

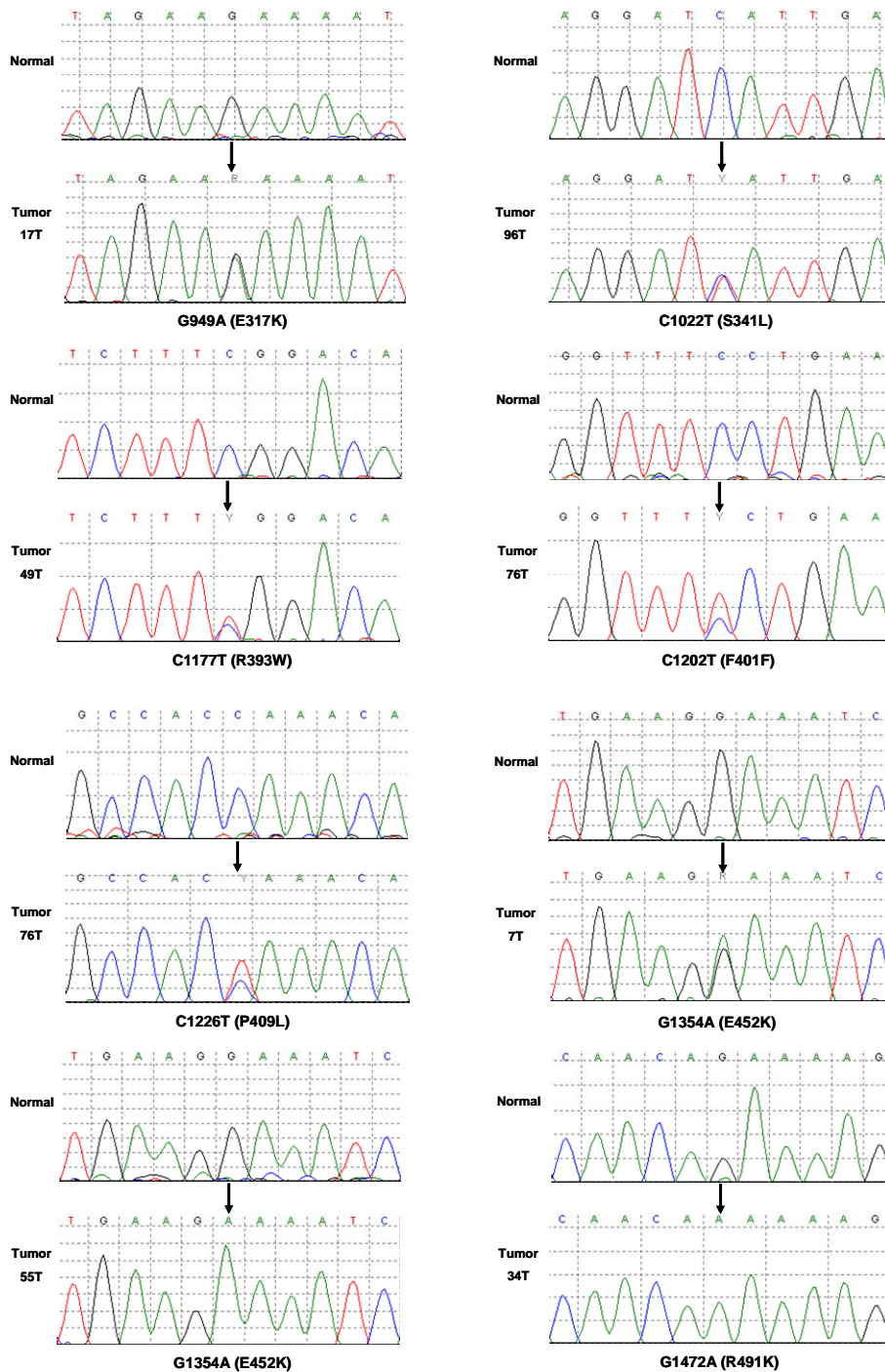
Supplementary Material:

Analysis of the Tyrosine Kinome in Melanoma Reveals Recurrent Mutations in *ERBB4*

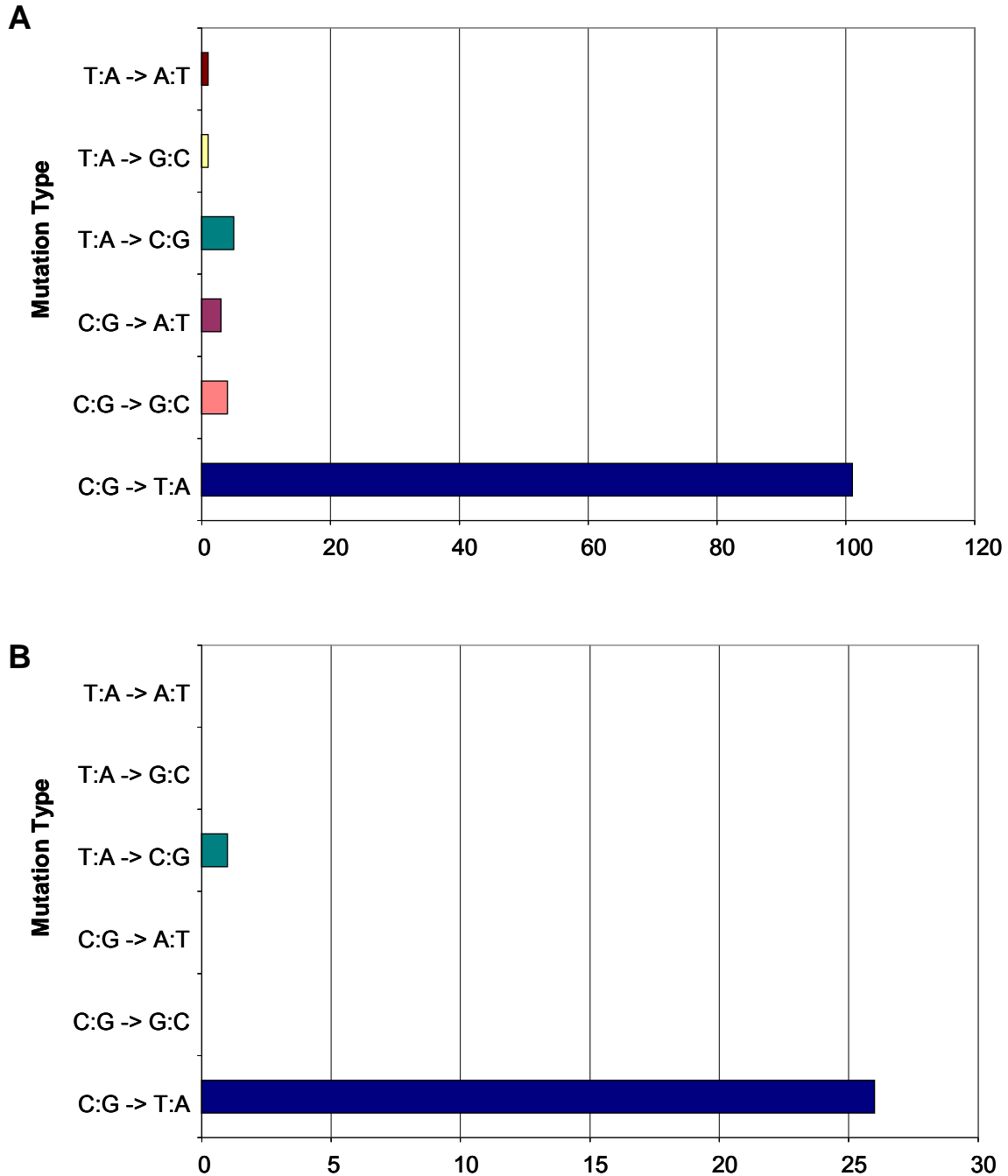
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Supplementary Figures

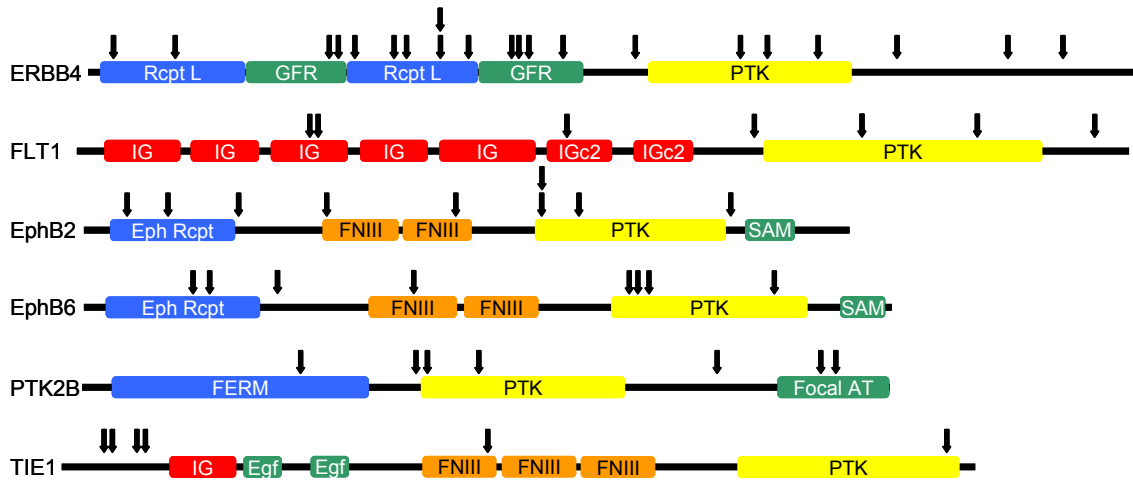


Supplementary Figure 1. Detection of mutations in ERBB4. Representative examples of mutations in ERBB4. In each case, the top sequence chromatogram was obtained from normal tissue and the lower sequence chromatogram from the indicated tumors. Arrows indicate the location of missense mutations. The nucleotide and amino acid alterations are indicated below the chromatograms.



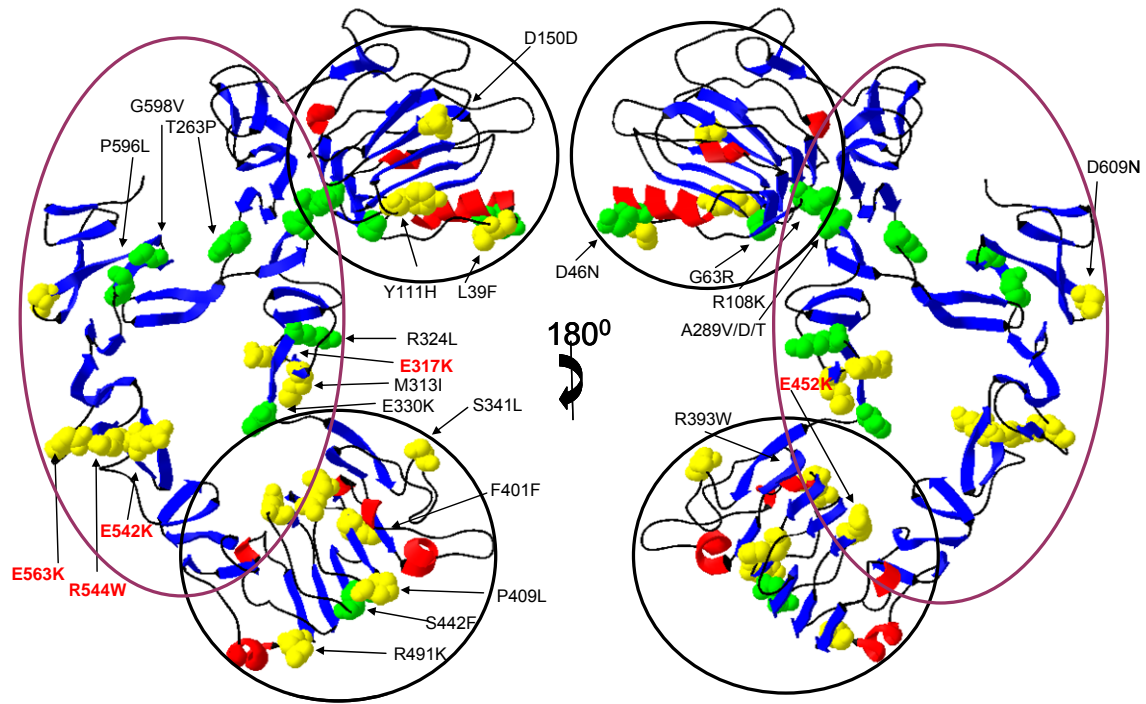
Supplementary Figure 2. Mutation spectra of single base pair substitutions.

A. Kinome mutation spectrum. The number of each of the six classes of base substitutions resulting in nonsynonymous changes in the kinome screen is shown. B. Mutation spectra of single base pair substitutions in ERBB4. The number of each of the six classes of base substitutions resulting in nonsynonymous changes in ERBB4 is shown.

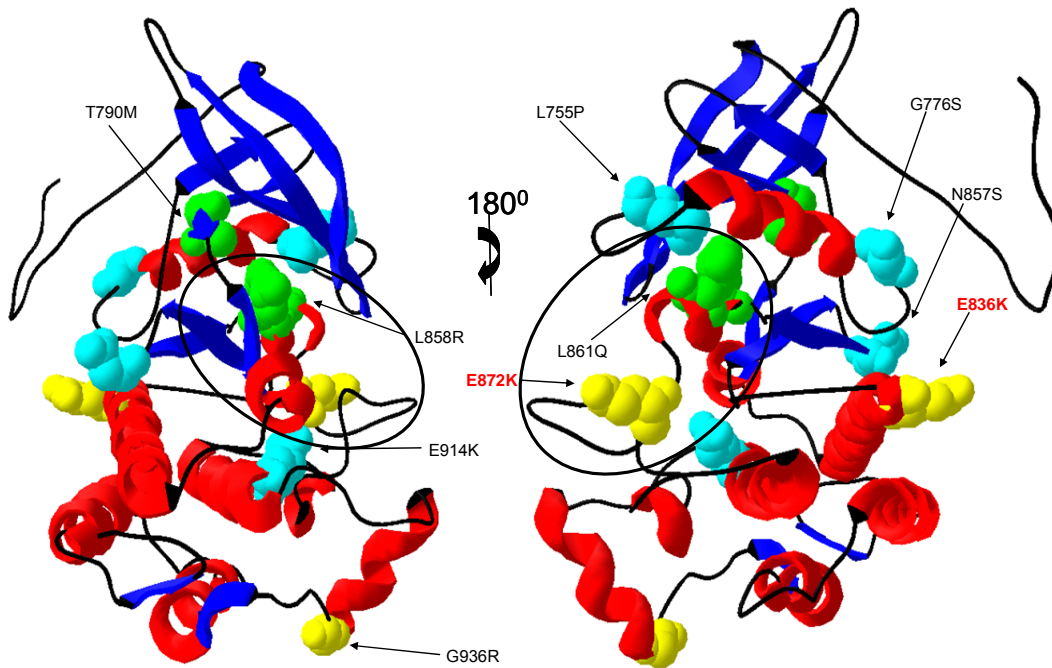


Supplementary Figure 3. Distribution of mutations in ERBB4, FLT1, EphB2, EphB6, PTK2B, and TIE1. Black arrows indicate positions of nonsynonymous mutations and boxes represent functional domains (Rcpt L, Receptor L; GFR, Growth Factor Receptor; PTK, Protein Tyrosine Kinase; IG, Immunoglobulin; IGc2, Immunoglobulin C-2 Type; Eph Rcpt, Ephrin Receptor; FNIII, Fibronectin Type III; SAM, Sterile Alpha Motif; FERM, Protein 4.1, Ezrin, Radixin, Moesin (FERM) Domain; Focal AT, Focal Adhesion Targeting Region)

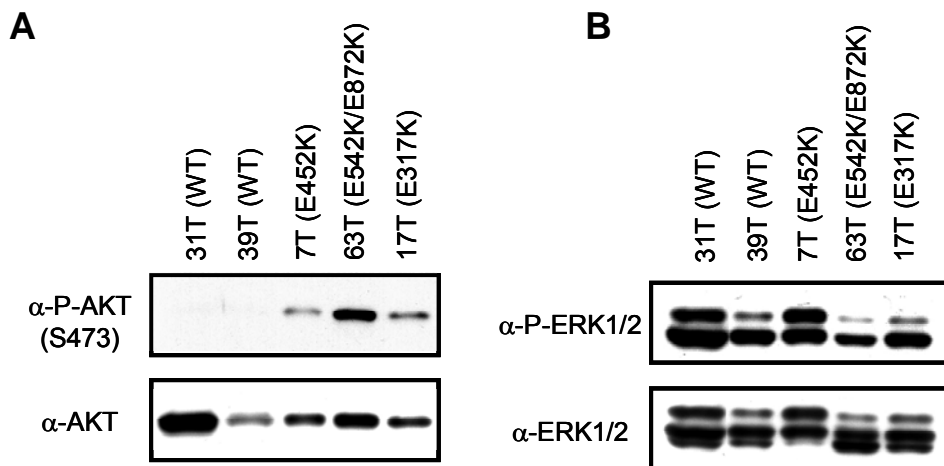
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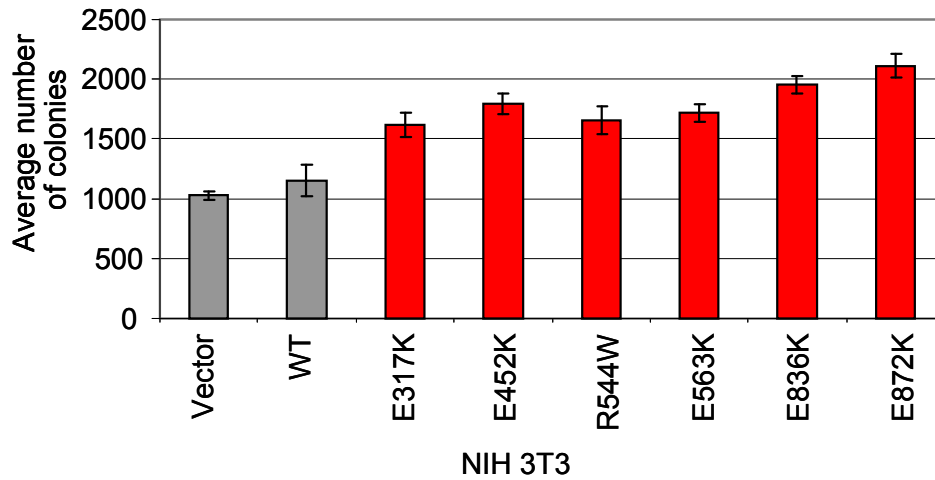
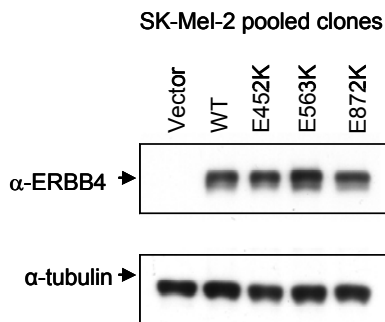
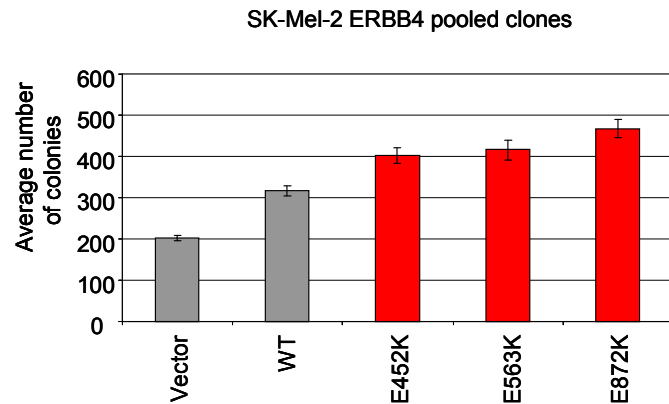
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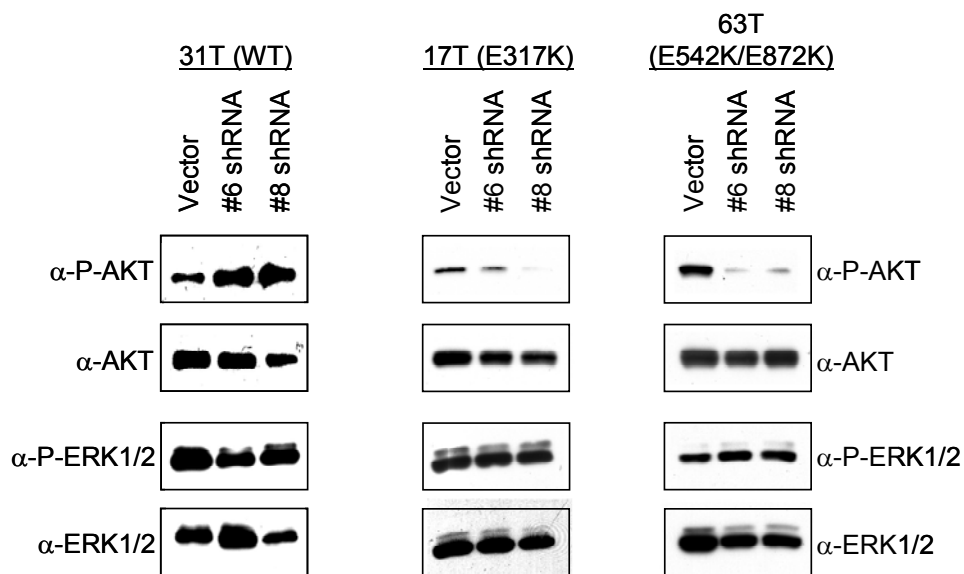
Supplementary Figure 4. Ribbon diagrams of ERBB4. A. Distribution of ERBB4 mutations in a ribbon diagram of its extracellular domain (PDB ID 2ahx). The two views are related to one another by a 180° rotation about a vertical axis. α -helices are shown in red, β -sheets are shown in blue, and loops are shown in black. All mutations are represented as space-filling molecules. ERBB4 mutations are shown in yellow, and the equivalent positions of known activating EGFR mutations are shown in green. The receptor L domain is denoted by black circles, the growth factor receptor domain is denoted by purple circles and cloned ERBB4 mutants are highlighted in red. B. Distribution of ERBB4 mutations in a ribbon of its kinase domain (PDB ID 2r4b). The two views are related to one another by a 180° rotation about a vertical axis. α -helices are shown in red, β -sheets are shown in blue, and loops are shown in black. All mutations are represented as space-filling molecules. ERBB4 mutations are shown in yellow, equivalent positions of known ERBB2 mutations are shown in cyan, and the equivalent positions of known activating EGFR mutations are shown in green. The activation loop is denoted by black circles. In A and B, Images were created by SWISS-MODEL. Determination of the location of previously reported EGFR and ERBB2 mutations is described in the experimental procedures and cloned ERBB4 mutants are highlighted in red.



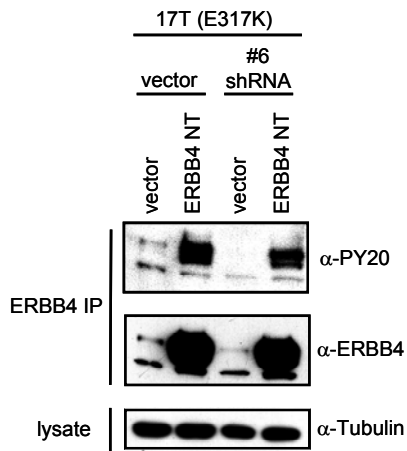
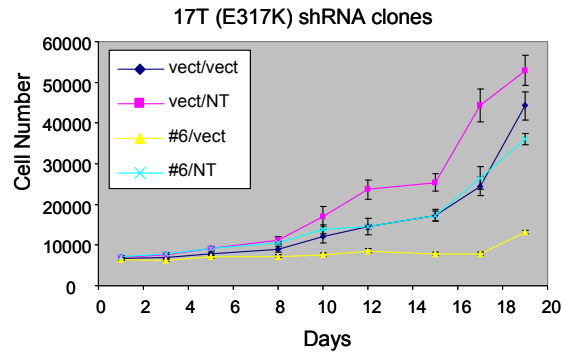
Supplementary Figure 5. Effects of ERBB4 mutation on AKT and ERK phosphorylation. Melanoma cell lines, containing either WT or mutant ERBB4, were harvested and analyzed by immunoblot. Shown are immunoblots of lysates probed with the indicated antibodies (α -P-ERK1/2 (recognizes phosphorylation of T202 and Y204 on ERK1 and T185 and Y187 on ERK2).

A**B****C**

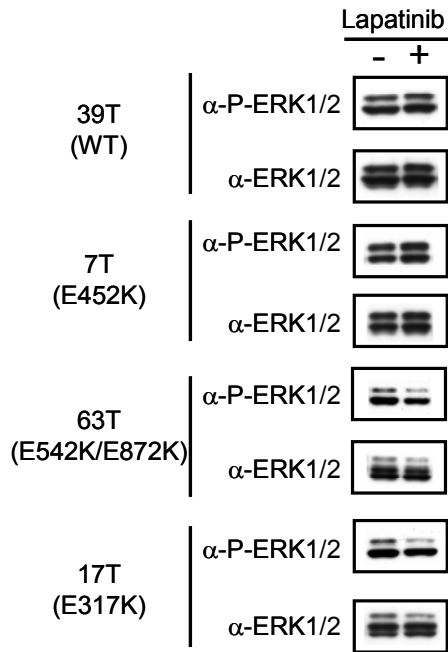
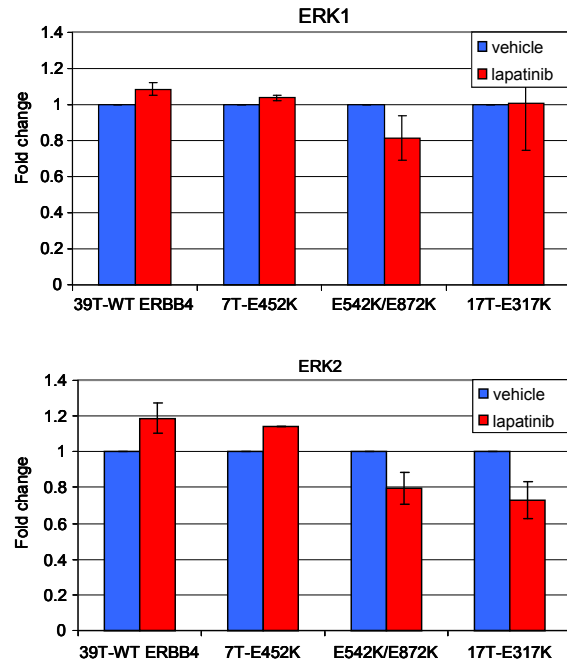
Supplementary Figure 6. Effect of ERBB4 mutations on cell growth in NIH 3T3 and SK-Mel-2 cells. A. Growth of NIH 3T3 cells expressing vector, WT ERBB4 or various ERBB4 missense mutants in soft agar. The graph indicates the number of colonies after 14 days. B. Detection of ERBB4 protein expression in stable transfectants of SK-Mel-2 melanoma cells by immunoblot analysis, lysates from the different clones stably transfected with an empty vector, human WT ERBB4 or the indicated ERBB4 mutants were immunoprecipitated and immunoblotted with ERBB4 antibody. C. Anchorage-independent proliferation of SK-Mel-2 cell clones expressing the indicated constructs was assessed by measuring colony formation in soft agar. Graph indicates number of colonies observed after 14 days of growth.



Supplementary Figure 7. Knockdown of ERBB4 protein causes reduced activation of the AKT pathway but not of the ERK pathway. Melanoma cell lines, containing either WT or mutant ERBB4, were harvested and analyzed by western blot. Shown are immunoblots of lysates probed with the indicated antibodies (α -P-ERK1/2 (recognizes phosphorylation of T202 and Y204 on ERK1 and T185 and Y187 on ERK2)).

A**B**

Supplementary Figure 8. Rescue of oncogene dependence by exogenous non-targetable ERBB4. A. Melanoma cells harboring mutant ERBB4 and stably expressing control or ERBB4 shRNA #6 transduced with either vector or non targetable (NT) ERBB4 were analyzed by immunoprecipitation of lysates with α -ERBB4. Samples were analyzed by immunoblotting with the indicated antibodies. As a loading control lysates were immunoblotted with α -Tubulin. B. Melanoma cells expressing vector or the ERBB4 shRNA #6 transduced with a vector or NT ERBB4 were evaluated for cell proliferation by measuring the average cell number at each time point by determining DNA content using SYBR Green I.

A**B**

Supplementary Figure 9. Effect of lapatinib on ERK1/2 signaling pathways.

A. Melanoma lines expressing mutant ERBB4 exhibit increased lapatinib sensitivity with respect to ERK1 and ERK2 phosphorylation. Cells were treated for 72 hrs with 5 μ M lapatinib or vehicle as control. The activity of ERK1 and ERK2 was determined by immunoblotting with phospho-specific antibodies (α -P-ERK1/2 - recognizes phosphorylation of T202 and Y204 on ERK1 and T185 and Y187 on ERK2). Total ERK protein was also determined by immunoblotting. Shown are representative blots. B. Quantitative assessment of data from one melanoma cell line harboring WT ERBB4 and three melanoma cell lines harboring mutant ERBB4. The ratio of band intensities of P-ERK1/ERK1 or P-ERK2/ERK2 was analyzed for each melanoma cell line.

Table S1. Tyrosine Kinase genes analyzed

CCDS accession and amplimer number	Ref Seq accession and amplimer number	Gene Name	Gene Description
CCDS35165.1	NM_007313.2	ABL1/ABL	v-abl Abelson murine leukemia viral oncogene homolog 1
CCDS30947.1	NM_007314.2	ABL2/ARG	v-abl Abelson murine leukemia viral oncogene homolog 2 (arg, Abelson-related gene)
CCDS33928.1	NM_005781.4	ACK1/TNK2	tyrosine kinase, non-receptor, 2
CCDS33172.1	NM_004304.3	ALK	anaplastic lymphoma kinase (Ki-1)
CCDS12575.1	NM_021913.3	AXL	AXL receptor tyrosine kinase
CCDS5982.1	NM_001715.2	BLK	B lymphoid tyrosine kinase
CCDS14168.1	NM_203281.2	BMX	BMX non-receptor tyrosine kinase
CCDS13524.1	NM_005975.2	BRK/PTK6	PTK6 protein tyrosine kinase 6
CCDS14482.1	NM_000061.1	BTK	Bruton agammaglobulinemia tyrosine kinase
CCDS4302.1	NM_005211.2	CSF1R	colony stimulating factor 1 receptor, formerly McDonough feline sarcoma viral (v-fms) oncogene homolog
CCDS10269.1	NM_004383.1	CSK	c-src tyrosine kinase
CCDS4690.1	NM_001954.4	DDR1	discoidin domain receptor family, member 1
CCDS1241.1	NM_006182.2	DDR2	discoidin domain receptor family, member 2
CCDS5514.1	NM_005228.3	EGFR	epidermal growth factor receptor (erythroblastic leukemia viral (v-erb-b) oncogene homolog, avian)
CCDS5984.1	NM_005232.3	EPHA1	ephrin receptor EphA1
CCDS169.1	NM_004431.2	EPHA2	ephrin receptor EphA2
CCDS2922.1	NM_005233.5	EPHA3	ephrin receptor EphA3 isoform a precursor
CCDS2447.1	NM_004438.3	EPHA4	ephrin receptor EphA4
CCDS3514.1	NM_182472.1	EPHA5	ephrin receptor EphA5 isoform b
N/A	NM_001080448.2	EPHA6	EPH receptor A6 isoform a
CCDS5031.1	NM_004440.2	EPHA7	ephrin receptor EphA7
CCDS30626.1	NM_001006943.1	EPHA8	EPH receptor A8 isoform 2 precursor
CCDS425.1	NM_173641.2	EPHA10	EPH receptor A10 isoform 2
N/A	NM_004441.3	EPHB1	ephrin receptor EphB1 precursor
CCDS230.1	NM_004442.6	EPHB2	ephrin receptor EphB2 isoform 2 precursor
CCDS3268.1	NM_004443.3	EPHB3	ephrin receptor EphB3 precursor
CCDS5706.1	NM_004444.4	EPHB4	ephrin receptor EphB4 precursor
CCDS5873.1	NM_004445.2	EPHB6	ephrin receptor EphB6 precursor
CCDS32642.1	NM_004448.2	ERBB2	v-erb-b2 erythroblastic leukemia viral oncogene homolog 2, neuro/glioblastoma derived oncogene homolog (avian)
CCDS31833.1	NM_001982.2	ERBB3	v-erb-b2 erythroblastic leukemia viral oncogene homolog 3 (avian)
CCDS2394.1	NM_005235.2	ERBB4	v-erb-a erythroblastic leukemia viral oncogene homolog 4 (avian)
CCDS6381.1	NM_153831.2	FAK/PTK2	PTK2 protein tyrosine kinase 2
CCDS4098.1	NM_005246.2	FER	fer (fps/fes related) tyrosine kinase (phosphoprotein NCP94)
CCDS10365.1	NM_002005.2	FES	V-FES feline sarcoma viral/V-FPS fujinami avian
CCDS6107.1	NM_023110.2	FGFR1	fibroblast growth factor receptor 1 (fms-related tyrosine kinase 2, Pfeiffer syndrome)
CCDS1298.1	NM_000141.3	FGFR2	fibroblast growth factor receptor 2 (bacteria-expressed kinase, keratinocyte growth factor receptor)
CCDS3353.1	NM_000142.2	FGFR3	fibroblast growth factor receptor 3 (achondroplasia, thanatophoric dwarfism)
CCDS4410.1	NM_002011.3	FGFR4	fibroblast growth factor receptor 4 isoform 1
CCDS305.1	NM_005248.2	FGR	Gardner-Rasheed feline sarcoma viral (v-fgr) oncogene homolog
CCDS9330.1	NM_002019.3	FLT1/VEGFR1	fms-related tyrosine kinase 1 (vascular endothelial growth factor/vascular permeability factor receptor)
CCDS31953.1	NM_004119.2	FLT3	fms-related tyrosine kinase 3
CCDS4457.1	NM_182925.3	FLT4/VEGFR3	fms-related tyrosine kinase 4
CCDS5103.1	NM_002031.2	FRK	fyn-related kinase
CCDS5094.1	NM_002037.3	FYN	FYN oncogene related to SRC, FGR, YES
CCDS33460.1	NM_002110.2	HCK	hemopoietic cell kinase
CCDS10378.1	NM_000875.3	IGF1R	insulin-like growth factor 1 receptor
CCDS12176.1	NM_000208.2	INSR	insulin receptor
CCDS1160.1	NM_014215.1	INSRR	insulin receptor-related receptor
CCDS4336.1	NM_005546.3	ITK	IL2-inducible T-cell kinase
N/A	NM_002227.2	JAK1	Janus kinase 1
CCDS6457.1	NM_004972.2	JAK2	Janus kinase 2
CCDS12366.1	NM_000215.2	JAK3	Janus kinase 3
CCDS3497.1	NM_002253.1	KDR/VEGFR2	kinase insert domain receptor (a type III receptor tyrosine kinase)
CCDS3496.1	NM_000222.2	KIT	v-kit Hardy-Zuckerman 4 feline sarcoma viral oncogene homolog
CCDS359.1	NM_005356.3	LCK	lymphocyte-specific protein tyrosine kinase
CCDS10078.1	NM_206961.1	LTK	leukocyte tyrosine kinase
CCDS6162.1	NM_002350.2	LYN	v-yes-1 Yamaguchi sarcoma viral related oncogene homolog
CCDS12113.1	NM_002378.3	MATK	megakaryocyte-associated tyrosine kinase
CCDS2094.1	NM_006343.2	MERTK/MER	c-mer proto-oncogene tyrosine kinase
N/A	NM_000245.2	MET	met proto-oncogene (hepatocyte growth factor receptor)
CCDS2807.1	NM_002447.2	MST1R/RON	macrophage stimulating 1 receptor (c-met-related tyrosine kinase)
N/A	NM_005592.1	MUSK	muscle, skeletal, receptor tyrosine kinase
CCDS1161.1	NM_002529.3	NTRK1	neurotrophic tyrosine kinase, receptor, type 1
CCDS35053.1	NM_001007097.1	NTRK2	neurotrophic tyrosine kinase receptor type 2
CCDS32322.1	NM_001012338.1	NTRK3	neurotrophic tyrosine kinase receptor type 3
CCDS3495.1	NM_006206.3	PDGFRA	platelet-derived growth factor receptor alpha
CCDS4303.1	NM_002609.3	PDGFRB	platelet-derived growth factor receptor beta
CCDS4884.1	NM_002821.3	PTK7	PTK7 protein tyrosine kinase 7
CCDS6057.1	NM_004103.3	PYK2/PTK2B	PTK2B protein tyrosine kinase 2 beta
CCDS7200.1	NM_020975.4	RET	ret proto-oncogene
CCDS626.1	NM_005012.2	ROR1	receptor tyrosine kinase-like orphan receptor 1
CCDS6691.1	NM_004560.2	ROR2	receptor tyrosine kinase-like orphan receptor 2
CCDS5116.1	NM_002944.2	ROS1	v-ros UR2 sarcoma virus oncogene homolog 1 (avian)
N/A	NM_001005861.2	RYK	RYK receptor-like tyrosine kinase
CCDS13294.1	NM_005417.3	SRC	v-src sarcoma (Schmidt-Ruppin A-2) viral oncogene homolog (avian)
CCDS13525.1	NM_080823.2	SRMS	src-related kinase lacking C-terminal regulatory tyrosine and N-terminal myristylation sites
CCDS6688.1	NM_003177.3	SYK	spleen tyrosine kinase
CCDS3481.1	NM_003215.2	TEC	tec protein tyrosine kinase
CCDS6519.1	NM_000459.2	TEK	TEK tyrosine kinase, endothelial (venous malformations, multiple cutaneous and mucosal)
CCDS482.1	NM_005424.2	TIE	tyrosine kinase with immunoglobulin-like and EGF-like domains 1
N/A	NM_003985.3	TNK1	tyrosine kinase, non-receptor, 1
CCDS3480.1	NM_003328.2	TXK	TXK tyrosine kinase
CCDS12236.1	NM_003331.3	TYK2	tyrosine kinase 2
CCDS10080.1	NM_006293.2	TYRO3	TYRO3 protein tyrosine kinase
CCDS11824.1	NM_005433.3	YES1	v-yes-1 Yamaguchi sarcoma viral oncogene homolog 1
CCDS33254.1	NM_001079.3	ZAP70	zeta-chain (TCR) associated protein kinase 70kDa

TNK1_Exon-6	N/A	NM_003985.3	AAAGAGGACTTTTGTGAAGATGG	TGTCAACGACTGCAACTG	TGTAACAGCAGGCCAGT	CAGGAACAGCTATGACC
TXK_Exon-10	CCDS3480.1	NM_003328.2	GACATTGGAGCTTGGGAAGTGT	TCCCAGGTCTAGTTTTGGTGG	TGTAACAGCAGGCCAGT	CAGGAACAGCTATGACC
TXK_Exon-11	CCDS3480.1	NM_003328.2	TAATTTGGACCATATCTTACCA	AGTCACTCTTTCCTCCCTCTTG	TGTAACAGCAGGCCAGT	CAGGAACAGCTATGACC
TXK_Exon-13	CCDS3480.1	NM_003328.2	ACGATGGGGATGATGGTAGAA	GCAGGCTCTTTGTAGAGTTAKGA	TGTAACAGCAGGCCAGT	CAGGAACAGCTATGACC
TXK_Exon-14	CCDS3480.1	NM_003328.2	TGCAGAACCACATCTCAACCA	AAAAAGAAAGAAATCTACTGG	TGTAACAGCAGGCCAGT	CAGGAACAGCTATGACC
TXK_Exon-15	CCDS3480.1	NM_003328.2	GTGCAATCCCTCTCACACATA	TCACCCCTCACTCACTG	TGTAACAGCAGGCCAGT	CAGGAACAGCTATGACC
TYK2_Exon-10;TYK2_Exon-11	CCDS12236.1	NM_003331.3	GGTCTGGTCACTCATCTCTG	CTACCCCTGATCCTCACAG	TGTAACAGCAGGCCAGT	CAGGAACAGCTATGACC
TYK2_Exon-10;TYK2_Exon-11;TYK2_Exon-12	CCDS12236.1	NM_003331.3	GGGATCAGAGTACAGGTGAG	GGGTTGACCAGAAGGAGATCA	TGTAACAGCAGGCCAGT	CAGGAACAGCTATGACC
TYK2_Exon-10;TYK2_Exon-11;TYK2_Exon-12	CCDS12236.1	NM_003331.3	AGGGTGGCTGTCTGGTAGAA	GGGTTGACCAGAAGGAGATCA	TGTAACAGCAGGCCAGT	CAGGAACAGCTATGACC
TYK2_Exon-14;TYK2_Exon-15	CCDS12236.1	NM_003331.3	TCAGTCTGTACTTTCGCTTCTG	ATAGTCAAGTTGCTCTTTTCA	TGTAACAGCAGGCCAGT	CAGGAACAGCTATGACC
TYK2_Exon-14;TYK2_Exon-15	CCDS12236.1	NM_003331.3	CTGTGTGTAGCCAGAGAGA	ATAGTCAAGTTGCTCTTTTCA	TGTAACAGCAGGCCAGT	CAGGAACAGCTATGACC
TYK2_Exon-16	CCDS12236.1	NM_003331.3	CTTATGAATGCCACTGGAAGAA	GCCCAGAGAGGTTTTATCTA	TGTAACAGCAGGCCAGT	CAGGAACAGCTATGACC
TYK2_Exon-17;TYK2_Exon-18	CCDS12236.1	NM_003331.3	GTACTGGGGAAGGATCG	CTGCTGGGGAAGGATCG	TGTAACAGCAGGCCAGT	CAGGAACAGCTATGACC
TYK2_Exon-17;TYK2_Exon-18	CCDS12236.1	NM_003331.3	CTGCCCTTGAGGGCTTTCAC	CTCCCAACAGGTTGTCAGT	TGTAACAGCAGGCCAGT	CAGGAACAGCTATGACC
TYK2_Exon-18;TYK2_Exon-19	CCDS12236.1	NM_003331.3	GTTGGGAGGTGAGTCAAG	CAGGAGAGATGATGAATACA	TGTAACAGCAGGCCAGT	CAGGAACAGCTATGACC
TYK2_Exon-19;TYK2_Exon-20	CCDS12236.1	NM_003331.3	CTTCACTGGGATCTAGCCAT	GCTCTGCCAGCAGAT	TGTAACAGCAGGCCAGT	CAGGAACAGCTATGACC
TYK2_Exon-20;TYK2_Exon-21	CCDS12236.1	NM_003331.3	ACCCCGAAGGACGAGAT	CGCCCACTGAACTCAC	TGTAACAGCAGGCCAGT	CAGGAACAGCTATGACC
TYK2_Exon-22;TYK2_Exon-23	CCDS12236.1	NM_003331.3	CCTCTCCAGCAGCAGTATG	CAAACCTCTGACTCACGCTTA	TGTAACAGCAGGCCAGT	CAGGAACAGCTATGACC
TYRO3_Exon-12;TYRO3_Exon-13	CCDS10080.1	NM_006293.2	ACAAATCGCTGTGTGGAGTT	AGGCTACAGCTATCCCACT	TGTAACAGCAGGCCAGT	CAGGAACAGCTATGACC
TYRO3_Exon-13	CCDS10080.1	NM_006293.2	GGATGTTGTGGGGAATGTT	AGGCTCTGTCTTCCAGATGCT	TGTAACAGCAGGCCAGT	CAGGAACAGCTATGACC
TYRO3_Exon-14	CCDS10080.1	NM_006293.2	TTCAACACATAAACCAACA	TCTTATGTGGAGACTTTG	TGTAACAGCAGGCCAGT	CAGGAACAGCTATGACC
TYRO3_Exon-15;TYRO3_Exon-16	CCDS10080.1	NM_006293.2	GGATGCCCAAGCAGTGT	CTGGAGGGTGTGTTCTAGTG	TGTAACAGCAGGCCAGT	CAGGAACAGCTATGACC
TYRO3_Exon-16;TYRO3_Exon-17	CCDS10080.1	NM_006293.2	GTCTCCACAGCTCCTCTA	CTGCTCACAGCTCCTCTG	TGTAACAGCAGGCCAGT	CAGGAACAGCTATGACC
TYRO3_Exon-17;TYRO3_Exon-18	CCDS10080.1	NM_006293.2	GGTAGCTTGGGAGCAAGATG	CCATCAGCAAACTCCAGAT	TGTAACAGCAGGCCAGT	CAGGAACAGCTATGACC
TYRO3_Exon-18	CCDS10080.1	NM_006293.2	CAGGACACTAGTGACAGTCT	CAGTTCCTCTTCCAGGAT	TGTAACAGCAGGCCAGT	CAGGAACAGCTATGACC
TYRO3_Exon-19	CCDS10080.1	NM_006293.2	AACCTCGGGCTCAATATACC	GCACAGCTTGGGAGACTAC	TGTAACAGCAGGCCAGT	CAGGAACAGCTATGACC
YES1_Exon-10	CCDS11824.1	NM_005433.3	TATCTCATGTCTCCACCTGTC	AGTATTAGGTGCCAGCTGTACTT	TGTAACAGCAGGCCAGT	CAGGAACAGCTATGACC
YES1_Exon-11	CCDS11824.1	NM_005433.3	TTGTTGGACCCCTGAATATCG	TAATCAATGGGTTGGCTTTT	TGTAACAGCAGGCCAGT	CAGGAACAGCTATGACC
YES1_Exon-6	CCDS11824.1	NM_005433.3	TTTTAAAGATTTTAGACAACTTCA	CGATCTGCTCAGCTCTCTTA	TGTAACAGCAGGCCAGT	CAGGAACAGCTATGACC
YES1_Exon-6;YES1_Exon-7	CCDS11824.1	NM_005433.3	ACAAAGTGTCAAGTTTCTCTATCC	AGAAGTCTGATGTTTATGCT	TGTAACAGCAGGCCAGT	CAGGAACAGCTATGACC
YES1_Exon-8	CCDS11824.1	NM_005433.3	TCCCACTTCACTACACATAC	CCGTAATCAGACTCTAGCCTTTT	TGTAACAGCAGGCCAGT	CAGGAACAGCTATGACC
YES1_Exon-9	CCDS11824.1	NM_005433.3	AGCTAAACAAATCTTATCTGGAAA	AAATTAATGGGATTAAGGTGG	TGTAACAGCAGGCCAGT	CAGGAACAGCTATGACC
ZAP70_Exon-12	CCDS3254.1	NM_001079.3	CTCTCACCCAGGCTGT	CTACTCTCACCCGGAAG	TGTAACAGCAGGCCAGT	CAGGAACAGCTATGACC
ZAP70_Exon-7	CCDS3254.1	NM_001079.3	GTGCAGGAAACAGCATGG	ATCCCAAAAGAAATCAAAATG	TGTAACAGCAGGCCAGT	CAGGAACAGCTATGACC
ZAP70_Exon-8	CCDS3254.1	NM_001079.3	CGTGGTGGCAGCTACAGTT	AATGGTGACTGAGGGTTAGC	TGTAACAGCAGGCCAGT	CAGGAACAGCTATGACC
ZAP70_Exon-9;ZAP70_Exon-10	CCDS3254.1	NM_001079.3	GTGTGGGTGATCCTCAAGC	GTGACCCATAGCTCCAGAC	TGTAACAGCAGGCCAGT	CAGGAACAGCTATGACC
ZAP70_Exon-9;ZAP70_Exon-10;ZAP70_Exon-11	CCDS3254.1	NM_001079.3	CTACGCCAAGATCAGCAGTTT	ACTGGAGGACAGCAAC	TGTAACAGCAGGCCAGT	CAGGAACAGCTATGACC

PTK7-20	CCDS4884.1	NM_02821.3	GTA AACGACGGCCAGTTC AAGTCC ACAAAGTTTG	AGACCTCAGCAATGCCTGTTG	GTA AACGACGGCCAGT
ROR2-1	CCDS6691.1	NM_004560.2	GTA AACGACGGCCAGTGGACGCATCGTAGAAAGGG	GCTTATTGTA ACCAGCCCGAG	GTA AACGACGGCCAGT
ROR2-2	CCDS6691.1	NM_004560.2	GTA AACGACGGCCAGTCCC GATTTGCTATTCTTGTG	AAACACAGGAATCAAGGTGGC	GTA AACGACGGCCAGT
ROR2-3	CCDS6691.1	NM_004560.2	GTA AACGACGGCCAGTTCCTTGG AAGAGTTCTAAGC	TGCTGACTGGTGTGTTCAG	GTA AACGACGGCCAGT
ROR2-4	CCDS6691.1	NM_004560.2	GTA AACGACGGCCAGTCTTCCCTGTGTGGTGTTC	CAAAATCGGCAAGACATGAG	GTA AACGACGGCCAGT
ROR2-5	CCDS6691.1	NM_004560.2	AATTGCTGGATCGCAAGATG	GTA AACGACGGCCAGTCCC ACTGACCATGCCATTAAC	GTA AACGACGGCCAGT
ROR2-6	CCDS6691.1	NM_004560.2	GTA AACGACGGCCAGTGTGGGCACTGGGATGC	CCTGGGCTTCA CCGACAC	GTA AACGACGGCCAGT
ROR2-7	CCDS6691.1	NM_004560.2	GTA AACGACGGCCAGTATTTGGGCTTGGGACTTC	GTCAGGACAGAAAGCCCTC	GTA AACGACGGCCAGT
ROR2-8	CCDS6691.1	NM_004560.2	GTA AACGACGGCCAGTAGGTGGAGAGTGGTGTGTAG	GCAGTGAATCCCAACAGC	GTA AACGACGGCCAGT
ROR2-9	CCDS6691.1	NM_004560.2	GTA AACGACGGCCAGTAGGAAGTCAAGTGTGTCCAGC	AGCTGAAGATCATGCTCAGGG	GTA AACGACGGCCAGT
ROR2-9	CCDS6691.1	NM_004560.2	GACCGGTTGGAAAGTCTAC	GTA AACGACGGCCAGTCA CACCCTAGGACAGATGTC	GTA AACGACGGCCAGT
ROR2-9	CCDS6691.1	NM_004560.2	AGTACCTATCCAGCCACACG	GTA AACGACGGCCAGTGTGCTCACATTTGCTCACTGG	GTA AACGACGGCCAGT
ROR2-9	CCDS6691.1	NM_004560.2	GTA AACGACGGCCAGTCAAGTTCCTCATCGACTCAG	ATCTGCATTTGGGATCTGCAC	GTA AACGACGGCCAGT
ROR2-9	CCDS6691.1	NM_004560.2	CCAGTGAGCAATGTGAGCAAC	GTA AACGACGGCCAGTCTCAAAGGTGACTGAGGTCCC	GTA AACGACGGCCAGT
TIE1-1	CCDS482.1	NM_005424.2	GTA AACGACGGCCAGTAGCCACCTATTCTCTTCC	AATTGATCCTCCCATCTCAGC	GTA AACGACGGCCAGT
TIE1-2	CCDS482.1	NM_005424.2	GTA AACGACGGCCAGTAGTAAGACCAGAGGGCTGGG	GACCTTGTCTGGAAGCAGGTG	GTA AACGACGGCCAGT
TIE1-3	CCDS482.1	NM_005424.2	ACAGCCCTGGAGGTGAGTTAG	GTA AACGACGGCCAGTCTACAGGAGAGGCCCTGG	GTA AACGACGGCCAGT
TIE1-4	CCDS482.1	NM_005424.2	AGCTGAGCAGAGGTGGACAG	GTA AACGACGGCCAGTCTGGAGGAAGGTGTCTTACG	GTA AACGACGGCCAGT
TIE1-5	CCDS482.1	NM_005424.2	GTA AACGACGGCCAGTGCCCTATGGGTACTCTCTGTG	CACCCACATGGTCTAGGGTC	GTA AACGACGGCCAGT
TIE1-6	CCDS482.1	NM_005424.2	GACCTTAGAACCTGTGGGTG	GTA AACGACGGCCAGTAA GTCAGAGGCAGAGTGGAGG	GTA AACGACGGCCAGT
TIE1-7	CCDS482.1	NM_005424.2	GTA AACGACGGCCAGTCCAGTCTTCTATCCTCAGCC	AGAGCTCCCTGTCCATCAGAC	GTA AACGACGGCCAGT
TIE1-8	CCDS482.1	NM_005424.2	GTA AACGACGGCCAGTCTCCACTGAGAAACAGAAACAG	CTGACCTCATGTGAGCAATGG	GTA AACGACGGCCAGT
TIE1-9	CCDS482.1	NM_005424.2	GTA AACGACGGCCAGTATCAGTCCAGAGGCACACCTG	AGTATCTCCAGCTGAGCAGC	GTA AACGACGGCCAGT
TIE1-10	CCDS482.1	NM_005424.2	CCAGCTTGA AACAAAGATGC	GTA AACGACGGCCAGTCTAGAGCTGGCATGTCTCC	GTA AACGACGGCCAGT
TIE1-11	CCDS482.1	NM_005424.2	GTA AACGACGGCCAGTGTAGTAGSAGAGAGCTGGG	CAAGSAAACAACTCCAGTG	GTA AACGACGGCCAGT
TIE1-12	CCDS482.1	NM_005424.2	GTA AACGACGGCCAGTTGTTCTCTTGTTCACCTGGG	CCTCCGCTGTCTCTAGTGC	GTA AACGACGGCCAGT
TIE1-13	CCDS482.1	NM_005424.2	GTA AACGACGGCCAGTCTAGAGCAGATGTGTCCAGCC	TGTGGAGGAGAGCCTGC	GTA AACGACGGCCAGT
TIE1-14	CCDS482.1	NM_005424.2	GTA AACGACGGCCAGTGGGATCTTCCACTCTCCCTC	CACACAAGGTTGACAAGTATTG	GTA AACGACGGCCAGT
TIE1-15	CCDS482.1	NM_005424.2	GTA AACGACGGCCAGTTGACACTGAAACCTCCTCTGTG	GTCAAATGCTGGAGGAAGCC	GTA AACGACGGCCAGT
TIE1-16	CCDS482.1	NM_005424.2	GTA AACGACGGCCAGTCTTGTCCCAAAATCCCAAG	GATGAAGGAGATCAAGGGTG	GTA AACGACGGCCAGT
TIE1-17	CCDS482.1	NM_005424.2	GTA AACGACGGCCAGTATCCCTGTCTGTACCATCGG	GGCTGACTTCTCCACTGACAC	GTA AACGACGGCCAGT
TIE1-18	CCDS482.1	NM_005424.2	CGAAGACTGACTCTTACTGGC	GTA AACGACGGCCAGTCCCTAGGAGAAATGGGTTTG	GTA AACGACGGCCAGT
TIE1-19	CCDS482.1	NM_005424.2	AGTCTCATTCAACCTCACCC	GTA AACGACGGCCAGTCCACTGTGTCCACCTGGAAC	GTA AACGACGGCCAGT
TIE1-20	CCDS482.1	NM_005424.2	GTA AACGACGGCCAGTTGGCCAAAGCCAGATACTTAC	ACACTTCACTGTCACAGTTTCG	GTA AACGACGGCCAGT
TIE1-21	CCDS482.1	NM_005424.2	GTA AACGACGGCCAGTGGGAGATAGTAGGCTTGGTG	CTAGCTGTAGCCCAATCTGGG	GTA AACGACGGCCAGT
TIE1-22	CCDS482.1	NM_005424.2	GTA AACGACGGCCAGTTCAGCTTGTATCCTGTGACC	GCCTGGTGTCAAACCAAGTGAAC	GTA AACGACGGCCAGT
TIE1-23	CCDS482.1	NM_005424.2	GTA AACGACGGCCAGTATGGAAGTCCAGGAGCTTGAAG	CACCTACAAGCATGGGAACC	GTA AACGACGGCCAGT

#The primer pair did not meet our quality criteria that 250% of bases in the target region have a Phred quality score of at least 20 in three quarters of the tumor samples analyzed.

Table S4. Characteristics of melanoma patients with PTK mutations

Sample	Patient Age (years)*	Patient Gender	Tumor Source	Matched normal source
1T	29	F	Lung	Blood
2T	30	M	Pectoral muscle	Blood
3T	18	M	Forehead, subcutaneous	Blood
4T	33	F	Supraclavicular, soft tissue	Blood
4T	34	F	Lung	Blood
5T	47	M	External iliac soft tissue	Blood
6T	42	M	Neck, soft tissue	Blood
7T	53	M	Stomach	Blood
8T	61	M	Inguinal soft tissue	Blood
9T	62	M	Back, subcutaneous	Blood
10T	55	M	Axillary soft tissue	Blood
12T	53	M	Upper arm, subcutaneous	Blood
13T	49	M	Chest wall, subcutaneous	Blood
15T	39	M	Thigh, subcutaneous	Blood
16T	62	M	Lung	Blood
17T	33	M	Shoulder, subcutaneous	Blood
18T	55	M	Clavicle, soft tissue	Blood
19T	49	M	Scapula, subcutaneous	Blood
20T	58	F	Axillary soft tissue	Blood
21T	59	M	Omentum	Blood
22T	51	M	Chest wall, subcutaneous	Blood
23T	44	M	Lung	Blood
24T	49	M	Axillary soft tissue	Blood
26T	48	F	Lung	Blood
28T	28	F	Axillary soft tissue	Blood
29T	51	M	Inguinal soft tissue	Blood
30T	53	F	Lung	Blood
31T	49	F	Thigh, subcutaneous	Blood
32T	58	M	Omentum	Blood
33T	33	M	Chest wall subcutaneous, & pleura **	Blood
34T	31	M	Shoulder, subcutaneous	Blood
35T	23	F	Thigh, subcutaneous	Blood
36T	25	M	Thigh, subcutaneous	Blood
37T	38	F	Omentum	Blood
38T	27	M	Skull	Blood
39T	56	M	Mesentery	Blood
41T	45	M	Neck, soft tissue	Blood
43T	19	F	Popliteal soft tissue	Blood
44T	56	M	Lung	Blood
45T	48	M	Mediastinum	Blood
49T	43	M	Thigh, subcutaneous	Blood
50T	49	F	Inguinal soft tissue	Blood
51T	50	F	Adnexa	Blood
52T	39	F	Lung	Blood
54T	27	M	Subcutaneous	Blood
55T	60	M	Lung	Blood
56T	52	M	Lung	Blood
58T	46	F	Hip, subcutaneous	Blood
59T	64	F	Abdomen, subcutaneous	Blood
60T	46	M	Flank, subcutaneous	Blood
62T	58	F	Thigh, subcutaneous	Blood
63T	30	M	Jejunum	Blood
64T	32	F	Ovary	Blood
68T	49	M	Lung	Blood
69T	36	M	Axillary soft tissue	Blood
71T	67	M	Lung	Blood
72T	53	M	Liver	Blood
73T	45	F	Breast	Blood
74T	40	F	Lower extremity, subcutaneous	Blood
76T	40	M	Neck, soft tissue	Blood
77T	39	M	Lung	Blood
78T	27	F	Lung	Blood
79T	53	M	Supraclavicular, soft tissue	Blood
80T	36	F	Popliteal soft tissue	Blood
81T	60	F	Upper arm, subcutaneous	Blood
83T	33	F	Back, subcutaneous	Blood
84T	60	F	Thigh, subcutaneous	Blood
85T	44	M	Chest wall, subcutaneous	Blood
86T	42	F	Liver	Blood
87T	27	M	Small bowel & mesentery **	Blood
88T	37	F	Chest wall, subcutaneous	Blood
90T	19	M	Neck, soft tissue	Blood
91T	55	F	Subcostal soft tissue	Blood
92T	37	F	Femur	Blood
93T	42	F	Axillary soft tissue	Blood
94T	44	M	Adrenal gland	Blood
95T	58	F	Inguinal soft tissue	Blood
96T	49	M	Inguinal soft tissue	Blood
99T	57	M	Liver	Blood

* Patient's age when tumor was surgically removed. ** Cell line generated by cells mixed from 2 tumors resected during the patient's surgery. Abbreviations: F, female; M, male.

Table S5. Mutations Identified in RAF and RAS isoforms in melanoma samples containing ERBB4 mutations

Sample	ERBB4	BRAF	NRAS	ARAF	CRAF	HRAS	KRAS
7T	E452K	wt	Q61R	wt	wt	wt	wt
12T	E563K	wt	Q61Q/R	wt	wt	wt	wt
17T	E317K	wt	Q61Q/K	wt	wt	wt	wt
31T	wt	wt	wt	wt	wt	wt	wt
34T	R491K	V600V/E	wt	wt	T362T/A	wt	wt
39T	wt	wt	wt	wt	wt	wt	wt
49T	R393R/W	V600V/E	wt	wt	wt	wt	wt
55T	E452K	V600V/E	wt	P216S, P254L	wt	wt	wt
56T	R544R/W	V600V/E	wt	wt	wt	wt	wt
63T	E542K, E872K	wt	Q61Q/K	wt	wt	wt	wt
68T	Splice Site / LOH	V600V/E	wt	wt	wt	wt	wt
71T	L39L/F, S1246S/N	V600V/M, V600V/E	wt	wt	wt	wt	wt
86T	E836E/K	V600V/E	wt	wt	wt	wt	wt
93T	wt	wt	wt	A345A/G	wt	wt	wt

Table S6. Primers used for PCR amplification and sequencing of RAS and Raf isoforms

Gene and Exon Name	Forward Primer	Reverse Primer	Sequencing Primer
ARAF-1	TGAGCAGGATCTTGGACTG	GTA AACGACGCGCCAGTCAGCTGATGTCCCATCCTTC	GTA AACGACGCGCCAGT
ARAF-2	GTCTATGGAAAGCGAATGCG	GTA AACGACGCGCCAGTCAGAAGGAATCAATGACTGAGG	GTA AACGACGCGCCAGT
ARAF-3	GTA AACGACGCGCCAGTTCATCCCTCTGAGCCTGTTCC	AGCAGGGAAGTTTGAGACTG	GTA AACGACGCGCCAGT
ARAF-4	GTA AACGACGCGCCAGTACCCAACTCCCACTCATT	CACGGGTCAGCTGTCTGAAG	GTA AACGACGCGCCAGT
ARAF-5	AGTACCAACCGCCCAACAGTG	GTA AACGACGCGCCAGTGAGAAATGAGGTGACTTGCCC	GTA AACGACGCGCCAGT
ARAF-6	CAATATGATGTTTATGGCTGG	GTA AACGACGCGCCAGTCATCAGTGTGAACTCTGGCC	GTA AACGACGCGCCAGT
ARAF-7	GTA AACGACGCGCCAGTCAACGTCCTCATATGTCAGCAC	CTGGTGACTTGGAAATGTGGG	GTA AACGACGCGCCAGT
ARAF-8	CCAGAGTTTCAGCACTGATGG	GTA AACGACGCGCCAGTCCAGATGGTGGCATCTAAG	GTA AACGACGCGCCAGT
ARAF-9	GTA AACGACGCGCCAGTGAAGAGTGGTATGCTCGAGGG	ATGTCCAGGAAGCACTCCAG	GTA AACGACGCGCCAGT
ARAF-10	GTA AACGACGCGCCAGTGAACAGTGCCTCACTGATG	TCTCGGTGATGATGTTCTTG	GTA AACGACGCGCCAGT
ARAF-11	GGATTTGCCATCATCACAG	GTA AACGACGCGCCAGTCCCTGTAATTCAGAAACCC	GTA AACGACGCGCCAGT
ARAF-12	GTA AACGACGCGCCAGTCAAGGTTGTGGATGTTTGGC	GTGGACATGAGGAGTCCAG	GTA AACGACGCGCCAGT
ARAF-13	GTGTGGGTGCTGTGAGTTG	GTA AACGACGCGCCAGTGGCATTTATCAGTGCAAAAGG	GTA AACGACGCGCCAGT
ARAF-14	GTA AACGACGCGCCAGTGCAGAGAATCCCTCCCAAGTC	TCACATCTGCCTCATCTCAG	GTA AACGACGCGCCAGT
ARAF-15 ^a	GTGTGTGTTCCACATGAGGC	GTA AACGACGCGCCAGTGGCAGAGCAACATTGATTG	GTA AACGACGCGCCAGT
BRAF-1	GTA AACGACGCGCCAGTAGCTCTCCGCTCCCTTC	AAGGTGGCTGAGGGCATC	GTA AACGACGCGCCAGT
BRAF-2	GTA AACGACGCGCCAGTGCAGCACTGGCAGTACTGTG	TCTCTTCCCAATCTATTCTAATCC	GTA AACGACGCGCCAGT
BRAF-3	TGSGTGTGATCTGACCTAGTAACCC	GTA AACGACGCGCCAGTCCATATGGCCCTACAGTATTTCTTC	GTA AACGACGCGCCAGT
BRAF-4	GTA AACGACGCGCCAGTCCCTCCCTCACTGTTACTAGCCC	TTACTTCCATATTTACATTTCC	GTA AACGACGCGCCAGT
BRAF-5	GTA AACGACGCGCCAGTGTAGTCCATCTATTACTTTGAACCC	GGGAGAAACTGCTCCATTTCC	GTA AACGACGCGCCAGT
BRAF-6	GTGTTTCTGAGAATGAAATTTGA	GTA AACGACGCGCCAGTCTGAGTGGTATGATAAGTTATTTGGG	GTA AACGACGCGCCAGT
BRAF-7	GTA AACGACGCGCCAGTGCCTTTGGCAGTATTGGATT	TCATCAGAGAAACAGCAAGC	GTA AACGACGCGCCAGT
BRAF-8	GTA AACGACGCGCCAGTGGTTTACATTGGCAAGTCTTC	GTACTTGAAGAGGCAAGTATAAAGG	GTA AACGACGCGCCAGT
BRAF-9	GTA AACGACGCGCCAGTTTGGCCCATCTCTTCCA	GCAGTGCCTGAGAAATGTGTT	GTA AACGACGCGCCAGT
BRAF-10	CTTCTCTATCCCTCTCAGGC	GTA AACGACGCGCCAGTGGCAAGTGAATATTTCCCTTGATG	GTA AACGACGCGCCAGT
BRAF-11	GTA AACGACGCGCCAGTCCATCGAACAAACAAAGTTGG	AATAGTTGCTACCCTGGGAAC	GTA AACGACGCGCCAGT
BRAF-12	CAATTGGCCAGTCTCTTAAATGATC	GTA AACGACGCGCCAGTTTACATCCTTATGTTCTGAC	GTA AACGACGCGCCAGT
BRAF-13	GTA AACGACGCGCCAGTCACTGGATAAATAGGCTTGAATGG	CTCATACATGCAACAATCC	GTA AACGACGCGCCAGT
BRAF-14	TCATCCTAACACATTTCAAGCC	GTA AACGACGCGCCAGTGTGTAATCTGGGAATCTGAAA	GTA AACGACGCGCCAGT
BRAF-15	GAATCTCTGGCAATGATGGT	GTA AACGACGCGCCAGTTCACGCTTACCCAGGATTA	GTA AACGACGCGCCAGT
BRAF-16	CCATCTATGATGGCATTGG	GTA AACGACGCGCCAGTCAACCTCATGGAAGCCATC	GTA AACGACGCGCCAGT
BRAF-17	GCTTTCTTGAAGTGTGATGGG	GTA AACGACGCGCCAGTCCACAGCAAGTGTCTTGAGTTC	GTA AACGACGCGCCAGT
RAF-1-1	GTA AACGACGCGCCAGTCTGGTCCCATTTTCCCTCATC	ATAGGGGTTGGGAAGGAAC	GTA AACGACGCGCCAGT
RAF-1-2	GTA AACGACGCGCCAGTATTTCTGTGCCACCTTTCC	AGGTATTGGTCTCAGGGCC	GTA AACGACGCGCCAGT
RAF-1-3	GTA AACGACGCGCCAGTGGCCTTGAGCAAAATACCTTC	TTGCTTACTGTAAACAGCAGCA	GTA AACGACGCGCCAGT
RAF-1-4	GTA AACGACGCGCCAGTGGAGGCCAAAGTAAGTTG	ATGAAATGCCCAACCTAGC	GTA AACGACGCGCCAGT
RAF-1-5	GTA AACGACGCGCCAGTAAAGCAAGCAGCATGATGG	CCAGAAAGCAGCAAGGG	GTA AACGACGCGCCAGT
RAF-1-6	GTA AACGACGCGCCAGTGGTGTACAGTGTAGATTTGGCC	TCCTTGATCAGATTTGAAACCC	GTA AACGACGCGCCAGT
RAF-1-7	GTA AACGACGCGCCAGTTGAAAAATCAGCCTTGAGA	TTGGCAGGAGTACTGTGTC	GTA AACGACGCGCCAGT
RAF-1-8	GGATCGAATTCGAAGTCACAG	GTA AACGACGCGCCAGTCTGTGATGCAAGTGTGCC	GTA AACGACGCGCCAGT
RAF-1-9	AACAGATGACATGGGTTGATCC	GTA AACGACGCGCCAGTCTCAGTCCCTCTCCCTCTGC	GTA AACGACGCGCCAGT
RAF-1-10	GTA AACGACGCGCCAGTTGAAATTTGCCGTATCTGTG	CGGGCACAGTCCCACTAATC	GTA AACGACGCGCCAGT
RAF-1-11	GTA AACGACGCGCCAGTGCAGCAGCAGAAACCACTGTC	GGCTTGTGCAAAATATCACAG	GTA AACGACGCGCCAGT
RAF-1-12	GTA AACGACGCGCCAGTCCCTGTGTGTAACACTCCTTGG	TTCTGCTCTCTGCCTTTTC	GTA AACGACGCGCCAGT
RAF-1-13	GCTGTGACGAGTAAAGTGG	GTA AACGACGCGCCAGTATGCAATTCGCCCTGAGGC	GTA AACGACGCGCCAGT
RAF-1-14	GTA AACGACGCGCCAGTCAATGAAAGGGACAGCCTGG	AGCCTTCTATTGTTTTGGG	GTA AACGACGCGCCAGT
RAF-1-15	CAGGTAAAGTGTGTGGTGG	GTA AACGACGCGCCAGTCCATCTGTAGAGGACCTGGG	GTA AACGACGCGCCAGT
RAF-1-16	CCAGGTCCTCTACAAGATGG	GTA AACGACGCGCCAGTAAACATGTGTTGTGCTCTGG	GTA AACGACGCGCCAGT
HRAS-1	TGGGTCTAATAGCAAGTGG	GTA AACGACGCGCCAGTTAGAGGAAGCAGGAGACAGG	GTA AACGACGCGCCAGT
HRAS-2	GTA AACGACGCGCCAGTGTGAGGCTGGCTGTGAAC	GACATCGCCAGAGGAGACAG	GTA AACGACGCGCCAGT
HRAS-3	TTCTGTGTGTGTTGCCATC	GTA AACGACGCGCCAGTGTGAGTGTGCTCCCTGG	GTA AACGACGCGCCAGT
HRAS-4	GCCTCTGTGCTCTCTGC	GTA AACGACGCGCCAGTGTGCTCTCCCAAGGACCTC	GTA AACGACGCGCCAGT
KRAS-1	GTA AACGACGCGCCAGTAAAGCCCACTGTGAAGCTGGT	AGAGAAGCAGGCTAAGTTG	GTA AACGACGCGCCAGT
KRAS-2	GTA AACGACGCGCCAGTGTCCGAGATGAGATATGG	TTTCAATGCTCTTCCCTC	GTA AACGACGCGCCAGT
KRAS-3	GTA AACGACGCGCCAGTAAAGCTTGGCATGTCCTGAC	ACTCGATCAGACAGCAGGC	GTA AACGACGCGCCAGT
KRAS-4	ATTTCCACATTCGAGGCTGAG	GTA AACGACGCGCCAGTGCAATGTCAACATGGGAAGG	GTA AACGACGCGCCAGT
NRAS-1	GTA AACGACGCGCCAGTCCAAATGGAAGGTCAACTAGG	GAACTCACTAGTATTGCAATG	GTA AACGACGCGCCAGT
NRAS-2	GTA AACGACGCGCCAGTAATAGCATTCATCCCTGTG	CTCTGGTTCCAGTCACTCC	GTA AACGACGCGCCAGT
NRAS-3	GTA AACGACGCGCCAGTAAAGCACTGTCGCCCTCTCAG	CAAGAGACAGGCTGCAAGT	GTA AACGACGCGCCAGT
NRAS-4	GCTCTGTGTGATCAATAGG	GTA AACGACGCGCCAGTTGTGAGAAAGGATAGGCAG	GTA AACGACGCGCCAGT

^aThe primer pair did not meet our quality criteria that ≥90% of bases in the target region have a Phred quality score of at least 20 in three quarters of the tumor samples analyzed.

Table S7. Primers used for ERBB4 plasmid construction

Gene Name	Forward Primer	Reverse Primer
ERBB4 into pCDF-MCS2-EF1-Puro™	CGGCTCTAGAGCCACATGAAAGCCGGCGAC	ATCGGGCGGCGCTTACACCACAGTATTCGG
Knockdown resistant ERBB4 primers into pCDF1-MCS2-EF1-puro	CACCAAAATCAAGGGGACTT	GGAGCCAGTACACGACATCA
Knockdown resistant ERBB4 primer set #1 into pcDNA3.1	GATTCTGTGGCCATTAAAGTTCT	AGAATCTTAATGGCCACAGGAATC
Knockdown resistant ERBB4 primer set #2 into pcDNA3.1	CCTGTGGCCATCAAGATTCT	AGAATCTTGTATGGCCACAGG
Knockdown resistant ERBB4 primer set #3 into pcDNA3.1	CCTGTGGCCATCAAAATCTTAATGAGAC	GTCTCTAAGAATTTTGTATGGCCACAGG

Supplementary Note
Tumor Tissue Collection

A panel of 79 pathology-confirmed metastatic melanoma tumor resections was assembled from patients referred to the Surgery Branch of the National Cancer Institute for enrollment in IRB-approved Surgery Branch clinical trials of immunotherapy. All of the patients had progressive metastatic melanoma that had failed available conventional treatments, were 16 years or older and passed eligibility testing for the relevant protocol. None had received therapy for 1 month prior to entering the protocol. Before tissue was acquired all patients signed written informed consents. The melanoma tissue panel was selected on the basis of an available pathology-confirmed, melanoma tissue culture line, paired with freshly frozen tissue from the resected melanoma metastasis and apheresis-collected, peripheral blood mononuclear cells.