

Supplementary material

Article title: Tumor kinase activity in locally advanced rectal cancer – angiogenic signaling and early systemic dissemination

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Author names: Marie Grøn Saelen, Kjersti Flatmark, Sigurd Folkvord, Rik de Wijn, Heidi Rasmussen, Øystein Fodstad, Anne Hansen Ree

Corresponding author: A. H. Ree, Department of Oncology, Akershus University Hospital, 1478 Lørenskog, Norway. E-mail: a.h.ree@medisin.uio.no.

Table 1 The 102 array substrates included in the sunitinib inhibition profile

Peptide substrate ^a	Gene name – Common name ^b	Difference [*]	P ^{**}
41_Y660	EPB41 - Protein 4.1	0.103	0.239
ACHD_Y383/Y390	CHRND - Acetylcholine receptor subunit delta	0.098	0.150
AMPE_Y12	ENPEP - Glutamyl aminopeptidase	0.014	0.840
ANXA1_Y21	ANXA1 - ANXA1 protein	0.078	0.265
ANXA2_Y24	ANXA2 - Annexin A2	0.198	0.060
ART_YAAPFAKKKXC	Artificial peptide	0.197	0.062
C1R_Y204/Y210	C1R - Complement C1r subcomponent	-0.008	0.881
CALM_Y100	CALM1 - Calmodulin	0.174	0.012
CBL_Y700	CBL - CBL E3 ubiquitin protein ligase	0.064	0.321
CD3Z_Y153	CD247 - T-cell surface glycoprotein CD3 zeta chain	0.280	0.018
CDK2_Y15/Y19	CDK2 - Cell division protein kinase 2	0.282	0.025
CDK7_Y169	CDK7 - Cell division protein kinase 7	0.020	0.796
CTNB1_Y86	Beta-catenin - CTNNB1	0.193	0.011
DCX_Y112	DCX - Neuronal migration protein doublecortin	0.097	0.290
DDR1_Y513	DDR1 - Epithelial discoidin domain receptor 1	0.216	0.055
DYR1A_Y319/Y321	DYRK1A - Dual-specificity tyrosine-phosphorylation regulated kinase 1A	0.067	0.361
EGFR_Y1110	EGFR - Epidermal growth factor receptor	0.145	0.110
EGFR_Y1172	EGFR - Epidermal growth factor receptor	0.051	0.315
EGFR_Y1197	EGFR - Epidermal growth factor receptor	0.101	0.036
EPHA1_Y781	EPHA1 - Ephrin type-A receptor 1	0.222	0.051
EPHA2_Y772	EPHA2 - Ephrin type-A receptor 2	0.205	0.078

EPHA4_Y596	EPHA3 - Ephrin type-A receptor 3	0.009	0.904
EPHA7_Y608/Y614	EPHA7 - Ephrin type-A receptor 7	0.236	0.033
EPHB1_Y778	EPHB1 - Ephrin type-B receptor 1	0.250	0.015
EPHB4_Y590	EPHB4 - Ephrin type-B receptor 4	0.008	0.866
EPOR_Y368	EPOR - Erythropoietin receptor	0.259	0.008
EPOR_Y426	EPOR - Erythropoietin receptor	0.265	0.013
ERBB2_Y1248	ERBB2 - Receptor tyrosine-protein kinase erbB-2	0.131	0.053
ERBB2_Y877	ERBB2 - Receptor tyrosine-protein kinase erbB-2	0.094	0.214
ERBB4_Y1284	ERBB4 - Receptor tyrosine-protein kinase erbB-4	0.073	0.240
FAK1_Y570/Y576/Y577	PTK2 - Focal adhesion kinase 1	0.100	0.130
FAK2_Y573/Y579/Y580	PTK2B - Focal adhesion kinase 2 beta	0.115	0.151
FER_Y714	FER - Proto-oncogene tyrosine-protein kinase FER	0.215	0.031
FES_Y713	FES - Proto-oncogene tyrosine-protein kinase Fes/Fps	0.261	0.025
FGFR1_Y766	FGFR1 - Basic fibroblast growth factor receptor 1	0.054	0.324
FGFR2_Y769	FGFR2 - Fibroblast growth factor receptor 2	0.095	0.135
FGFR3_Y760	FGFR3 - Fibroblast growth factor receptor 3	0.066	0.274
FRK_Y387	FRK - Tyrosine-protein kinase FRK	0.248	0.040
INSR_Y1355	INSR - Insulin receptor	-0.059	0.298
INSR_Y992/Y999	INSR - Insulin receptor	0.050	0.304
JAK1_Y1022/Y1023	JAK1 - Tyrosine-protein kinase JAK1	0.173	0.030
JAK2_Y570	JAK2 - Tyrosine-protein kinase JAK2	0.160	0.052
K2C6E_Y62	KRT6E - Keratin, type II cytoskeletal 6E	0.173	0.042
KSYK_Y525/Y526	SYK - Tyrosine-protein kinase SYK	0.087	0.071
LAT_Y200	LAT - Linker for activation of T cells	0.075	0.143
LAT_Y255	LAT - Linker for activation of T cells	0.123	0.101
LCK_Y394	LCK - Proto-oncogene tyrosine-protein kinase LCK	0.152	0.017
MBP_Y203	MBP - Myelin basic protein	0.032	0.700
MBP_Y261/Y268	MBP - Myelin basic protein	0.020	0.705
MBP_Y268	MBP - Myelin basic protein	0.060	0.338
MET_Y1230/Y1234/Y1235	MET - Hepatocyte growth factor receptor	0.133	0.142
MK01_Y187	MAPK1 - Mitogen-activated protein kinase 1	0.051	0.460
MK07_Y220	MAPK7 - Mitogen-activated protein kinase 7	0.087	0.321
MK10_Y223	MAPK10 - Mitogen-activated protein kinase 10	0.104	0.156
MK12_Y185	MAPK12 - Mitogen-activated protein kinase 12	-0.014	0.758
NCF1_Y324	NCF1 - Neutrophil cytosol factor 1	0.008	0.921
NPT2_Y511	SLC34A1 - Renal sodium-dependent phosphate transport protein 2	0.020	0.784
NTRK1_Y496	NTRK1 - High affinity nerve growth factor receptor	-0.105	0.055
NTRK2_Y702/Y706/Y707	NTRK2 - BDNF/NT-3 growth factors receptor	0.160	0.059
ODBA_Y345	BCKDHA - 2-oxoisovalerate dehydrogenase alpha subunit, mitochondrial PPP2CB - Serine/threonine protein phosphatase 2A, catalytic subunit, beta isoform	0.085	0.319
P2AB_Y307		0.029	0.597
P85A_Y607	PIK3R1 - Phosphatidylinositol 3-kinase regulatory alpha subunit	0.300	0.014
PAXI_Y118	PXN - Paxillin	0.252	0.025
PAXI_Y31/Y33	PXN - Paxillin	0.260	0.020
PDGFB_Y1021	PDGFRB - Beta platelet-derived growth factor receptor	0.111	0.058
PDPK1_Y373/Y376	PDPK1 - 3-phosphoinositide dependent protein kinase 1	0.117	0.169
PDPK1_Y9	PDPK1 - 3-phosphoinositide dependent protein kinase 2	0.212	0.016

PECA1_Y713	PECAM1- Platelet endothelial cell adhesion molecule	0.272	0.021
PGFRB_Y1009	PDGFRB - Beta platelet-derived growth factor receptor	0.241	0.002
PGFRB_Y579/Y581	PDGFRB - Beta platelet-derived growth factor receptor	0.223	0.044
PGFRB_Y716	PDGFRB - Beta platelet-derived growth factor receptor	0.179	0.029
PGFRB_Y771/Y775/Y778	PDGFRB - Beta platelet-derived growth factor receptor	0.182	0.020
PGFRB_Y771/Y775/Y778	PDGFRB - Beta platelet-derived growth factor receptor	0.177	0.036
PLCG1_Y771/775	PLCG1 - 1-phosphatidylinositol-4,5-bisphosphate phosphodiesterase gamma 1	0.233	0.050
PRGR_Y795	PGR - Progesterone receptor	0.108	0.122
PRRX2_Y208/Y214	PRRX2 -Paired mesoderm homeobox protein 2	0.111	0.105
PTN11_Y546/551	PTPN11 - Tyrosine-protein phosphatase, non-receptor type 11	0.086	0.199
RAF1_Y340/Y341	RAF1 - RAF proto-oncogene serine/threonine-protein kinase	0.088	0.297
RASA1_Y460	RASA1 - Ras GTPase-activating protein 1	0.171	0.086
RB_Y805/Y813	RB1 - Retinoblastoma-associated protein	0.066	0.402
RET_Y1029	RET - Proto-oncogene tyrosine-protein kinase receptor ret	0.250	0.035
RON_Y1353	MST1R - Macrophage-stimulating protein receptor	0.133	0.177
RON_Y1353/Y1360	MST1R - Macrophage-stimulating protein receptor	0.154	0.017
SRC8_Y477	CTTN - Src substrate protein p85	0.091	0.201
SRC8_Y477/Y483	CTTN - Src substrate protein p86	0.297	0.021
SRC8_Y479/486/489	CTTN - Src substrate protein p87	0.149	0.227
TEC_Y513/519	TEC - Tyrosine-protein kinase Tec	0.132	0.078
TNNT1_Y9	TNNT1 - TNNT1 protein	0.020	0.809
TYRO3_Y681/685/686	TYRO3 - Tyrosine-protein kinase receptor TYRO3	0.049	0.430
VEGFR1_Y1048	FLT1 - Vascular endothelial growth factor receptor 1	0.106	0.164
VEGFR1_Y1053	FLT1 - Vascular endothelial growth factor receptor 1	0.055	0.445
VEGFR1_Y1242	FLT1 - Vascular endothelial growth factor receptor 1	-0.020	0.707
VEGFR1_Y1327/Y1333	FLT1 - Vascular endothelial growth factor receptor 1	0.156	0.042
VEGFR2_Y1054/Y1059	KDR - Vascular endothelial growth factor receptor 2	0.063	0.268
VEGFR2_Y1063	KDR - Vascular endothelial growth factor receptor 2	0.068	0.340
VEGFR2_Y1175	KDR - Vascular endothelial growth factor receptor 2	0.122	0.023
VEGFR2_Y951	KDR - Vascular endothelial growth factor receptor 2	0.035	0.605
VEGFR2_Y996	KDR - Vascular endothelial growth factor receptor 2	0.250	0.025
VEGFR3_Y1063/Y1068	FLT4 - Vascular endothelial growth factor receptor 3	0.018	0.749
VINC_Y822	VCL - Vinculin	0.033	0.587
ZAP70_Y492/Y493	ZAP70 - Tyrosine-protein kinase ZAP-70	0.153	0.087
ZBT16_Y630	ZBTB16 - Zinc finger and BTB domain containing protein 16	0.032	0.617

^a Substrate names from the Tyrosine Kinase PamChip96 Array (PamGene International B.V, 's-Hertogenbosch, The Netherlands). For each substrate, position of phosphorylation sites within the protein is indicated

^b Gene names and common names, retrieved from UniProtKB/SwissProt (<http://au.expasy.org/sprot>)

* Calculated as mean \log_2 inhibition in patients positive for disseminated tumor cells to bone

marrow minus mean \log_2 inhibition in negative patients

** By two-sample *t*-test

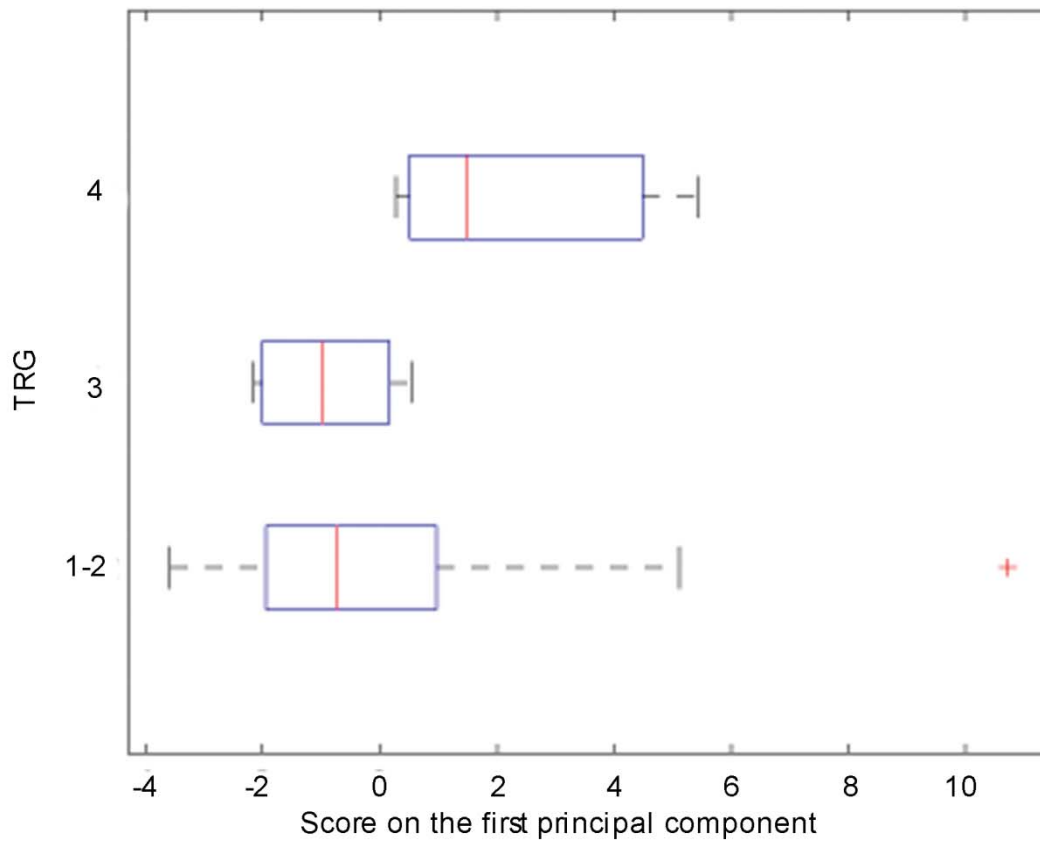


Fig. 1 Distribution of the score of the first principal component of the 102 peptide substrates among the groups of patients with different histomorphologic tumor regression grade (TRG). $P = 0.049$

TRG 1–2, good responders; TRG 3, intermediate responders; TRG 4, poor responders

Boxes, 25th, 50th, and 75th percentiles; bars, 10th and 90th percentiles; crosses, outlier values

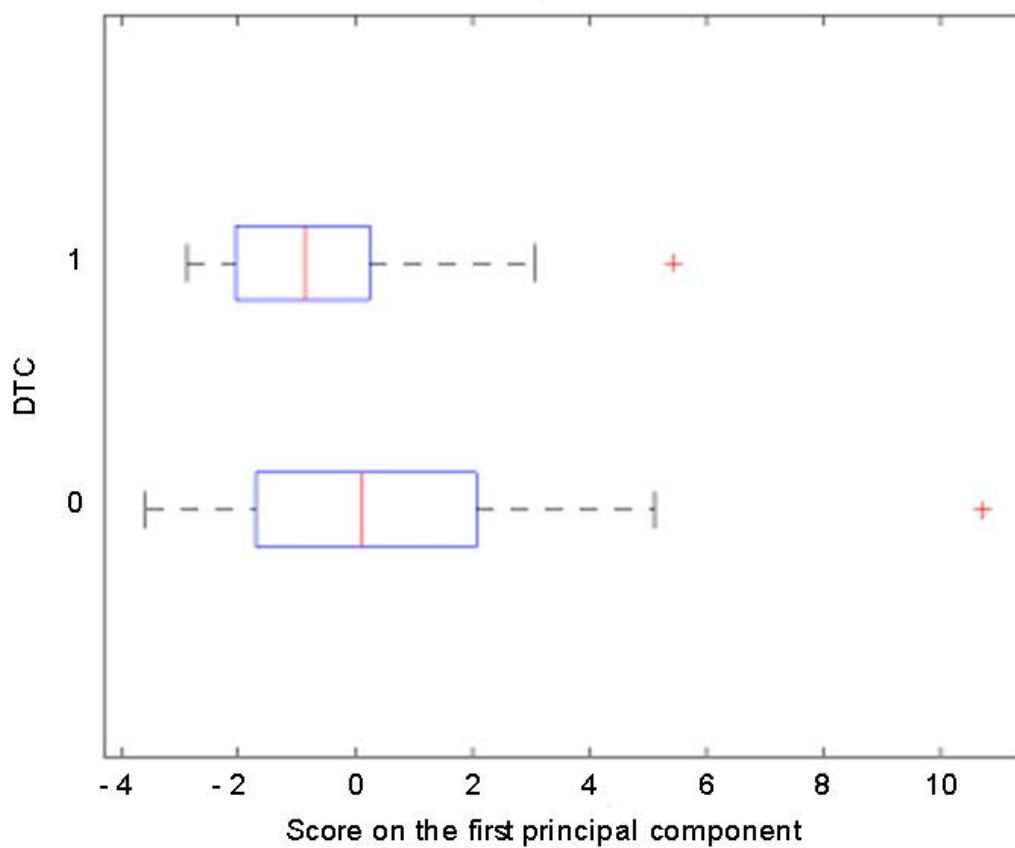


Fig. 2 Distribution of the score of the first principal component of the 102 peptide substrates among patients with positive and negative status for disseminated tumor cells (DTC) to bone marrow. $P = 0.042$

DTC 0, negative DTC status; DTC 1, positive DTC status

Boxes, 25th, 50th, and 75th percentiles; bars, 10th and 90th percentiles; crosses, outlier values

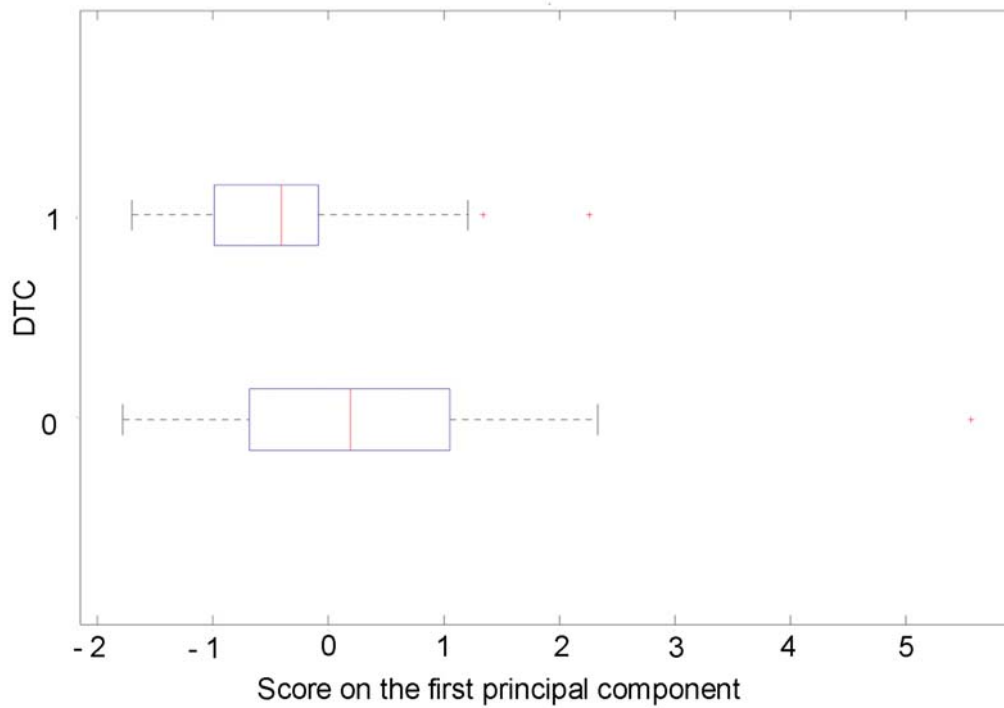


Fig. 3 Distribution of the score of the first principal component of the 23 angiogenesis-related peptide substrates among patients with positive and negative status for disseminated tumor cells (DTC) to bone marrow. $P = 0.019$

DTC 0, negative DTC status; DTC 1, positive DTC status

Boxes, 25th, 50th, and 75th percentiles; bars, 10th and 90th percentiles; crosses, outlier values