Molecular Medicine

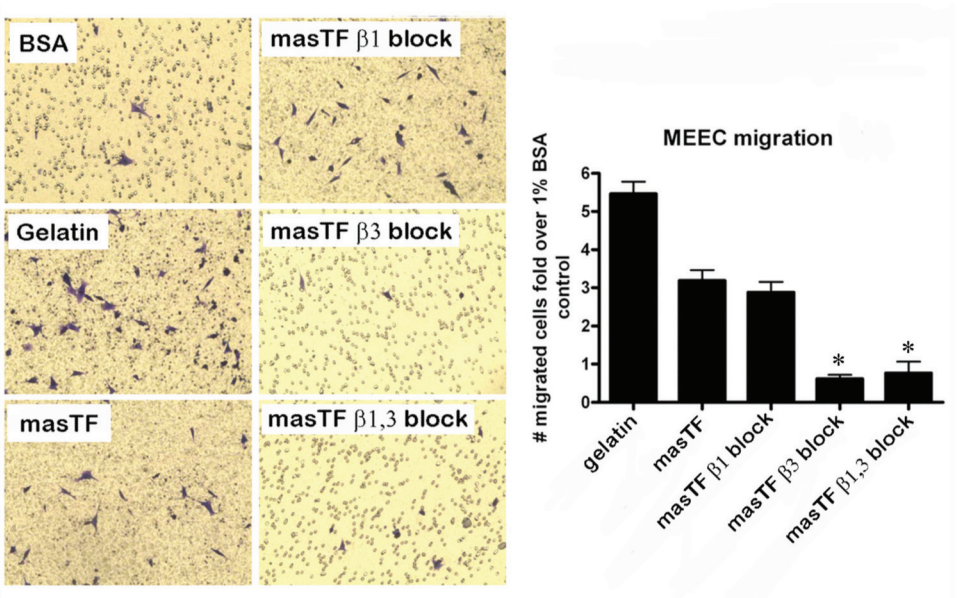
Supplemental Data

Nonproteolytic Properties of Murine Alternatively Spliced Tissue Factor: Implications for Integrin-Mediated Signaling in Murine Models

Richard C Godby,[1] Yascha W van den Berg,[2*] Ramprasad Srinivasan,[1] Robert Sturm,[1] David Y Hui,[3] Stephen F Konieczny,[4] Bruce J Aronow,[5] Evgeny Ozhegov,[1] Wolfram Ruf,[6] Henri H Versteeg,[2*] and Vladimir Y Bogdanov[1*]*

Online address: http://www.molmed.org

The Feinstein Institute for Medical Research North Shore LIJ



Supplementary Figure S1. masTF elicits β3 integrin-dependent migration of MEEC. Transwell inserts were coated with BSA (negative control), 1% gelatin (positive control), and 50 μg/mL masTF, following which MEEC were pre-incubated with integrin-blocking antibodies and seeded onto the wells (n≥3). Flattened cells were counted. Cells were fixed with 4% formaldehyde and stained with 0.1% Crystal Violet; quantifications are shown in the graph, representative images are on the left (n≥3). *p<0.01 vs masTF.

Supplementary Table S1. Genes expressed in murine ECs (b.End.3) whose transcripts were upregulated by at least 1.2 fold or downregulated by at least 0.9 fold in response to hasTF and masTF.

No.	Gene Symbol	Gene Description	Fold Change
1	Cxcl2	chemokine (C-X-C motif) ligand 2	29.3
2	Vcam1	vascular cell adhesion molecule 1	6.5
3	Cxcl1	chemokine (C-X-C motif) ligand 1	5.2
4	Ccl2	chemokine (C-C motif) ligand 2	3.7
5	Csf1	colony stimulating factor 1 (macrophage)	3.7
6	Cxcl10	chemokine (C-X-C motif) ligand 10	3.1
7	Ccl7	chemokine (C-C motif) ligand 7	2.4
8	Ripk2	receptor (TNFRSF)-interacting serine-threonine kinase 2	2.4
9	Cebpd	CCAAT/enhancer binding protein (C/EBP), delta	2.2
10	Casp4	caspase 4, apoptosis-related cysteine peptidase	2.1
11	Nfkbia	nuclear factor of kappa light polypeptide gene enhancer in B-cells inhibitor, alpha	1.8
12	Birc3	baculoviral IAP repeat-containing 3	1.8
13	Gbp1	guanylate binding protein 1	1.8
14	Tslp	thymic stromal lymphopoietin	1.7
15	Snord49b	small nucleolar RNA, C/D box 49B	1.6
16	Ch25h	cholesterol 25-hydroxylase	1.6
17	Ccl5	chemokine (C-C motif) ligand 5	1.6
18	Cenpw	centromere protein W	1.6
19	Mrpl52	mitochondrial ribosomal protein L52	1.5
20	Ddx28	DEAD (Asp-Glu-Ala-Asp) box polypeptide 28	1.5
21	Endog	endonuclease G	1.5
22	Purb	purine rich element binding protein B	1.5
23	Rpia	ribose 5-phosphate isomerase A	1.5
24	Samd4	sterile alpha motif domain containing 4	1.5
25	Lclat1	lysocardiolipin acyltransferase 1	1.4
26	Cdkn2b	cyclin-dependent kinase inhibitor 2B	1.4
27	Chd7	chromodomain helicase DNA binding protein 7	1.4
28	SNORA38	Small nucleolar RNA 38	1.4
29	Bach1	BTB and CNC homology 1	1.4
30	Zadh2	zinc binding alcohol dehydrogenase, domain containing 2	1.4
31	Tusc1	tumor suppressor candidate 1	1.4
32	Ndufa13	NADH dehydrogenase (ubiquinone) 1 alpha subcomplex, 13	1.4
33	E2f3	E2F transcription factor 3	1.4
34	Chst7	carbohydrate (N-acetylglucosamino) sulfotransferase 7	1.3
35	Chic2	cysteine-rich hydrophobic domain 2	1.3
36	Jund	Jun proto-oncogene related gene d	1.3
37	Cebpb	CCAAT/enhancer binding protein (C/EBP), beta	1.3
38	Mfsd7b	major facilitator superfamily domain containing 7B	1.3
39	Mafb	v-maf musculoaponeurotic fibrosarcoma oncogene family, protein B	1.3
40	Ntn1	netrin 1	1.3
41	Taf1d	TATA box binding protein (Tbp)-associated factor, RNA polymerase I, D	1.3
42	Gas5	SNORD77	1.3
43	Magohb	mago-nashi homolog B	1.3
44	Aim2	absent in melanoma 2	1.3
45	Rtn4rl2	reticulon 4 receptor-like 2	1.3
46	Ccdc9	coiled-coil domain containing 9	1.3
47	Dmwd	dystrophia myotonica-containing WD repeat motif	1.3
48	Ocel1	occludin/ELL domain containing 1	1.3
49	H2-Ke2	H2-K region expressed gene 2	1.3
50	Mrpl47	mitochondrial ribosomal protein L47	1.3
51	Atf7	activating transcription factor 7	1.3
52	Dpysl3	dihydropyrimidinase-like 3	1.3

Continued

Supplementary Table S1. ***Continued.***

53	Nudt3	nudix (nucleotide diphosphate linked moiety X)-type motif 3	1.3
54	Prr23a	proline rich 23A	1.3
55	Strn4	striatin, calmodulin binding protein 4	1.3
56	Polr2c	polymerase (RNA) II polypeptide C	1.3
57	Rhou	ras homolog gene family, member U	1.3
58	Ifi204	interferon activated gene 204	1.3
59	Tshz1	teashirt zinc finger family member 1	1.3
60	Brp44	brain protein 44	1.3
61	Chd7	chromodomain helicase DNA binding protein 7	1.2
62	Sash1	SAM and SH3 domain containing 1	1.2
63	Prkrip1	Prkr interacting protein 1 (IL11 inducible)	1.2
64	Nkiras1	NFKB inhibitor interacting Ras-like protein 1	1.2
65	Map1lc3a	microtubule-associated protein 1 light chain 3 alpha	1.2
66	Rala	v-ral simian leukemia viral oncogene homolog A (ras related)	1.2
67	Chd2	chromodomain helicase DNA binding protein 2	1.2
68	Zeb2	zinc finger E-box binding homeobox 2	1.2
69	Junb	Jun-B oncogene	1.2
70	Pbld2	phenazine biosynthesis-like protein domain containing 2	1.2
71	Frmd6	FERM domain containing 6	1.2
72	Ssbp4	single stranded DNA binding protein 4	1.2
73	Aqp2	aquaporin 2	1.2
74	Mrps16	mitochondrial ribosomal protein S16	1.2
75	Trav2	T cell receptor alpha variable 2	1.2
76	Polr3d	polymerase (RNA) III polypeptide D	1.2
77	Ccdc66	coiled-coil domain containing 66	1.2
78	Ccdc90b	coiled-coil domain containing 90B	1.2
79	Med26	mediator complex subunit 26	1.2
80	Nfya	nuclear transcription factor-Y alpha	1.2
81	Golph3	golgi phosphoprotein 3	1.2
82	Wdfy1	WD repeat and FYVE domain containing 1	1.2
83	Cdc42ep4	CDC42 effector protein (Rho GTPase binding) 4	1.2
84	Zar1	zygote arrest 1	1.2
85	Nosip	nitric oxide synthase interacting protein	1.2
86	Polr3gl	polymerase (RNA) III polypeptide G like	1.2
87	Gm5939	predicted pseudogene 5939	1.2
88	Vax1	ventral anterior homeobox containing gene 1	1.2
89	Igfbp7	insulin-like growth factor binding protein 7	1.2
90	Gm7312	predicted gene 7312	1.2
91	Sdc4	syndecan 4	1.2
92	Akap12	A kinase (PRKA) anchor protein (gravin) 12	1.2
93	Mir615	microRNA 615	1.2
94	Rpl14	ribosomal protein L14	1.2
95	Mrps34	mitochondrial ribosomal protein S34	1.2
96	Cars2	cysteinyl-tRNA synthetase 2 (mitochondrial)	0.9
97	Gm16410	predicted gene 16410	0.9
98	Sh3d19	SH3 domain protein D19	0.8
99	Stard7	START domain containing 7	0.8
100	Klf3	Kruppel-like factor 3 (basic)	0.8
101	Dnalc4	dynein, axonemal, light chain 4	0.8
102	Ccng2	cyclin G2	0.8
103	Gm4831	predicted gene 4831	0.8
104	Ltbp3	latent transforming growth factor beta binding protein 3	0.8
105	Marveld1	MARVEL (membrane-associating) domain containing 1	0.8
106	Itpripl1	inositol 1,4,5-triphosphate receptor interacting protein-like 1	0.8
107	Nans	N-acetylneuraminic acid synthase	0.8

Continued

Supplementary Table S1. ***Continued.***

108	Crtc1	CREB regulated transcription coactivator 1	0.8
109	Rassf3	Ras association (RalGDS/AF-6) domain family member 3	0.8
110	Fam102a	family with sequence similarity 102, member A	0.8
111	Nefl	neurofilament, light polypeptide	0.8
112	Cyb561d2	cytochrome b-561 domain containing 2	0.8
113	Rgs3	regulator of G-protein signaling 3	0.8
114	Ssna1	Sjogren's syndrome nuclear autoantigen 1	0.8
115	Mrpl51	mitochondrial ribosomal protein L51	0.8
116	Zfp362	zinc finger protein 362	0.8
117	Acvrl1	activin A receptor, type II-like 1	0.8
118	Lrrcc1	leucine rich repeat coiled-coil domain containing 1	0.8
119	Fbxl2	F-box and leucine-rich repeat protein 2	0.8
120	Slc46a3	solute carrier family 46, member 3	0.8
121	Zdhhc8	zinc finger, DHHC domain containing 8	0.8
122	Dock8	dedicator of cytokinesis 8	0.8
123	Rassf9	Ras association (RalGDS/AF-6) domain family (N-terminal) member 9	0.8
124	Cep57l1	centrosomal protein 57-like 1	0.8
125	Cxx1c	CAAX box 1 homolog C (human)	0.8
126	Ncoa7	nuclear receptor coactivator 7	0.8
127	Gnptab	N-acetylglucosamine-1-phosphate transferase, alpha and beta subunits	0.8
128	Hdac5	histone deacetylase 5	0.8
129	Acaa2	acetyl-Coenzyme A acyltransferase 2 (mitochondrial 3-oxoacyl-Coenzyme A thiolase)	0.8
130	Caskin2	CASK-interacting protein 2	0.8
131	Tfdp2	transcription factor Dp 2	0.8
132	Tgfbr2	transforming growth factor, beta receptor II	0.8
133	Gpr137	G protein-coupled receptor 137	0.8
134	Tcta	T-cell leukemia translocation altered gene	0.8
135	St6gal1	beta galactoside alpha 2,6 sialyltransferase 1	0.8
136	Zmym5	zinc finger, MYM-type 5	0.8
137	Nicn1	nicolin 1	0.8
138	Rfc2	replication factor C (activator 1) 2	0.8
139	Ephb4	Eph receptor B4	0.8
140	Uvrag	UV radiation resistance associated gene	0.8
141	Thsd1	thrombospondin, type I, domain 1	0.8
142	Ncapd2	non-SMC condensin I complex, subunit D2	0.8
143	Zdhhc3	zinc finger, DHHC domain containing 3	0.8
144	Znf512b	zinc finger protein 512B	0.8
145	Pik3r3	phosphatidylinositol 3 kinase, regulatory subunit, polypeptide 3 (p55)	0.8
146	Map2k6	mitogen-activated protein kinase kinase 6	0.8
147	Grasp	GRP1 (general receptor for phosphoinositides 1)-associated scaffold protein	0.8
148	Cpt1a	carnitine palmitoyltransferase 1a, liver	0.8
149	Gbf1	golgi-specific brefeldin A-resistance factor 1	0.8
150	Tmem64	transmembrane protein 64	0.7
151	Epha4	Eph receptor A4	0.7
152	Zfp238	zinc finger protein 238	0.7
153	Rbms2	RNA binding motif, single stranded interacting protein 2	0.7
154	Dock6	dedicator of cytokinesis 6	0.7
155	Ddit4	DNA-damage-inducible transcript 4	0.7
156	Ggta1	glycoprotein galactosyltransferase alpha 1, 3	0.7
157	Zfp161	zinc finger protein 161	0.7
158	Dock6	dedicator of cytokinesis 6	0.7
159	Slc5a3	solute carrier family 5 (inositol transporters), member 3	0.7
160	Tob1	transducer of ErbB-2.1	0.7
161	Nfia	nuclear factor I/A	0.7
162	Tm6sf1	transmembrane 6 superfamily member 1	0.7

Continued

Supplementary Table S1. ***Continued.***

163	Ptprb	protein tyrosine phosphatase, receptor type, B	0.7
164	Sumf2	sulfatase modifying factor 2	0.7
165	Ube2cbp	ubiquitin-conjugating enzyme E2C binding protein	0.7
166	Stk10	serine/threonine kinase 10	0.7
167	Wdr85	WD repeat domain 85	0.7
168	Klhdc10	kelch domain containing 10	0.7
169	Pfkfb3	6-phosphofructo-2-kinase/fructose-2,6-biphosphatase 3	0.7
170	Tmem44	transmembrane protein 44	0.7
171	Asph	aspartate-beta-hydroxylase	0.7
172	Bpgm	2,3-bisphosphoglycerate mutase	0.7
173	Lrrk2	leucine-rich repeat kinase 2	0.7
174	Mavs	mitochondrial antiviral signaling protein	0.7
175	Rtp3	receptor transporter protein 3	0.7
176	Slc22a4	solute carrier family 22 (organic cation transporter), member 4	0.7
177	Ppcs	phosphopantothenoylcysteine synthetase	0.7
178	Ceacam1	carcinoembryonic antigen-related cell adhesion molecule 1	0.7
179	Sorbs2	sorbin and SH3 domain containing 2	0.7
180	Gm10482	predicted gene 10482	0.7
181	Gm10482	predicted gene 10482	0.7
182	Fam198b	family with sequence similarity 198, member B	0.7
183	Gm6981	predicted pseudogene 6981	0.7
184	Ttll5	tubulin tyrosine ligase-like family, member 5	0.7
185	Nfia	nuclear factor I/A	0.7
186	Gad2	glutamic acid decarboxylase 2	0.7
187	Herpud1	homocysteine-inducible, endoplasmic reticulum stress-inducible, ubiquitin-like domain member 1	0.7
188	Sema3g	sema domain, immunoglobulin domain (Ig), short basic domain, secreted, (semaphorin) 3G	0.7
189	Mecom	MDS1 and EVI1 complex locus	0.7
190	Mms22l	MMS22-like, DNA repair protein	0.7
191	Tenc1	tensin like C1 domain-containing phosphatase	0.7
192	Podxl	podocalyxin-like	0.7
193	Mef2c	myocyte enhancer factor 2C	0.7
194	Mkl2	MKL/myocardin-like 2	0.7
195	Trim25	tripartite motif-containing 25	0.7
196	Man1c1	mannosidase, alpha, class 1C, member 1	0.7
197	Fam101b	family with sequence similarity 101, member B	0.7
198	Gm10482	predicted gene 10482	0.7
199	Klf4	Kruppel-like factor 4 (gut)	0.7
200	Idh1	isocitrate dehydrogenase 1 (NADP+), soluble	0.7
201	Reps2	RALBP1 associated Eps domain containing protein 2	0.7
202	Top3b	topoisomerase (DNA) III beta	0.7
203	Epas1	endothelial PAS domain protein 1	0.7
204	Zfp787	zinc finger protein 787	0.7
205	Sema6a	sema domain, transmembrane domain (TM), and cytoplasmic domain, (semaphorin) 6A	0.7
206	Pdcd4	programmed cell death 4	0.7
207	Fam63b	family with sequence similarity 63, member B	0.7
208	Cln6	ceroid-lipofuscinosis, neuronal 6	0.7
209	Pias3	protein inhibitor of activated STAT 3	0.7
210	Ets2	E26 avian leukemia oncogene 2, 3′ domain	0.7
211	Abca1	ATP-binding cassette, sub-family A (ABC1), member 1	0.7
212	Foxp1	forkhead box P1	0.7
213	Rab27a	RAB27A, member of RAS oncogene family	0.7
214	Actr6	ARP6 actin-related protein 6 homolog (yeast)	0.7
215	Npnt	nephronectin	0.6
216	Kctd12b	potassium channel tetramerisation domain containing 12b	0.6
217	Lmo2	LIM domain only 2	0.6

Continued

Supplementary Table S1. ***Continued.***

218	Hadh	hydroxyacyl-Coenzyme A dehydrogenase	0.6
219	Mkl2	MKL/myocardin-like 2	0.6
220	Arhgef3	Rho guanine nucleotide exchange factor 3	0.6
221	Myct1	myc target 1	0.6
222	Cc2d2a	coiled-coil and C2 domain containing 2A	0.6
223	Sepp1	selenoprotein P, plasma, 1	0.6
224	Zfp626	zinc finger protein 626	0.6
225	Mal	myelin and lymphocyte protein, T-cell differentiation protein	0.6
226	Meis2	Meis homeobox 2	0.6
227	Dnm3	dynamin 3	0.6
228	Snapin	SNAP-associated protein	0.6
229	Ttll1	tubulin tyrosine ligase-like 1	0.6
230	Cyp2j6	cytochrome P450, family 2, subfamily j, polypeptide 6	0.6
231	Cd97	CD97 antigen	0.6
232	Cyyr1	cysteine and tyrosine-rich protein 1	0.6
233	Brca2	breast cancer 2	0.6
234	Tmtc2	transmembrane and tetratricopeptide repeat containing 2	0.6
235	Ctnnal1	catenin (cadherin associated protein), alpha-like 1	0.6
236	Abhd14b	abhydrolase domain containing 14b	0.6
237	Crybg3	beta-gamma crystallin domain containing 3	0.6
238	Scin	scinderin	0.6
239	Dusp6	dual specificity phosphatase 6	0.6
240	Mansc1	MANSC domain containing 1	0.6
241	Car5b	carbonic anhydrase 5b, mitochondrial	0.6
242	Sox17	SRY-box containing gene 17	0.6
243	Galntl4	UDP-N-acetyl-alpha-D-galactosamine:polypeptide N-acetylgalactosaminyltransferase-like 4	0.6
244	Entpd1	ectonucleoside triphosphate diphosphohydrolase 1	0.6
245	Pla2g16	phospholipase A2, group XVI	0.6
246	Ddx6	DEAD (Asp-Glu-Ala-Asp) box polypeptide 6	0.6
247	Rb1	retinoblastoma 1	0.6
248	Dusp19	dual specificity phosphatase 19	0.6
249	Timp3	tissue inhibitor of metalloproteinase 3	0.5
250	Rnf125	ring finger protein 125	0.5
251	Orc5	origin recognition complex, subunit 5	0.5
252	Scd2	stearoyl-Coenzyme A desaturase 2	0.5
253	Txnip	thioredoxin interacting protein	0.5
254	Mras	muscle and microspikes RAS	0.5
255	Adamts5	a disintegrin-like and metallopeptidase (reprolysin type) with thrombospondin type 1 motif, 5 (aggrecanase-2)	0.5
256	Tmem200a	transmembrane protein 200A	0.5
257	Gja5	gap junction protein, alpha 5	0.5
258	Cdc42ep1	CDC42 effector protein (Rho GTPase binding) 1	0.5
259	Nynrin	NYN domain and retroviral integrase containing	0.5
260	Ebf1	early B-cell factor 1	0.5
261	Stxbp4	syntaxin binding protein 4	0.4
262	Suox	sulfite oxidase	0.4
263	Arrdc3	arrestin domain containing 3	0.4
264	Chic1	cysteine-rich hydrophobic domain 1	0.3