

SUPPLEMENTARY INFORMATION

Suppression of Angiopoietin-like 4 Reprograms Endothelial Cell Metabolism and Inhibits Angiogenesis

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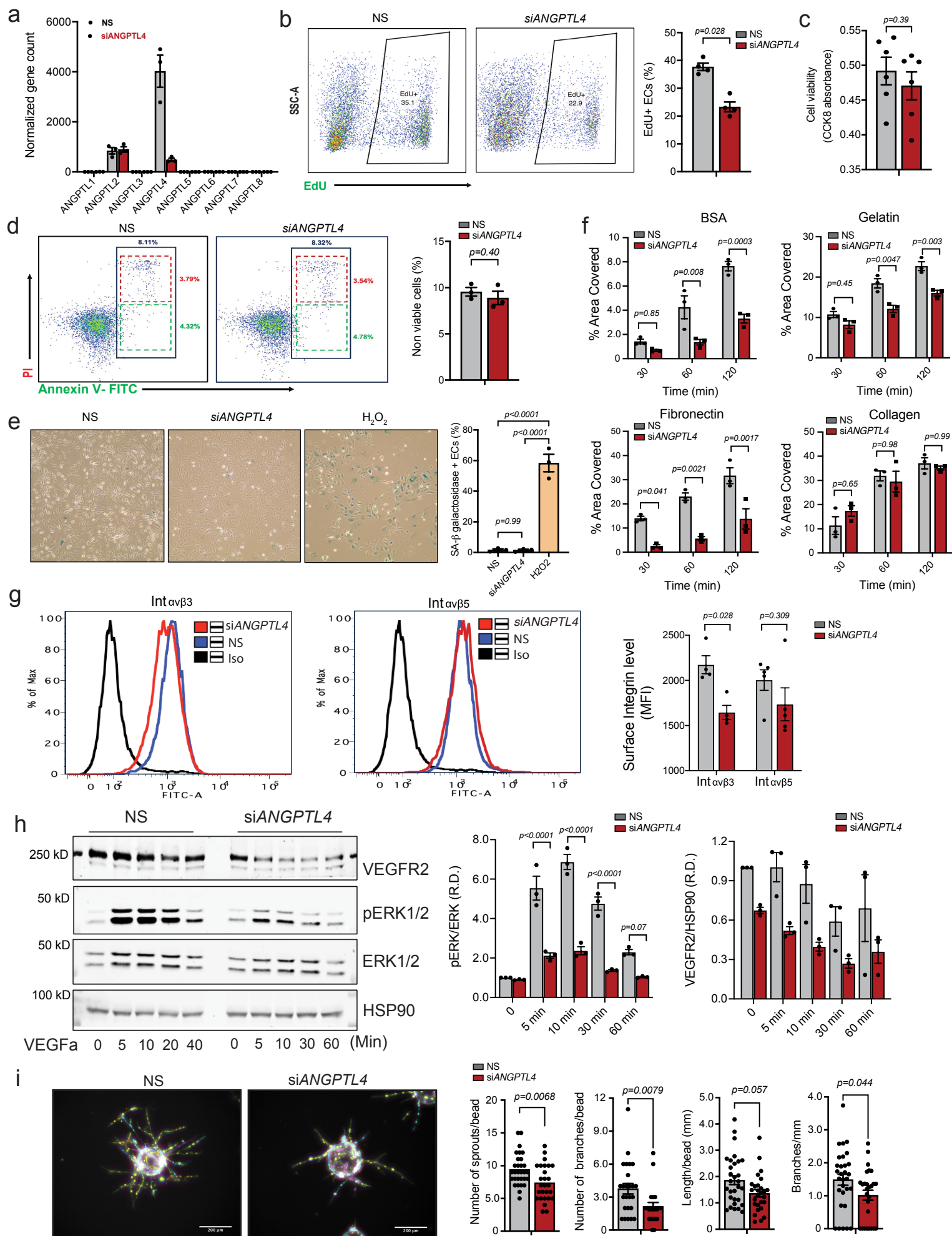
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Supplementary Figures 1-12 with legends

Supplementary Table 1-3



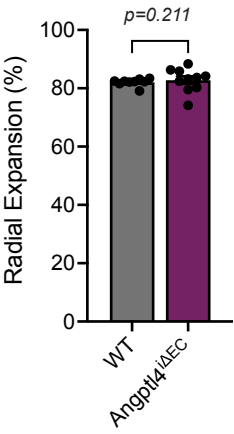
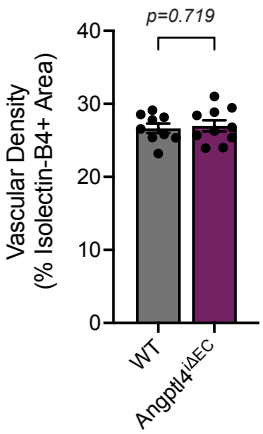
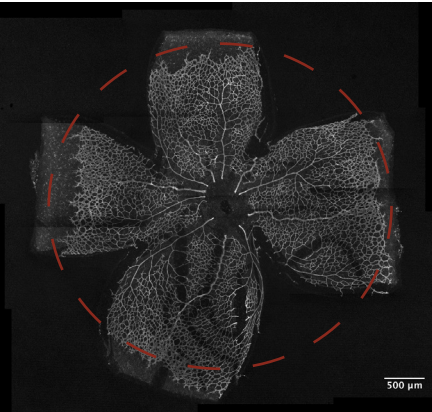
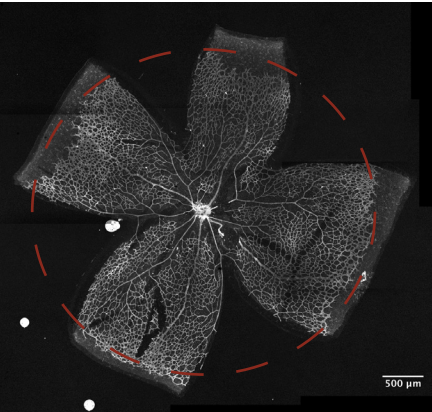
Supplementary Fig. 1. ANGPTL4 is Required for EC Angiogenic Function *in vitro*. **a-h**, HUVECs were transfected for 6h with siRNA against *ANGPTL4* or NS control. Cells were harvested 60h post-transfection unless otherwise indicated. **a**, RNAseq analysis as shown in Fig. 1c (n=3 biological replicates). Normalized log2 expression of ANGPTL family genes in HUVECs. **b**, EdU incorporation in HUVECs assessed by flow cytometry at 48h post-transfection. Representative dot plot (left) and the bar graph showing the % EdU positive cells (right). **c**, At 60h post-transfection, cell viability was assessed by measuring the absorbance of CCK8 at 450nm. **d**, Apoptosis in HUVECs was assessed by staining with Annexin V and propidium iodide (PI), by flow cytometry. Right panel displays the percentage of total non-viable cells, which is the sum of both Annexin V+ and PI-Annexin V double-positive cells, for each group. **e**, Senescence-associated β -galactosidase staining was conducted. HUVECs treated with 100 μ M H₂O₂ used as a positive control. Representative images show the β -galactosidase staining (left). Right panel displays the quantification of β -galactosidase positive cells. **f**, Cell adhesion was evaluated on glass coverslip coated with indicated matrix protein at the indicated time points. Bar graphs represent %area covered on glass coverslips. **g**, Representative histograms for integrins α v β 3 and α v β 5 surface protein levels as accessed by flow cytometry (Left), M.F.I was used for quantification on the right. (**b-g**) n=3-6 independent experiments as shown, performed in duplicate. **h**, Representative immunoblot images showing the level of VEGFR2, pERK1/2 and ERK1/2. HSP90 was used as a loading control. Right panel shows the densitometric quantitation of indicated immunoblots (Representative blots of 1 experiment out of 3 with similar results). **i**, Representative micrograph showing the 3-D HUVECs sprouting in fibrin gels. Right panels showing the quantitation of different parameters as indicated (Data representative of a single experiment. The dots represent the mean quantification derived from four images per technical replicate). Data are represented as means $\pm$ SEM. Mann–Whitney U test in (**b-d**) (**e**) (**g**) and (**i**). One-way and two way ANOVA with Tukey's multiple comparisons test in (**e**) and (**f**) respectively. Scale bar, 200 μ m (**i**). Exact *p* values were shown for each comparison. Source data are provided as a Source data file.

a

P7 Retina

WT

Angptl4^{ΔEC}

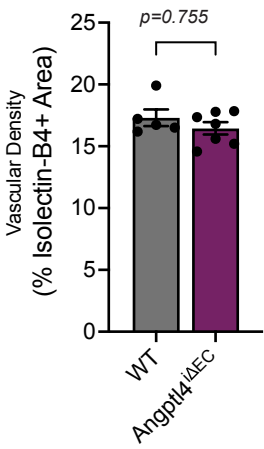
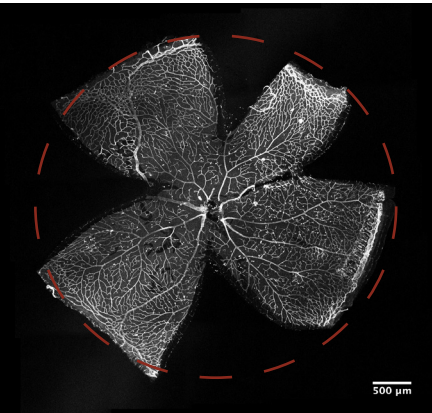
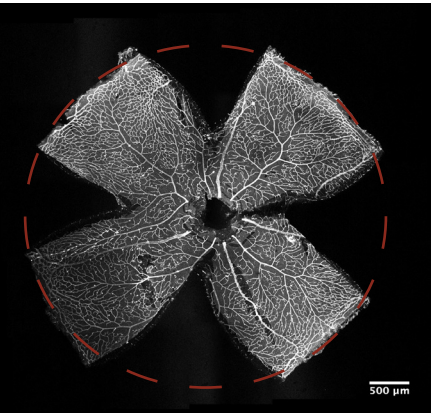


b

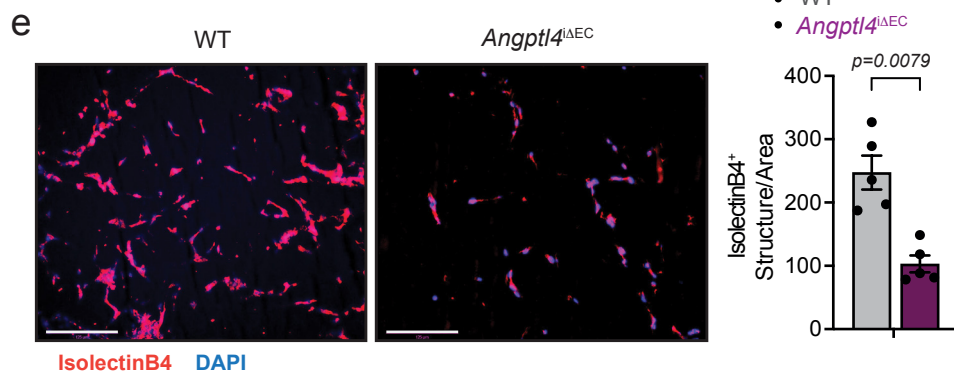
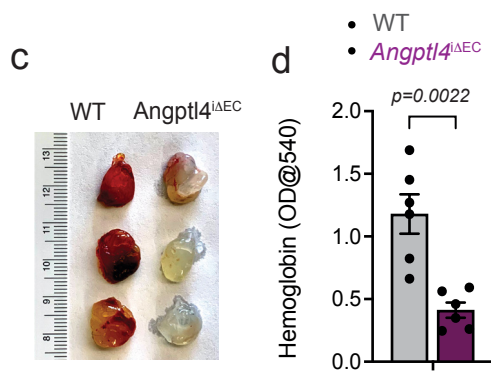
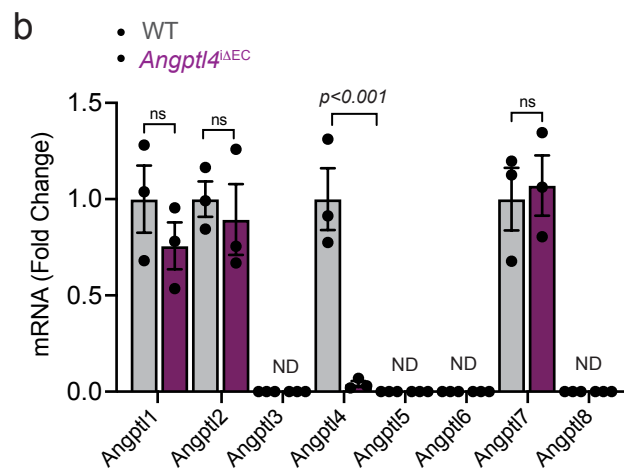
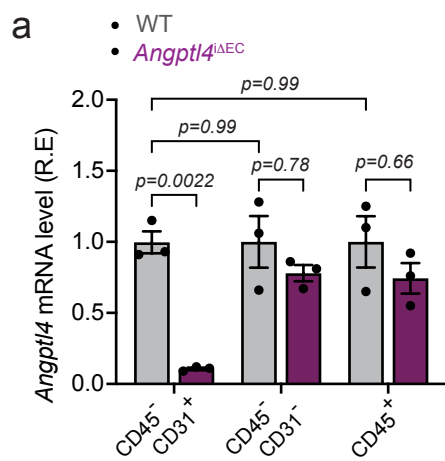
P14 Retina

WT

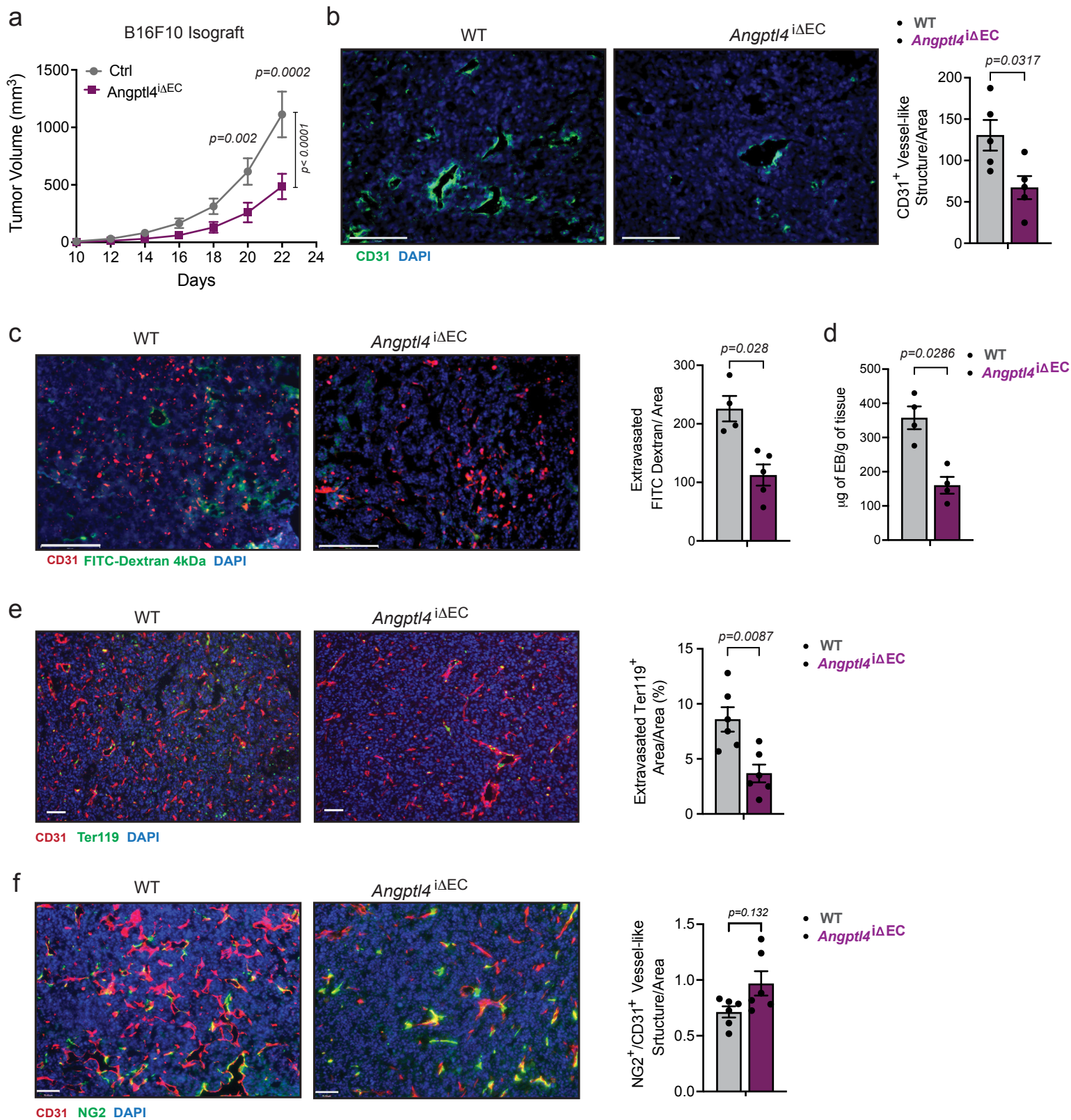
Angptl4^{ΔEC}



Supplementary Fig. 2. EC specific deletion of *Angptl4* does not affect physiological retinal angiogenesis. *Angptl4* deletion on retinal vasculature (mice injected with tamoxifen P1–P3, assessed at P7 and P14). **a**, Retinal whole mounts. Left, representative immunofluorescence of IB4-AF647 staining (grey) in P7 mouse retinas showing no apparent change in vascular density or radial expansion in *Angptl4*^{iΔEC} mice (n= 10) as compared to WT mice (n= 9). Right, quantification of vascular density and radial expansion (n= 10 or 9 per genotype, at least 4 images per mouse was quantified). **b**, Retinal whole mounts. Left, representative immunofluorescence staining of IB4-AF647 (grey) in P14 mouse retinas showing no apparent change in vascular density in *Angptl4*^{iΔEC} mice (n= 7) as compared to WT mice (n= 5). Right, quantification of vascular density (n= 7 or 5 per genotype, at least 4 images per mouse was quantified). All data are represented as means ± SEM. Student's t test (two-tailed, unpaired, nonparametric) in (**a** and **b**). Scale bars, 500 μm. Exact *p* values were shown for each comparison. Source data are provided as a Source data file.

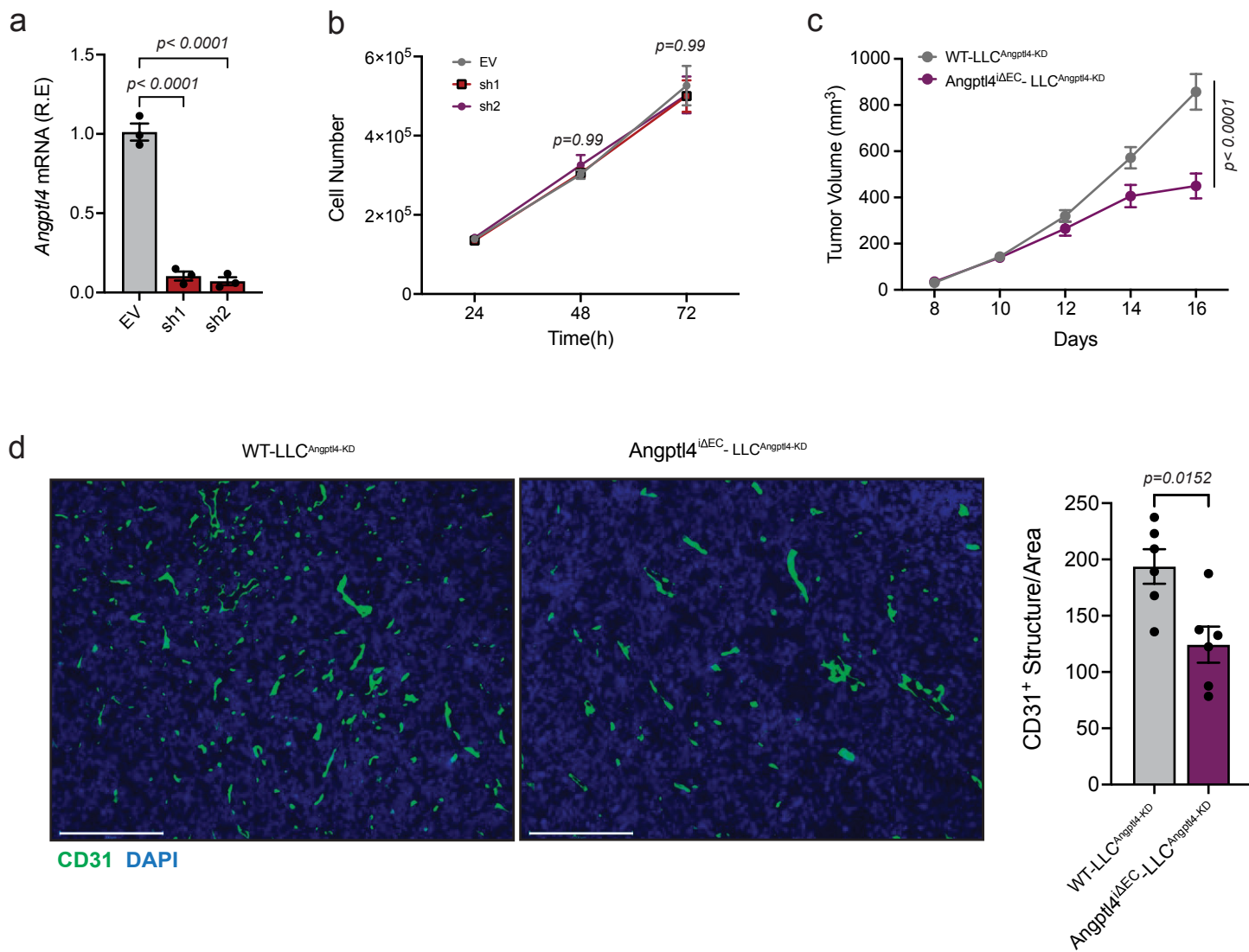


Supplementary Figure 3: EC specific deletion of *Angptl4* Impairs angiogenesis. **a**, Bar graph showing the mRNA levels of *Angptl4* in different cells isolated from lungs of WT and *Angptl4*^{ΔEC} mice (n= 3 for each genotype) via FACS sorting. **b**, qRT-PCR analysis of *Angptl* family genes in MLECs isolated from WT and *Angptl4*^{ΔEC} mice (n= 3 for each genotype). **c**, Representative images showing 3 Matrigel plugs isolated from WT and *Angptl4*^{ΔEC} mice. **d**, Quantification of hemoglobin content in the Matrigel plugs. (n=6 per genotype). **e**, Left, representative images of Matrigel plugs sections stained with isolectin B4. Right, quantitation of EC positive area/vessel area in the plugs (n=5 per genotype). Scale bars, 125μm (**e**) Data are represented as means ± SEM. One-way ANOVA with Tukey's multiple comparisons test in (**a**) Student's t test (two-tailed, unpaired, nonparametric) in (**b**) and Mann–Whitney U test (**d** and **e**). Exact *p* values were shown for each comparison. Source data are provided as a Source data file.

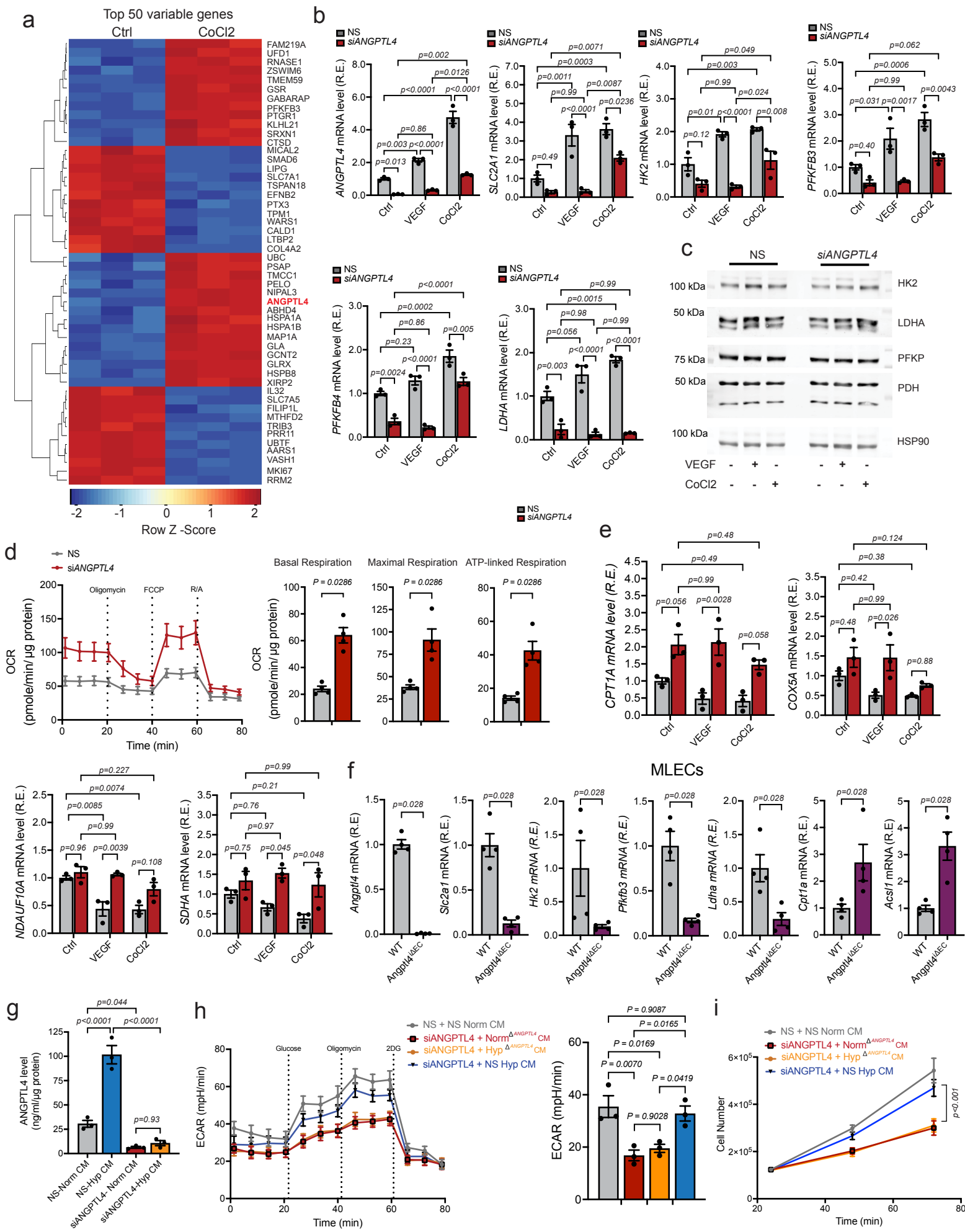


Supplementary Fig. 4. EC specific deletion of *Angptl4* Impairs tumour angiogenesis

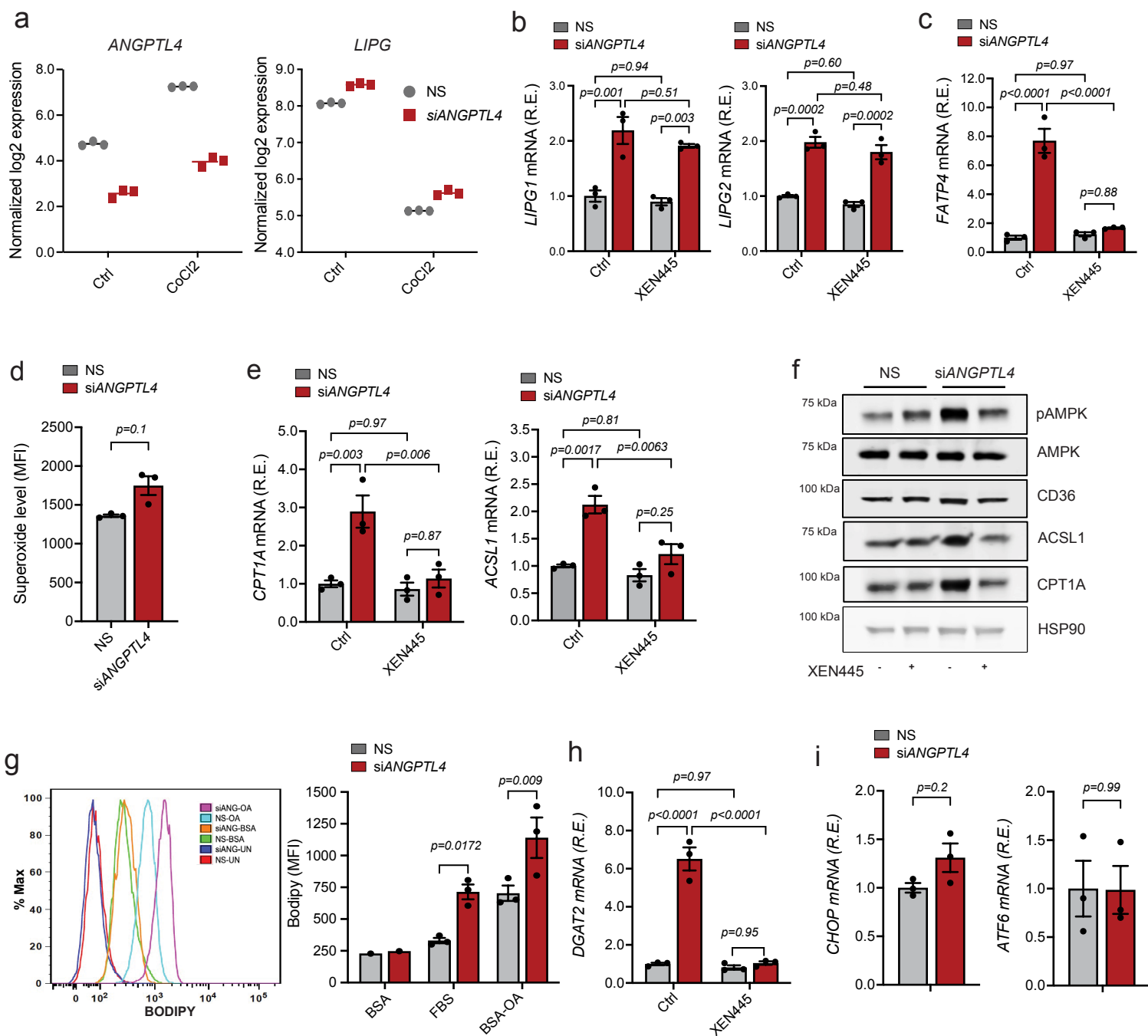
a and **b**, Tumor growth analysis of WT and *Angptl4*^{iΔEC} mice with s.c. injection of B16F10 cells in the dorsal flank (n=9 for each genotype). **a**, Tumor volume. **b**, Left, representative micrographs of CD31 (green) and DAPI immunostaining. Right, Quantification of CD31⁺ vessel-like structures. **c**, Left, representative images of cross-sections of LLC tumours from WT and *Angptl4*^{iΔEC} mice stained for FITC-Dextran (4kDa) (green) and EC marker CD31 (red). Right, quantification of extravasated FITC-Dextran. **d**, LLC tumours bearing WT and *Angptl4*^{iΔEC} mice were intravenously injected with EB. Bar graph represents the quantitation of extravasated EB extracted from the overnight incubation of tumours in formamide solution and absorbance was recorded at 580 nm (n=4 per genotype). **e**, Left, representative images of cross-sections of LLC tumours from WT and *Angptl4*^{iΔEC} mice, stained for Ter119 (green) and EC marker CD31 (red). Right, quantification of extravasated Ter119. **f**, Left, representative micrographs of LLC tumours from WT and *Angptl4*^{iΔEC} mice, stained for NG2 (green) and EC marker CD31 (red). Right, quantification of NG2 covered CD31⁺ vessel-like structures. Right panels (**b**, **c**, **e** and **f**) represent quantification of at least 4 different images from each mouse (**b**, n= 5 quantified per genotype out of 9 randomly selected, **c**, n=4 or 5 quantified per genotype. **e** and **f**, n=6 quantified per genotype out of 6 or 9 randomly selected). Scale bars, 125 μm (**b**), 200 μm (**c**), 70 μm (**e** and **f**). All data are represented as means ± SEM. Two-way ANOVA with Tukey's multiple comparisons test in (**a**) and Mann–Whitney U test in (**b-f**). Exact *p* values were shown for each comparison. Source data are provided as a Source data file.



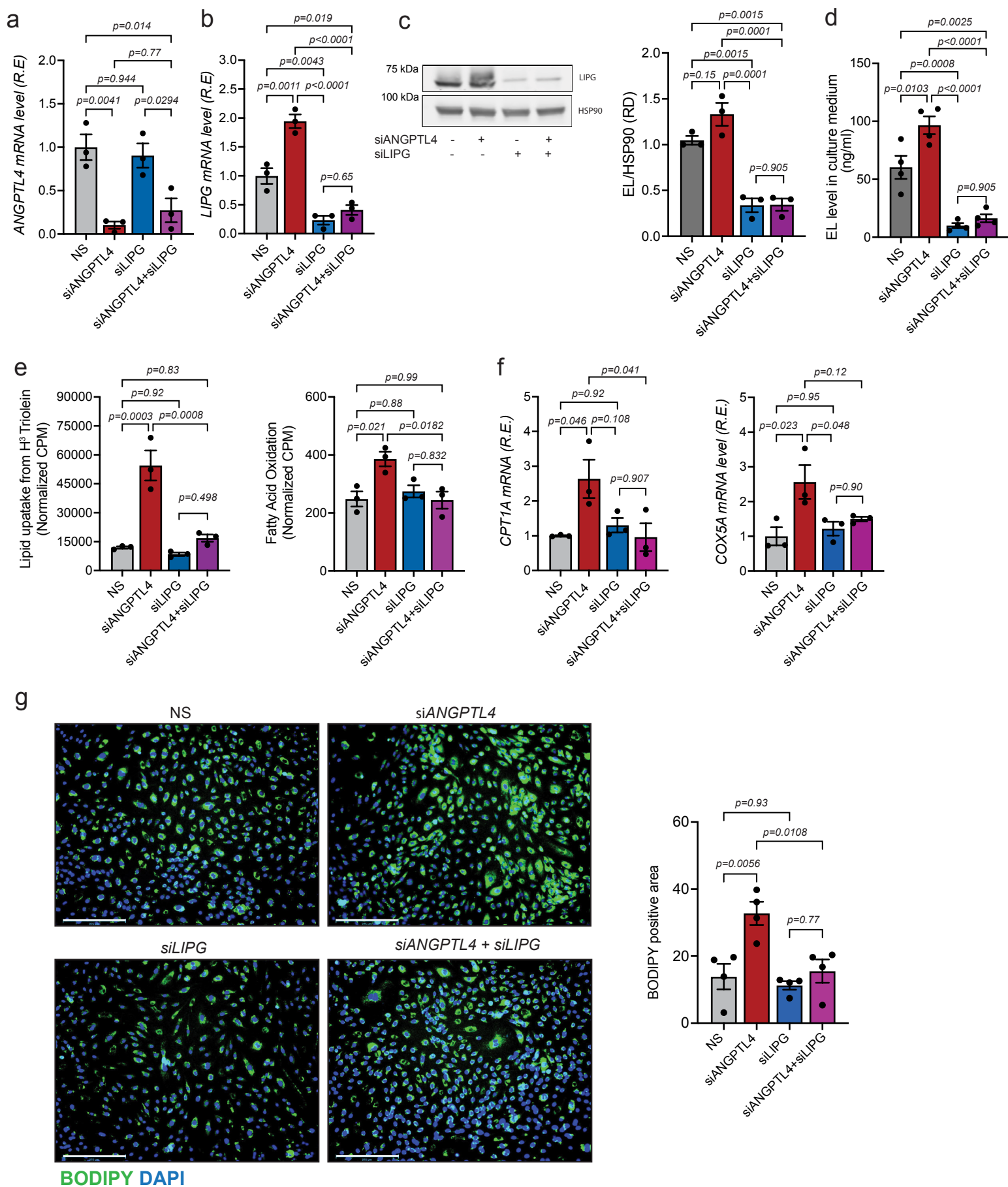
Supplementary Fig. 5. EC specific ANGPTL4 is required for endothelial tumour. **a**, LLCs were transduced with lentivirus carrying shRNAs against *Angptl4*, and stable clones were selected using puromycin. qRT-PCR data show reduced expression levels of *Angptl4* in LLC cells after achieving stable knockdown through lentiviral-mediated shRNA transduction (n= 3 independent experiments). **b**, Cell count analyses for LLC stable clones (EV vs. sh*Angptl4* referred to as LLC-*Angptl4*-KD) reveal no effects on cell proliferation (n= 3 independent experiments). LLC clones with the highest knockdown efficiency (sh2) were selected for the tumor isograft analysis in WT and *Angptl4*^{iΔEC} mice. **c**, Growth curve of LLC lacking ANGPTL4 (LLC-*Angptl4*-KD) isograft in WT and *Angptl4*^{iΔEC} mice (n= 6 for each genotype). **d**, Bar graph shows the end-stage tumour weight of LLC-*Angptl4*-KD derived tumours in WT and *Angptl4*^{iΔEC} mice (n= 6 for each genotype). **e**, Left, representative micrographs of LLC-*Angptl4*-KD derived tumours in WT and *Angptl4*^{iΔEC} mice, stained for CD31 (green) and DAPI (n= 6 for each genotype). Right panels represent quantification of at least 4 different images from each mouse (n=6 per genotype). Scale bars, 200 μm (**e**). All data are represented as means ± SEM. Mann–Whitney U test in (**d** and **e**). One way or Two-way ANOVA with Tukey’s multiple comparisons test in (**a**) and (**b**, **c**) respectively. Exact *p* values were shown for each comparison. Source data are provided as a Source data file.



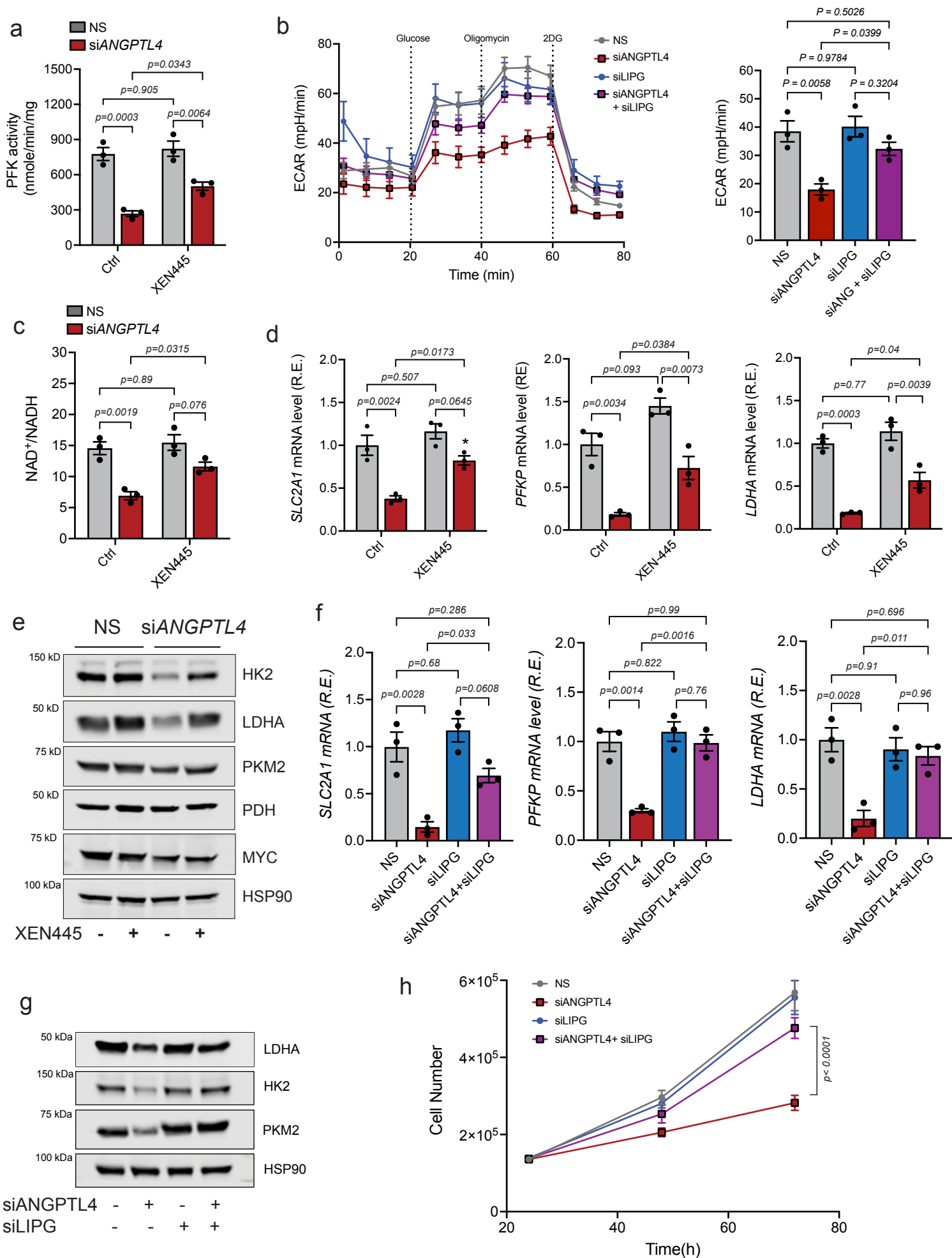
Supplementary Fig. 6. ANGPTL4 modulates VEGF or CoCl₂ induced metabolic response in HUVECs. HUVECs were transfected for 6h with siRNA against *ANGPTL4* or NS control and then treated with VEGF, CoCl₂ or vehicle control as indicated, cells were harvested 60h post-transfection. **a**, Heatmap representing the transcript levels of top 50 variable genes in HUVECs treated with 100 μ M CoCl₂ from the RNAseq analysis, as shown in (Fig. 1c) (n=3 biological replicates). Color key: red, high correlation; blue, low correlation. **b**, HUVECs were transfected with siRNAs against NS or *ANGPTL4* as described above. qRT-PCR analysis of indicated genes. Expression of these genes was normalized with 18S rRNA (n=3 independent experiments). **c**, Immunoblots showing the protein levels of glycolytic enzymes in HUVECs grown under similar conditions as used in (**b**). (Immunoblots represent a single experiment). **d**, Seahorse analysis showing Oxygen consumption rate (OCR). HUVECs were transfected as described above. 48h post-transfection, they were replated on Seahorse plates. Right panel represents the quantification of basal, maximal and ATP linked respiration (n=4 independent experiments). **e**, qRT-PCR analysis of indicated genes related to FAO in HUVECs grown under similar conditions as in (**b**) (n= 3 independent experiments performed in duplicates). **f**, qRT-PCR analysis of indicated genes related to glycolysis and FAO in MLECs isolated from WT and *Angptl4*^{iΔEC} mice (n= 4 for each genotype). Expression of these genes was normalized with 18S rRNA. **g**, HUVECs were transfected with siRNA against *ANGPTL4* as described above and grown in either normoxia or hypoxia for 48h. Quantitative ELISA analysis of secreted levels of ANGPTL4 in the CM (n= 3 independent experiments performed in duplicates). **h**, (Left panel) Seahorse analysis in *ANGPTL4* KD HUVECs grown in the presence of CM enriched (NS-Hyp CM) or depleted (*siANGPTL4*-Hyp CM refers to as Hyp^{ΔANGPTL4} CM) for ANGPTL4. Right panel shows the quantification of ECAR (glycolysis) (n=3 independent experiments). **i**, Proliferation of *ANGPTL4* KD HUVECs grown in the presence of CM enriched or depleted for ANGPTL4, expressed as relative cell number to NS at indicated time points (n=3 independent experiments). Unless indicated above, all data are represented as means $\pm$ SEM. Mann–Whitney U test in (**d** and **f**). Two-way ANOVA with Tukey's multiple comparisons test in (**b**, **e**, and **g-i**). Hyp- Hypoxia, Norm-Normoxic condition, CM- Conditioned Medium. Exact *p* values were shown for each comparison. Source data are provided as a Source data file.



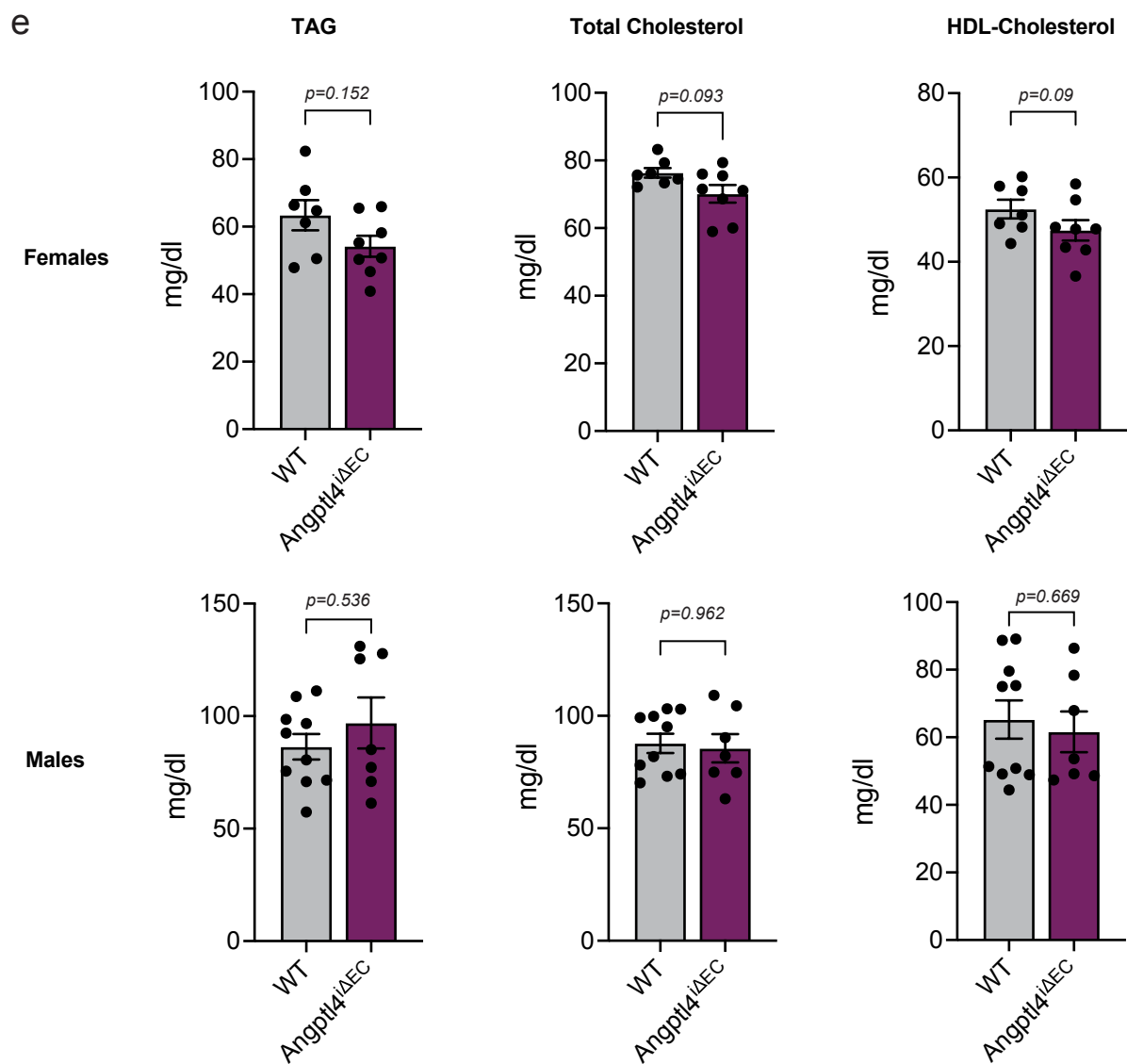
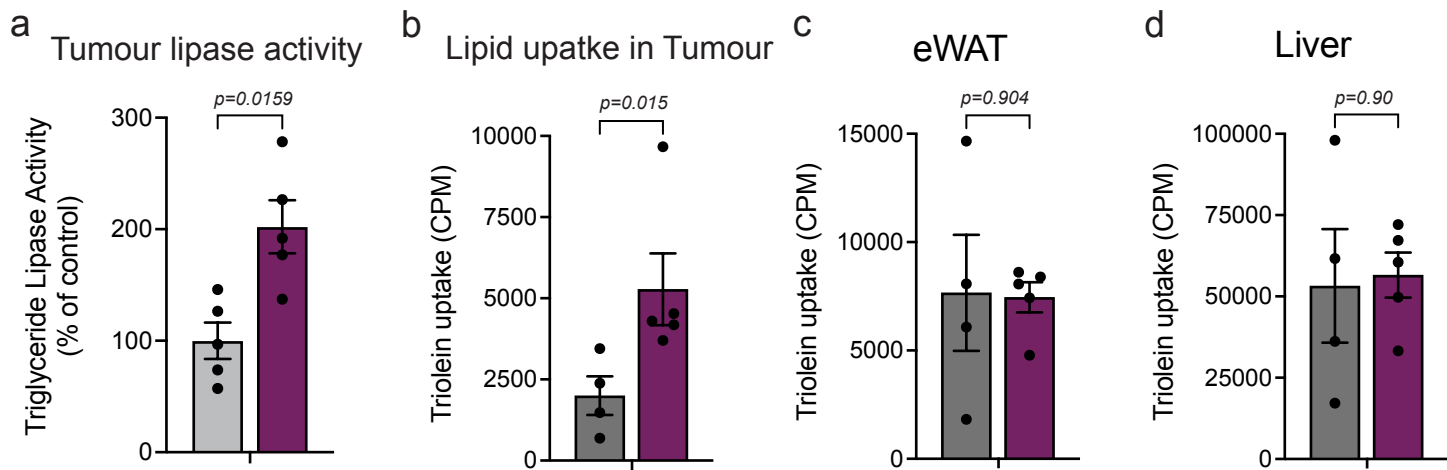
Supplementary Fig. 7. ANGPTL4 regulates EC lipase activity and FA metabolism. **a**, RNAseq analysis showing the normalized log2 expression of *ANGPTL4* and *LIPG* in HUVECs as in (Fig 3a) (n=3 biological replicates). **b** and **c**, HUVECs were transfected for 6h with siRNA against *ANGPTL4* or NS control and then treated with XEN445 or vehicle control, cells were harvested 60h post-transfection. qRT-PCR analysis of showing expression of *LIPG1*, *LIPG2* genes (**b**) and *FATP4* (**c**). Expression of these genes was normalized with 18S rRNA (**b** and **c**, n= 3 independent experiments as shown, performed in duplicate). **d**, Superoxide level in HUVECs post 60h transfection with *ANGPTL4* siRNA as determined by mean fluorescence intensity (MFI) of DHE (dihydroethidium) via Flow cytometry (n=3 independent experiments). **e**, qRT-PCR analysis of *CPT1A* and *ACSL1* genes in HUVECs grown under the conditions described above. Expression of these genes was normalized with 18S rRNA (n=3 independent experiments). **f**, Immunoblots of indicated molecules in whole cell lysate of HUVECs grown under the conditions described above. HSP90 was used as loading control (Immunoblots represent a single experiment). **g**, HUVECs were transfected as described above. 60h post-transfection, HUVECs were FBS-starved for 12h followed by loading with either BSA, 20% FBS or BSA-Oleic acid for 6h. Left, representative histogram showing the neutral lipid storage in HUVECs as determined by staining with BODIPY via Flow cytometry. Right, M.F.I was used for quantification on the right. (n=3 independent experiments). **h**, mRNA level of *DGAT2* gene in HUVECs grown under the similar conditions used in (**b**). Expression of *DGAT2* gene was normalized with 18S. **i**, mRNA levels of ER stress marker genes in HUVECs grown under the similar conditions used in (**d**) (n=3 independent experiments performed in triplicate). All data are represented as means $\pm$ SEM. Two-way ANOVA with Tukey's multiple comparisons test in (**b**, **c**, **e**, **g** and **h**). Mann–Whitney U test in (**d** and **i**). Exact *p* values were shown for each comparison. Source data are provided as a Source data file.



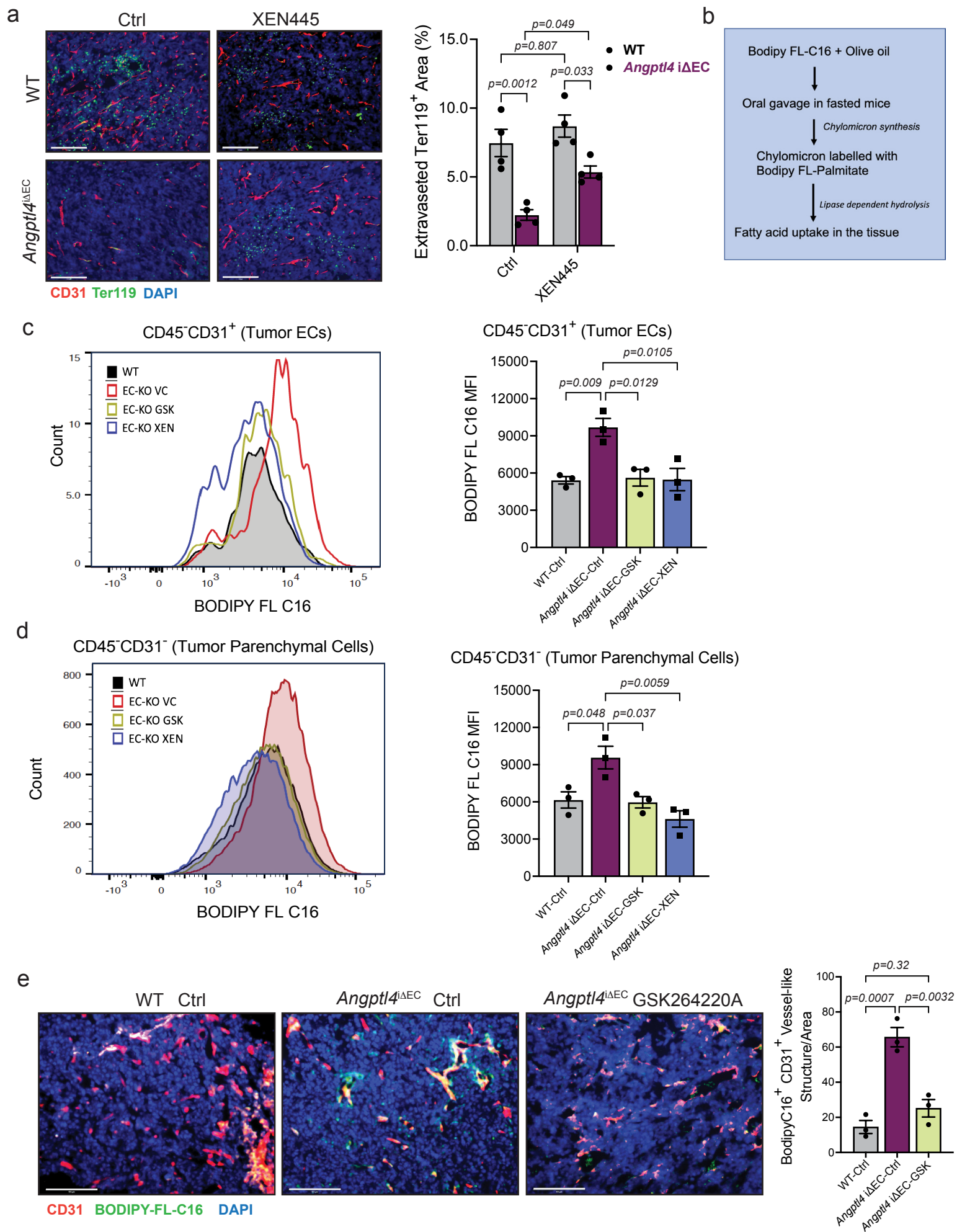
Supplementary Fig. 8. ECs lacking *ANGPTL4* exhibit elevated FA uptake and storage in lipase dependent manner. **a-e**, HUVECs were co-transfected with siRNAs against *ANGPTL4* and/or *LIPG*, or with a NS control, for 6h. Cells were then harvested 60h post-transfection. **a** and **b**, mRNA levels of *ANGPTL4* and *LIPG* showing the knockdown efficiency of these genes (n= 3 independent experiment). **c**, Representative immunoblots of EL in whole cell lysate of HUVECs grown under the conditions described above (Representative blots of 1 experiment out of 3 with similar results). HSP90 was used as loading control. Right panel shows the densitometric quantitation of immunoblots (n=3 independent experiments). **d**, Bargraph shows the secreted level of EL in the heparinized culture medium from the HUVECs grown in the similar conditions as used in (**a**), determined by quantitative ELISA (n= 4 independent experiment). **e**, Lipid uptake from [³H]-triolein in HUVECs under the similar growth condition used in (**a**). Right panel shows FAO as determined by oxidation of C¹⁴-Palmitate (n= 3 independent experiment). **f**, qRT-PCR analysis of *CPT1A* and *COX5A* genes in HUVECs grown under similar conditions used in (**a**) (n= 3 independent experiment). **g**, Representative micrograph showing the neutral lipid accumulation in HUVECs stained with BODIPY (Representative experiment out of 2 with similar results, dots are the mean of the quantification of 4 images per technical replicates). Scale bars, 275 μ m (**g**). All data are represented as means $\pm$ SEM. Two-way ANOVA with Tukey's multiple comparisons test in (**a-g**). Exact *p* values were shown for each comparison. Source data are provided as a Source data file.



Supplementary Fig. 9. *ANGPTL4* knockdown in HUVECs rewire glucose metabolism via activating LIPG. **a**, HUVECs were transfected for 6h with siRNA against *ANGPTL4* or NS control and then treated with XEN445 or vehicle control, cells were harvested 60h post-transfection. Enzyme activity of PFK (n= 3 independent experiment performed in duplicates). **b**, HUVECs were co-transfected with siRNAs against *ANGPTL4* and/or LIPG, or with a NS control, for 6h. 48h post-transfection, they were replated on Seahorse plates. Seahorse analysis evaluates the extracellular acidification rate (glycolysis). Bar graph on the right is quantification of extracellular acidification rate (n=3 independent experiments). **c**, Determination of NAD⁺/NADH in HUVECs upon silencing *ANGPTL4* as described in (a) (n= 3 independent experiment performed in triplicates). **d**, qRT-PCR analysis of glycolytic metabolism genes in HUVECs grown under similar conditions as in (a). Expression of these genes was normalized with 18S (n= 3 independent experiment performed in duplicates). **e**, Representative immunoblots showing the protein levels of glycolytic enzymes in HUVECs grown under similar conditions as in (a). (Representative blots of 1 experiment out of 3 with similar results). **f**, qRT-PCR analysis of glycolytic metabolism genes in HUVECs grown under similar conditions used in (b). Expression of these genes was normalized with 18S (n= 3 independent experiment performed in duplicates). **g**, Immunoblots showing the protein levels of glycolytic enzymes in HUVECs (Immunoblots represent a single experiment). **h**, Proliferation of HUVECs grown under similar conditions used in (b), expressed relative cell number to NS at indicated time points post-transfection (n= 3 independent experiment). All data are represented as means $\pm$ SEM. Two-way ANOVA with Tukey's multiple comparisons test in (a-h). Exact *p* values were shown for each comparison. Source data are provided as a Source data file.

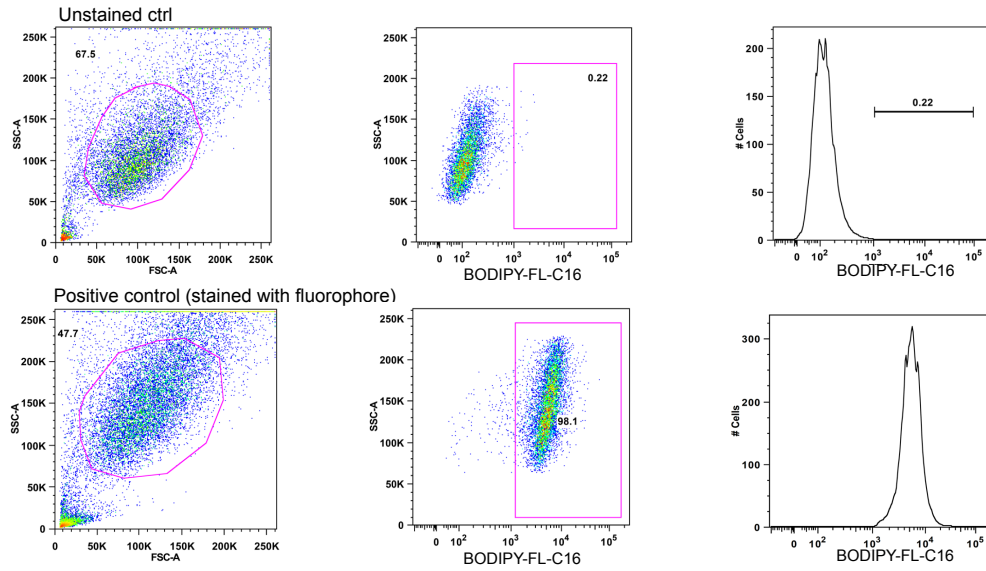


Supplementary Fig. 10. EC specific deletion of *Angptl4* does not affect plasma lipid levels *in vivo*. **a**, Triglyceride lipase activity in the tumour from WT and *Angptl4*^{iΔEC} mice. **b-d**, Lipid uptake in the tumour (**b**), eWAT (**c**), and liver (**d**) from [³H]-triolein in WT and *Angptl4*^{iΔEC} mice (n= 4-5 mice per genotype). **e**, Levels of triglycerides (TAG), total cholesterol and HDL cholesterol in the plasma from females (upper panel) and males (lower panel) WT and *Angptl4*^{iΔEC} mice (females n= 7-8 mice in each group; males n= 7-10 mice in each group). All data are represented as means ± SEM. Mann–Whitney U test in (**a-e**). Exact *p* values were shown for each comparison. Source data are provided as a Source data file.

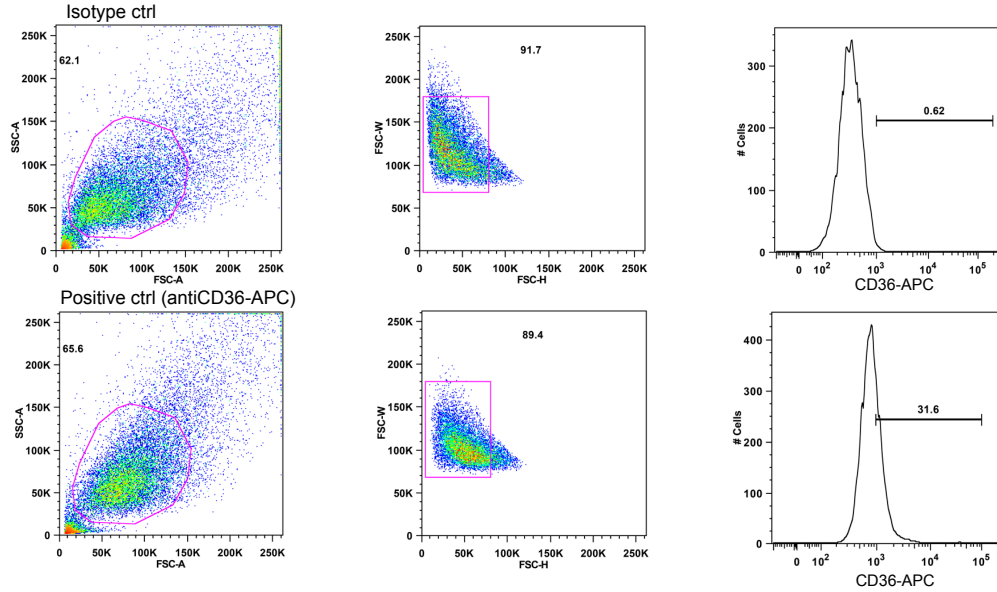


Supplementary Fig. 11. EC specific ANGPTL4 regulates vascular lipolysis *in vivo* via inhibiting. Tumor analysis of WT Ctrl (n=4), WT XEN445 treated (n=4), *Angptl4*^{ΔEC} Ctrl (n=4) mice and *Angptl4*^{ΔEC} XEN445 treated (n=4) with s.c. injection of LLCs in the dorsal flank as described in (Fig. 2) **a**, Left, representative micrographs Ter119 (green) and CD31 (red) immunostaining. Right panel represent quantification of extravascular Ter119 in tumours.(n=4 quantified per genotype, at least 4 different images from each mouse were quantified). **b**, Scheme showing the experimental strategy. **c** and **d**, Flow cytometry analysis of BODIPY-FL-C16 uptake in tumour parenchymal (CD45⁻ CD31⁻ cells) (**c**), and tumour ECs (CD45⁻ CD31⁺) (**d**) from WT and *Angptl4*^{ΔEC} treated with or without XEN445 or GSK264220A (n= 3 mice per group). Right panels, MFI of BODIPY-FL-C16 is quantified for the indicated groups. **e**, Left, representative micrographs of LLC tumours from WT and *Angptl4*^{ΔEC} mice treated with GSK264220A, stained for EC marker CD31 (red) and fluorescent FA (BODIPY-FL-C16) (green). Right panel represents quantitation of CD31 and BODIPY-FL-C16 double positive vessel like structures in tumours. (n= 3 mice per group, at least 4 different images from each mouse were quantified). Scale bars, 125 μm (**a** and **e**). All data are represented as means ± SEM. Two-way ANOVA with Tukey's multiple comparisons test in (**a**). One-way ANOVA with Tukey's multiple comparisons test in (**c-e**). Exact *p* values were shown for each comparison. Source data are provided as a Source data file.

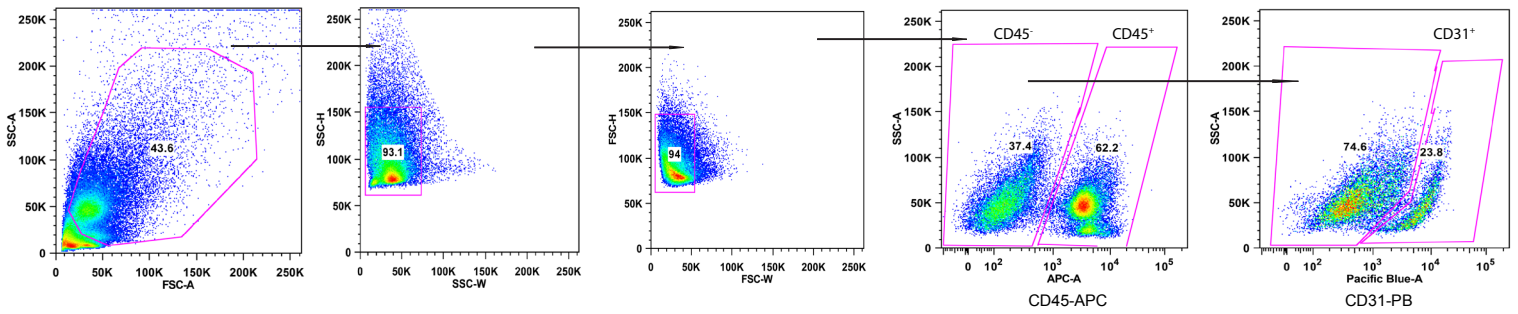
a Gating strategy for analysing labeled Fatty acids (BODIPY-FL-C16) or glucose (2NBDG)



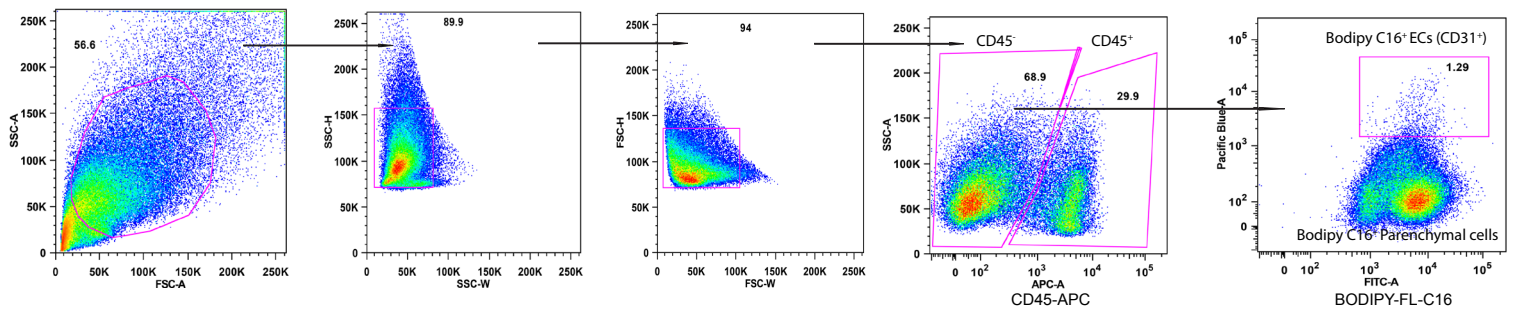
b Gating strategy for analysing surface expression of CD36



C Gating strategy for isolating ECs from mouse lungs



d Gating strategy for BODIPY FL C16 uptake in tumor ECs and Parenchymal cells



Supplementary Fig. 12. **a**, Gating strategy for the FACS tests applied for the detection of fluorescently labelled fatty acid (BODIPY-FL-C16) and glucose (2NBDG) in HUVECs as shown in Fig 4, Fig. 5, Fig. 6 and Supplementary Fig.7. **b**, Gating strategy for the FACS tests applied for the detection of surface expression of CD36 as shown in Fig. 5. **c**, Gating strategy for sorting ECs (CD45⁻CD31⁺) from the mouse lungs as shown in Supplementary Fig. 4. **d**, Gating strategy for the FACS tests applied for the detection of fluorescently labelled fatty acid (BODIPY-FL-C16) in the tumor ECs and other parenchymal cells as shown in Supplementary Fig.11.

Supplementary Table 1: List of primers

Primers

Human qRT PCR Primers

Gene name	Forward (5'-3')	Reverse (5'-3')
hANGPTL4	CTCCACTTGGGACCAGGATCA	ATGGCTGCAGGTGCCAAAC
hLIPG1	CCACCCAATGGAAGGATTC	GCCATCACAGGAGACCAGTT
hLIPG2	GGCCTCTCAGTCTTGGTACAAC	CGGGGTTGAGACAGGTAGC
hHK2	ATCCCCCAGCACTGATGTT	TCACATCCTCAGTGGAGACCT
hPFKFB3	TCTCTCCTCCCACCTGGAC	GGTCACACTTCTGCCATCAA
hPFKFB4	CTTCAGGCCTGCTCTCACA	CTCGAGCACTTCGGTGTGT
hLDHA	TTTTCCCAGTGAGTCACATCC	TTGGAAGAATTATGCACAAGACA
hSLC2A1	CAGATGATGCGGGAGAAGA	ACAGCTGCAGCACCCACAG
hSDHA	CAGCACAGGGAGGAATCAAT	TCACGGTGTCGTAGAAATGC
hNDAUF10A	TTCCTTCAACCTTGGTGGTT	CTGACCACAGGGCAGACC
hCPT1A	TGGAAAGTCTCAGCATATGGAA	CCAGCACATGAACTTGGTGA
hCOX5A	CAAAGTGTAACCGCATGGAT	TCCAGGTAAGTGTTCACACTCAA
hFATP4	AGCAGTGTAGCCAACTTCCT	CACGAACTCATTGCGGTTCT
hDGAT2	GTTTTTCTTGGGTGGCTGAT	CTGAGGCCACCAGTAAGCAT
hATF6	GAGTCATCGCGTCTCTCCAC	GGCCTCAGAGTTGACGGAAG
hCHOP	AAGGCACTGAGCGTATCATGT	TGAAGATACACTTCCTTCTTGAACA
hACSL1	AACAGACGGAAGCCCAAGC	TCGGTGAGTGACCATTGCTC

Mouse qRT PCR Primers

Gene name	Forward (5'-3')	Reverse (5'-3')
<i>mAngptl1</i>	TCTTCTCTGCTGCCCAC	GTGAGCCTCTGCACAATC

<i>mAngptl2</i>	GGGACCTTAACTGTGC	GAATGGCTACAGGTACCA
<i>mAngptl3</i>	GATGGCTCTGTCAATT	CATCAATGTTTCCAAACCC
<i>mAngptl4</i>	ACTTCAGATGGAGGCTGGAC	TCCGAAGCCATCCTTGTAGG
<i>mAngptl5</i>	GTCACCTAAGGCCAGATGCTT	TGCACTCCCTGACTTGCATT
<i>mAngptl6</i>	TATCCACCGGCTCACC	TCTGCGTAGCGTGCATTG
<i>mAngptl7</i>	CACCGTGAGGCATGTG	GGTTGGTCTTTATGAACTG
<i>mAngptl8</i>	GCTTTACACCTTCGAGCTGA	ATCCAGGTAGTCTCAGGCTG
<i>mCPT1a</i>	TTGATCAAGAAGTGCCGGACGAGT	GTCCATCATGGCCAGCACAAAGTT
<i>mLdha</i>	CCTGCACCAAACATGCCTA	CACTGGAGCCAGGTTATACGA
<i>mSlc2a1</i>	ATCATGGGCAATGCAGACTT	GTAGCAGGGCTGGGATGA
<i>mHk2</i>	GTTTGTACATTTATTTGCCCTCCT	AGCGTGCATCTCGTGTA AAA
<i>mPfkfb3</i>	CAGAGGACAGCTTGACAGGA	GAGGCCTGGGCTCAATCT
<i>mAcs1</i>	TGGGGTGGAATCATCAGCC	CACAGCATTACACACTGTACAACG
m18S	TTCTGGCCAACGGTCTAGACAAC	CCAGTGGTCTTGGTGTGCTGA
<i>mAngptl4</i> Exon4	GGGACTGCCAGGAACTCTTC	AATGGTGGAGACCCCAGAGG
<i>mAngptl4</i> Exon5	GAGGCTGGACAGTGATTCAGAGA	GGGATCTCCGAAGCCATCCTT

Genotyping Primers

	Forward (5'-3')	Reverse (5'-3')
<i>Angptl4</i> Flox (genotyping)	TGTGGGGTAGGCTTTAGGTG	GCTTCTCTCCCTGAGCCTTT
ERT2 (genotyping)	ATACCGGAGATCATGCAAGC	ATGTGAACCAGCTCCCTGTC

Supplementary Table 2. List of siRNAs and shRNAs

siRNAs		
ON-TARGETplus Human <i>ANGPTL4</i> siRNA (SMARTPool)	Horizon Discovery	Catalog # L-007807-00-0020
Accell Human <i>LIPG</i> siRNA (SMARTPool)	Horizon Discovery	Catalog # E-009601-00-00010
ON-TARGETplus Non-targeting Pool	Horizon Discovery	Catalog# D-001810-10-20
SMARTvector mouse <i>Angptl4</i> Lentiviral shRNA (set of 3)	Horizon Discovery	Catalog ID:V3SM11244- 01EG57875 SET1: V3SM11241-233353210 Clone Id: V3SVMM01_13290866 SET2: V3SM11241-237020981 Clone Id: V3SVMM01_16958618 SET3: V3SM11241-231728343 Clone Id: V3SVMM01_11665979
SMARTvector Empty Vector Control	Horizon Discovery	Catalog ID:VSC11649

Supplementary Table 3: List of Antibodies

Antibodies		
Immunoblots		
Phospho-AMPK α (Thr172) (40H9) Rabbit mAb	Cell Signaling Technology	Cat #2535
AMPK α (D5A2) Rabbit mAb	Cell Signaling Technology	Cat #5831
Phospho-p44/42 MAPK (Erk1/2) (Thr202/Tyr204) (D13.14.4E) XP [®] Rabbit mAb	Cell Signaling Technology	Cat #4370
p44/42 MAPK (Erk1/2) (137F5) Rabbit mAb	Cell Signaling Technology	Cat #4695
HSP90	BD Biosciences	Cat# 610419
VEGF Receptor 2 (55B11) Rabbit mAb	Cell Signaling Technology	Cat #2479
Hexokinase II (C64G5) Rabbit mAb	Cell Signaling Technology	Cat #2867
LDHA (C4B5) Rabbit mAb	Cell Signaling Technology	Cat #3582
PFKP (D4B2) Rabbit mAb	Cell Signaling Technology	Cat #8164
PKM2 (D78A4) XP [®] Rabbit mAb	Cell Signaling Technology	Cat #4053

PDH (C54G1) Rabbit mAb	Cell Signaling Technology	Cat #3205
ACSL1 (D2H5) Rabbit mAb	Cell Signaling Technology	Cat# 9189
CPT1A [8F6AE9] Mouse mAb	Abcam	Cat# ab128568
CD36 Rabbit pAB (Clone 948)	Proteintech	Cat# 18836-1-AP
NG2 (clone: 546930) mAb	Invitrogen	Cat# MA5-24247
SECONDARY ANTIBODIES		
Goat anti-Rabbit IgG, Alexa Fluor 800	Invitrogen	Cat# A32735
Goat anti-Mouse IgG, Alexa Fluor 800	Invitrogen	Cat# A32730
Goat anti-Mouse IgG, Alexa Fluor 680	Invitrogen	Cat# A21058
Goat anti-Rabbit IgG, Alexa Fluor 680	Invitrogen	Cat# A21109
Immunofluorescence/Flowcytometry		
CD31	BD Bioscience	Cat# 550274
CD31-PE, Clone MEC 13.3	BD Bioscience	Cat# 553373
CD31-BV421, Clone MEC 13.3	BD Bioscience	Cat# 562939
Ter119-FITC	BioLegend	Cat# 116205
α -SMA-eFluor™ 660	Invitrogen	Cat# 50-9760-82
Ki67-APC, Clone: 16A8	BioLegend	Cat# 652405
Ki67 PE	BioLegend	Cat# 652404
CD36-FITC	Invitrogen	Cat# 11-0369-42
CD36-APC	BioLegend	Cat# 102611
CD45-PE,	BioLegend	Cat# 157603

CD45-APC	BioLegend	Cat# 103112
Anti-BrdU-PE	eBioscience	Cat# 12-5071-41
Integrin alpha v beta 3	Invitrogen	Cat# 11-0519-41
Anti-ERG	Abcam	Cat# ab92513