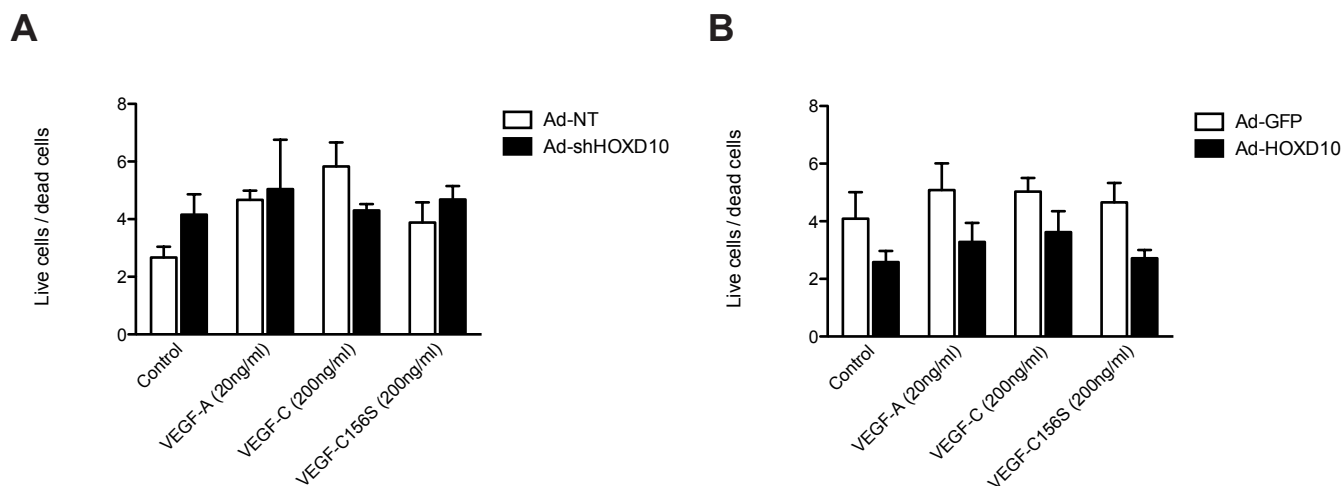
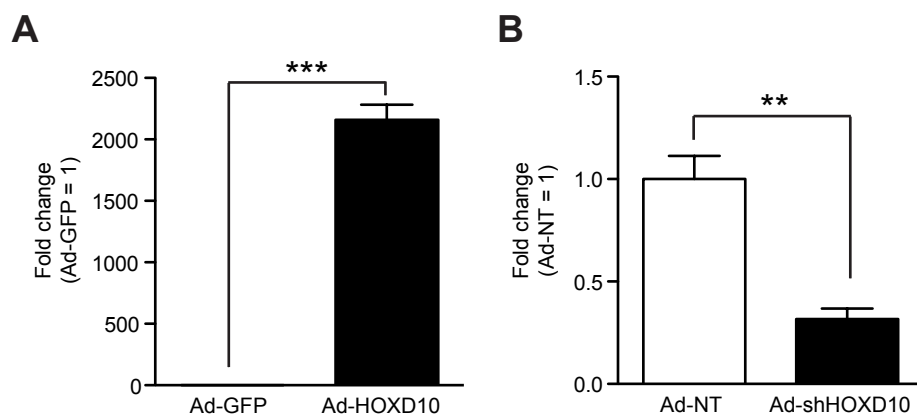


Supplementary Figure 1: HOXD10 expression in VEGF-C156S stimulated LEC

(A) Western Blot detection of HOXD10 in LEC treated with VEGF-C156S for 45 min or 240 min. (B) Quantification of HOXD10 band intensity normalized to b-actin band intensity. Bars represent means + SD (n =3). There were no significant changes in the HOXD10 protein level (one-way ANOVA with Dunnett's post-test).



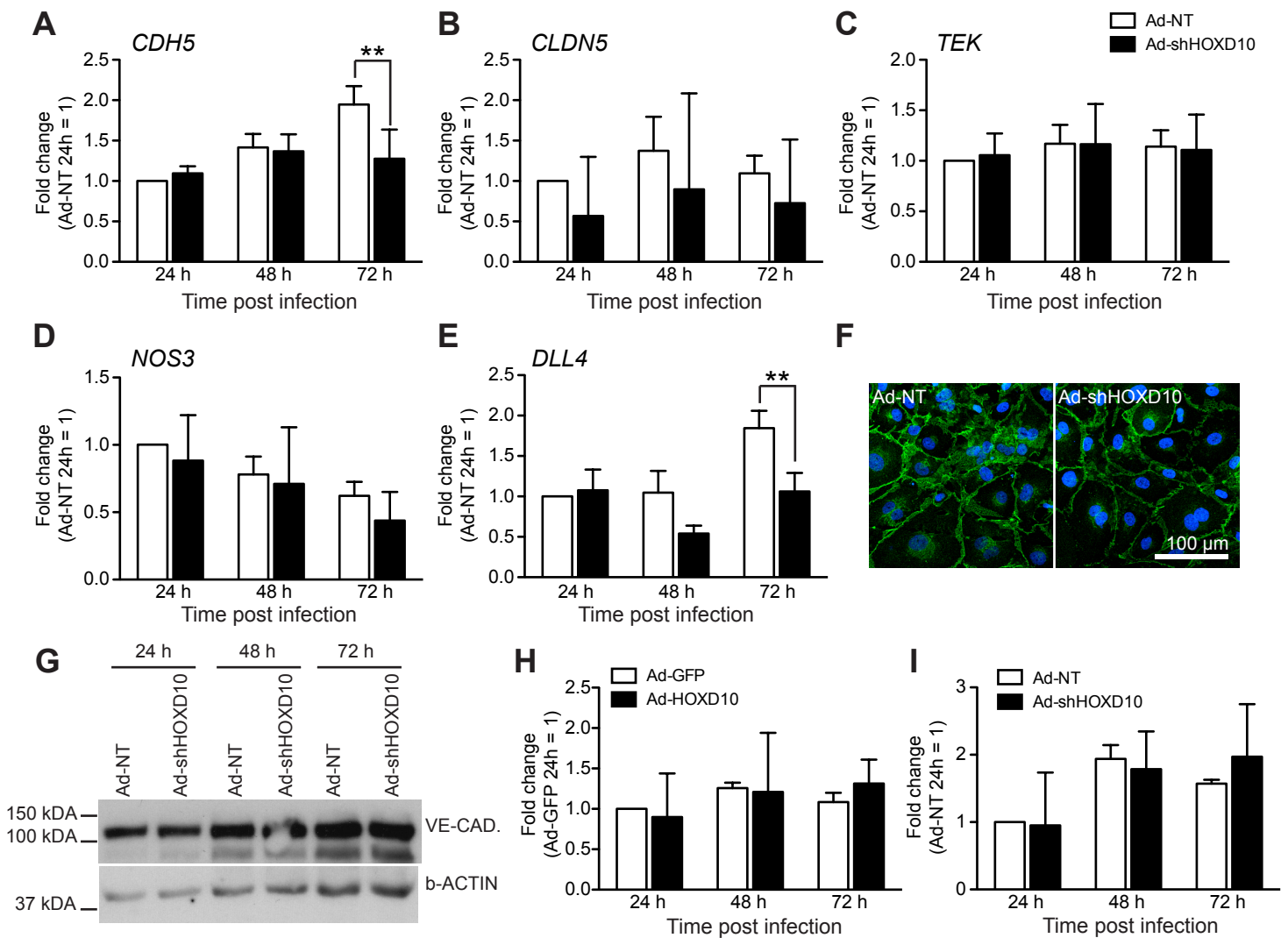
Supplementary Figure 2: Cell death assessment in LEC after knockdown and overexpression of HOXD10. LEC were infected with adenovirus to knock down (A) or overexpress HOXD10 (B), and incubated with the indicated growth factors for 72 h. Cell death was assessed by manual counting using trypan blue-excluding viable cells and trypan blue-stained dead cells.



Supplementary Figure 3: Confirmation of HOXD10 overexpression / knockdown in samples used for CAGE RNA sequencing

qPCR confirmation of efficient overexpression (A) and knockdown (B) of HOXD10 in LEC 24h after infection with Ad-HOXD10 and Ad-shHOXD10, respectively. The same samples were used for the CAGE RNA sequencing in Figure 6. Bars represent means + SD (n = 3).

p<0.01, *p<0.001 (Student's t-test).



Supplementary Figure 4: HOXD10 knockdown does not significantly affect expression of CDH5, CLDN5, TEK, NOS3, DLL4 and HOXD8 in LEC

(A-E) LEC were infected with adenovirus to knock down HOXD10 (Ad-shHOXD10) or control virus (Ad-NT), and expression of selected target genes CDH5 (A), CLDN5 (B), TEK (C), NOS3 (D) and DLL4 (E) was determined by qPCR after 24 h, 48 h, and 72 h. Bars represent mean + SD (pooled data from 4 individual experiments). ** $p < 0.01$ (Two-way ANOVA with bonferroni post-test).

(F) Representative microscopic images of Ve-Cadherin staining in LEC monolayers 72 h after infection with Ad-NT and Ad-shHOXD10. (G) Western Blot analysis of total cellular Ve-Cadherin in LEC 24 h, 48 h, and 72 h after infection with Ad-NT and Ad-shHOXD10. (H-I) LEC were infected with adenovirus to overexpress (H) or knock down HOXD10 (I), and expression of HOXD8 was determined by qPCR after 24 h, 48 h, and 72 h. Bars represent mean + SD. (pooled data from 3 individual experiments).

Supplementary Table 1: Table of predicted transcription factor activity according to oPOSSUM3 Z-scores and Fisher-Scores

List of predicted active transcription factors identified by oPOSSUM3 analysis, showing significant enrichment of corresponding TFBS in promoters of differentially expressed genes (Z-score) and significant enrichment of the number of promoters with at least one corresponding TFBS (Fisher-Score) at all time points of the analysis.

above significance threshold (>mean+1.5xSD)

time point	TF	Z-score	Fisher -score	time point	TF	Z-score	Fisher -score	
15 min	HOXD10	24.909	12.582	150 min	SRF	27.17	6.967	
	EGR1	22.816	10.527		HOXD8	10.918	3.046	
	NKX3-1	22.093	7.343		TBP	10.705	3.943	
	ELK1	21.164	6.878		HOXA7_2	10.554	3.856	
	E2F1	20.657	14.561		PDX1	7.03	3.918	
	FOXD1	20.348	5.778		HOXA7-2	10.554	3.856	
	NKX2-5	18.322	1.62		LHX3	5.067	3.551	
	SRY	18.044	5.494		GFI	3.436	3.455	
	ELK4	15.077	8.978		FOXD1	6.52	3.393	
	IRF1	13.16	8.874		NKX2-5	5.235	3.387	
	MAFB_2	13.647	8.504		180 min	SRF	18.52	5.705
	ARNT	11.084	8.185			TBP	16.898	9.18
	TBP	7.527	7.378			PAX4	15.643	1.751
	GABPA	15.44	7.359			SRY	13.797	2.042
	HOXA9	13.639	7.05			HOXD8	13.427	4.785
	MIZF	12.956	7.023			HIF1A::ARNT	8.387	3.719
	NFIL3	10.551	6.49			ARNT	6.042	5.004
30 min	SRF	31.177	12.142	NFIL3	2.838	3.191		
	HOXD8	13.034	4.836	210 min	SRF	25.144	7.966	
	E2F1	11.462	2.972		MAFB_1	14.107	3.405	
	NFKB1	10.593	1.149		TBP	12.062	6.992	
	IRF1	9.173	2.207		NFKB1	10.197	2.394	
	HOXA7_2	9.166	2.255		PLAG1	9.117	2.039	
	PBX1	8.973	4.477		MAX	8.515	2.514	
	PLAG1	8.901	1.856		E2F1	8.164	1.815	
	TBP	8.197	8.206	FOXO3	5.393	4.125		
HOXD10	5.249	3.545	HOXD8	6.925	3.041			
45 min	SRF	34.767	13.54	GFI	6.681	2.924		
	TBP	19.344	9.684	FOXQ1	0.007	2.823		
	EWSR1-FLI1	13.41	0.151	PDX1	-0.17	2.809		
	MAFB-1	11.033	2.88	240 min	SRF	34.478	11.435	
	STAT1	9.088	3.372		REST	20.384	4.783	
60 min	SRF	26.22	10.201		NFATC2	13.148	1.648	
	TBP	13.236	8.11		FOXD1	11.775	2.914	
	MAFB 1	11.023	2.314		TBP	11.112	6.038	
	FOXI1	8.716	1.451		FOXD3	4.705	4.171	
	ARNT	5.216	2.93		STAT3	6.161	3.571	
RELA	5.476	2.905	PBX1	5.53	3.084			
80 min	SRF	11.53	4.179	300 min	EGR1	28.073	23.332	
	CTCF	10.637	1.951		ELK1	26.499	5.726	
	TBP	9.753	2.037		NKX2-5	24.604	1.876	
	TAL1::TCF3	8.035	2.653		SRY	23.808	6.324	
	HNF1B	7.0404	2.554		NKX3-1	22.268	7.673	
	NFATC2	8.134	1.071		ARID3A	21.874	2.636	
	PLAG1	7.894	1.378		TBP	21.094	11.499	
	ZFP423	6.39	2.17		E2F1	20.7	16.347	
	MYC::MAX	4.477	2.902		HOXD10	19.956	7.024	
	RELA	3.762	2.634		SOX5	13.945	10.564	

	TP53	N/A	2.389		ELK4	16.723	9.728
	HAND1::TCFE2A	1.472	1.882		FOXI1	13.351	9.726
	HOXD10	0.925	1.81		GABPA	19.492	9.094
100 min	SRF	32.054	6.194		FOXD3	12.02	8.868
	TBP	17.482	2.839		HIF1A::ARNT	14.447	8.284
	RREB1	13.221	3.335	360 min	SRF	27.25	10.135
	NR3C1	7.829	0.697		MAFB_1	22.316	7.642
	NKX3-2	7.623	0		TBP	13.939	6.933
	FOXQ1	4.439	3.853		NFKB1	10.202	4.662
	SOX5	-0.245	2.01		RELA	9.416	5.148
					HOXD10	9.147	2.013
120 min	SRF	15.871	3.585		NR3C1	8.62	2.426
	MZF1_5-13	11.436	0.862		PBX1	7.037	3.432
	NFATC2	9.782	0.512	420 min	SRF	23.465	9.49
	E2F1	9.617	0.707		RELA	15.047	11.23
	HIF1A::ARNT	8.641	2.746		TBP	12.703	8.259
	NFKB1	8.443	2.152		NFKB1	12.176	3.518
	REST	7.302	1.642		NFKB	8.221	0.494
	NFYA	6.867	3.844		NHLH1	7.558	1.882
	FOXO3	-1.354	3.531	480 min	SRF	20.836	7.884
	SRY	3.554	2.886		MZF1_5_13	13.466	0.67
	FOXD1	-1.421	2.745		HOXD8	12.747	4.292
	ELK4	1.743	2.462		PPARG	12.627	1.751
	PDX1	0.611	2.441		PLAG1	10.741	2.811
	LHX3	4.697	2.213		EWSR1-FLI1	9.808	0.637
	TBP	5.089	2.145		NFKB1	7.722	1.593
	EGR1	3.81	2.142		TBP	6.958	6.754
					MEF2A	5.956	2.959
					SRY	2.083	2.783
					SOX5	1.684	2.666

Supplementary Table 2: Predicted HOXD10 target genes

List of HOXD10 target genes (identified by the presence of HOXD10 TFBS in the core promoter) that were found up-regulated at the respective time points of predicted HOXD10 activity, indicated by “X”.

Gene	Time point [min] with significant HOXD10 activity				
	15	30	80	300	360
CSRNP1		X		X	X
DDX60	X	X			X
DNAJB4	X	X		X	
DUSP5		X		X	X
FOSB		X	X	X	X
GIMAP7	X	X		X	
HES1		X		X	X
MMRN1	X	X		X	
NR4A1		X	X	X	X
SPRY4		X		X	X

Supplementary Table 3

[Click here to Download Table S3](#)

Supplementary Table 4: GO annotation of up- and downregulated genes in cells overexpressing HOXD10

List of the 10 most significantly enriched GO "biologic process" (BP) terms among the up- and downregulated genes identified by CAGE RNA sequencing in LEC overexpressing HOXD10.

Upregulated genes:

Term	P-value	Genes
Positive regulation of I-kappaB kinase/NF-kappaB cascade	3.11E-04	LITAF, BST2, TBK1, TFG, NDFIP2, TLR3, TLR4, MAP3K7, TRIM38, TMEM9B, ATP2C1, IL1B, HTR2B
Negative regulation of cell death	6.41E-04	MEF2C, YWHAZ, XIAP, CLU, TGFB3, HSPA1A, NFKB1, BCL2L1, HSPA1B, MAP3K7, BAG3, CDKN2D, IL1B, NOS3, TAF9, FAS, RASA1, PIK3CG, IL6, SLC25A4, IL7, IGF1, CSDA, FURIN, ATM, AMIGO2, DHRS2, UBB, PRNP
Regulation of I-kappaB kinase/NF-kappaB cascade	7.67E-04	LITAF, BST2, TBK1, TFG, NDFIP2, TLR3, TLR4, MAP3K7, TRIM38, TMEM9B, ATP2C1, IL1B, HTR2B
Positive regulation of cellular biosynthetic process	8.98E-04	MEF2C, ELF1, TGFB3, TLR3, TLR4, NFKB1, FOS, SORBS1, IL1B, PDGFC, TAF9, YAP1, RARB, FAM129A, ASF1A, ETV4, RNF14, EGR1, IL6, SMAD9, KLF12, CCNH, PCBD1, CD276, IGF1, MED14, CDK7, TOPORS, SMAD1, KAT5, WWTR1, MECOM, STAT3, ATF6, PTHLH, PLA2G4A, PKNOX1, NCOA2, NCOA4, GTF2F1, UBB, SMARCA2, RNF20, MAPRE3
Negative regulation of apoptosis	0.001084321	MEF2C, YWHAZ, XIAP, CLU, TGFB3, HSPA1A, NFKB1, BCL2L1, HSPA1B, MAP3K7, BAG3, CDKN2D, IL1B, NOS3, TAF9, FAS, RASA1, PIK3CG, IL6, IL7, IGF1, CSDA, FURIN, ATM, AMIGO2, DHRS2, UBB, PRNP
Positive regulation of biosynthetic process	0.001174285	MEF2C, ELF1, TGFB3, TLR3, TLR4, NFKB1, FOS, SORBS1, IL1B, PDGFC, TAF9, YAP1, RARB, FAM129A, ASF1A, ETV4, RNF14, EGR1, IL6, SMAD9, KLF12, CCNH, PCBD1, CD276, IGF1, MED14, CDK7, TOPORS, SMAD1, KAT5, WWTR1, MECOM, STAT3, ATF6, PTHLH, PLA2G4A, PKNOX1, NCOA2, NCOA4, GTF2F1, UBB, SMARCA2, RNF20, MAPRE3
Positive regulation of macromolecule biosynthetic process	0.001209855	MEF2C, ELF1, TGFB3, TLR3, TLR4, NFKB1, FOS, SORBS1, IL1B, TAF9, PDGFC, YAP1, RARB, FAM129A, ASF1A, ETV4, RNF14, EGR1, IL6, SMAD9, KLF12, CCNH, PCBD1, CD276, IGF1, MED14, CDK7, TOPORS, SMAD1, KAT5, WWTR1, MECOM, STAT3, ATF6, PKNOX1, NCOA2, NCOA4, GTF2F1, UBB, SMARCA2, RNF20, MAPRE3
Negative regulation of programmed cell death	0.001330821	MEF2C, YWHAZ, XIAP, CLU, TGFB3, HSPA1A, NFKB1, BCL2L1, HSPA1B, MAP3K7, BAG3, CDKN2D, IL1B, NOS3, TAF9, FAS, RASA1, PIK3CG, IL6, IL7, IGF1, CSDA, FURIN, ATM, AMIGO2, DHRS2, UBB, PRNP
Regulation of growth	0.001380527	EXTL3, TSG101, LTBP4, ZMAT3, GREM1, DDR2, TSPYL2, CDKN2C, TRO, CDKN2D, CAMK2D, TAF9, SEMA3A, MUC12, MAP2K5, IL7, SOCS1, IGF1, KAT5, CSDA, STAT3, DDR1,

		CTH, RUVBL2, UBB, KCTD11
Negative regulation of cell proliferation	0.001439549	RARRES1, PTGS2, TSG101, TGFB3, IFI30, CHEK1, IL15, MXI1, CDKN2C, KIFAP3, CDKN2D, IL1B, NOS3, RARB, TINF2, FGFBP1, IL6, IL8, AIMP1, CD276, SF1, SMAD1, PTHLH, DHRS2, DDR1, CTH, SMARCA2

Downregulated genes:

Term	P-value	Genes
Regulation of cell migration	1.26E-06	DLC1, ACVRL1, PDGFB, PDGFA, IL6ST, BCAR1, JAG2, KIT, CXCL12, SPAG9, CXCR4, DLL4, TEK, ROBO4
Cell proliferation	1.82E-06	BMP4, BCAT1, ERG, LIPA, TNFSF4, PDGFB, PDGFA, BCAR1, E2F8, MOV10L1, KIT, CXCL12, CTNNB1, GPC4, ZFP36L2, EVI5, IRF6, CXCR4, TXNRD1, HHIP, MYC, MYH10
Regulation of phosphorus metabolic process	2.73E-06	DLC1, BMP4, CAV1, PDGFB, PDGFA, IL6ST, C13ORF15, TNFSF15, PKMYT1, TLR4, KIT, PRKCE, INHBA, SPAG9, ADRB2, SPRY1, CCND2, CXCR4, RGS3, RGS4, TNK2, GADD45B, ENG
Regulation of phosphate metabolic process	2.73E-06	DLC1, BMP4, CAV1, PDGFB, PDGFA, IL6ST, C13ORF15, TNFSF15, PKMYT1, TLR4, KIT, PRKCE, INHBA, SPAG9, ADRB2, SPRY1, CCND2, CXCR4, RGS3, RGS4, TNK2, GADD45B, ENG
Regulation of cell proliferation	4.32E-06	DLC1, CAV1, ACVRL1, LST1, PDGFB, PDGFA, IL6ST, PPARG, TNFSF15, JAG2, GJA1, BCL2L1, KIT, PAWR, CDH5, CTNNB1, WARS, TEK, SERPINE1, TGM2, CSK, MYC, BMP4, TNFSF4, PDCD1LG2, TNS3, ADRB2, CCND2, IRF6, ENG
Regulation of phosphorylation	5.16E-06	BMP4, CAV1, PDGFB, PDGFA, IL6ST, C13ORF15, TNFSF15, PKMYT1, TLR4, KIT, PRKCE, INHBA, SPAG9, ADRB2, SPRY1, CCND2, CXCR4, RGS3, RGS4, TNK2, GADD45B, ENG
Regulation of locomotion	5.20E-06	DLC1, ACVRL1, PDGFB, PDGFA, IL6ST, BCAR1, JAG2, KIT, CXCL12, SPAG9, CXCR4, DLL4, TEK, ROBO4
Regulation of cell motion	5.51E-06	DLC1, ACVRL1, PDGFB, PDGFA, IL6ST, BCAR1, JAG2, KIT, CXCL12, SPAG9, CXCR4, DLL4, TEK, ROBO4
Blood vessel development	1.58E-05	BMP4, CAV1, ACVRL1, PDGFA, ELK3, GJA4, CXCL12, CDH5, CTNNB1, CXCR4, DLL4, ROBO4, TGM2, SOX17, ENG
Vasculature development	2.07E-05	BMP4, CAV1, ACVRL1, PDGFA, ELK3, GJA4, CXCL12, CDH5, CTNNB1, CXCR4, DLL4, ROBO4, TGM2, SOX17, ENG

Supplementary Table 5: List of qPCR primers

Gene names are provided as well as their corresponding forward and reverse primer sequence used in qPCR analyses.

qPCR primers used for detection of human genes:

Gene	Forward	Reverse
RPLP0	CAGATTGGCTACCCAAGTGGT	GGGAAGGTGTAATCCGTCTCC
HOXD10	GACATGGGGACCTATGGAATGC	CGGATCTGTCCAAGTGTCTACT
FOSB	GCTGCAAGATCCCCTACGAAG	CCGGCAAATCTCTCACCTCC
NR4A1	CCCTGAAGTTGTTCCCCTCAC	CCGTAGGCATGGAATAGCTCT
CDH5	GATCAAGTCAAGCGTGAGTCG	AGCCTCTCAATGGCGAACAC
CLDN5	GGGAACCTCCTGAAGTGGTGT	TCCCATGGCAAACAGAGAGG
TEK	TTAGCCAGCTTAGTTCTCTGTGG	AGCATCAGATACAAGAGGTAGGG
NOS3	TGATGGCGAAGCGAGTGAAG	ACTCATCCATACACAGGACCC
DLL4	TGGGTCAGAACTGGTTATTGGA	GTCATTGCGCTTCTTGACACAG
HOXD8	TCAAATGTTTCCGTGGATGAG	AGAGTTTGGAAGCGACTGTAG

qPCR primers used for detection of mouse genes

Gene	Forward	Reverse
RPLP0	AGATTTCGGGATATGCTGTTGG	TCGGGTCCTAGACCAGTGTTTC
HOXD10	GGCCTTCCAGAAGACAGGAG	AGCCAATTGCTGGTTGGAGT