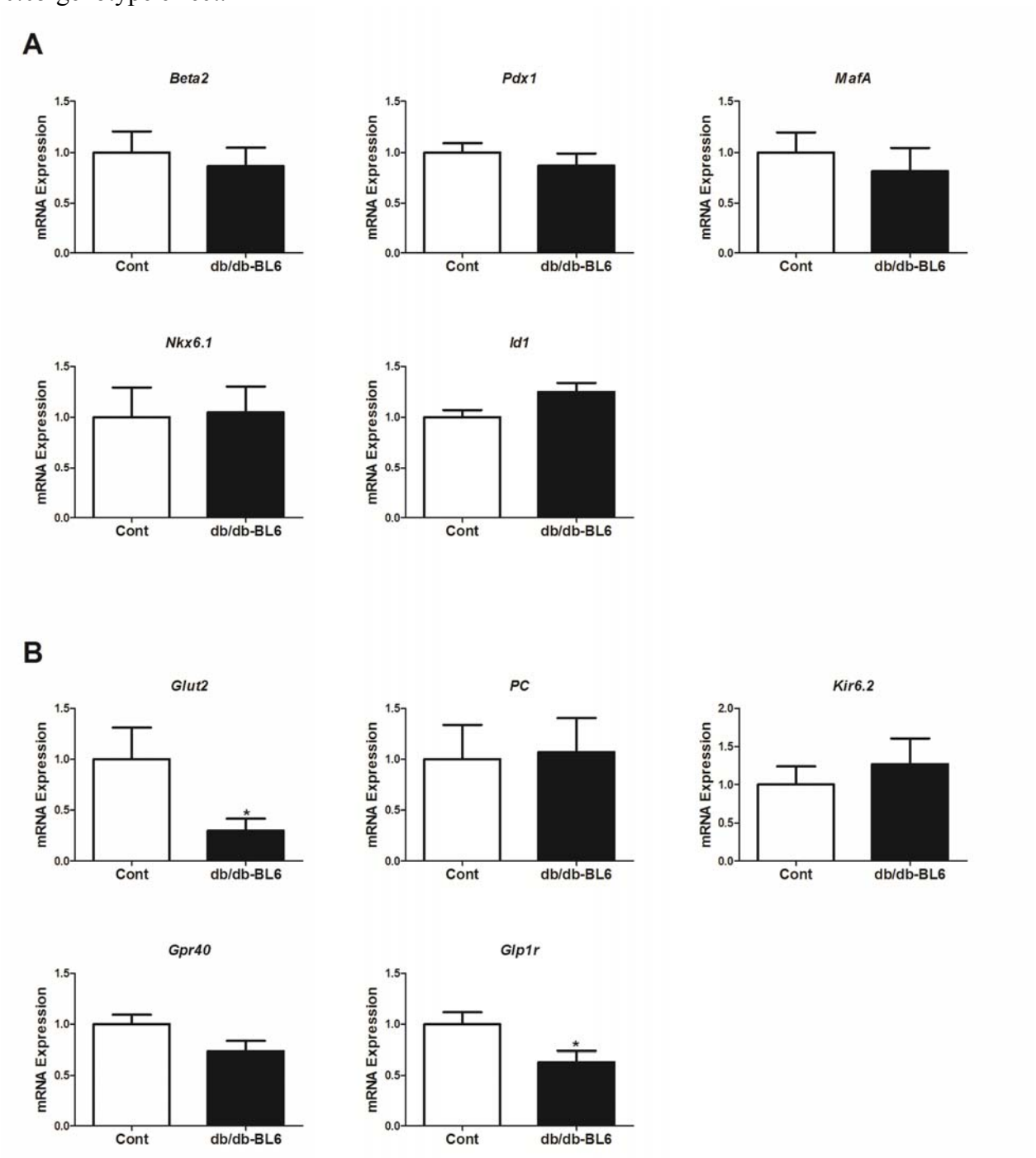
SUPPLEMENTARY DATA

Supplementary Figure 2. Impaired glucose tolerance in *db/db* (A) and *ob/ob* (B) mice at 6 weeks of age. Blood glucose levels during an intraperitoneal glucose tolerance test. A: C57BL/KsJ control (n = 6, *empty circle, solid line*) and *db/db* (n = 6, *filled squares/dashed line*) mice. ANOVA: $p<0.001$ genotype effect. B: C57BL/6J control (n = 6, *empty circle, solid line*) and *ob/ob* (n = 6, *filled squares/dashed line*) mice. ANOVA: $p<0.001$ genotype effect. All results shown are mean±SEM.



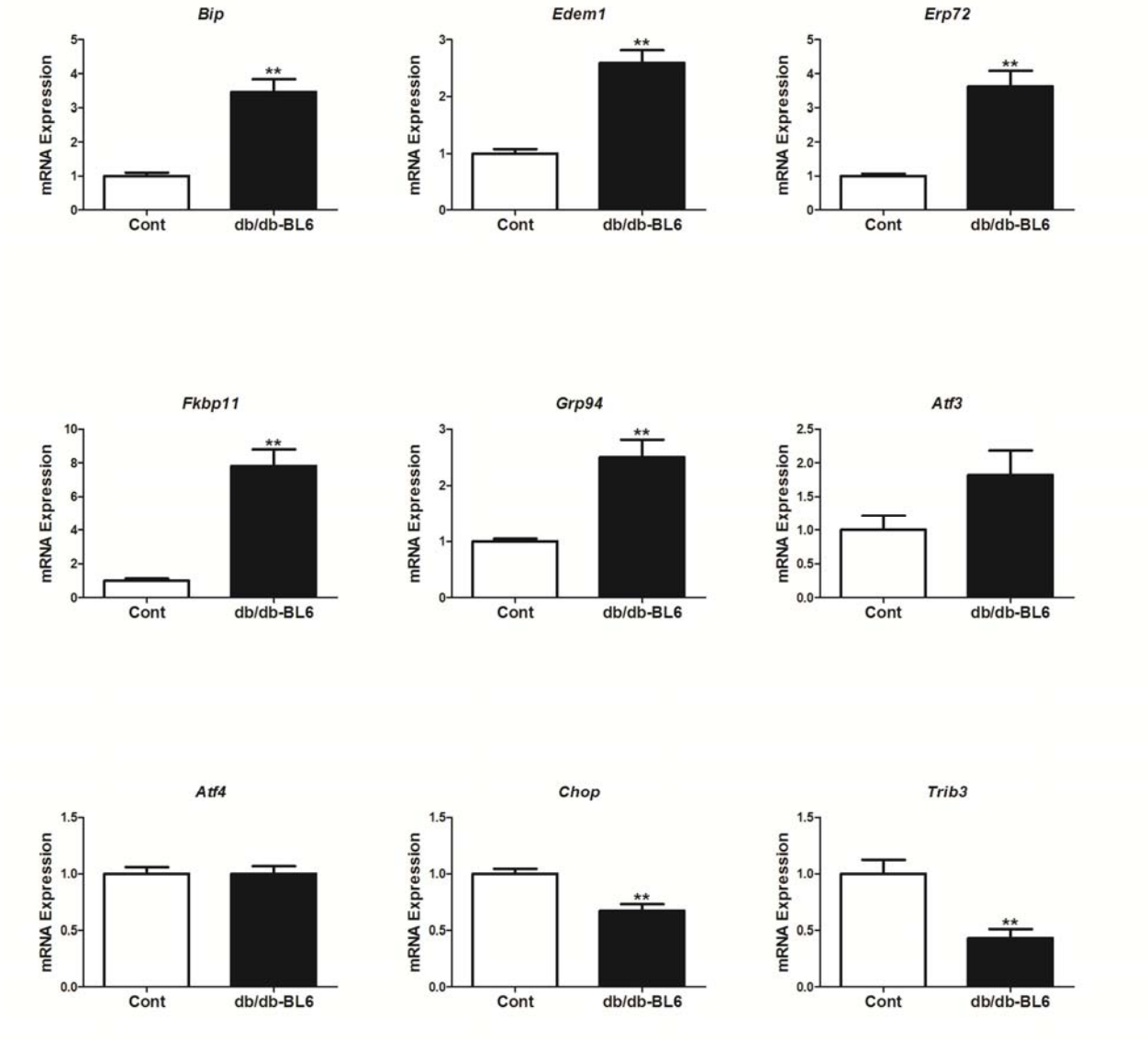
SUPPLEMENTARY DATA

Supplementary Figure 3. Changes in mRNA expression of islet-associated transcription factors (A) and genes that optimize β-cell function (B) in islets of *db/db* mice on the C57BL/6J background. Islets were isolated from C57BL/6J control and *db/db* mice (n = 6 in each group) at 6 weeks of age. Total RNA was extracted, reverse-transcribed and analyzed by real-time RT-PCR. mRNA levels were expressed as fold-change of the levels in respective age-matched controls. All results are mean±SEM; **p*<0.05 genotype effect.



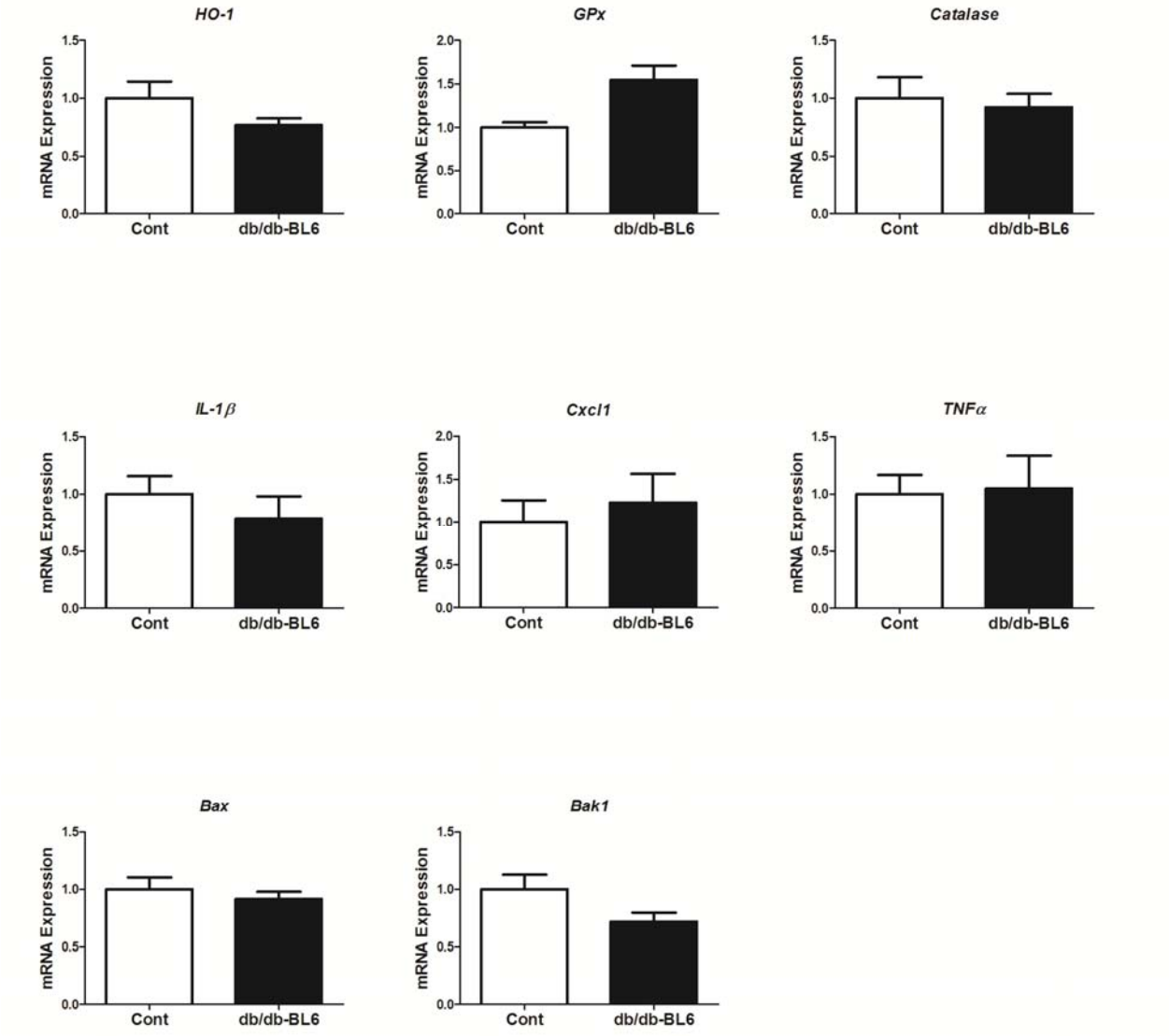
SUPPLEMENTARY DATA

Supplementary Figure 4. Changes in mRNA expression of UPR genes in islets of *db/db* mice on the C57BL/6J background. Islets were isolated from C57BL/6J control and *db/db* mice (n = 6 in each group) at 6 weeks of age. Total RNA was extracted, reverse-transcribed and analyzed by real-time RT-PCR. mRNA levels were expressed as fold-change of the levels in respective age-matched controls. All results are mean±SEM; ***p*<0.01 genotype effect.



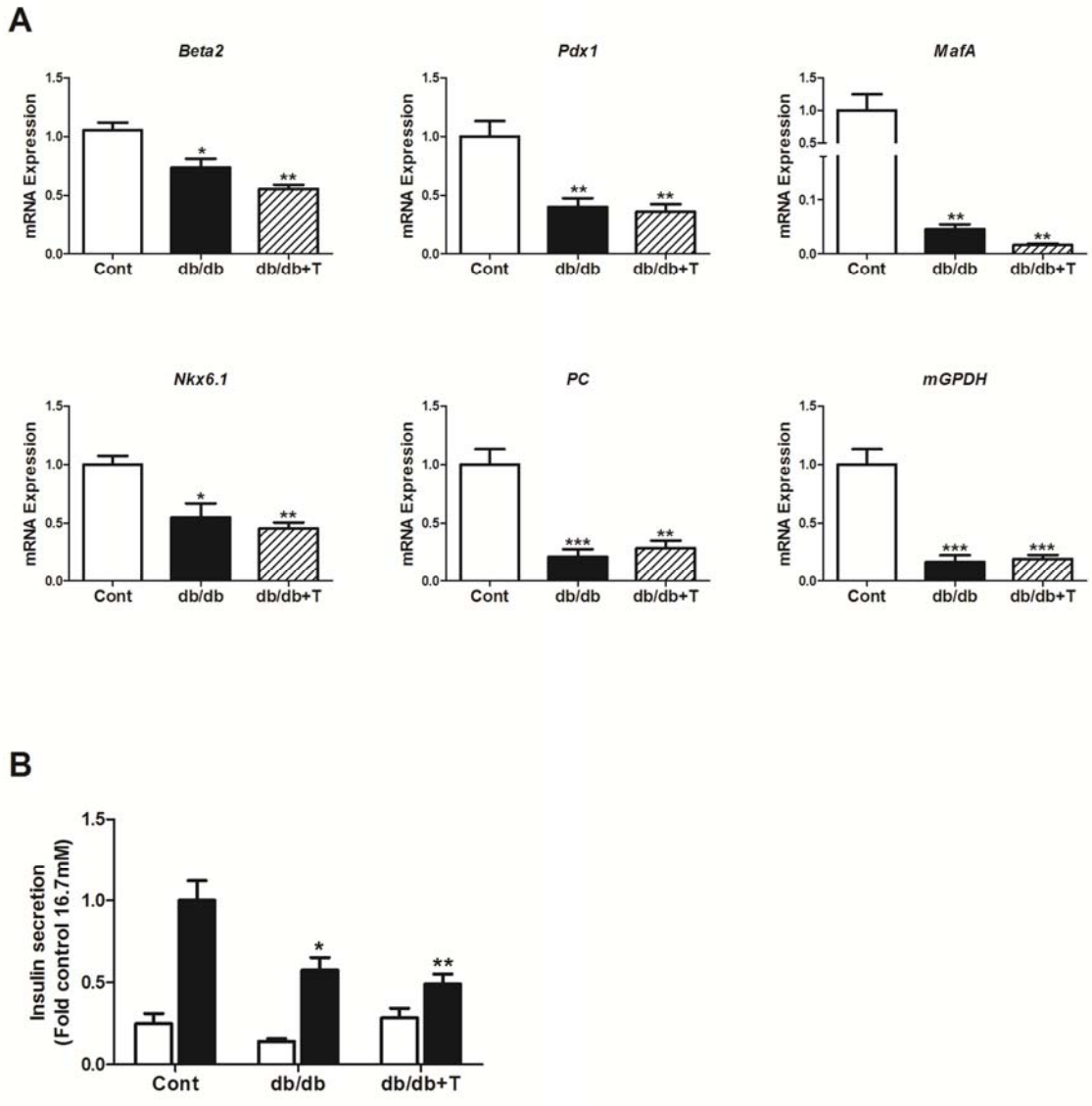
SUPPLEMENTARY DATA

Supplementary Figure 5. Changes in mRNA expression of anti-oxidant and inflammation genes in islets of *db/db* mice on the C57BL/6J background. Islets were isolated from C57BL/6J control and *db/db* mice (n = 6 in each group) at 6 weeks of age. Total RNA was extracted, reverse-transcribed and analyzed by real-time RT-PCR. mRNA levels were expressed as fold-change of the levels in respective age-matched controls. All results are mean±SEM.



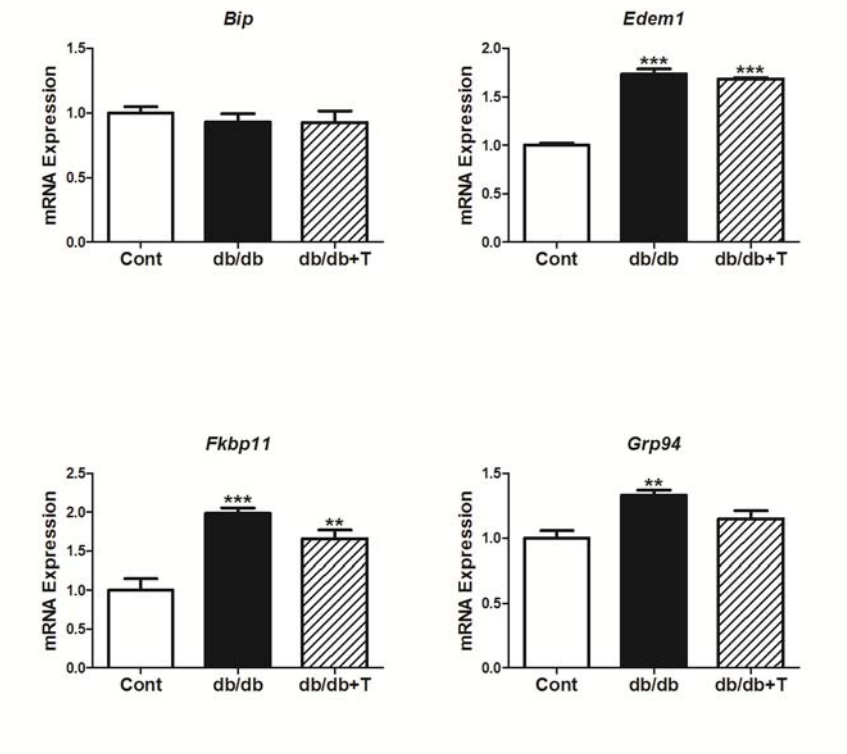
SUPPLEMENTARY DATA

Supplementary Figure 6. Effect of TMAO treatment on the changes in gene expression and insulin secretion in islets of *db/db* mice. A: Islets isolated from diabetic *db/db* and age-matched non-diabetic control mice (12-14 weeks of age) were cultured in the absence (control, *white bars*; and *db/db*, *black bars*) or presence (*db/db*, *striped bars*) of TMAO (100 mmol/l) for 24 h. Total RNA was extracted, reverse-transcribed and analyzed by real-time RT-PCR. mRNA levels were expressed as fold-change of the level in control islets. n = 4 in each group. B: Batches of islets were cultured in Krebs-Ringer HEPES buffer containing 0.1% BSA and 2.8 mmol/l (*white bars*) or 16.7 mmol/l glucose (*black bars*) for 1 h. Insulin was measured in an aliquot of the buffer by radioimmunoassay. Insulin secretion was expressed as fold change of the level in control islets cultured in 16.7 mmol/l glucose. n = 4 in each group. All results are mean±SEM; **p*<0.05, ***p*<0.01, ****p*<0.001 compared to control.



SUPPLEMENTARY DATA

Supplementary Figure 7. Effect of TMAO on the expression of UPR genes in islets of *db/db* mice. Islets isolated from diabetic *db/db* and age-matched non-diabetic control mice were cultured in the absence (control, *white bars*; and *db/db*, *black bars*) or presence (*db/db*, *striped bars*) of TMAO (100 mmol/l) for 24 h. Total RNA was extracted, reverse-transcribed and analyzed by real-time RT-PCR. mRNA levels were expressed as fold-change of the level in control islets. All results are mean±SEM, n = 4 in each group; **p<0.01, ***p<0.001 compared to control.



SUPPLEMENTARY DATA

Supplementary Figure 8. Effect of chemical chaperone treatment on the changes in anti-oxidant or inflammatory stress gene expression in islets of *db/db* mice. Islets isolated from diabetic *db/db* and age-matched non-diabetic control mice (12-14 weeks of age) were cultured in the absence (control, *white bars*; and *db/db*, *black bars*) or presence (*db/db*, *striped bars*) of the chemical chaperone PBA (2.5 mmol/l) for 24 h. Total RNA was extracted, reverse-transcribed and analyzed by real-time RT-PCR. mRNA levels were expressed as fold-change of the level in control islets. All results are mean±SEM, n = 7 in each group; *p<0.05, **p<0.01 genotype effect.