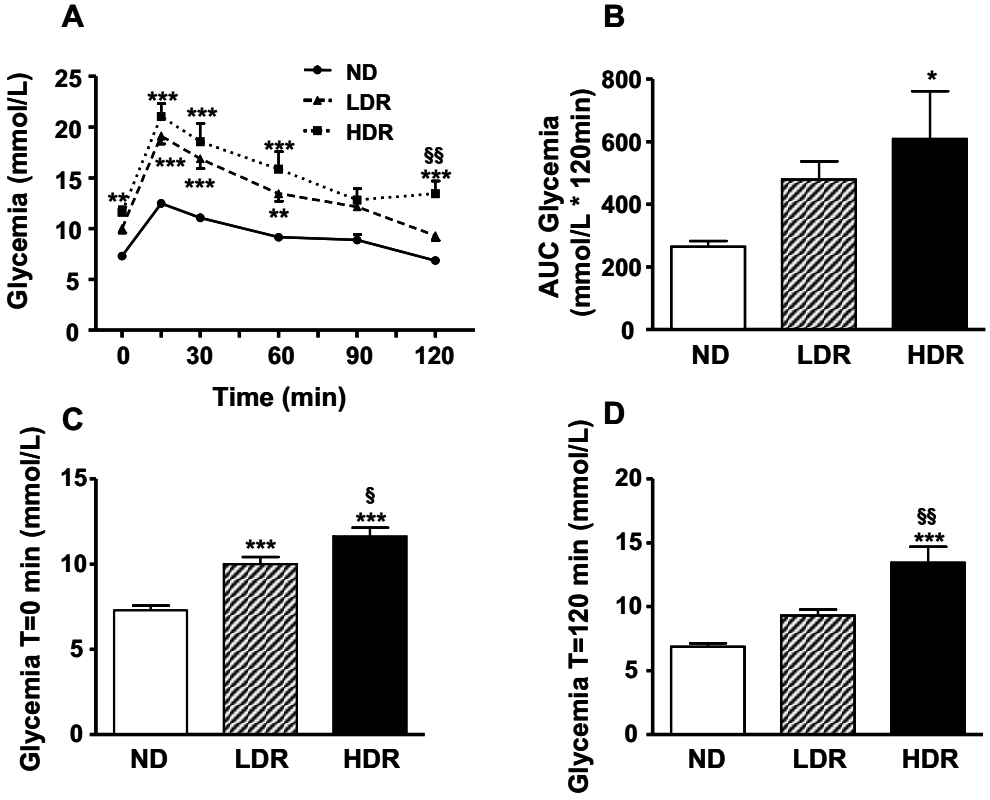
Supplemental research design and methods

Quantitative RT-PCR. Total RNA was extracted from islets as previously described (1). Gene expression was determined by a standard curve method and normalized to the expression of β-actin. Real-time PCR analysis was performed on Rotor-Gene R3000 (Corbett Research, Mortlake, NSW, Australia) using Quantitech Sybrgreen (Qiagen, Mississauga, ON, Canada). Primers, listed in supplementary Table 1, were designed using Primer3 software. Results are expressed as the ratio of target mRNA to β-actin mRNA.

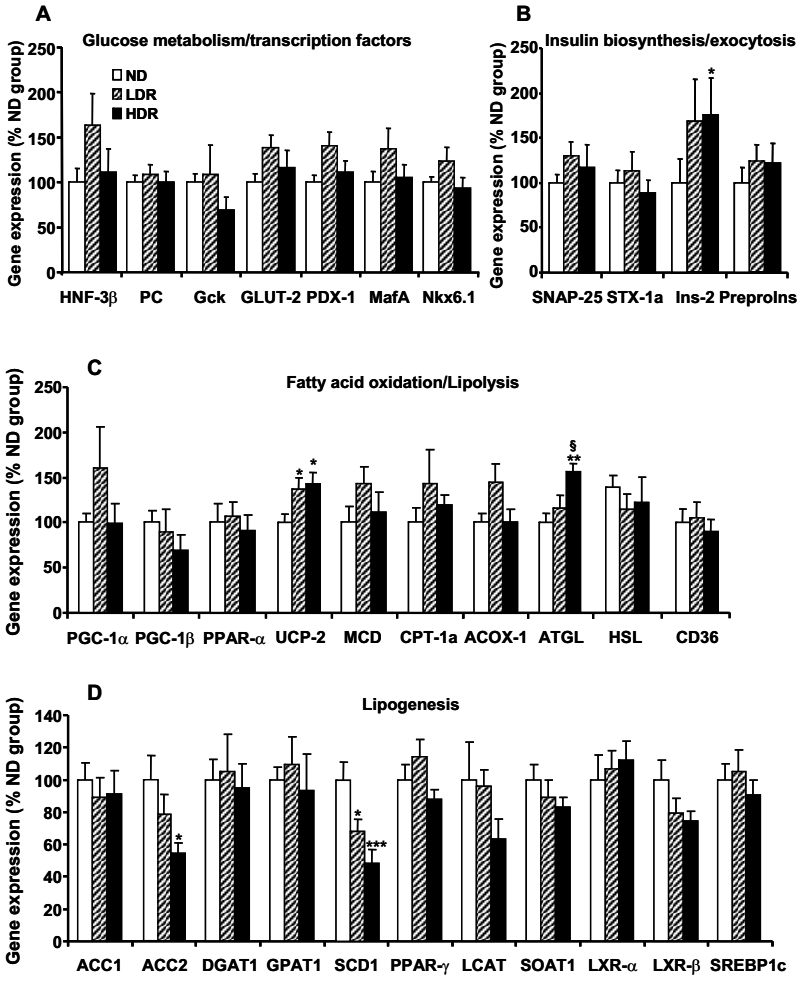
Supplemental reference

1. Delghingaro-Augusto V, Nolan CJ, Gupta D, Jetton TL, Latour MG, Peshavaria M, Madiraju SR, Joly E, Peyot ML, Prentki M, Leahy J: Islet beta cell failure in the 60% pancreatectomised obese hyperlipidaemic Zucker fatty rat: severe dysfunction with altered glycerolipid metabolism without steatosis or a falling beta cell mass. *Diabetologia* 52:1122-1132, 2009

Suppl. Fig.1. Oral glucose tolerance in ND, LDR and HDR mice. (*A-D*) Oral glucose tolerance test (OGTT). Blood glucose (*A*) was measured at times 0, 15, 30, 60, 90 and 120 min in response to an oral glucose administration (1g/kg) in 6h-fasted mice. (*B*) Area under the curves (AUC) of the 0-120 min glycemic responses. (*C-D*) Blood glucose levels at time 0 (*C*) and a time 120 min (*D*) of the OGTT. Means ± SE of 13-14 animals per group. **P*<0.05, ***P*<0.01, ****P*<0.001 ND *versus* LDR or HDR; §*P*<0.05, §§*P*<0.01 LDR *versus* HDR; one-way (*B-D*) or two-way ANOVA (*A*), with Bonferroni post hoc test.



Suppl. Fig.2. mRNA expression of selected genes related to glucose and lipid metabolism. Gene expression was measured in islets from ND, LDR and HDR mice using real-time RT-PCR. Genes were grouped by function. (*A*) Glucose metabolism and transcription factors: forkhead box A2 (Foxa2/HNF-3β), pyruvate carboxykase (PC), glucokinase (Gck), glucose transporter 2 (GLUT-2), pancreas-duodenum homeobox-1 (PDX-1), v-maf musculoaponeurotic fibrosarcoma oncogene homolog A (Maf-A) and NK6 homeobox-1 (Nkx6.1). (*B*) Insulin biosynthesis and exocytosis: preproinsulin-2 (preproins), insulin-2 (Ins-2), synaptosomal-associated protein 25 (SNAP-25) and syntaxin-1a (STX-1a). (*C)* Fatty acid oxidation and lipolysis: peroxisome proliferator-activated receptor-α (PPAR-α), PPAR-γ-coactivator-1α and -1β (PGC-1α and -1β), uncoupling protein-2 (UCP-2), malonyl-CoA-decarboxylase (MCD), carnitine palmitoyltransferase-1a (CPT-1a) and acyl-CoA oxydase-1 (ACOX-1), adipose triglyceride lipase (ATGL) and hormone-sensitive lipase (HSL). (*D*) lipogenesis: acetyl-CoA carboxylase-1 and -2 (ACC-1 and ACC-2), diacylglycerol-O-acyltransferase-1 (DGAT-1), glycerol-3-phosphate acyltransferase-1 (GPAT-1), stearoyl-CoA deasturase-1 (SCD-1), fatty acid transporter (FAT/CD36), lecithin-cholesterol acyltransferase (LCAT), sterol-O-acyltransferase (SOAT-1), liver-X-receptor-α and -β (LXR-α and -β), sterol regulatory element binding protein-1c (SREBP-1c) and PPAR-γ. Results are expressed as percent of the ND group values and were normalized to actin. Means ± SE of 9-18 animals per group. LDR or HDR *versus* ND: **P*<0.05, ***P*<0.01, ****P*<0.001; LDR *versus* HDR: §*P*<0.05; one-way ANOVA- Bonferroni's multiple comparison test.



Supplemental Table 1. PCR primer sequences used for Quantitative Real Time PCR. Primers: S, sense; AS, antisense.

GENE	GenBank accession no	Primer sequences (5'-3')
ACC-1	NM_133360	S: ATGTCCGCACTGACTGTAACC AS: TCCATAGCCGACTTCCATAGC
ACC-2	NM_133904	S: GAAGCCTCCTTCGAGTACCTG AS: GAGGAAGATGTGGTTGCAGTC
ACOX-1	NM_015729	S: GCGTGGAACTTGACTTCTGTC AS: ACGGCTCTGTCTTGAATCTTG
ATGL	NM_025802	S: TCCAAGGATGAGCTCATCCA AS: CCTCCTAAGGAATCGAAGTC
β-ACTIN	NM_007393	S: CATGGATGACGATATCGCTGC AS: GTACGACCAGAGGCATACAGG
CD36	NM_007643	S: AGGTCTATCTACGCTGTGTTCG AS: CAATGGTTGTCTGGATTCTGG
CPT1a	NM_013495	S: GGTTCAAGCTGTTCAAGATAGC AS: ACCACATAGAGGCAGAAGAGG
DGAT-1	NM_010046	S: GAGCTATCCAGACAACCTGACC AS: AGCATCTCAAGAACTCGTCGT
GCK	NM_010292	S: ATCTTCTGTTCCACGGAGAGG AS: GATGTTAAGGATCTGCCTTCG
GLUT-2	NM_031197	S: CTTGGTTCATGGTTGCTGAAT AS: GCAATGTACTGGAAGCAGAGG
GPAT-1	NM_008149	S: CGGAACTGAACTGGAGAAGTG AS: GATGAATTGCTGGTGCTCCTT
HNF-3β	NM_010446	S: CTACGCCAACATGAACTCGAT AS: GGTGATGAGCGAGATGTACGA
HSL	NM_001039507	S: GGCTCACAGTTACCATCTCACC AS: GAGTACCTTGCTGTCCTGTCC
INS-2	NM_008387	S: TGGAGGCTCTCTACCTGGTG AS: TCTACAATGCCACGCTTCTG
LCAT	NM_008490	S: GGCTGCACTCTATGAAGATGG AS: TGAGGTGATCTGTCTCGTTCA
LXR-α	NM_013839	S: GTTCTCCAGAGCCATGAATGA AS: GTTGCAGCCTCTCTACTTGGA
LXR-β	NM_009473	S: TCACCTACAGCAAGGACGACT AS: ATGGCGATAAGCAAGGCATAC
Maf-A	NM_194350	S: GTGCTGGAGGATCTGTACTGG AS: ATGGTGGTGATGGTGATGG
MCD	NM_019966	S: AGAAGATCAGCGAGTGTGAGG AS: AGTCAGAGCCACATGCAGAAC
Nkx6.1	NM_144955	S: CACGTTCTCTGGACAGCAAAT AS: TTGACCTGACTCTCCGTCATC
PC	NM_008797	S: TGACTCTCTGCTCGTCAAGGT AS: AGGAACTGCTGGTTGTTGAGA
PDX-1	NM_008814	S: GGTATAGCCGGAGAGATGC AS: CTGGTCCGTATTGGAACG
PGC-1α	NM_008904	S: TAGAGTGTGCTGCTCTGGTTG

		AS: GATTGGTCGCTACACCACTTC
PGC-1β	NM_133249	S: GCTCTGGAAGGTGAAGACCTG AS: TCAAGCAGGAAGCTACTCTCG
PPAR-α	NM_011144	S: AGAGAATCCACGAAGCCTACC AS: GGCCATACACAAGGTCTCCAT
PPAR-γ	NM_011146	S: ATCAGCTCTGTGGACCTCTCC AS: GGTCAGCTCTTGTGAATGGAA
PREPROINS-2	NT_039437	S: CAGTGCCAAGGTCTGAAGGT AS: TGTGTCCATCCATGACCAGT
SCD-1	NM_009127	S: GAAGCTGGTGATGTTCCAGAG AS: CCAGAGTGTATCGCAAGAAGG
SNAP-25	NM_01142	S: ATGGAGAAGGCTGATTCCAAC AS: CCATGAGAGAAGCATGAAGGA
SOAT-1	NM_009230	S: CCACGGTTACAGCAAGAGTTC AS: TGGTGGCAGTGTGTATGCTAA
SREBP-1c	NM_011480	S: ATGCTCCAGCTCATCAACAAC AS: GAGGCCAGAGAAGCAGAAGAG
STX-1a	NM_016801	S: GTGAGGAATTGGAAGACATGC AS: GCCTTGCTCTGGTACTTGACG
UCP-2	NM_011671	S: AGAGCACTGTCGAAGCCTACA AS: AGGCAGAAGTGAAGTGGCAAG