SUPPLEMENTARY INFORMATION

Prolyl-4-hydroxylase 3 maintains β-cell glucose metabolism during fatty acid excess in mice

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Supplementary figure 1. ACACB and ACACA gene regulation in human islets. (A) ACACB is expressed in human islets and purified β-cells but levels are lower than ACACA (B) The ACACB promoter is regulated by multiple β-cell transcription factors, with the presence of an antisense-transcribed long non-coding RNA. All data are previously described (1-4), and publicly available via EMBL-EBI and www.isletregulome.com. Data visualization was performed using the open source University of California Santa Cruz (UCSC) Genome Browser (5).
Supplementary figure 2. PHD3 knockout does not induce a hypoxic gene expression
or Ca\(^{2+}\) signaling phenotype. (A) *Egln3* expression is highly upregulated in wild-type islets
following exposure of islets to hypoxic (1% O\(_2\)) conditions for 24 hrs (n = 4-5 replicates; 9-11
animals, unpaired t-test). (B-D) Expression of the HIF1α-target genes *Bnip* (B), *Car9* (C) and
*Gls* (D), is similar or decreased in βPHD3KO versus βPHD3CON islets exposed to normoxia
(21% O\(_2\)) or hypoxia (1% O\(_2\)) for 24 hrs (n = 4 animals, Kruskal-Wallis test, Dunn’s multiple
comparison test). (E and F) Glucose- (E) and KCl- (F) stimulated Ca\(^{2+}\) fluxes are not
significantly different in βPHD3KO versus βPHD3CON islets exposed to normoxia or
hypoxia (n = 10-27 islets, 2 animals/genotype, two-way ANOVA; Sidak’s multiple
comparison test). (G and H) Mean Ca\(^{2+}\) traces from βPHD3CON and βPHD3KO islets
exposed to (G) normoxia or (H) hypoxia. (I-K) Expression of the HIF1α-target genes *Gls* (I),
*Bnip* (J) and *Car9* (K) is unchanged or decreased in 4 weeks HFD βPHD3KO islets (n = 3-4
animals/genotype, unpaired t-test). (L-M) Expression of the HIF2α-targets (L) *Ccnd1* and (M)
*Dll4* is unchanged or downregulated, respectively, in 8 weeks HFD βPHD3KO islets (n = 3
animals/genotype, unpaired t-test). Bar graphs (scatter plot) and line graphs show mean ±
SEM. *P<0.05, **P<0.01 and NS, non-significant. PHD3, prolyl-hydroxylase 3.
### Supplementary Table 1. Primer sequences used for qPCR.

| Gene name | Forward sequence (5' – 3') | Reverse sequence (5' - 3') |
|-----------|-----------------------------|---------------------------|
| Ppia      | AAGACTGAGTGGTTGGATGG        | ATGGTGATCTTCTTGCTGGT     |
| Actb      | CGAGTCCGCTCCACCC            | CATCCATGGCGAACTGTTGG     |
| Egln3 (Exon 2) | GCTTGCTATCCAGGAATGG | GCGTCCAATTCTTATCAG      |
| Egln3 (Exon 1) | GGCTGGGGATAATCTATGCAAA   | GGTTGTCCACATGGCGAACA     |
| Egln1     | TAAACGGGCGAAGCGAAG        | GGGTTATCAACGTGACGGCA     |
| Egln2     | CATCAATGGGCACCA            | GATTGTCAACAGCTCAGTAC     |
| BNip3     | CTGGGTAGAACTGCACTTCAG      | CTGGGTAGAACTGCACTTCAG    |
| Car9      | GGAGCTACTTCGTCCAGATTCAAT  | CCGGAACTGAGCTCATCAAC     |
| Glis      | TCCGCCCCTGGAGATCTTAC      | CCAAGCTAGTAAAGACCCCT     |
| Ldha      | TTCCAGCAGGTTGGGTTA        | CCGGCCACATTGACACCAC      |
| Cpt1a     | CTCCGCGCTGAAGCAGTAGAAG    | CACCAGTATGAGCTCATTCT     |
| Acaca     | TTTCCTGACAAGCAGTAGTCTG    | CTGCCGAAACACATCTCTGGGA   |
| Acacb     | CCTTTGGCAACAAGCAGGAAGTA   | AGTCGTACACATAGTGTTGTC    |
| Pdx1      | CCAAAAGCTCAGCGGTTGGA      | TGTTTTCTCGGGTTTCCG      |
| Nkx6-1    | GGCTGTACCCCATCAAG         | GTGGGTCTGTTGGTTTCTTCTT  |
| Mafa      | TCCAGCAAGGAGGGAGTCTAC     | CTTAGCCGGTTTTCTTCT      |
| Ddit3     | CTGGAGCACTGTGATGAGGAT    | CAGGGTAAAGATGTAAGGTTAG  |
| Xbp1      | AGCAGCAAAGTGCTGGATTG      | GAGTTTTCTCCGTAAGAGCTGA  |
| Hspa5     | ACTTGGCGACCACCTATTTCCT   | GTTGCCCTGATCGTTGGCTA     |
| Ccnd1     | GCGTACCCCTGACACCAATTC    | CCTCTTTCGACCTTTGTGCTC   |
| Dll4      | TTCAGCGCAACCTTTCTCCGA    | ACTGCGGCTATTCTTGTC      |
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