Supplementary information

Materials and methods

RNA Sequencing and analysis

Three biological replicates from CT, HSD and HSD+AA were subjected to RNA-seq. Reads were pair-ended to generate 150-bp-long fragments. RNA-seq reads were assessed for quality using FastQC (v0.12.1), aligned to Ensembl Drosophila melanogaster (BDGP6.46) genome using STAR (v2.7.11a). Read counts were generated using featureCounts from Rsubread Biconductor R package (v2.18.0**)**. Gene counts were rounded using the R function “round” and differential expression analysis was performed using DESeq2 (v1.46.0) with a pre-filtering step, in which only genes with more than one count when summing up across all samples were considered. Differential expression analysis between conditions was carried out by Wald test implemented by DESeq2 pairwise comparison, with default parameters. Differentially expressed genes (DEG) with FDR <0.05, calculated by DEseq2 pairwise comparisons, were considered significant and used for further downstream functional analysis. The significant DEG between conditions were plotted using EnchancedVolcano R package (v1.24.0). The count plots for specific genes were plotted using plotCounts function from Deseq2.

Functional Analyses

Functional analysis of DEG across conditions was conducted using *enrichKEGG* and *enrichGO* from the ClusterProfiler R package (v3.18.1). Pathways and biological processes with an adjusted P-value < 0.05 (Benjamini-Hochberg correction) were considered significant and visualized with ggpubr and ggplot2 (ggplot2.tidyverse.org).

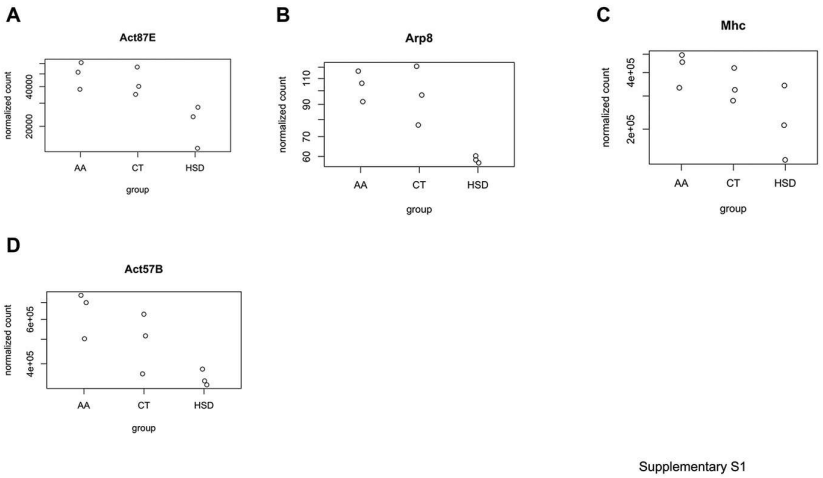
Immuno Histochemistry

The pancreas was fixed using 4% paraformaldehyde and then sectioned to thin sections. The FFPE (formalin-fixed, paraffin-embedded) sections on the slide were deparaffinized, which was followed by rehydration using sequential alcohol gradient washes (100%, 90%, 70%, 50%). Tris EDTA buffer was used for antigen retrieval, slides were immersed in Tris-EDTA buffer and boiled. To enhance the membrane permeability, the slides were permeabilized using 0.5% triton X (in PBS) for 15 minutes. 3% Bovine Serum Albumin (BSA) was used as a blocking reagent for the tissue sections. Anti-insulin primary antibody raised in rabbit, and anti-rabbit alexa 488 was used as the secondary antibody to stain the β-cells (green in color). Hoechst staining was done, following which the sections were mounted using an antifade and coverslip. The slides were then imaged using the Nikon Ti2 Eclipse confocal microscope through specific channels based on the fluorophores used.

Table 1. List of primers

SI No	Gene	Forward primer (5'-3')	Reverse primer (5'-3')
1	InR	AAGCGTGGGAAAATTAAGATGGA	GGCTGTCAACTGCTTCTACTG
2	Thor	CAGATGCCCGAGGTGTACTC	TTCATGAAAGCCCGCTCGTA
3	dilp2	ATGAGCAAGCCTTTGTCCTTC	ACCTCGTTGAGCTTTTCACTG
4	akh	GAGCTGGTCCTGGAACCTTTT	GCCTGAGATTGCACGAAGC
5	Mhc	CCAAGACGGTCAAAAACGAT	GATGTTGGCTCCCGAGATAA
6	mef2	GAAGCCGAAACGGACTACACA	GTTGTCGCCGTAAGATCCCG
7	dTET (Exon 7)	GACCTCAATAAGACGATGTCGG	CGCCTGAATACCCGTTACAGT
8	Tet-R1 (-38 to -339 promoter)	TTCGTGCTCGAAGCGAACAG	TCGGTCGTCATGCTTGTTGTA
9	Tet-R2 (-691 to -892 promoter)	AGCATGCATTATATGGTAATTGGCG	CGAACTGCGAATGCCCTGAA
10	SOD1	CGCACTTCAATCCGTATGGC	TGATGTTGACCTTGGTGGGG
11	SOD2	CAAGAAGTCGGGCAAACTGC	GAGGGACGCACGTTCTTGTA
12	CAT	GTGAACTTCCTGGATGAGATGTC	CACCTCAAAGTAACCAAAAGCAC
13	RPL32	AAGCGGCGACGCACTCTGTT	GCCCAGCATACAGGCCCAAG

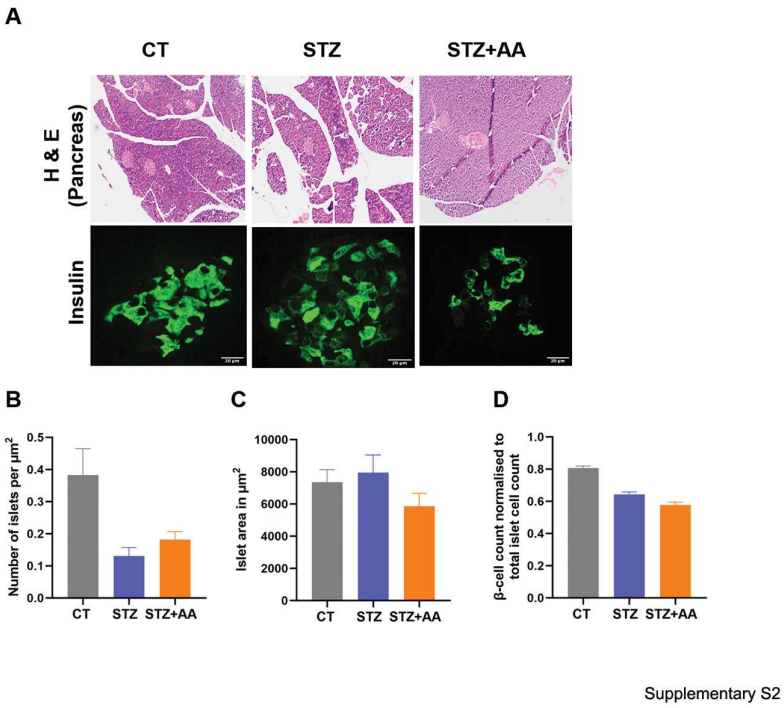
Supplementary figure S1



Supplementary S1

Supplementary Figure S1. Expression of mef2 target genes obtained from RNA-seq data

Supplementary figure S2



Supplementary Figure S2**.** (A) Pancreatic islets of the TA muscle injured mice stained with Haematoxylin and eosin (top panel) and immunohistochemistry images of pancreatic islets probed with insulin antibody (green, lower panel). (B, C&D) Quantification of number of islets, islet area and β-cell count. n=4 mice each group.