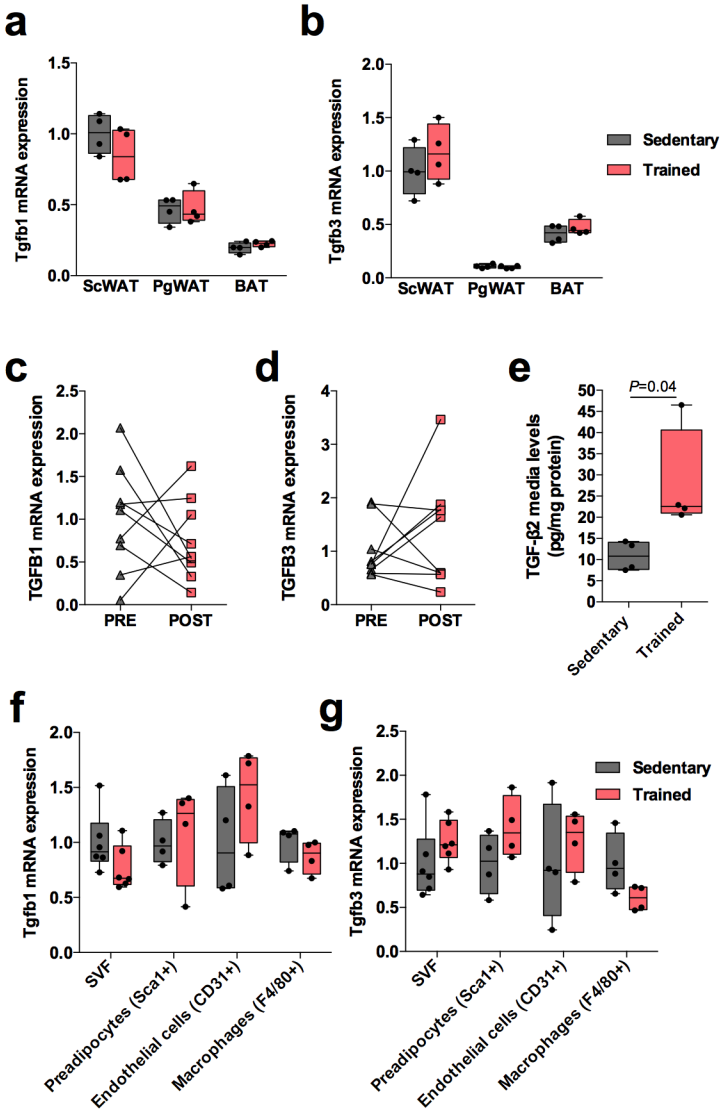
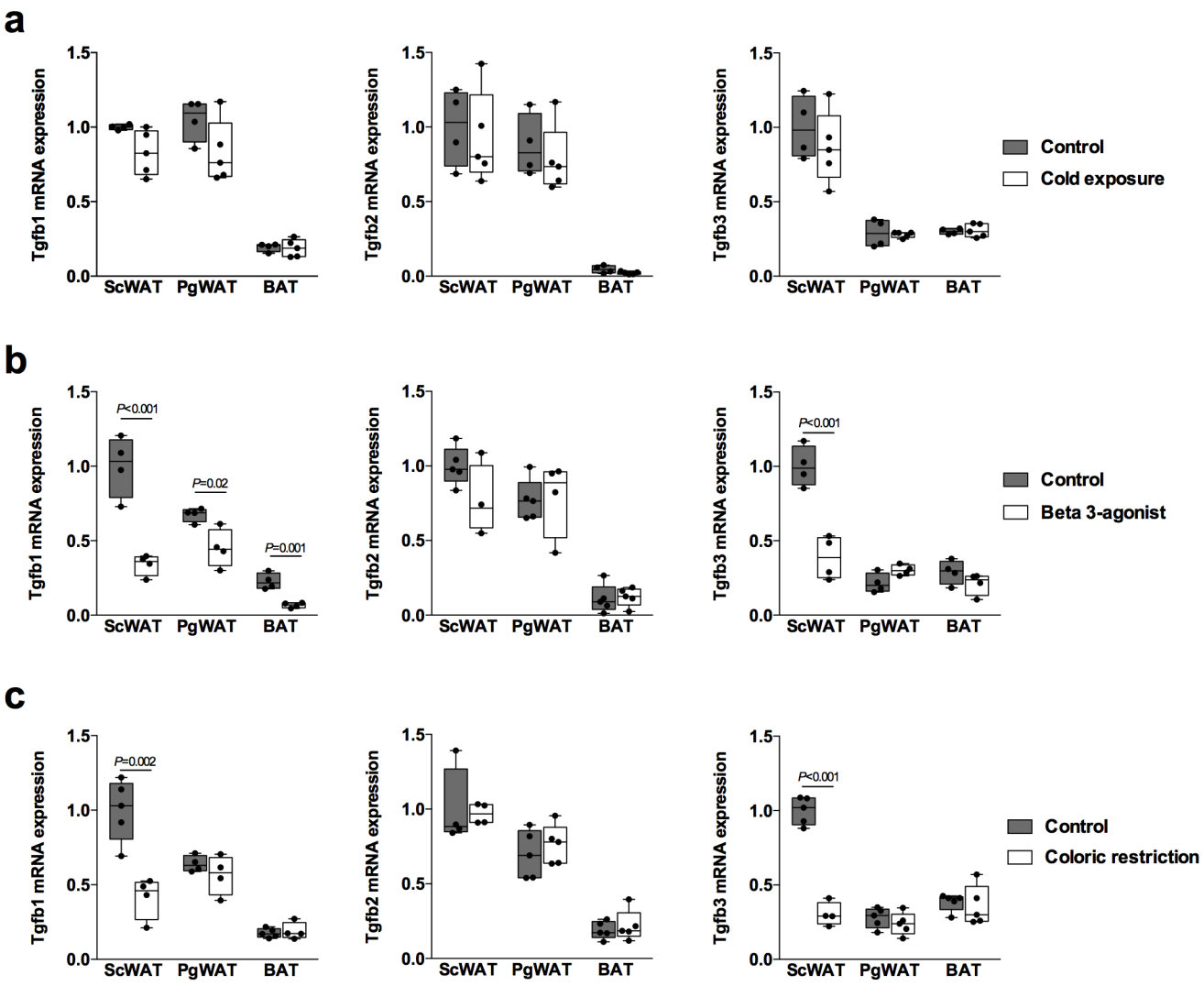
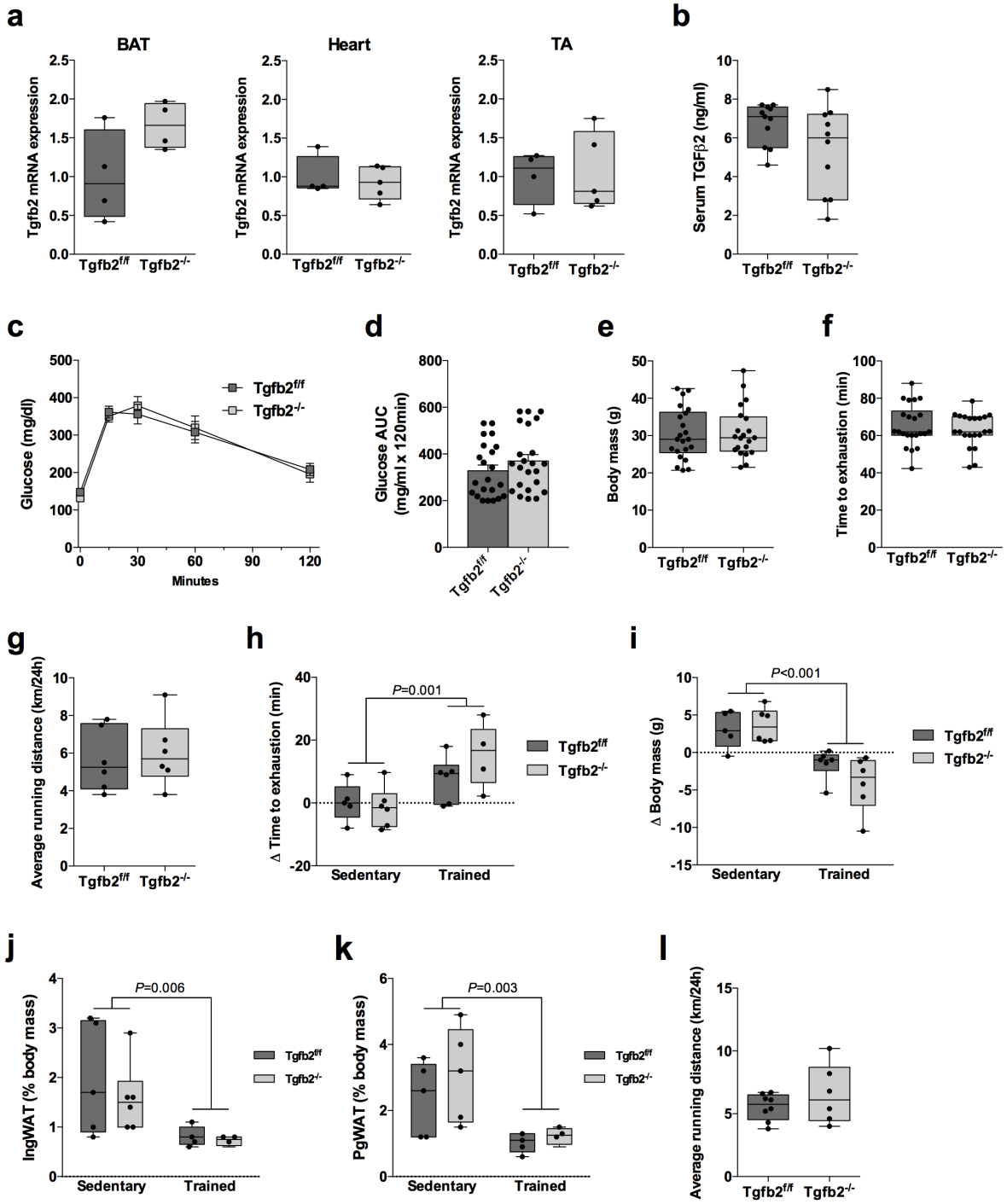
Supplementary Figures



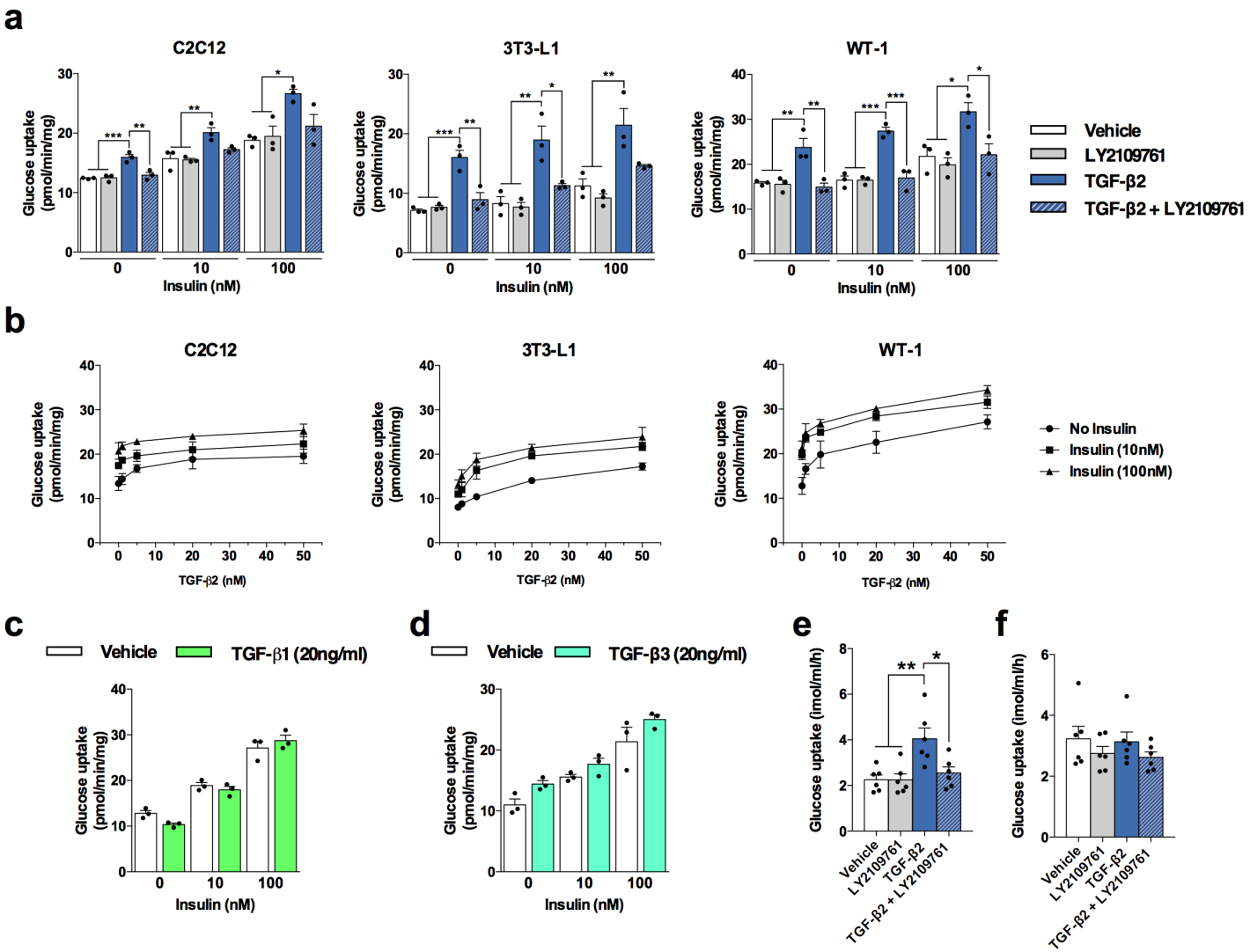
Supplementary Figure 1. Exercise training does not affect Tgfb1 and Tgfb3 expression in adipose tissue. (**a**) Tgfb1 and (**b**) Tgfb3 mRNA relative expression in scWAT, pgWAT, and BAT in trained mice; *n*=4 mice. (**c**) TGFB1 and (**d**) TGFB3 mRNA relative expression in human scWAT pre-and post-aerobic exercise training. *n*=9 mice. (**e**) Mature adipocytes isolated from trained pgWAT were incubated in serum-free media and TGF-β2 concentrations were determined in the media. *n*=4. (**f**) Tgfb1 and (**g**) Tgfb3 mRNA relative expression in stromal vascular fraction, preadipocytes, endothelial cells, and macrophages isolated from scWAT of trained mice; *n*=4 mice. Data are presented as box plots (min, max, median, and 25th and 75th percentiles) with dots as individual values (**a**, **b**, **e**, **f** and **g**), or individual values (**c** and **d**). Unpaired two-tailed Student's t-tests were used for **a**, **b**, **e**, **f** and **g**. Paired two-tailed Student's t-tests were used for **c** and **d**.



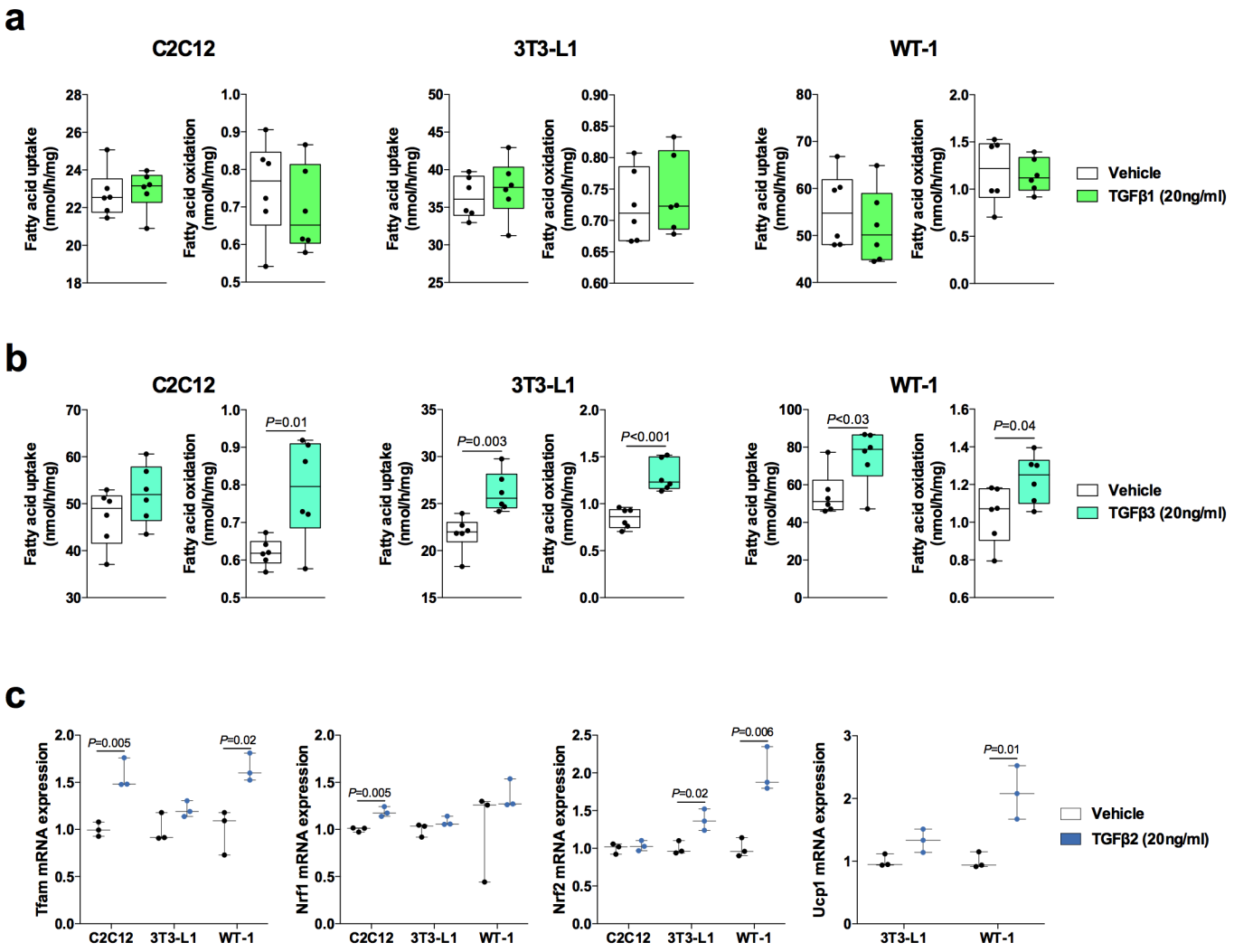
Supplementary Figure 2. Effects of cold exposure, calorie restriction, and thermogenic beta 3-agonist on Tgfb1, Tgfb2, and Tgfb3 relative expression in adipose tissue. (**a,b,c**) Tgfb1, Tgfb2 and Tgfb3 mRNA relative expression in scWAT, pgWAT, and BAT in response to (**a**) 5 days of cold exposure, (**b**) 5 days of beta 3-agonist (CL-316,243) treatment, and (**c**) 14 days of calorie restriction; *n*=4 or 5 mice. Data are presented as box plots (min, max, median, and 25th and 75th percentiles) with dots as individual values. Unpaired two-tailed Student's t-tests were used.



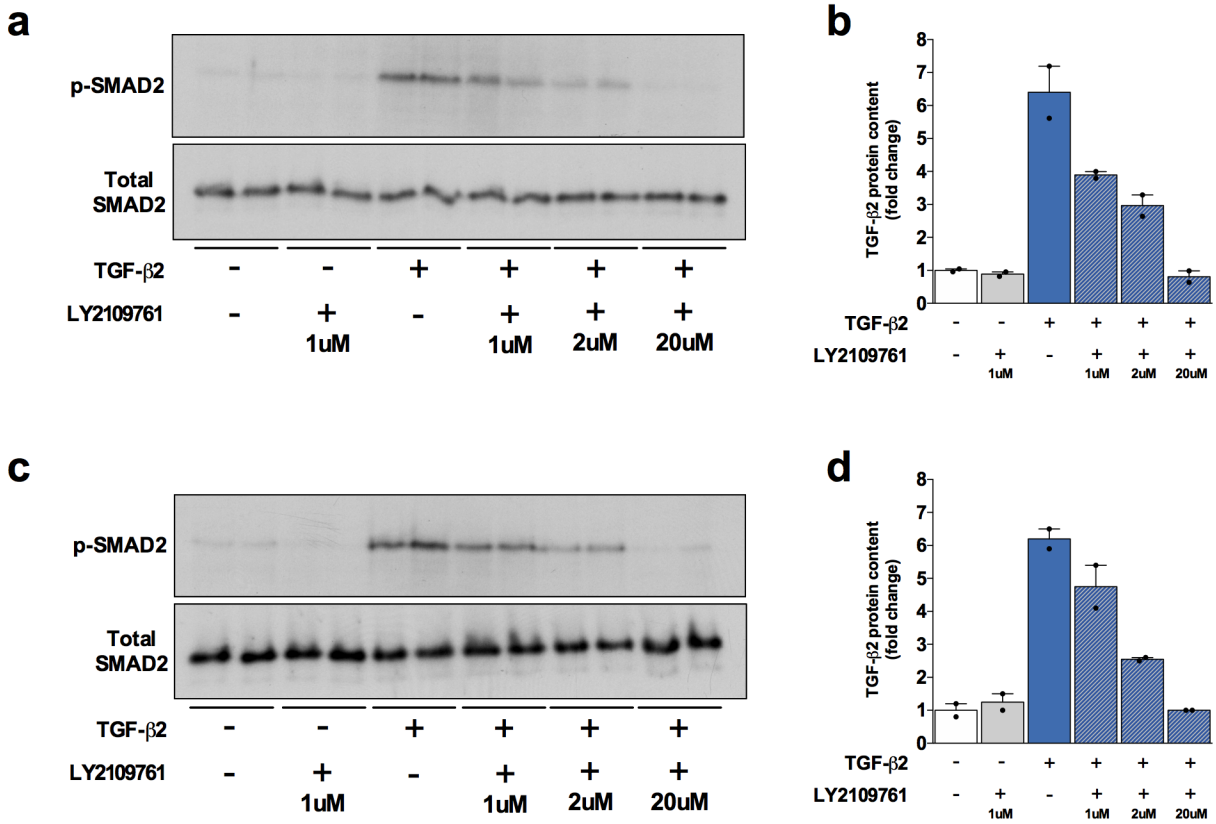
Supplementary Figure 3. Adipose-specific Tgfb2 knockout mice (Tgfb2$^{-/-}$) and control (Tgfb2$^{f/f}$) mice present similar TGF-β2 serum levels, glucose metabolism, and exercise capacity. (**a**) Tgfb2 mRNA relative expression in BAT, heart, and *tibialis anterior* (*TA*); *n*=4 mice. (**b**) TGF-β2 serum concentration; *n*=11 Tgfb2$^{f/f}$ mice, 10 Tgfb2$^{-/-}$ mice. (**c**) Glucose tolerance test (GTT) and (**d**) area under the curve (GTT AUC); *n*=22 Tgfb2$^{f/f}$, *n*=23 Tgfb2$^{-/-}$ mice. (**e**) Body mass; *n*=22 Tgfb2$^{f/f}$, *n*=21 Tgfb2$^{-/-}$ mice. (**f**) Exercise capacity; *n*=22 Tgfb2$^{f/f}$ mice, *n*=21 Tgfb2$^{-/-}$ mice. (**g,h,i**) Tgfb2$^{-/-}$ and Tgfb2$^{f/f}$ mice had access to running wheel for 28 days. (**g**) Average running distance and delta changes in (**h**) exercise capacity and (**i**) body mass were analyzed. *n*=5 or 6 mice. (**j,k**) percentage of (**j**) scWAT and (**k**) pgWAT mass in trained Tgfb2$^{-/-}$ mice. *n*=4 or 5 mice. (**l**) Average running distance in Tgfb2$^{-/-}$ and Tgfb2$^{f/f}$ trained mice. scWAT was collected for transplantation experiments in **Figure 1q,r**; *n*=8 Tgfb2$^{f/f}$ mice, *n*=5 Tgfb2$^{-/-}$ mice. Data are presented as box plots (min, max, median, and 25th and 75th percentiles) with dots as individual values (**a**, **b**, **d**, **e**, **f**, **g**, **h**, **i**, **j**, **k** and **l**) or mean ± s.e.m (**c**). Unpaired two-tailed Student's t-tests were used for **a**, **b**, **d**, **e**, **f**, **g** and **l**. ANOVA was used for **c**, **h**, **i**, **j**, and **k**.



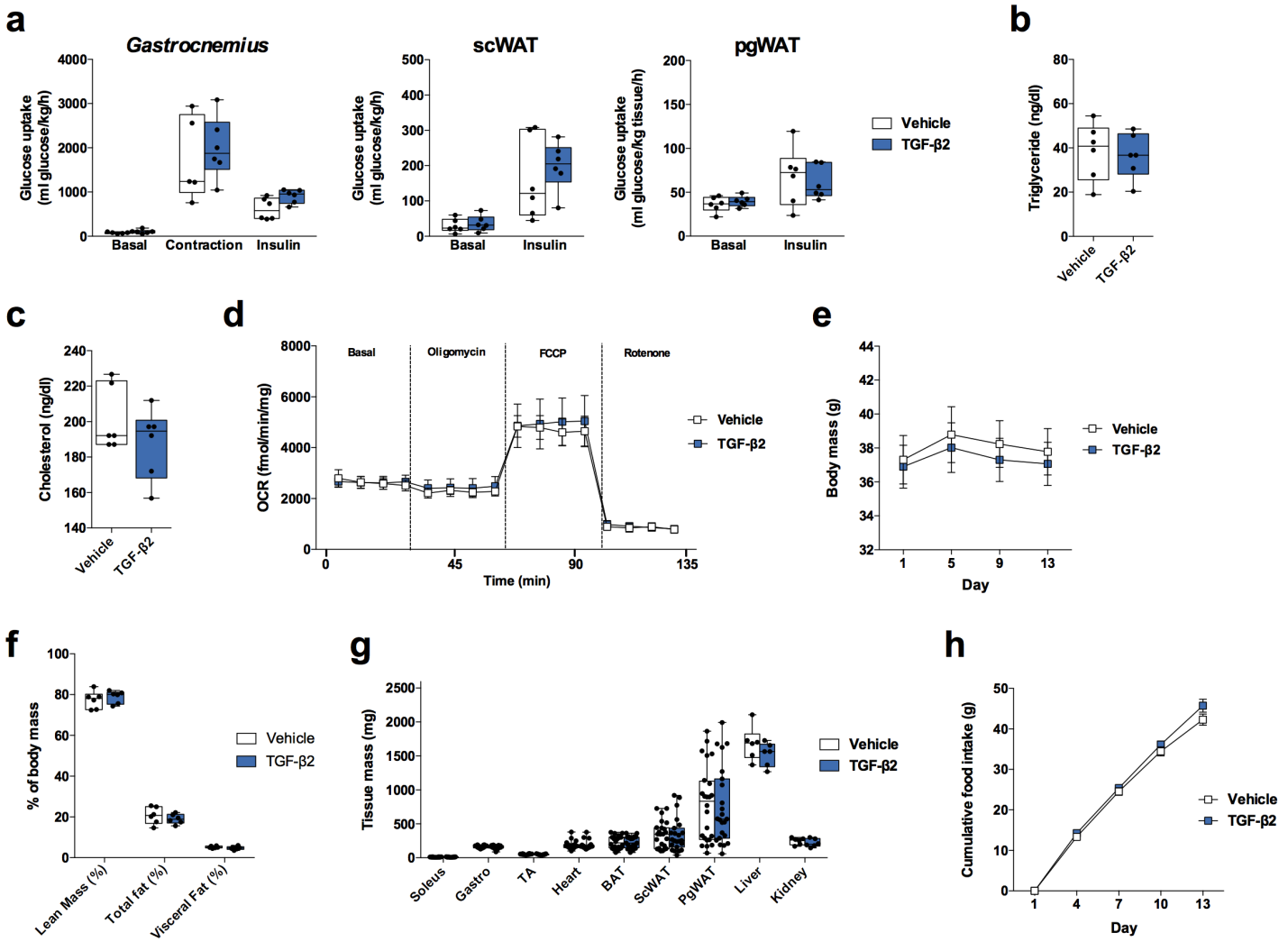
Supplementary Figure 4. Effects of recombinant TGF-β1, TGF-β2 or TGF-β3 incubation on glucose uptake *in vitro*. (**a**) Glucose uptake in C2C12 myotubes, 3T3-L1 adipocytes, and WT-1 brown adipocytes treated with TGF-β2 and/or TGF-β receptor inhibitor (LY2109761). *n*=3 biological replicates. (**b**) Glucose uptake in C2C12 myotubes, 3T3-L1 adipocytes, and WT-1 brown adipocytes treated with different concentrations of TGF-β2; *n*=3 biological replicates. (**c,d**) Glucose uptake in C2C12 myotubes treated with (**c**) TGF-β1 or (**d**) TGF-β3; *n*=3 biological replicates. (**e,f**) Glucose uptake in incubated (**e**) *soleus* and (**f**) EDL muscle treated with TGF-β2 and/or TGF-β receptor inhibitor (LY2109761); *n*=6 biological replicates. Data are presented as mean ± s.e.m. Unpaired two-tailed Student's t-tests were used for **c** and **d**. ANOVA was used for **a**, **b**, **e** and **f**. When ANOVA showed P<0.05, Tukey's multiple comparisons tests were used with *P<0.05; **P<0.01; ***P<0.001.



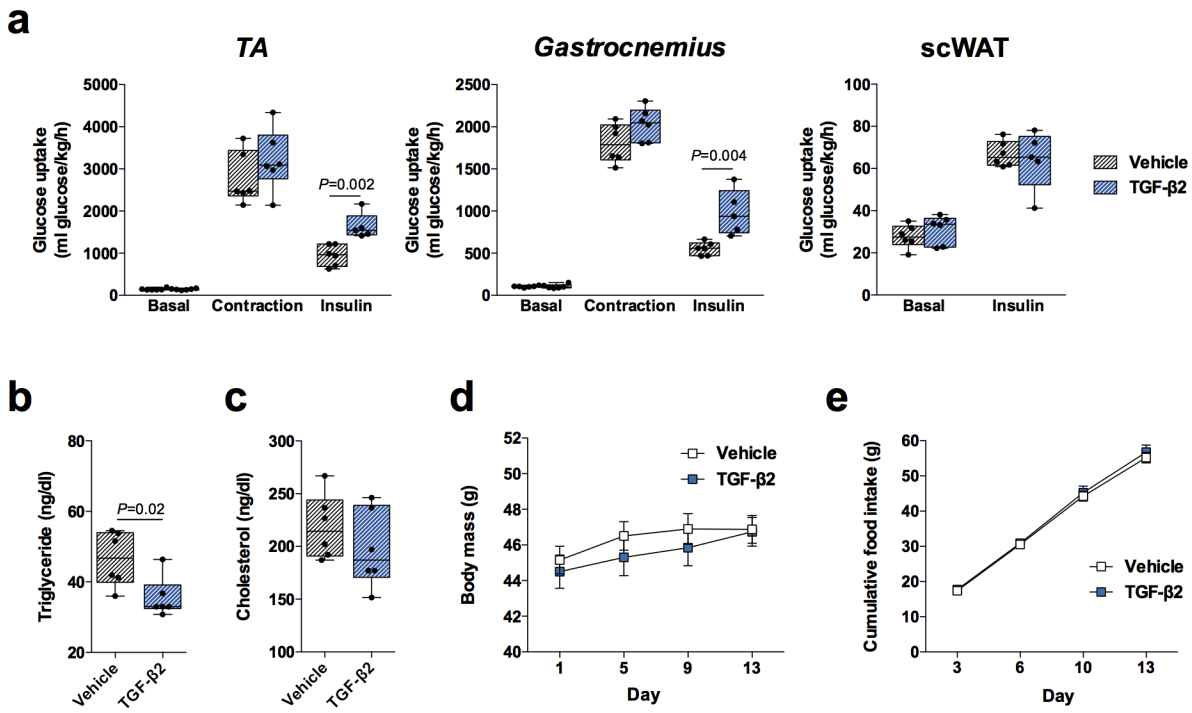
Supplementary Figure 5. Effects of recombinant TGF-β1 or TGF-β3 incubation on fatty acid uptake and oxidation *in vitro*. (**a,b**) [^{14}C] palmitic acid uptake and oxidation in C2C12 myotubes, 3T3-L1 adipocytes and WT-1 brown adipocytes incubated with TGF-β1 or TGF-β3; *n*=6 biological replicates. (**c**) Tfam, Nrf1, Nfr2, and Ucp1 mRNA relative expression in C2C12 myotubes, 3T3-L1 adipocytes, and WT-1 brown adipocytes treated with TGF-β2; *n*=3 biological replicates. Data are presented as box plots (min, max, median, and 25th and 75th percentiles) with dots as individual values. Unpaired two-tailed Student's t-tests were used.



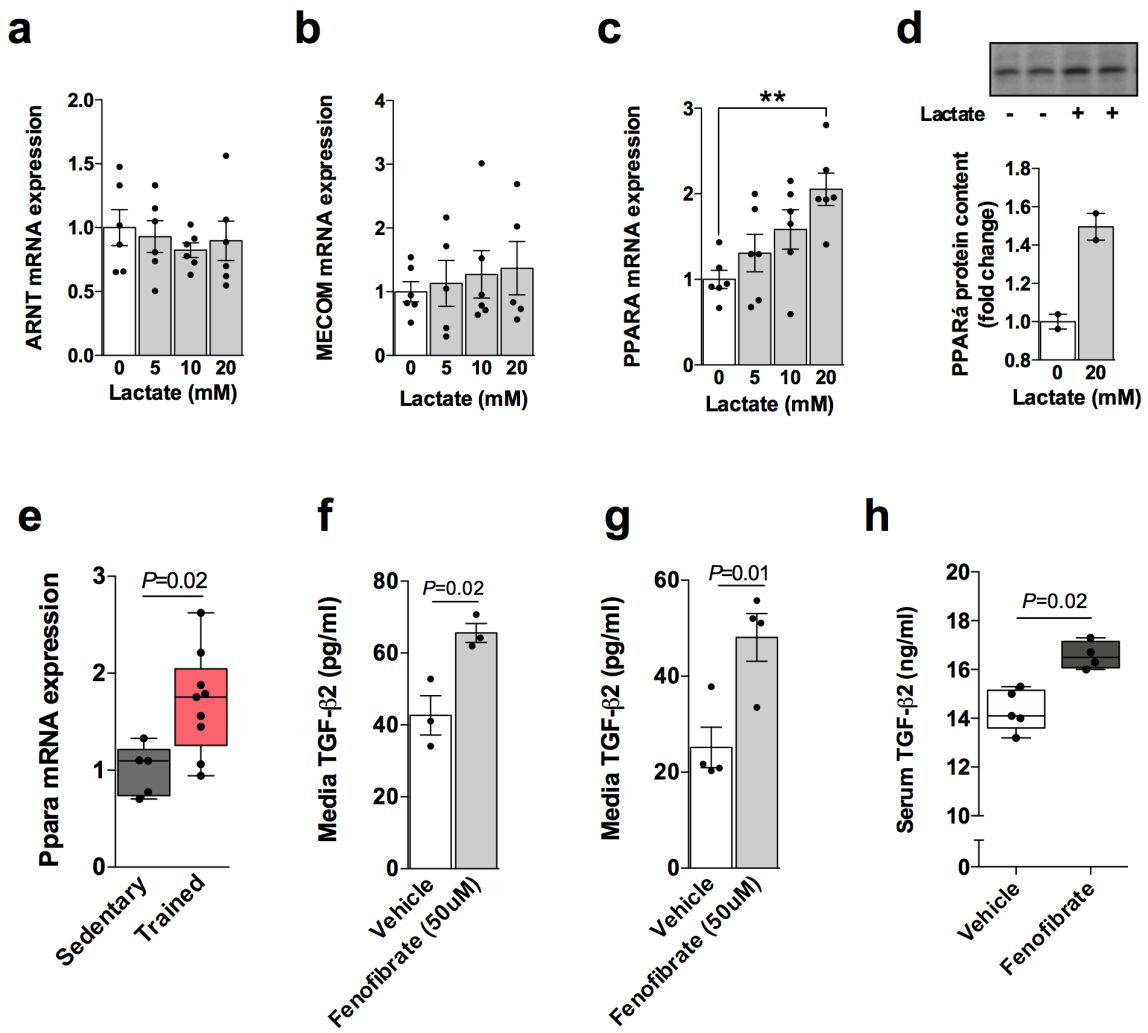
Supplementary Figure 6. Effects of recombinant TGF-β2 incubation on SMAD2 phosphorylation. (**a**) Representative immunoblot and (**b**) quantification of p-SMAD2/total SMAD2 ratio in C2C12 myotubes treated with TGF-β2 and/or TGF-β receptor inhibitor (LY2109761) for 1 hour. n = 2 biological replicates. (**c**) Representative immunoblot and (**d**) quantification of p-SMAD2/total SMAD2 ratio in C2C12 myotubes treated with TGF-β2 and/or TGF-β receptor inhibitor (LY2109761) for 5 hours. n = 2 biological replicates. Data are presented as mean ± s.e.m.



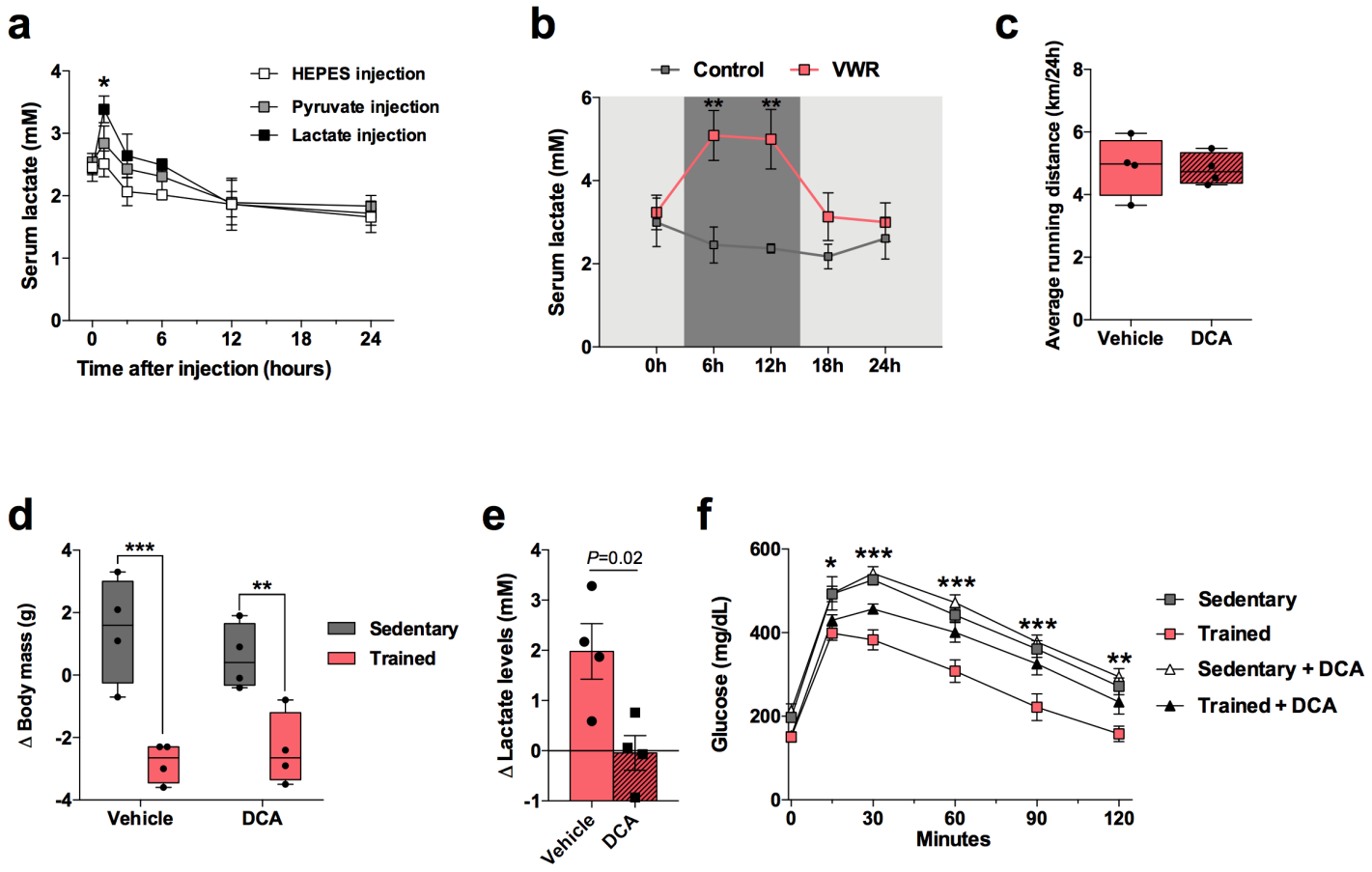
Supplementary Figure 7. TGF-β2 infusion via osmotic pump does not affect white adipose tissue glucose uptake and body composition in normal chow diet-fed mice. (**a**) [^{3}H]-2-deoxyglucose uptake in *gastrocnemius*, scWAT, and pgWAT after TGF-β2 treatment; n = 5 or 6 mice. (**b**) Serum triglyceride and (**c**) cholesterol concentrations in mice; n = 6 mice. (**d**) Extracellular flux analysis in *flexor digitorum brevis* (FDB); n = 5 mice. (**e**) Body mass; n = 25 mice. (**f**) Percentage of lean, total fat, and visceral fat mass; n = 6 mice. (**g**) Tissue mass. n = 6 mice. (**h**) Food intake; n = 7 mice. Data are presented as box plots (min, max, median, and 25th and 75th percentiles) with dots as individual values (**a**, **b**, **c**, **f** and **g**) or mean ± s.e.m (**d**, **e** and **h**). Unpaired two-tailed Student's t-tests were used for **a**, **b**, **c**, **f** and **g**. ANOVA was used for **d**, **e** and **h**.



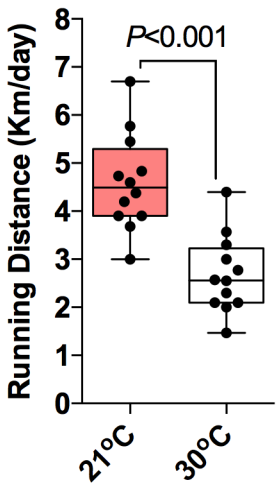
Supplementary Figure 8. Effects of TGF-β2 infusion via osmotic pump on skeletal muscle and white adipose tissue glucose uptake, body mass, and food intake in high-fat diet-fed (HFD). TGF-β2 treatment attenuates inflammation in adipose tissue of HFD mice. (**a**) [^{3}H]-2-deoxyglucose uptake of *tibialis anterior* (TA), *gastrocnemius*, and scWAT after TGF-β2 treatment in HFD mice; n = 5 or 6 mice. (**b**) Serum triglyceride and (**c**) cholesterol concentrations in HFD mice; n = 6 mice. (**d**) Body mass; n = 25 mice. (**e**) Food intake; n = 7 mice. Data are presented as box plots (min, max, median, and 25th and 75th percentiles) with dots as individual values (**a**, **b** and **c**) or mean ± s.e.m (**d** and **e**). Unpaired two-tailed Student's t-tests were used for **a**, **b** and **c**. ANOVA was used for **d** and **e**.



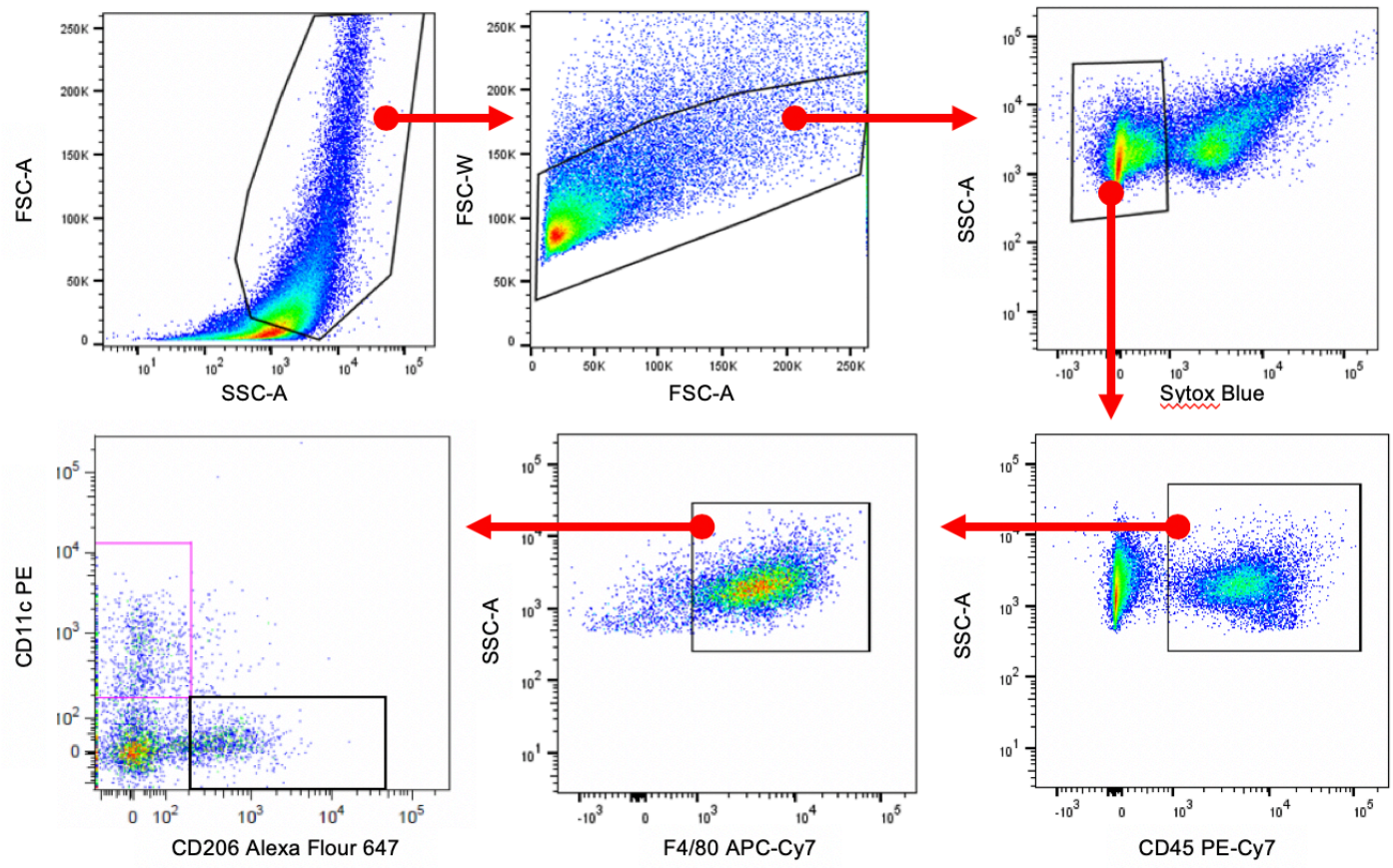
Supplementary Figure 9. Lactate and exercise training increase PPARa expression. (**a**) ARNT, (**b**) MECOM, and (**c**) PPARA mRNA relative expression in human adipocytes treated with different concentrations of lactate; *n*=6 biological replicates. (**d**) Immunoblot for PPARα in human adipocytes treated with lactate. *n*=2 biological replicates (**e**) Ppara mRNA expression in scWAT of trained mice. *n*=5 sedentary mice, *n*=9 trained mice. (**f**) TGF-β2 media concentration in 3T3-L1 adipocytes treated with fenofibrate; *n*=3 technical replicates. (**g**) TGF-β2 media concentration in *ex vivo* scWAT treated with fenofibrate for 24 hours; *n*=4 biological replicates. (**h**) Serum TGF-β2 concentration in mice injected with fenofibrate intraperitoneally. Data are presented as box plots (min, max, median, and 25th and 75th percentiles) with dots as individual values (**e** and **h**) or mean ± s.e.m (**a**, **b**, **c**, **d**, **f** and **g**). Unpaired two-tailed Student's t-tests were used for **e**, **f**, **g** and **h**. ANOVA was used for **a**, **b** and **c**. When ANOVA showed P<0.05, Tukey's multiple comparisons tests were used with **P<0.01.



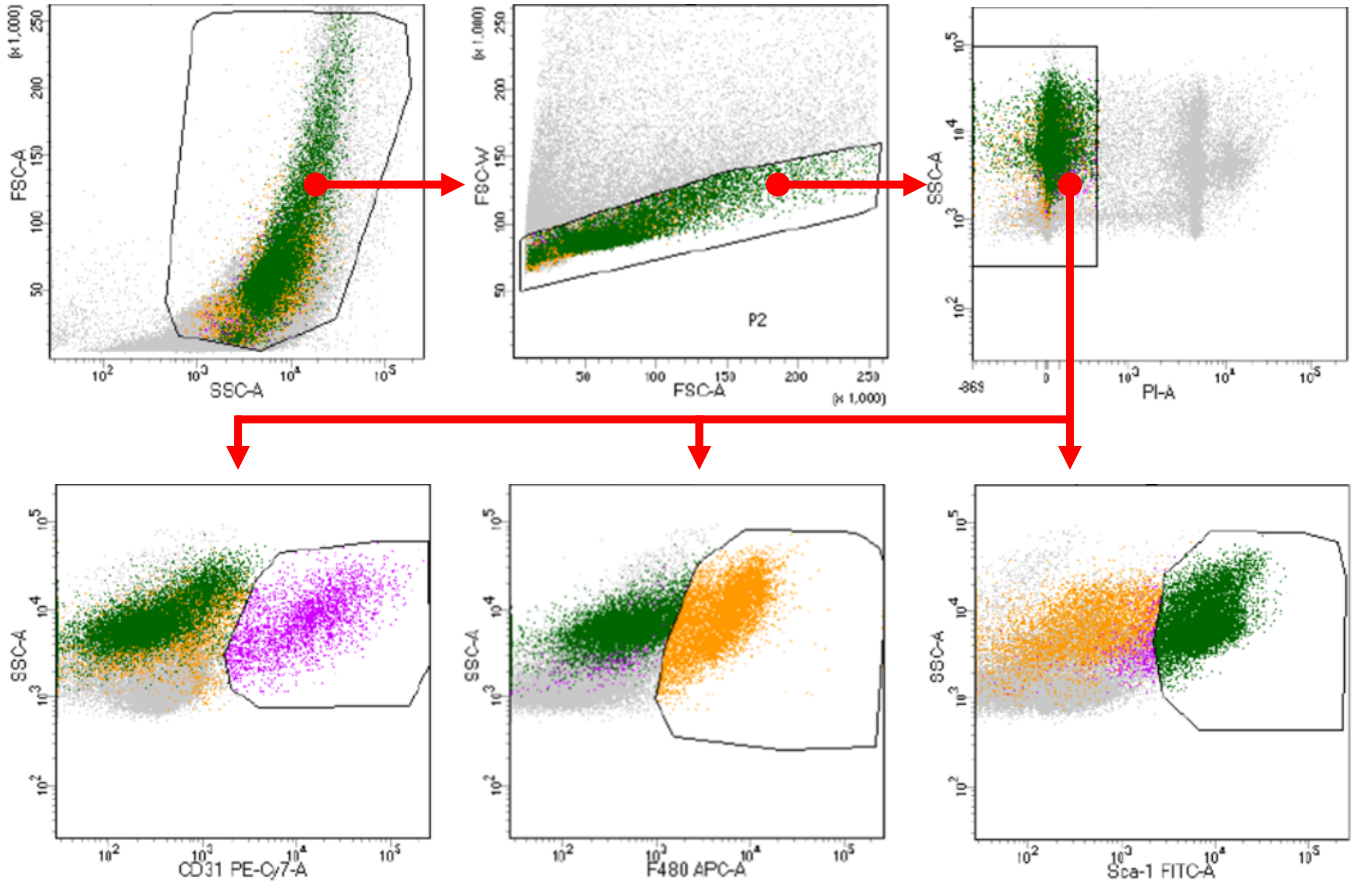
Supplementary Figure 10. (**a**) Time course of lactate serum concentration in mice injected with lactate, pyruvate, or vehicle (HEPES) intraperitoneally; *n*=4 mice. (**b**) Time course of serum lactate concentration during voluntary wheel running (VWR); *n*=6 mice. (**c**) Average running distance of mice treated daily with dichloroacetate (DCA) injections; n=4 mice. (**d**) Delta changes in body mass, (**e**) lactate levels, and (**f**) glucose tolerance test (GTT) in trained mice after daily dichloroacetate (DCA) injections; n=4 mice. Data are presented as box plots (min, max, median, and 25th and 75th percentiles) with dots as individual values (**c** and **d**) or mean ± s.e.m (**a**, **b**, **e** and **f**). Unpaired two-tailed Student's t-tests were used for **c** and **e**. ANOVA was used for **a**, **b**, **d** and **f**. When ANOVA showed P<0.05, Tukey's multiple comparisons tests were used with *P<0.05; **P<0.01, ***P<0.001.



Supplementary Figure 11. Running distance in mice housed at 21°C and mice housed in a thermoneutral environment (30°C); *n* = 12 mice. Data are presented as box plots (min, max, median, and 25th and 75th percentiles) with dots as individual values. Unpaired two-tailed Student's t-test was used.



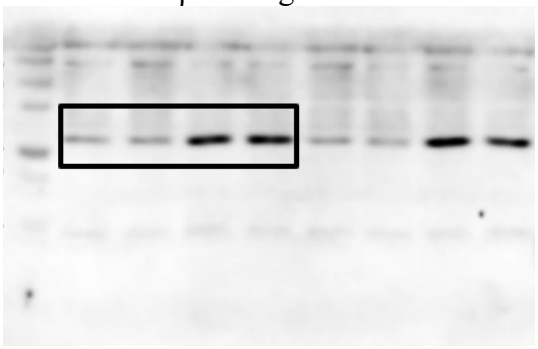
Supplementary Figure 12. Gating strategy to identify M1 and M2 macrophages. Cells were incubated with CD45-PE-Cy7, F4/80-APC-Cy7, CD206-Alex647 and CD11c-PE antibodies and suspended in solution with Sytox Blue. M1 or M2 macrophages were identified as F4/80-positive/CD11c-positive/CD206-negative or F4/80-positive/CD11c-negative/CD206-positive cells, respectively.



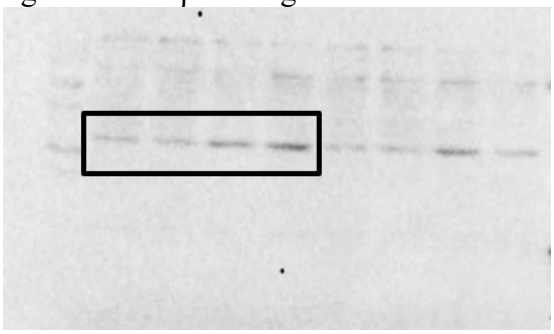
Supplementary Figure 13. Gating strategy for sorting preadipocytes, endothelial cells and macrophages. Cells were incubated with CD31-PE-Cy7, F4/80-APC, and Sca-1 FITC antibodies and resuspended in solution with Propidium Iodide Staining Solution (PI).

Supplementary Figure 14. Uncropped Blots.

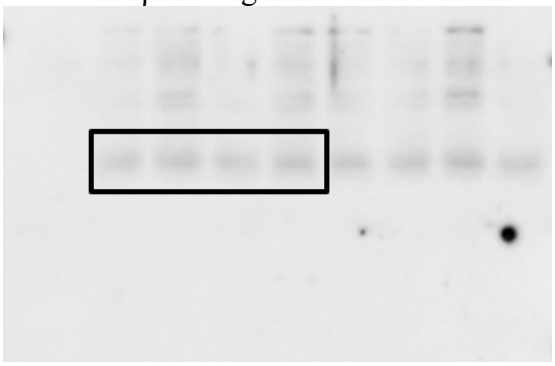
ScWAT TGF-β2 in Figure 1d



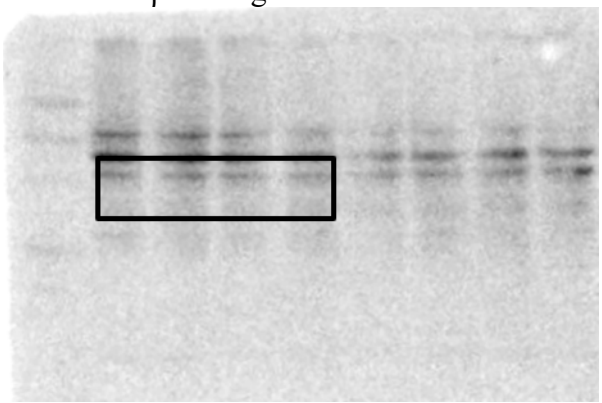
PgWAT TGF-β2 in Figure 1d



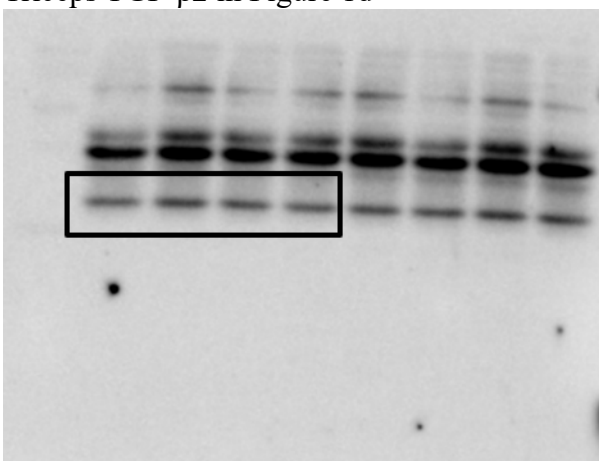
BAT TGF-β2 in Figure 1d



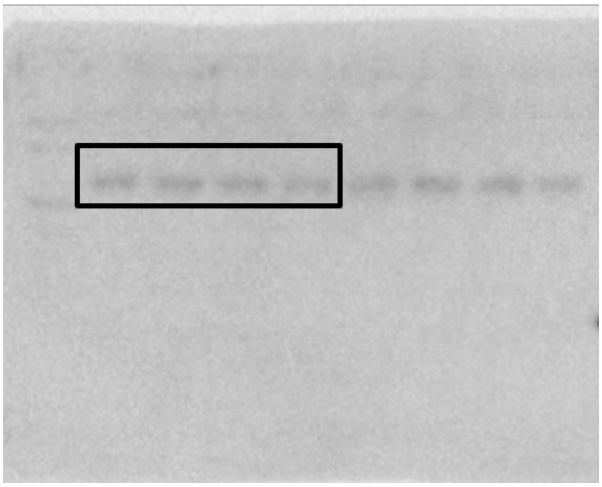
Liver TGF-β2 in Figure 1d



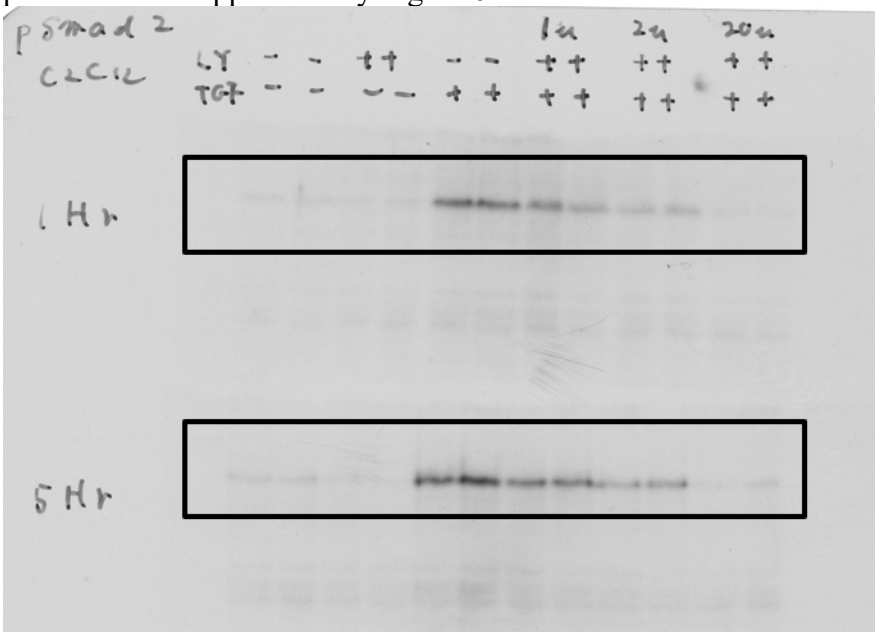
Triceps TGF-β2 in Figure 1d



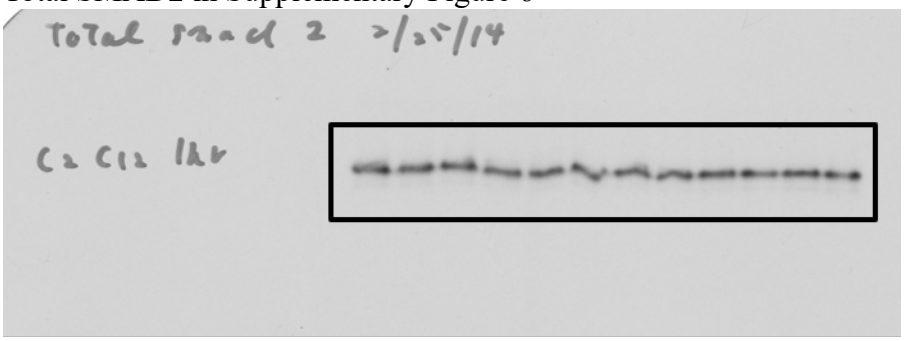
Heart TGF-β2 in Figure 1d

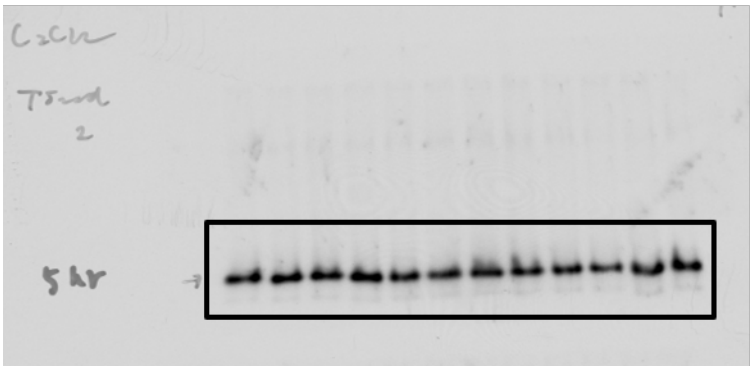


p-SMAD2 in Supplementary Figure 6

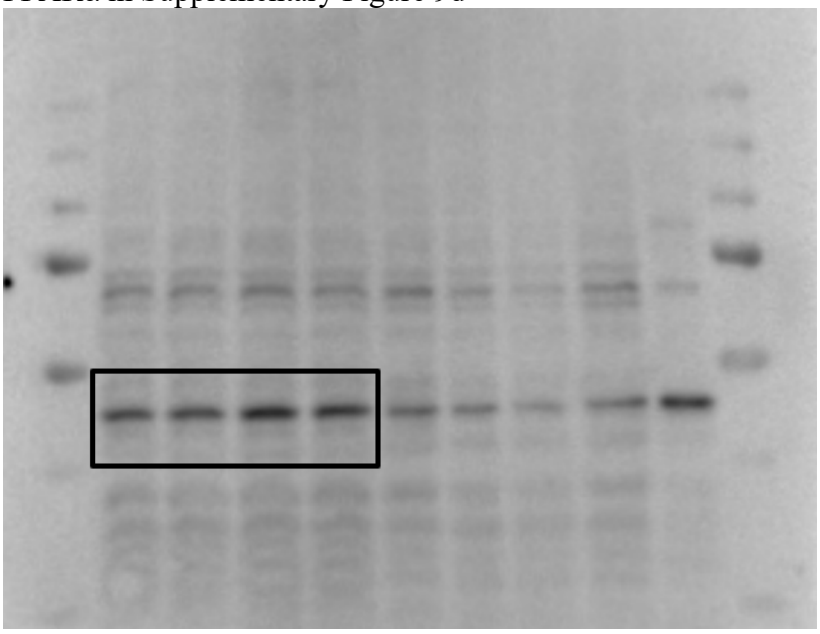


Total SMAD2 in Supplementary Figure 6





PPARα in Supplementary Figure 9d



Supplementary Tables

Supplementary Table 1. Separate excel file **"**Human_gene_stats"

Supplementary Table 2. List of primer sequences used for RT-qPCR in humans' samples.

Gene	Forward (5' > 3')	Reverse (5' > 3')
ACTB	CAGCCATGTACGTTGCTATCCAGG	AGGTCCAGACGCAGGATGGCATG
ARNT	GAACCTCACTTCGTGGTGGT	CAATGTTGTGTCGGGAGATG
ADAMTS12	ACTTTGGGGCGCTTTGCTAT	GCAGGCCCTTGATAAAATGCT
ANGPTL2	GAACCGAGTGCATAAGCAGGA	GTGACCCGCGAGTTCATGTT
APLP2	TGAGCCTCAAATCGCAATGTT	CCTGTTGGATCAGGTTCCCAT
APOM	TGCCCCGGAAATGGATCTA	CAGGGCGGCCTTCAGTT
COL11A2	GCTCCCCTCCTGACTCTCTAC	CCGGGTGACTCGCTTCTTG
CXCL12	ACCGCGCTCTGCCTCA	CATGGCTTTCGAAGAATCGG
CYR61	ACCGCTCTGAAGGGGATCT	ACTGATGTTTACAGTTGGGCTG
DAG1	AGCAAAGGATTGACCTCCTGC	CCACCGGCACTAATTTCATGTT
DKK2	CTCACAGATCGGCAGTTCG	ATGCCAGTCCTTGGTACATGC
FGF5	CACTGATAGGAACCCTAGAGGC	CAGATGGAAACCGATGCCC
GAPDH	GTCTCCTCTGACTTCAACAGCG	ACCACCCTGTTGCTGTAGCCAA
GAS6	CCGGGGACTTGTTCCAACC	CTGCACGAGGTCCTTCTCAT
GPX3	GAGCTTGCACCATTCGGTCT	GGGTAGGAAGGATCTCTGAGTTC
HAPLN1	TTGAGAGCATCCGAACTCCT	GATGATCAGCCCAGCAGATT
HFE2	GGCTGAGGTGGATAATCTTCCT	CCCAGGGTTAGCAGTTTGAAT
IGFBP5	TGACCGCAAAGGATTCTACAAG	CGTCAACGTACTCCATGCCT
LOXL1	CTGTGCTGCGGAGGAGAAG	GTAGTGGCTGAACTCGTCCA
MASP1	GTCCATCACTTTCCGGTCAGA	GTGGAGGATGTAGCCGAAGC
MECOM	TGAAGGAAACCCTGAGGATG	CGGCTGCTTAAGTTCCTCTG
NTF3	GAACTGCTGCGACAACAGAGA	CCCACGTAATCCTCCATGAGA
PPARA	GTTTGAGGGGGTAACAGCAA	GCTAACTGCAGAGGGTGAGG
SFRP4	ACGAGCTGCCTGTCTATGAC	TGTCTGGTGTGATGTCTATCCAC
SOD1	GGTGGGCCAAAGGATGAAGAG	CCACAAGCCAAACGACTTCC
TGFB1	GGCCAGATCCTGTCCAAGC	GTGGGTTTCCACCATTAGCAC
TGFB2	AAGAAGCGTGCTTTGGATGCGG	ATGCTCCAGCACAGAAGTTGGC
TGFB3	ACTTGCACCACCTTGGACTTC	GGTCATCACCGTTGGCTCA
TIMP3	CATGTGCAGTACATCCATACGG	CATCATAGACGCGACCTGTCA
TCN2	CAGAACAGTGCGAGAGGAGATC	TCGCCTTGAGACATGCTGTTCC
VEGF	CATGCAGATTATGCGGATCAA	TTTGTTGTGCTGTAGGAAGCTCAT

Supplementary Table 3. List of primer sequences used for RT-qPCR in mice' samples.

Gene	Forward (5' > 3')	Reverse (5' > 3')
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Adipoq	GGCAGGAAAGGAGAACCTGG	CAGGCTGTCTTTTGTCAACGA
Arg1	AGACCACAGTCTGGCAGTTG	CCACCCAAATGACACATAGG
Ccl2	GGCTCAGCCAGATGCAGT	GCTGCTGGTGATCCTCTTGT
Ccr2	TGCCATCATAAAGGAGCCA	AGCACATGTGGTGAATCCAA
Itgax (CD11c)	CAGAACTTCCCAACTGCACA	TCTCTGAAGCTGGCTCATCA
Gapdh	AACTTTGGCATTGTGGAAGG	ACACATTGGGGGTAGGAACA
Ifng	CAGCTCTTCCTCATGGCTGT	ATCTGGCTCTGCAGGATTTT
Il10	AGTGGAGCAGGTGAAGAGTG	CACTGCAGGTGTTTTAGCTTT
Il18	GACTCTTGCGTCAACTTCAAGG	CAGGCTGTCTTTTGTCAACGA
Il6	TCCAGTTGCCTTGGGAC	GTGTAATTAAGCCTCCGACTTG
Ilb	AGTTGACGGACCCCAAAAG	GCGAGATTTGAAGCTGGATG
Mcp1	AGGTCCCTGTCATGCTTCTG	TCATTGGGATCATCTTGCTG
Mrc1 (CD206)	TGATTACGAGCAGTGGAAGC	GTTCACCGTAAGCCCAATTT
Nrf1	CAACAGGGAAGAAACGGAAA	GCACCACATTCTCCAAAGGT
Nfe2l2	AGGTTGCCCACATTCCCAAACAAG	TTGCTCCATGTCCTGCTCTATGCT
Ppara	AGAGCCCCATCTGTCCTCTC	ACTGGTAGTCTGCAAAACCAAA
Ppargc1a	GCAGGCCTAACTCCTCCCACGA	TGGTGGAGTGGCTGCCTTGG
Sfrp5	CACTGCCACAAGTTCCCCC	TCTGTTCCATGAGGCCATCAG
Tfam	GTCCATAGGCACCGTATTGC	CCCATGCTGGAAAAACACTT
Tgfb1	CTCCCGTGGCTTCTAGTGC	GCCTTAGTTTGGACAGGATCTG
Tgfb2	TTGTTGCCCTCCTACAGACTGG	GTAAAGAGGGCGAAGGCAGCAA
Tgfb3	CCTGGCCCTGCTGAACTTG	TTGATGTGGCCGAAGTCCAAC
Tnfa	CCACCACGCTCTTCTGTCT	GCTCCTCCACTTGGTGGTTT
Ucp1	AGGCTTCCAGTACCATTAGGT	CTGAGTGAGGCAAAGCTGATTT
Ucp3	CCTACGACATCATCAAGGAGAA	GCCACCATCTTCAGCATACA