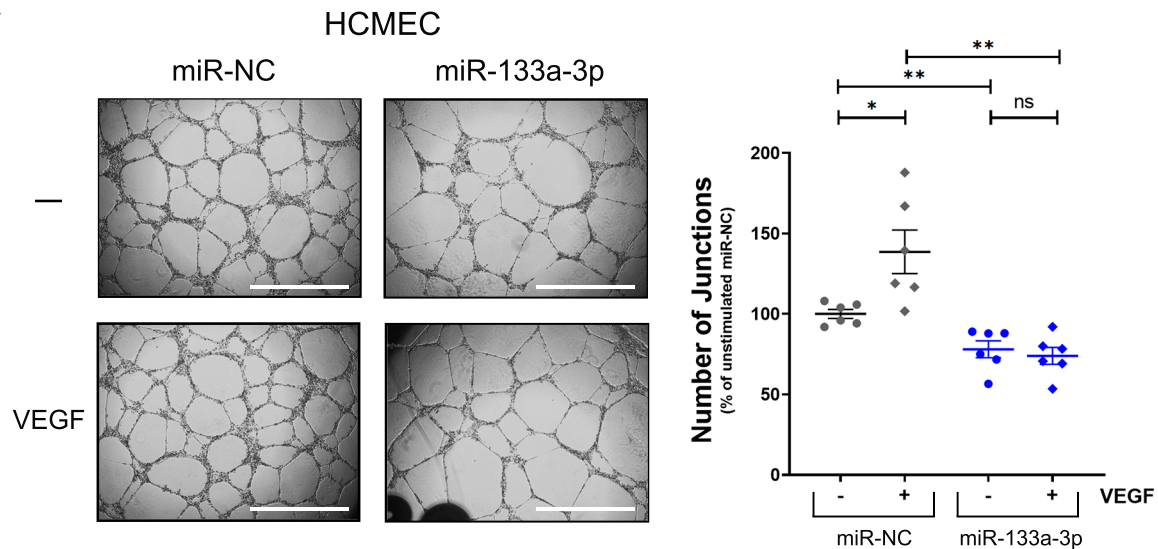


A



B

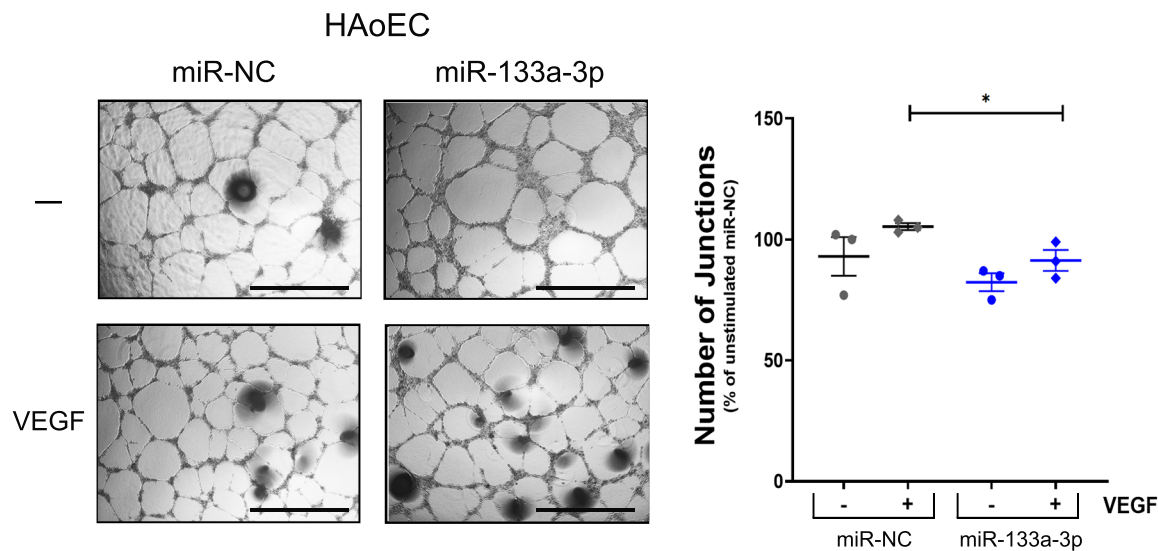


Figure S1. Ectopic expression of miR-133a-3p in HCMEC or HAoEC inhibits VEGF-induced angiogenesis. Human Cardiac Microvascular Endothelial Cells (HCMEC) (A) or Human Aortic Endothelial Cells (HAoEC) (B) transfected with miR-133a-3p mimics or a negative control (miR-NC) were seeded on Growth-Factor Reduced Matrigel (Geltrex) in ECGM medium containing 1% FBS and were left untreated (-) or were stimulated with VEGF-A165 at 50 ng/ml (+) as indicated. Images show representative fields from experiments quantified in the histogram. Data are shown as mean \pm SE, $n = 6$ (A) or $n = 3$ (B). ns = non-significant; *, $P \leq 0.05$; **, $P \leq 0.01$ (unpaired two-tailed Student's *t*-test when comparing the indicated groups). Scale bars, 1000 μ m. Image J for windows bundled with 64-bit Java 1.8.0_172 was used in this figure (<https://imagej.nih.gov/ij/>).

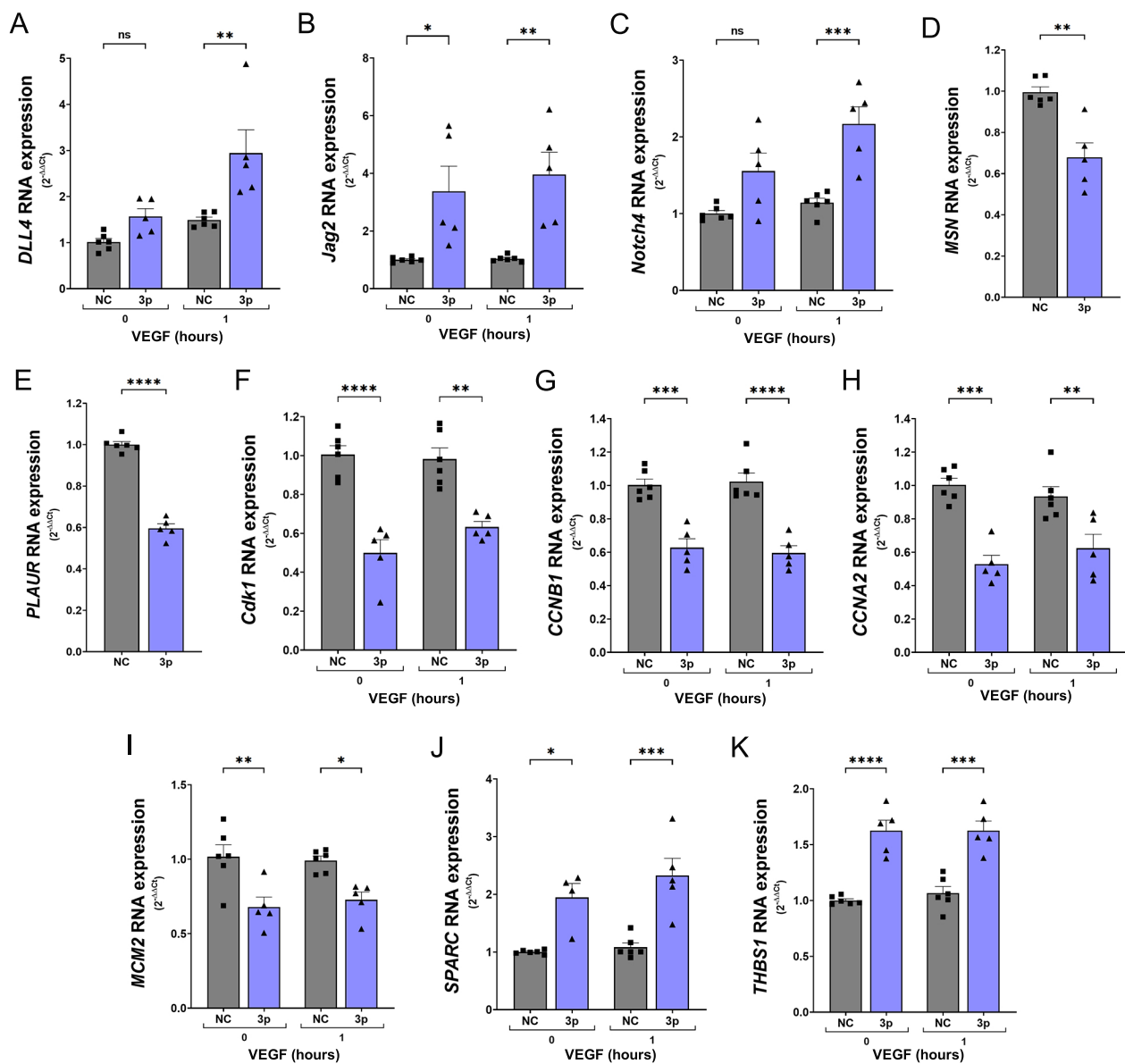


Figure S2. Aberrant expression of miR-133a-3p in primary Human Aortic Endothelial Cells (HAoEC) significantly alters the expression of genes encoding components of Notch signalling, cell migration, cell cycle regulators, and extracellular matrix. HAoEC were transfected with a control miR mimic (NC, grey bars) or a mimic for human miR-133a-3p (3p, blue bars) and RNA levels for the indicated genes were determined by qPCR. Expression of genes encoding Notch-signalling components (A-C), motility proteins (D, E), cell-cycle regulators (F-I) or extracellular matrix proteins (J-K) were significantly altered in cells expressing miR-133a-3p. Data are shown as mean \pm SE, $n = 5-6$. Data were analysed for statistical differences by two-way ANOVA with *post hoc* Tukey's comparison test. ns = non-significant; *, $P \leq 0.05$; **, $P \leq 0.01$; ***, $P \leq 0.001$; ****, $P \leq 0.0001$. VEGF = VEGF-A165.

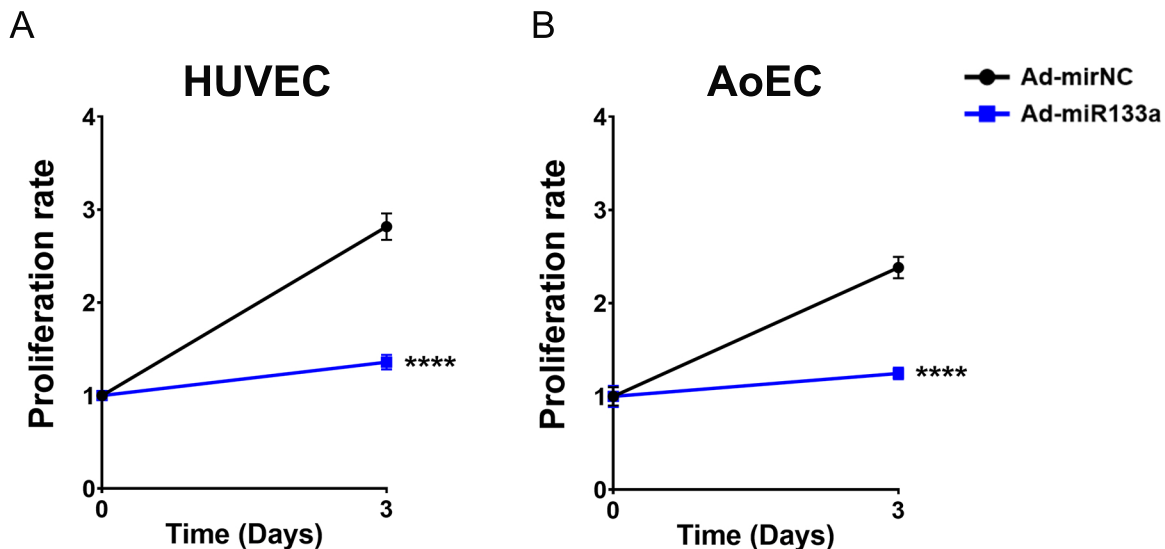
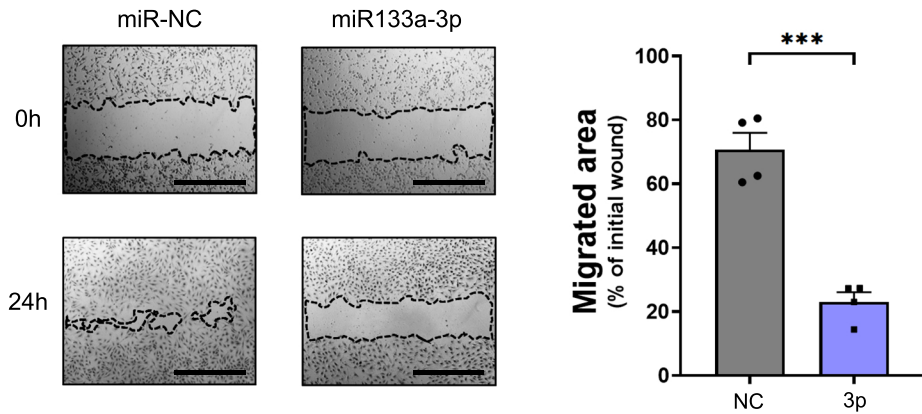


Figure S3. Ectopic expression of miR-133a in HUVEC or AoEC significantly inhibits endothelial cell proliferation. HUVEC (A) or HAoEC (B) infected with adenovirus negative control (AdmiR-NC, black line) or with an adenovirus expressing miR-133a (AdmiR-133a, blue line) were cultured in ECGM containing VEGF-A165 (50 ng/ml) for the indicated times. Number of cells were determined at the indicated time by detachment with trypsin and cell counting using a hemacytometer. Proliferation rate was calculated as number of cells after 3 days of culture divided by number of cells at 0 days. Data are shown as mean \pm SE, n = 5. ****, $P \leq 0.0001$ AdmiR-133a vs AdmiR-NC at 3 Days analysed by two-way ANOVA with *post hoc* Bonferroni's comparison test.

A

HUVEC hydroxyurea



B

HAoEC hydroxyurea

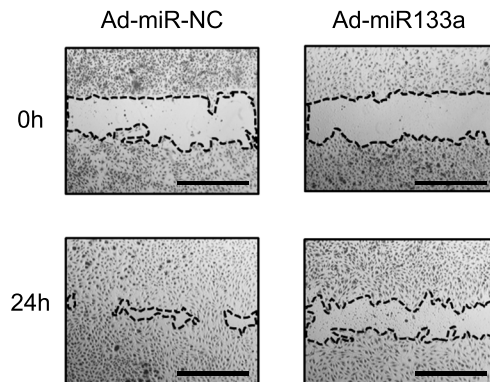


Figure S4. miR-133a inhibits HUVEC and HAoEC migration in the presence of the proliferation inhibitor hydroxyurea. (A) Representative images of wound-healing migration assays performed with HUVEC transfected with microRNA mimics for miR-133a-3p, or a negative control (miR-NC). Images were taken at time zero (0h) and after incubation for 24 hours in endothelial cell medium in the presence of 2 mM hydroxyurea (24h). The migrated area was calculated by subtracting the value of the non-migrated area from the wound area at time zero and expressing this as a percentage of the total area at time zero. Scale bars, 1000 μm. Data are shown as mean ± SE, n = 4. Data were analysed for statistical differences by one-way ANOVA with *post hoc* Tukey's comparison test. ***, P ≤ 0.001. (B) Human Aortic Endothelial Cells were infected with control adenovirus (Ad-miR-NC) or an adenovirus expressing miR-133a. (Ad-miR-133a) at MOI=200 for 72 hours. Motility of the infected cells was analysed in wound-healing migration assays as described in (A). Images are representative of two independent experiments. Scale bars, 1000 μm. Image J for windows bundled with 64-bit Java 1.8.0_172 was used in this figure (<https://imagej.nih.gov/ij/>).

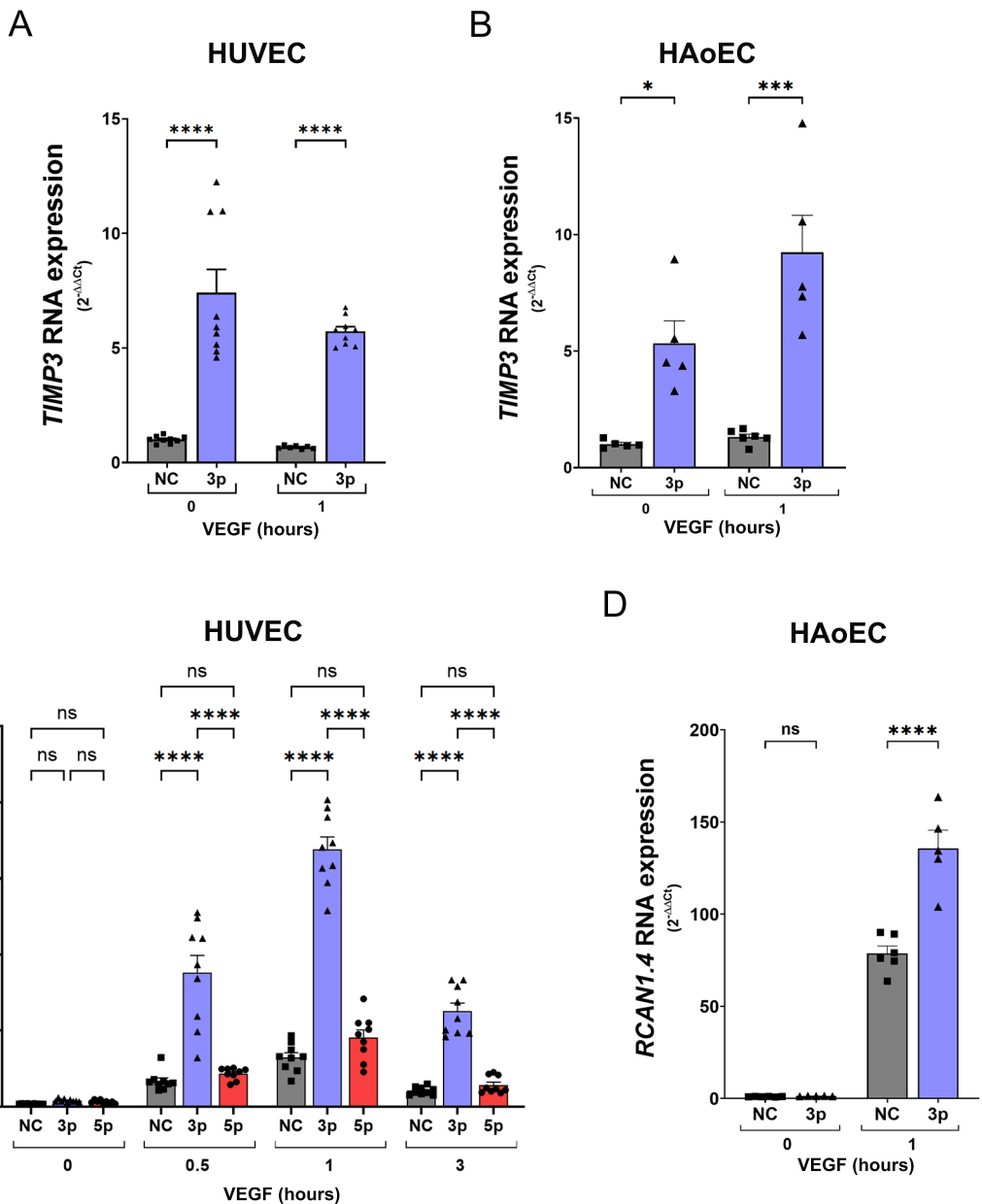
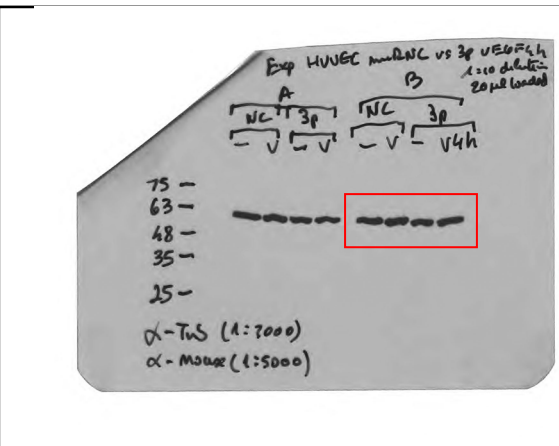
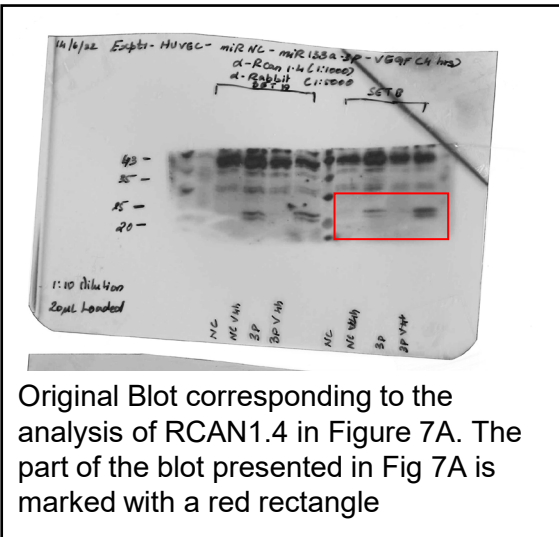
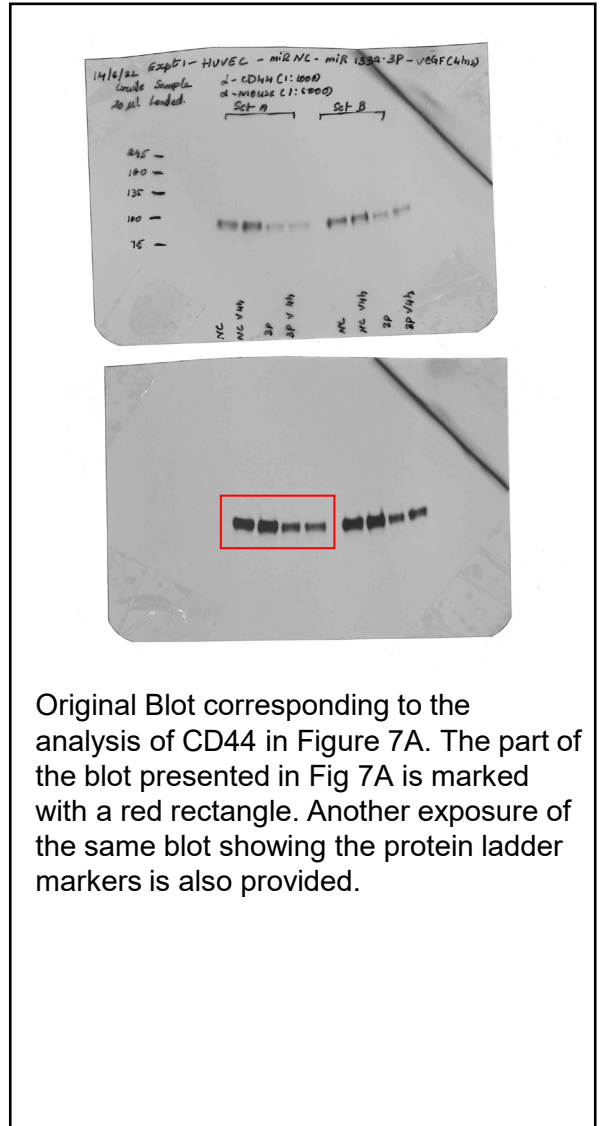
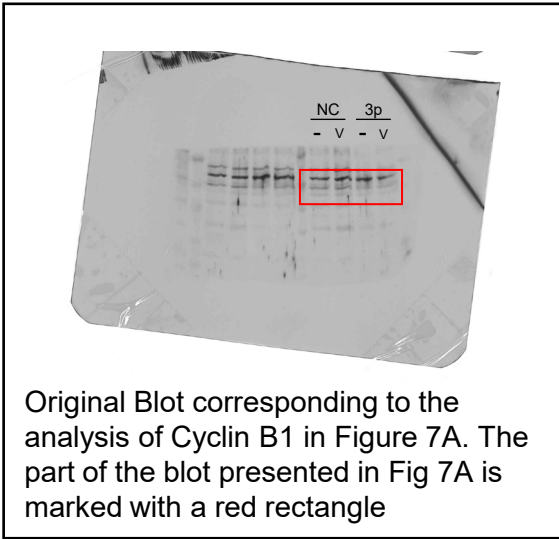
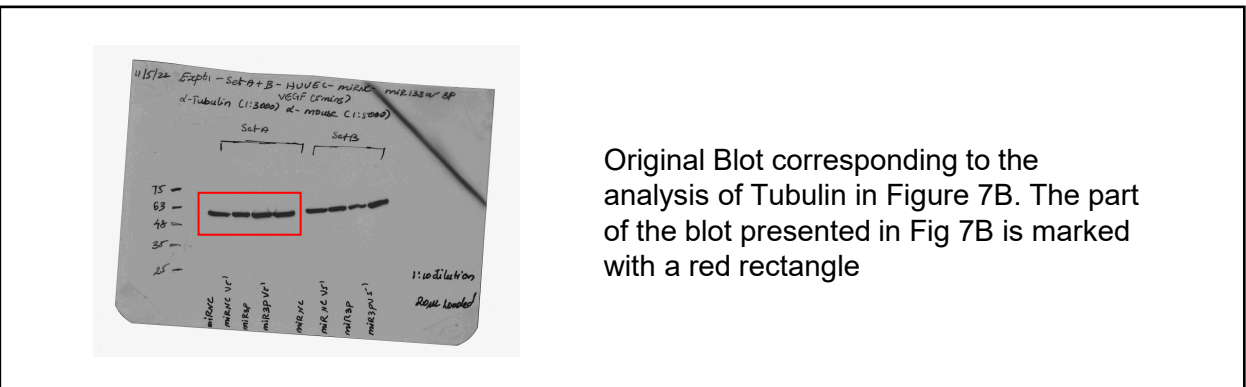
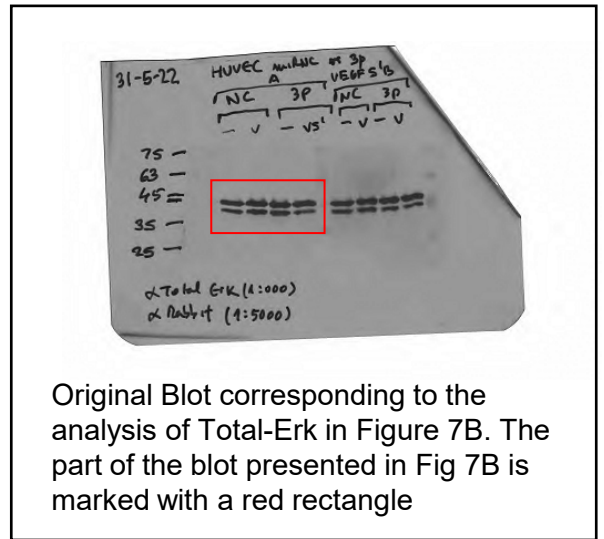
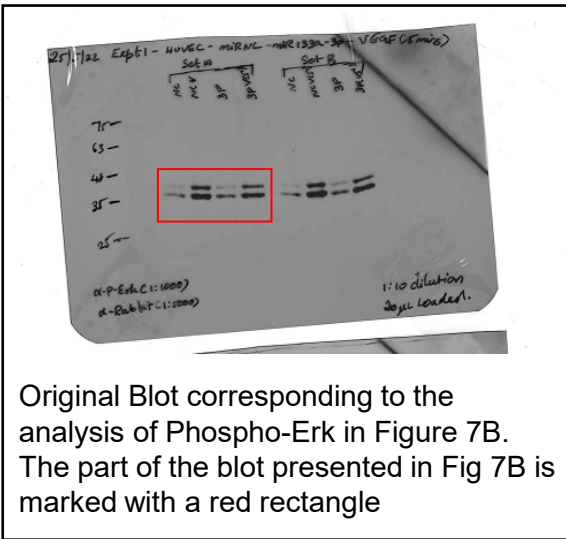
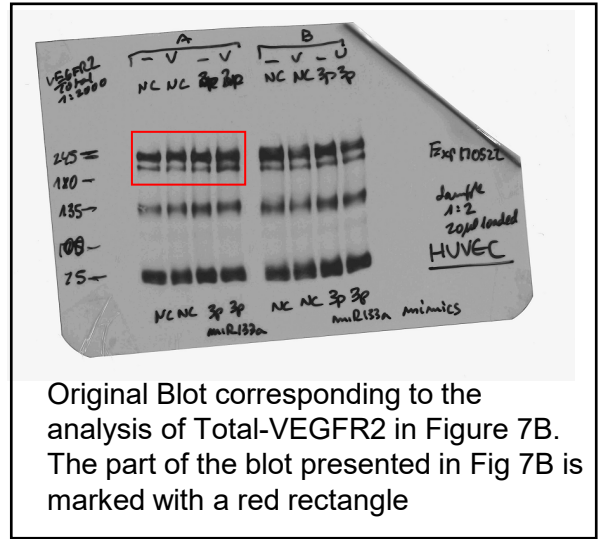
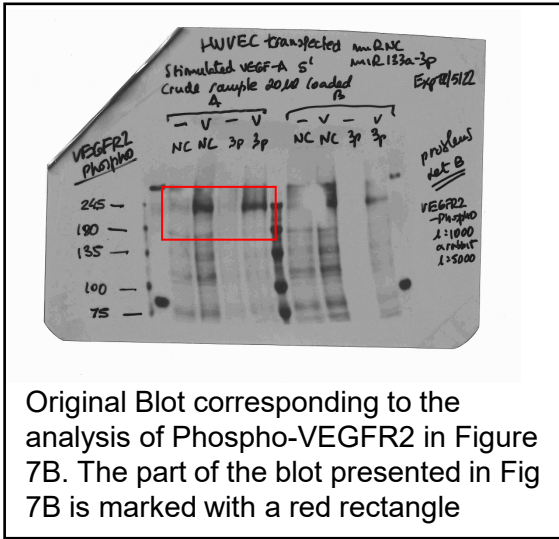


Figure S5. Changes in gene expression of *TIMP3* and *RCAN1.4* in endothelial cells expressing miR-133a. HUVEC (A, C) or HAoEC (B, D) transfected with miR-133a-3p (blue columns), miR-133a-5p (red columns), or negative control miR-NC (grey columns) were left unstimulated (0h) or were stimulated with VEGF-A165 as indicated. RNA levels of *TIMP3* (A, B) and *RCAN1.4* (C, D) significantly increased in miR-133a-3p transfected cells. Ct values were normalised using the Ct values for the housekeeping gene *Hprt-1*. Analysis of data was carried out using the comparative $2^{-\Delta\Delta Ct}$ method referred to miR-NC (0h). Data are shown as mean \pm SE, $n = 7$ in HUVEC or $n = 5$ in AoEC. ns = non-significant; ****, $P \leq 0.0001$; ***, $P \leq 0.001$ two-way ANOVA with *post hoc* Tukey's comparison test for the indicated groups.

Original blots used in figure 7A



Original blots used in figure 7B



Position	Gene Symbol	AVG ΔCt		2 ^{-ΔCt}		Fold Change		p-value
		miR133a-3p VEGF 1h	miR-NC VEGF 1h	miR133a-3p VEGF 1h	miR-NC VEGF 1h	miR133a-3p VEGF 1h/miR-NC VEGF 1h	miR-NC VEGF 1h/miR-NC VEGF 1h	
A01	ADAM10	2.37	2.87	0.19382	0.136999		1.41	0.010688
A02	ADAM17	4.33	4.38	0.049602	0.047951		1.03	0.967439
A03	AES	2.39	2.54	0.191036	0.172456		1.11	0.373270
A04	AXIN1	6.09	6.06	0.014722	0.014697		0.98	0.833499
A05	CBL	6	6.03	0.015696	0.015344		1.02	0.802729
A06	CCND1	3.04	2.06	0.121559	0.23953		0.51	0.032068
A07	CCNE1	7.54	6.74	0.005356	0.009343		0.57	0.003667
A08	CD44	4.81	3.01	0.035679	0.124391		0.29	0.002287
A09	CKN1A	1.98	2.12	0.254345	0.229741		1.11	0.622333
A10	CFIAR	3.68	4.29	0.078231	0.051042		1.53	0.001597
A11	CHUK	4.52	5.16	0.043592	0.028058		1.55	0.474799
A12	CTNNB1	2.52	3.11	0.174488	0.116168		1.5	0.165019
B01	DLL1	7.63	7.76	0.005062	0.004599		1.1	0.868859
B02	DLL3	13.89	14.94	0.000066	0.000032		2.08	0.500788
B03	DLL4	3.54	6.41	0.086118	0.01174		7.34	0.002103
B04	EP300	5.11	5.08	0.028991	0.029642		0.98	0.744398
B05	ERBB2	6	6.48	0.015619	0.011181		1.4	0.084305
B06	FIGF	12.68	12.13	0.000152	0.000224		0.68	0.427723
B07	FOS	7.3	7.76	0.006344	0.004814		1.37	0.312468
B08	FOSL1	3.97	3.84	0.063655	0.069966		0.91	0.347284
B09	FZD7	11.95	12.28	0.000253	0.000201		1.26	0.608977
B10	GLI1	11.23	12.12	0.000415	0.000225		1.85	0.430816
B11	GSK3B	4.14	4.15	0.056579	0.056255		1.01	0.985468
B12	HDAC1	3.91	4.37	0.066313	0.04834		1.37	0.042302
C01	HES1	4.06	3.9	0.059933	0.066897		0.9	0.748074
C02	HEY1	6.71	6.12	0.009553	0.003066		2.65	0.003644
C03	HOXB4	6.25	6.84	0.002276	0.00381		0.96	0.863624
C04	HIF1	11.48	13.02	0.00035	0.000121		2.9	0.200304
C05	HD	6.15	5.63	0.014127	0.030772		0.46	0.011005
C06	IFNG	15.93	16.78	0.000016	0.000009		1.81	0.333035
C07	IL2RA	15.83	16.69	0.000017	0.000009		1.81	0.355301
C08	JAG1	4.13	5.18	0.056924	0.027556		2.07	0.060391
C09	JAG2	4.1	5.45	0.058398	0.022886		2.55	0.008049
C10	KRT1	16.16	16.78	0.000014	0.000009		1.55	0.308032
C11	LFNG	6.84	6.92	0.00875	0.008239		1.06	0.840951
C12	LMO2	2.91	3.61	0.133194	0.081731		1.63	0.038285
D01	LOR	16.15	16.31	0.000014	0.000012		1.12	0.688941
D02	LRP5	7.55	7.59	0.005347	0.00519		1.03	0.851724
D03	MAML1	6.31	5.87	0.012626	0.017067		0.74	0.198066
D04	MAML2	6.44	6.51	0.011547	0.010951		1.05	0.889567
D05	MFNG	3.07	2.83	0.119359	0.140923		0.85	0.021107
D06	MMP7	8.32	8.22	0.003136	0.003351		0.94	0.664009
D07	NCOR2	4.53	4.27	0.04326	0.052003		0.83	0.495544
D08	NCSTN	3.27	3.61	0.103865	0.081666		1.27	0.000225
D09	NFKB1	5.71	6.01	0.01913	0.015483		1.24	0.441816
D10	NFKB2	5.4	5.68	0.023755	0.019526		1.22	0.387127
D11	NOTCH1	5.99	6.37	0.015696	0.012083		1.3	0.521508
D12	NOTCH2	6.14	5.67	0.014193	0.019697		0.72	0.141066
D13	NOTCH3	9.74	9.88	0.001168	0.001064		1.1	0.905228
E01	NOTCH4	4.25	5.81	0.044167	0.017865		2.47	0.011642
E03	NUMB	4.29	4.21	0.051168	0.054054		0.95	0.244417
E04	PAX5	15.62	16.36	0.00002	0.000012		1.67	0.028959
E05	POU1F1	7.7	7.9	0.0048	0.004199		1.14	0.366457
E06	PPARG	8.36	10.18	0.003044	0.000965		3.52	0.000338
E07	PSEN1	4.63	4.42	0.040491	0.046872		0.86	0.032967
E08	PSEN2	6	6.02	0.015611	0.0154		1.01	0.502805
E09	PSENEN	4.33	4.4	0.049687	0.047374		1.05	0.620692
E10	PTCRA	14.15	15.3	0.000055	0.000025		2.22	0.400245
E11	RBPJL	15.59	15.66	0.00002	0.000019		1.05	0.763113
E12	RFNG	5.21	5.44	0.027062	0.023039		1.17	0.088466
F01	RUNX1	6.81	6.06	0.008936	0.014977		0.6	0.033440
F02	SELL	3.35	3.84	0.097817	0.068881		1.4	0.006913
F03	SHH	11.97	11.35	0.00025	0.000382		0.65	0.007158
F04	SMO	7.62	7.37	0.005075	0.006603		0.84	0.105797
F05	SNW1	5.2	4.82	0.027124	0.035378		0.77	0.170943
F06	STAT6	4.16	3.93	0.055907	0.065676		0.85	0.108075
F07	TLE1	4.7	4.75	0.03838	0.037252		1.03	0.761570
F08	WISP1	12.63	15.4	0.000158	0.000023		6.83	0.000754
F09	ADA	4.92	4.89	0.033064	0.033743		0.96	0.912166
F10	AFAP112	9.3	12.01	0.001592	0.000242		6.57	0.058412
F11	DTX1	15.26	16.07	0.000025	0.000015		1.75	0.428849
F12	H19	11.49	13.47	0.000347	0.000088		3.93	0.032924
G01	Hes4	4.95	6.64	0.032401	0.010001		3.24	0.044112
G02	HES5	13.99	15.62	0.000062	0.00002		3.11	0.407130
G03	HEY2	10.92	10.17	0.000966	0.000686		1.11	0.872876
G04	HEY1	16	16.68	0.000015	0.00001		1.6	0.230238
G05	NIBARP	5.7	7.11	0.019182	0.007259		2.64	0.046670
G06	SCGB1A1	15.64	16.78	0.00002	0.000009		2.22	0.048320
G07	SERPINA3	13.27	13.9	0.000101	0.000065		1.55	0.348460
G08	SLC6A12	16.66	16.78	0.00001	0.000009		1.09	0.629893
G09	SNAI2	10.11	9.59	0.000902	0.001301		0.69	0.321168
G10	TFE1	16.3	16.58	0.000012	0.00001		1.21	0.481258
G11	TNFSF10	4.19	4.23	0.054855	0.053357		1.03	0.877834
G12	UBD	10.53	10.94	0.000678	0.000051		1.33	0.547084
H01	ACTB	-2.73	-2.65	6.644773	6.274732		1.06	0.419120
H02	B2M	-0.76	-0.36	1.696719	1.28043		1.33	0.182271
H03	GAPDH	-1.15	-1.33	2.22983	2.508923		0.89	0.269595
H04	HPRT1	5.96	5.59	0.016051	0.020728		0.77	0.007666
H05	RPLP0	-1.31	-1.26	2.48292	2.393297		1.04	0.704859

Supplementary Table S1. Differential gene expression analysis of RT2 Profiler PCR Array Human Notch Signalling Pathway Plus Kit. HUVEC transfected with miR-133a-3p or negative control miR-NC were stimulated with VEGF-A165 for 1 hour. Upregulated and downregulated genes are highlighted in red and green, respectively. Genes showing values of AVG ΔCt ≥ 10 indicating high number of amplification cycles required in the PCR analysis were not considered in further experiments to improve accuracy and reproducibility of data.

Position	Gene Symbol	AVG ΔCt		2 ^{-ΔΔCt}		Fold Change		p-value
		miR133a-3p VEGF 1h	miR-NC VEGF 1h	miR133a-3p VEGF 1h	miR-NC VEGF 1h	miR133a-3p VEGF 1h/miR-NC VEGF 1h	miR133a-3p VEGF 1h / miR-NC VEGF 1h	
A01	ABL1	5.34	6.03	0.024515	0.015268	1.61		
A02	ANAPC2	6.51	7.12	0.011003	0.007179	1.53		0.163561
A03	ATM	7.71	7.98	0.004776	0.003964	1.2		0.872675
A04	ATR	8.28	8.4	0.003216	0.002964	1.08		0.764296
A05	AURKA	9.76	5.73	0.009201	0.018801	0.49		0.193957
A06	AURKB	6.17	7.13	0.001741	0.007153	0.24		0.117954
A07	BCCIP	4.74	4.44	0.037322	0.046224	0.81		0.385842
A08	BCL2	8.81	10.45	0.002229	0.000715	3.12		0.002742
A09	BIRC5	12.19	10.18	0.002015	0.000603	0.25		0.113855
A10	BRC1A	10.06	8.42	0.000936	0.00291	0.32		0.126832
A11	BRC1A2	9.49	9.05	0.001395	0.001889	0.74		0.510409
A12	CASP3	4.08	4.58	0.05904	0.041737	1.41		0.568454
B01	CCNA2	7.19	4.63	0.006844	0.040488	0.17		0.007077
B02	CCNB1	6.88	4.77	0.008902	0.036538	0.23		0.014929
B03	CCNB2	8.04	5.97	0.003788	0.019924	0.24		0.101785
B04	CCNC	6.14	6.9	0.014162	0.003999	1.69		0.310072
B05	CCND1	2.69	2.97	0.151598	0.127381	1.22		0.750907
B06	CCND2	6.84	8.46	0.008759	0.00283	3.09		0.280935
B07	CCND3	3.61	5.03	0.082101	0.036607	2.68		0.029723
B08	CCNE1	7.52	7.47	0.005459	0.005632	0.97		0.907534
B09	CCNF	7.33	7.27	0.008214	0.006487	0.96		0.573206
B10	CCNG1	1.48	3.26	0.388997	0.120426	3.44		0.037799
B11	CCNG2	5.08	5.59	0.029496	0.020746	1.42		0.194174
B12	CCNH	6.44	7.4	0.015121	0.00594	1.94		0.570288
C01	CNT1	6.45	7.22	0.011417	0.009717	1.7		0.805792
C02	CDC16	4.48	5.2	0.044827	0.027234	1.65		0.533739
C03	CDC20	8.37	6.42	0.003019	0.011709	0.26		0.08117
C04	CDC25A	8.32	8.1	0.00313	0.003657	0.86		0.57382
C05	CDC25C	12.28	10.35	0.000201	0.000764	0.26		0.286847
C06	CDC34	5.74	6.49	0.018961	0.011092	1.68		0.240041
C07	CDK6	7.69	6.54	0.004951	0.010772	0.6		0.223499
C08	CDK1	6.57	3.44	0.010568	0.0921	0.11		0.000561
C09	CDK2	8.22	8.2	0.013418	0.013595	0.99		0.963534
C10	CDK4	4.09	4.72	0.058805	0.038073	1.54		0.545356
C11	CDK5R1	9.49	9.87	0.001387	0.001667	1.3		0.530816
C12	CDK5RAP1	9.72	10.39	0.001187	0.000743	1.6		0.596426
D01	CDK6	4.61	6.94	0.04084	0.008172	5		0.007356
D02	CDK7	5.02	5.13	0.030758	0.028553	1.08		0.98945
D03	CDK8	7.4	7.42	0.009916	0.005848	1.01		0.915495
D04	CDKN1A	2.25	3.54	0.209834	0.085955	2.44		0.157594
D05	CDKN1B	4.96	5.77	0.032056	0.018345	1.75		0.486002
D06	CDKN2A	11.48	12.12	0.000349	0.000224	1.55		0.482816
D07	CDKN2B	7.18	8.71	0.00689	0.002394	2.88		0.094351
D08	CDKN3	7.19	6.41	0.008688	0.011727	0.59		0.33057
D09	CHEK1	7.44	6.66	0.00574	0.009899	0.58		0.283328
D10	CHEK2	8.09	8.63	0.003957	0.00253	1.45		0.432738
D11	CKS1B	6.14	4.94	0.014219	0.032558	0.44		0.159196
D12	CKS2	4.19	4.44	0.054895	0.040506	1.19		0.769768
E01	CUL1	5.12	5.19	0.02882	0.027368	1.05		0.918052
E02	CUL2	5.1	5.55	0.029246	0.02138	1.37		0.205873
E03	CUL3	3.82	4.62	0.070815	0.040534	1.75		0.448347
E04	E2F1	8.13	7.39	0.003577	0.005964	0.6		0.307408
E05	E2F4	4.36	5.41	0.048904	0.023558	2.06		0.501406
E06	GADD45A	6.33	5.55	0.012446	0.021391	0.58		0.133284
E07	GTSF1	6.37	6.45	0.003024	0.014404	0.27		0.807804
E08	HUS1	5.26	6.23	0.025811	0.013311	1.94		0.541133
E09	KNTC1	8.79	8.1	0.002266	0.003652	0.62		0.428603
E10	KPNA2	2.09	2.75	0.234331	0.148985	1.57		0.794139
E11	MAD2L1	7.9	6.16	0.004173	0.014016	0.3		0.130544
E12	MAD2L2	9.12	8.91	0.001795	0.002077	0.86		0.564885
F01	MCM2	7.9	6.7	0.004176	0.009641	0.43		0.060139
F02	MCM3	6.32	5.57	0.012524	0.021959	0.59		0.182696
F03	MCM4	7.4	6.29	0.009935	0.012751	0.46		0.307947
F04	MCM5	6.26	5.84	0.012896	0.017422	0.74		0.579795
F05	MDM2	2.68	3.82	0.155521	0.070568	2.2		0.104164
F06	MKI67	7	5.16	0.007788	0.0279	0.28		0.245959
F07	MNAT1	6.4	7.08	0.011809	0.007386	1.6		0.551554
F08	MRE11A	6.51	6.8	0.010994	0.008984	1.22		0.966266
F09	NBN	4.84	5.69	0.034921	0.019065	1.8		0.092656
F10	RAD1	6.89	7.16	0.019111	0.009099	1.59		0.476448
F11	RAD17	6.4	7.3	0.011683	0.006341	1.87		0.241411
F12	RAD51	9.4	8.35	0.001476	0.003064	0.48		0.236303
G01	RAD9A	8.4	8.51	0.002964	0.002752	1.08		0.936191
G02	RB1	5.49	5.56	0.022225	0.021268	1.05		0.933481
G03	RBBP8	8.25	8.15	0.00329	0.003516	0.94		0.936822
G04	RBL1	7.71	7.21	0.004772	0.006754	0.71		0.415562
G05	RBL2	5.35	5.93	0.024592	0.016415	1.5		0.548759
G06	SERTAD1	6.5	7.16	0.011949	0.006976	1.58		0.297238
G07	SFP2	7.69	7.14	0.007346	0.007102	1.03		0.731383
G08	STMN1	2.5	1.52	0.176889	0.348554	0.51		0.032378
G09	TFDP1	4.22	3.63	0.053994	0.080782	0.66		0.455342
G10	TFDP2	7.18	7.67	0.008887	0.004909	1.4		0.922733
G11	TP53	3.04	3.74	0.121287	0.074857	1.62		0.148892
G12	WEE1	5.01	5.49	0.031	0.022296	1.39		0.373908
H01	ACTB	2.49	2.53	5.637138	5.76735	0.98		0.847267
H02	B2M	-0.12	0.22	1.839723	3.280933	1.26		0.422386
H03	GAPDH	1.18	1.71	2.269950	3.280933	0.7		0.385742
H04	HPRT1	4.65	4.97	0.039753	0.032014	1.24		0.485624
H05	RPLP0	-0.85	-0.95	1.807953	1.929933	0.94		0.734311

Supplementary Table S2. Differential gene expression analysis of RT2 Profiler PCR Array Human Cell Cycle Kit. HUVEC transfected with miR-133a-3p or negative control miR-NC were stimulated with VEGF-A165 for 1 hour. Upregulated and downregulated genes are highlighted in red and green, respectively. Genes showing values of AVG ΔCt ≥ 10 indicating high number of amplification cycles required in the PCR analysis were not considered in further experiments to improve accuracy and reproducibility of data.

Position	Gene_Symbol	AVG ΔCt		2 ^{-ΔCt}		Fold Change		p-value
		miR133a-3p VEGF th	miR-NC VEGF th	miR133a-3p VEGF th	miR-NC VEGF th	miR133a-3p VEGF th/miR-NC	miR133a-3p VEGF th / miR-NC	
A01	ADAMT51	3.51	4.63	0.007613	0.004552	2.17	0.009728	
A02	ADAMT513	12.78	13.14	0.000145	0.000111	1.30	0.425539	
A03	ADAMT58	16.85	16.41	0.000008	0.000011	0.73	0.557347	
A04	CD44	3.12	2.32	0.115291	0.289893	0.57	0.032911	
A05	CDH1	16.85	17.27	0.000008	0.000006	1.33	0.752007	
A06	CLEC3B	13.88	13.13	0.000007	0.000112	0.60	0.842833	
A07	CNTN1	14.68	13.55	0.000039	0.000084	0.46	0.347898	
A08	COL1A1	14.84	16.46	0.000032	0.000011	2.87	0.291711	
A09	COL12A1	6.17	7.21	0.013542	0.006753	2.05	0.153185	
A10	COL14A1	13.18	11.44	0.000215	0.000359	0.60	0.288410	
A11	COL15A1	14.64	11.91	0.000039	0.000016	2.40	0.472859	
A12	COL16A1	11.06	11.62	0.000289	0.000318	0.85	0.625143	
B01	COL1A1	14.86	12.47	0.000034	0.000178	0.19	0.054788	
B02	COL4A2	0.35	1.17	0.785597	0.44353	1.77	0.185808	
B03	COL5A1	2.22	3.33	0.214854	0.099276	2.16	0.141926	
B04	COL6A1	5.58	6.02	0.020913	0.015454	1.35	0.467934	
B05	COL6A2	10.59	10.13	0.000651	0.000892	0.73	0.975060	
B06	COL7A1	14.68	14.35	0.000038	0.000048	0.80	0.765503	
B07	COL8A1	4.25	3.45	0.05246	0.09175	0.57	0.251567	
B08	CTGF	-0.35	-0.8	1.272852	1.735088	0.73	0.601233	
B09	CTNNA1	2.02	1.99	0.245921	0.251472	0.98	0.961821	
B10	CTNNB1	2.57	3.55	0.188494	0.085373	1.97	0.024149	
B11	CTNND1	3.42	3	0.093283	0.125032	0.75	0.264605	
B12	CTNND2	16.8	15.62	0.000009	0.00002	0.44	0.336888	
C01	ECM1	9.44	9.05	0.001437	0.001885	0.76	0.640300	
C02	FN1	-2.43	-1.94	5.375817	3.837224	1.40	0.256214	
C03	HAS1	13.49	13.91	0.000087	0.000065	1.34	0.941511	
C04	ICAM1	3.17	4	0.111119	0.062804	1.77	0.116271	
C05	ITGA1	7.13	8.1	0.00716	0.003852	1.96	0.251296	
C06	ITGA2	3.84	3.48	0.096607	0.089804	0.78	0.014392	
C07	ITGA3	3.25	2.89	0.105097	0.134514	0.78	0.497416	
C08	ITGA4	6.49	7.9	0.011127	0.004184	2.66	0.345427	
C09	ITGA5	0.44	0.9	0.739286	0.535757	1.38	0.188920	
C10	ITGA6	2.1	3.27	0.232914	0.103672	2.25	0.004711	
C11	ITGA7	13.45	13.93	0.00009	0.000064	1.40	0.191173	
C12	ITGA8	11.79	15.66	0.000282	0.000019	14.60	0.001639	
D01	ITGAL	15.52	16.38	0.000021	0.000012	1.81	0.231277	
D02	ITGAM	16.59	16.47	0.00001	0.000011	0.92	0.850826	
D03	ITGAV	1.72	2.31	0.302769	0.202065	1.50	0.097940	
D04	ITGB1	-0.32	-0.06	1.250113	1.039281	1.20	0.095719	
D05	ITGB2	10.98	12.45	0.000495	0.000179	2.76	0.153679	
D06	ITGB3	4.55	4.41	0.042643	0.048962	0.91	0.731497	
D07	ITGB4	6.89	6.58	0.008432	0.010462	0.81	0.652303	
D08	ITGB5	2.54	3.01	0.171767	0.123762	1.39	0.366620	
D09	ANOS1	14.11	12.67	0.000056	0.000154	0.37	0.124574	
D10	LAMA1	15.86	17.27	0.000017	0.000006	2.65	0.229367	
D11	LAMA2	9.66	10.19	0.001232	0.000854	1.44	0.430870	
D12	LAMA3	9.18	8.99	0.001719	0.001964	0.88	0.678911	
E01	LAMB1	1.13	1.58	0.456631	0.334105	1.37	0.201049	
E02	LAMB3	9.75	8.26	0.001163	0.003255	0.38	0.256610	
E03	LAMC1	0.89	1.36	0.538577	0.389106	1.38	0.056254	
E04	MMP1	-0.53	-0.02	1.4419	1.017202	1.42	0.238318	
E05	MMP10	4.58	4.12	0.041799	0.057384	0.73	0.637562	
E06	MMP11	7.68	7.11	0.00487	0.007256	0.67	0.424430	
E07	MMP12	10.63	14.78	0.00063	0.000036	17.71	0.373191	
E08	MMP13	16.48	16.23	0.000011	0.000013	0.84	0.628353	
E09	MMP14	2.4	2.19	0.189256	0.216887	0.87	0.871463	
E10	MMP15	12.49	13.47	0.000174	0.000088	1.98	0.231498	
E11	MMP16	5.69	6.27	0.019349	0.012915	1.50	0.183187	
E12	MMP2	1.11	0.97	0.462092	0.50978	0.91	0.573546	
F01	MMP3	13.73	13.89	0.000074	0.000086	1.12	0.731364	
F02	MMP7	10.01	9.54	0.000971	0.001345	0.72	0.818732	
F03	MMP8	16.38	16.57	0.000012	0.00001	1.13	0.813477	
F04	MMP9	16.32	15.66	0.000012	0.000019	0.63	0.402307	
F05	NCAM1	13.29	15.91	0.0001	0.000016	6.17	0.046454	
F06	PECAM1	3.31	3.93	0.106651	0.065422	1.54	0.621359	
F07	SELE	5.1	5.97	0.029148	0.015907	1.83	0.402759	
F08	SELL	9.08	7.93	0.001846	0.004089	0.45	0.074673	
F09	SELP	7.77	6.92	0.004583	0.00828	0.55	0.205867	
F10	SGCE	4.99	5.51	0.031477	0.021936	1.43	0.251018	
F11	SPARC	-1.58	-0.56	2.994117	1.47938	2.02	0.001120	
F12	SPG7	5.56	5.51	0.021163	0.021955	0.96	0.954220	
G01	SPP1	9.38	10.76	0.001504	0.000578	2.60	0.302247	
G02	TGFB1	4.75	5.62	0.037102	0.020382	1.82	0.287751	
G03	THBS1	-3.9	-2.88	14.896099	7.378099	2.02	0.015251	
G04	THBS2	10.27	11.16	0.000811	0.000437	1.85	0.406394	
G05	THBS3	9.68	8.72	0.001221	0.002372	0.51	0.121761	
G06	TMP1	4.58	4.18	0.041804	0.055082	0.76	0.243837	
G07	TMP2	0.64	0.84	0.642928	0.559352	1.15	0.565337	
G08	TMP3	7.99	11.04	0.003943	0.000476	8.29	0.386948	
G09	TNC	16.48	14.38	0.000011	0.000047	0.24	0.288267	
G10	VCAM1	9.01	8.13	0.001942	0.00357	0.54	0.812977	
G11	VCAN	14.3	11.39	0.000049	0.000373	0.13	0.242396	
G12	VTN	11.24	13.16	0.000413	0.000109	3.77	0.325615	
H01	ACTB	-2.91	-2.88	7.510101	7.375274	1.02	0.827371	
H02	B2M	-0.27	-0.37	1.207778	1.288915	0.94	0.742964	
H03	GAPDH	-1.44	-1.29	2.718774	2.444897	1.11	0.801017	
H04	HPRT1	5.66	5.36	0.019748	0.024336	0.81	0.299149	
H05	RPLP0	-1.04	-0.82	2.05343	1.787895	1.16	0.687373	

Supplementary Table S3. Differential gene expression analysis of RT2 Profiler PCR Array Human Extracellular Matrix & Cell Adhesion Molecules Kit. HUVEC transfected with miR-133a-3p or negative control miR-NC were stimulated with VEGF-A165 for 4 hour. Upregulated and downregulated genes are highlighted in red and green, respectively. Genes showing values of AVG ΔCt ≥ 10 indicating high number of amplification cycles required in the PCR analysis were not considered in further experiments to improve accuracy and reproducibility of data.

GENES	miR-NC 0h									miR-133a-3p 0h								
	0.64	1.49	1.05	1.04	0.94	1.03	1.02	0.96	1.02	0.29	0.35	0.38	0.26	0.30	0.31	0.51	0.59	0.59
PLAUR	0.64	1.49	1.05	1.04	0.94	1.03	1.02	0.96	1.02	0.29	0.35	0.38	0.26	0.30	0.31	0.51	0.59	0.59
MSN	0.91	1.26	0.87	0.98	0.97	1.05	0.85	1.27	0.92	0.51	0.70	0.50	0.58	0.65	0.62	0.59	0.61	0.59
RCAN1.4	1.01	0.91	1.09	0.94	1.10	0.97	0.99	0.98	1.04	2.06	1.81	2.37	1.17	1.39	1.38	1.33	1.46	1.45
CD44	1.00	1.31	0.76	0.91	1.00	1.09	1.06	0.95	0.99	0.18	0.34	0.21	0.24	0.21	0.20	0.47	0.33	0.50
ID1	0.79	2.47	0.51	1.12	0.94	0.95	1.71	0.35	1.69	0.19	0.30	0.22	0.38	0.34	0.37	0.70	0.76	0.58
CCNE1	0.75	0.95	1.41	0.85	1.08	1.09	0.78	1.25	1.03	0.86	1.07	1.06	0.63	0.70	0.69	0.64	0.71	0.68
CCND1	0.80	0.99	1.25	0.92	1.07	1.01	0.80	1.24	1.01	0.95	1.14	1.29	0.83	0.91	0.73	0.59	0.79	0.83
DLL4	1.02	0.84	1.17	0.71	1.11	1.26	0.90	1.13	0.98	8.28	8.05	13.28	9.04	11.88	10.08	8.16	7.74	6.74
HEY1	0.97	1.09	0.95	1.00	0.97	1.03	1.04	1.01	0.95	4.65	4.59	5.04	3.46	3.76	3.97	2.67	1.47	5.81
JAG2	0.69	1.29	1.13	0.70	1.01	1.40	0.73	1.23	1.11	3.69	4.07	3.96	3.53	3.41	3.41	2.81	3.30	2.82
NOTCH4	0.79	1.20	1.06	0.79	1.03	1.22	1.04	1.01	0.96	3.72	4.01	4.02	4.35	6.35	4.92	4.19	3.84	2.69
NRARP	0.87	1.04	1.10	0.79	0.94	1.34	0.82	1.09	1.12	3.48	3.59	3.09	2.98	3.66	4.23	2.99	2.52	2.65
HES4	0.83	1.24	0.97	0.92	0.94	1.15	0.75	1.11	1.19	4.84	5.74	5.02	4.10	5.33	4.79	4.59	4.82	3.27
TIMP3	1.15	0.78	1.12	0.95	1.02	1.03	1.26	0.81	0.98	10.98	10.9	12.24	5.156	5.643	4.86	5.92	6.38	4.59
ADAMTS1	0.99	1.41	0.72	nd	nd	nd	0.91	1.28	0.86	0.70	1.02	0.85	nd	nd	nd	0.66	0.49	0.73
ITGa6	1.09	0.82	1.11	0.99	1.03	0.97	0.90	1.40	1.12	2.07	2.29	2.31	2.03	1.84	2.06	1.59	2.41	1.59
THBS1	1.07	1.01	0.92	0.96	0.97	1.08	0.78	1.40	0.91	1.75	2.48	1.70	1.90	2.21	1.89	1.90	1.47	1.50
SPARC	0.86	0.98	1.19	0.74	1.19	1.13	0.80	1.07	1.17	2.79	2.64	3.45	2.76	2.67	2.65	2.25	2.34	2.25
CDK1	0.89	nd	1.13	0.85	0.98	1.20	0.76	1.08	1.22	0.22	0.26	0.22	0.19	0.19	0.22	0.22	0.23	0.28
CCNA2	0.89	nd	1.12	0.77	1.21	1.07	0.88	1.00	1.13	0.21	0.22	0.22	0.14	0.15	0.16	0.14	0.19	0.24
CCNB1	0.86	nd	1.16	0.83	1.04	1.16	0.95	1.01	1.05	0.27	0.30	0.25	0.18	0.22	0.23	0.20	0.26	0.28
MCM2	0.84	1.03	1.15	0.90	1.00	1.10	1.00	0.98	1.02	0.38	0.42	0.42	0.37	0.42	0.42	0.60	0.53	0.51
MCM4	0.80	1.07	1.17	0.80	1.11	1.13	0.85	0.89	1.32	0.43	0.47	0.40	0.35	0.43	0.38	0.35	0.40	0.58
CCND3	0.65	1.72	0.89	0.76	1.14	1.16	0.87	1.05	1.09	1.22	1.32	1.41	1.73	1.95	1.86	1.71	1.86	1.79
CCNG1	1.24	0.87	0.93	0.80	1.05	1.20	0.82	1.08	1.13	2.59	2.87	2.63	2.48	2.53	2.84	2.20	2.44	2.33

GENES	miR-NC VEGF 1h									miR-133a-3P VEGF 1h								
	0.90	0.91	1.06	0.85	0.94	1.02	1.02	0.95	1.01	0.39	0.49	0.51	0.34	0.40	0.24	0.47	0.50	0.58
PLAUR	0.90	0.91	1.06	0.85	0.94	1.02	1.02	0.95	1.01	0.39	0.49	0.51	0.34	0.40	0.24	0.47	0.50	0.58
MSN	1.11	1.10	1.13	0.85	1.02	1.00	1.04	0.88	1.07	0.69	0.64	0.59	0.32	0.40	0.53	0.66	0.67	0.72
RCAN1.4	14.95	6.75	18.70	13.23	10.52	9.45	13.19	12.79	16.98	76.10	68.17	58.90	63.43	78.58	69.36	62.67	80.67	51.54
CD44	0.90	0.98	0.86	1.01	0.97	0.92	1.15	1.21	0.95	0.28	0.25	0.29	0.14	0.19	0.21	0.34	0.36	0.39
ID1	0.76	0.45	0.42	0.87	0.94	1.25	0.56	0.50	0.48	0.40	0.25	0.53	0.29	0.30	0.43	0.45	0.46	0.39
CCNE1	1.05	1.77	0.98	1.78	1.14	1.02	0.96	1.20	0.88	0.80	0.96	0.93	0.69	0.99	0.76	0.64	0.61	0.72
CCND1	1.11	1.84	0.99	nd	1.72	0.84	1.32	1.69	1.04	0.99	1.30	1.29	0.81	0.78	0.85	0.62	0.70	0.85
DLL4	1.76	1.95	1.96	1.30	2.69	1.74	2.45	3.58	2.40	14.57	17.91	15.95	11.73	21.50	21.22	14.39	14.08	16.60
HEY1	4.31	6.20	4.73	3.28	9.32	2.07	4.11	10.89	3.21	19.52	21.93	15.03	13.20	11.10	12.63	17.99	14.11	16.80
JAG2	1.01	1.76	0.91	1.12	1.96	0.87	0.96	1.66	1.30	3.48	3.23	3.36	3.58	3.02	4.18	2.72	2.48	2.69
NOTCH4	1.43	1.36	0.70	1.22	2.41	1.01	0.81	1.30	0.89	2.59	3.12	2.79	4.34	4.94	4.19	2.69	2.89	2.92
NRARP	1.17	2.25	1.18	1.60	3.31	1.30	2.35	3.39	2.73	4.40	4.58	4.80	4.85	5.02	6.68	5.03	4.47	5.59
HES4	0.91	1.62	0.88	1.19	2.18	0.91	1.18	1.85	0.84	4.97	4.03	4.60	4.18	4.57	5.60	4.48	3.77	4.18
TIMP3	0.702	0.576	0.733	0.7719	0.63	0.65	1.164	0.614	1.225	6.51	5.79	6.77	5.44	5.18	5.79	5.01	5.06	5.95
ADAMTS1	3.37	3.05	5.27	nd	nd	nd	3.06	3.15	3.24	9.64	8.71	8.31	nd	nd	nd	6.14	4.75	5.91
ITGa6	0.83	0.90	0.90	0.87	0.82	0.71	0.95	0.87	1.09	1.26	1.50	1.59	2.16	1.67	1.61	1.50	0.92	1.50
THBS1	1.18	1.17	1.17	0.97	1.01	0.94	1.28	1.34	1.27	2.25	1.87	2.17	1.53	1.93	2.66	1.89	2.12	2.15
SPARC	1.15	1.00	1.07	0.88	1.41	0.65	1.09	1.57	1.35	2.21	2.18	2.4	2.08	1.78	2.27	2.24	2.56	2.21
CDK1	1.06	1.93	1.04	1.20	1.55	0.64	1.13	2.15	0.96	0.25	0.23	0.30	0.17	0.16	0.20	0.22	0.25	0.28
CCNA2	1.12	1.98	1.09	1.73	1.08	1.09	0.74	1.34	0.65	0.20	0.23	0.27	0.13	0.18	0.13	0.12	0.11	0.15
CCNB1	1.15	2.05	0.93	1.04	1.59	1.01	1.11	1.44	0.82	0.29	0.25	0.32	0.18	0.14	0.19	0.19	0.17	0.26
MCM2	1.01	1.49	0.99	1.05	1.46	0.78	1.14	1.89	1.16	0.36	0.37	0.44	0.49	0.43	0.50	0.36	0.39	0.47
MCM4	1.21	1.83	0.92	1.03	1.69	0.91	0.88	1.18	0.76	0.50	0.48	0.30	0.37	0.33	0.40	0.29	0.25	0.35
CCND3	0.73	1.15	0.64	1.24	1.98	1.05	0.69	1.01	0.55	1.00	1.03	1.09	1.73	1.76	2.02	0.86	0.85	0.90
CCNG1	1.23	1.98	1.07	0.99	1.58	0.97	0.74	1.38	0.87	2.49	2.51	2.62	1.71	2.31	2.10	1.89	1.60	1.81

Supplementary Table S4. Differential gene expression of angiogenic-related genes in HUVEC expressing miR-133a-3p or miR-NC. HUVEC transfected with miR-133a-3p (blue columns) or negative control miR-NC (grey columns) were left unstimulated (0h) or were stimulated with VEGF-A165 for 1 hour. Rows show fold-induction RNA levels for each gene referred to miR-NC 0h. Ct values were normalised using the Ct values for the housekeeping gene *Hprt-1*. Analysis of data was carried out using the comparative $2^{-\Delta\Delta Ct}$ method. nd = non-determined.

Genes	miR-133a-3p	miR-133a-3p	miR-133a-3p	miR-133a-5p	miR-133a-5p	miR-133a-5p
	miRWalk	TargetScan	miRDB	miRWalk	TargetScan	miRDB
<i>VEGF-A</i>	YES/5UTR	NO	NO	NO	NO	NO
<i>VEGF-B</i>	NO	NO	NO	YES	YES	NO
<i>VEGF-C</i>	NO	NO	NO	NO	NO	NO
<i>VEGFR1</i>	NO	NO	NO	NO	NO	NO
<i>VEGFR2</i>	NO	NO	NO	NO	NO	NO
<i>VEGFR3</i>	NO	NO	NO	NO	NO	NO
<i>CALCINEURIN-A</i>	YES	NO	NO	NO	NO	NO
<i>CALCINEURIN-B</i>	YES	NO	NO	NO	NO	NO
<i>NFAT1</i>	NO	NO	NO	NO	NO	NO
<i>NFAT2</i>	NO	NO	NO	NO	NO	NO
<i>NFAT3</i>	YES	NO	NO	YES	NO	NO
<i>NFAT4</i>	YES	NO	NO	NO	NO	NO
<i>NFAT5</i>	YES	YES	YES	YES	YES	YES
<i>JNK</i>	NO	NO	NO	YES	NO	NO
<i>p38</i>	NO	NO	NO	NO	NO	NO
<i>ERK</i>	NO	NO	NO	NO	NO	NO

Supplementary Table S5. Bioinformatic analysis of miR-133a predicted binding sites in candidate genes encoding well-known mediators of VEGF-induced angiogenesis.

The sequences of the indicated genes were analysed for potential binding sites for miR-133a-3p, or miR-133a-5p using miRWalk, Targetscan, and miRDB data bases.

Genes	miR-133a-3p miRWalk	miR-133a-3p TargetScan	miR-133a-3p miRDB	miR-133a-5p miRWalk	miR-133a-5p TargetScan	miR-133a-5p miRDB
<i>PLAUR</i>	YES	NO	NO	NO	NO	NO
<i>MSN</i>	NO	YES	YES	NO	NO	NO
<i>RCAN1.4</i>	YES	NO	NO	YES	YES	NO
<i>CD44</i>	YES	NO	NO	YES	YES	NO
<i>ID1</i>	NO	NO	NO	NO	NO	NO
<i>CCNE1</i>	YES	NO	NO	YES	NO	NO
<i>CCND1</i>	YES	NO	NO	NO	NO	NO
<i>DLL4</i>	YES	NO	NO	YES	NO	NO
<i>HEY1</i>	YES	NO	NO	YES	YES	NO
<i>JAG2</i>	YES	NO	NO	YES	NO	NO
<i>NOTCH4</i>	YES	NO	NO	YES	NO	NO
<i>NRARP</i>	NO	NO	NO	NO	NO	NO
<i>HES4</i>	YES	NO	NO	NO	YES	NO
<i>TIMP3</i>	YES	NO	NO	YES	NO	NO
<i>ADAMTS1</i>	YES	NO	NO	YES	YES	NO
<i>ITGA6</i>	NO	NO	NO	YES	NO	NO
<i>THBS1</i>	YES	NO	NO	YES	NO	NO
<i>SPARC</i>	YES	NO	NO	NO	NO	NO
<i>CDK1</i>	NO	NO	NO	NO	YES	NO
<i>CCNA2</i>	NO	NO	NO	NO	NO	NO
<i>CCNB1</i>	YES	NO	NO	NO	NO	NO
<i>MCM2</i>	NO	NO	NO	YES	NO	NO
<i>MCM4</i>	NO	NO	NO	NO	NO	NO
<i>CCND3</i>	YES	NO	NO	NO	YES	YES
<i>CCNG1</i>	NO	NO	NO	NO	NO	NO

Supplementary Table S6. Bioinformatic analysis of miR-133a predicted binding sites in the differentially expressed group of genes described in this study. The sequences of the indicated genes were analysed for potential binding sites for miR-133a-3p, or miR-133a-5p using miRWalk, Targetscan, and miRDB data bases. None of the genes was predicted to contain miR-133a binding sites by all the data bases used.

Genes	Taqman Assay ID
<i>MSN</i>	Hs00741306_mH
<i>PLAUR</i>	Hs00958880_m1
<i>CD44</i>	Hs01075864_m1
<i>ID1</i>	Hs03676575_s1
<i>CCNE1</i>	Hs01026536_m1
<i>CCND1</i>	Hs00765553_m1
<i>DLL4</i>	Hs00184092_m1
<i>HEY1</i>	Hs05047713_s1
<i>JAG2</i>	Hs00171432_m1
<i>NOTCH4</i>	Hs00965889_m1
<i>NRARP</i>	Hs04183811_s1
<i>HES4</i>	Hs00368353_g1
<i>TIMP3</i>	Hs00165949_m1
<i>ITGA6</i>	Hs01041011_m1
<i>ADAMTS1</i>	Hs00199608_m1
<i>THBS1</i>	Hs00962908_m1
<i>SPARC</i>	Hs00234160_m1
<i>CDK1</i>	Hs00938777_m1
<i>CCNA2</i>	Hs00996788_m1
<i>CCNB1</i>	Hs01030099_m1
<i>MCM2</i>	Hs01091564_m1
<i>MCM4</i>	Hs00907398_m1
<i>CCND3</i>	Hs01017690_g1
<i>CCNG1</i>	Hs00171112_m1
<i>RCAN1.4</i>	Hs01120957_m1
<i>HPRT1</i>	Hs99999909_m1

Supplementary Table S7. References of TaqMan Gene Expression Assays (Applied Biosystems) used in this work.