Supplementary Online Content

Kris MG, Johnson BE, Berry LD, et al. Using Multiplexed Assays of Oncogenic Drivers in Lung Cancers to Select Targeted Drugs. *JAMA*.
doi:10.1001/jama.2014.3741

This supplementary material has been provided by the authors to give readers additional information about their work

eTable 1. Trials of targeted therapies available within the Lung Cancer Mutation Consortium

Gene target	Trial title (identifier)	Patient eligibility
ALK	Crizotinib (Xalkori®) Versus Standard Of Care In Patients With Advanced Non-Small Cell Lung Cancer (NSCLC) With A Specific Gene Profile Involving The Anaplastic Lymphoma Kinase (ALK) (NCT00932893)	Patients with NSCLC who have previously been treated with chemotherapy, including at least one platinum agent (for example, carboplatin, cisplatin), and have the EML4-ALK gene rearrangement
	Crizotinib (Xalkori®) In Patients With Advanced Non-Small Cell Lung Cancer With A Specific Gene Profile Involving The Anaplastic Lymphoma Kinase (ALK) Gene (NCT00932451)	Patients from study NCT00932893 whose disease progressed while receiving standard of care chemotherapy
PIK3CA	Safety and Efficacy of BKM120 in Patients With Metastatic Non-small Cell Lung Cancer (NCT01297491)	Patients with non-small cell lung cancer who have been previously treated with one or more therapies, and have a PIK3CA mutation
BRAF V600E	A Phase II Study of the Selective BRAF Kinase Inhibitor GSK2118436 in Subjects With Advanced Non-small Cell Lung Cancer and BRAF Mutations (NCT01336634)	Patients with non-small cell lung cancer who have been previously treated with one or more therapies, and have the V600E BRAF mutation
BRAF (non V600E) ***KRAS*** ***NRAS*** ***MEK***	An Open-label Study of GSK1120212 Compared With Docetaxel in Stage IV KRAS-mutant Non-small Cell Lung Cancer (NCT01362296)	Patients with NSCLC who have previously been treated with chemotherapy, including at least one platinum agent (for example, carboplatin, cisplatin), and have a mutation in the KRAS, NRAS, MEK, or BRAF (non-V600E) gene
KRAS	Erlotinib Plus ARQ 197 Versus Single Agent Chemotherapy in Locally Advanced or Metastatic Non-Small Cell Lung Cancer (NCT01395758)	Patients with non-small cell lung cancer who have been previously treated with one or more therapies, and have a mutation in the KRAS gene
EGFR	A Study of MM-121 Combination Therapy in Patients With Advanced Non-Small Cell Lung Cancer (NCT00994123)	Patients with metastatic NSCLC and (1) EGFR mutation-positive NSCLC, with no prior EGFR-directed treatment; (2) EGFR mutation-negative NSCLC, with no prior EGFR-directed treatment; or (3) EGFR mutation-positive NSCLC with acquired resistance to prior EGFR-directed treatment
	Study of Erlotinib (Tarceva®) in Combination With OSI-906 in Patients With Advanced Non-small Cell Lung Cancer (NSCLC) With Activating Mutations of the Epidermal Growth Factor Receptor (EGFR) Gene (NCT01221077)	Patients with stage IIIB or IV NSCLC who have a mutation in the EGFR gene and have not received chemotherapy for advanced cancer; Patients who received chemotherapy before or after surgery, but had their disease progress anyway
HER2	PF-00299804 As A Single Oral Agent In Selected Patients With Adenocarcinoma Of The Lung (NCT00818441)	Patients with locally advanced or metastatic adenocarcinoma with or without prior treatment who have a tumor with a mutation in or amplification of the HER2 gene
MET	A Study Of Oral PF-02341066 (crizotinib), A c-Met/Hepatocyte Growth Factor Tyrosine Kinase Inhibitor, In Patients With Advanced Cancer (NCT00585195)	Patients with locally advanced or metastatic lung cancer and a verified amplification of or mutation in the MET gene

eTable 2. Numbers of patients enrolled for each oncogenic driver and specific agents used

Gene target (n=patients with testing)	Patients with mutation identified	Patients on targeted therapy	Distribution of targeted therapy		
AKT1 (n=941)	0	0	N/A		
BRAF V600E (n=949)	14	2	AZD6244 (2)		
BRAF non-V600E (n=950)	4	1	AZD6244 (1)		
EGFR (sensitizing) (n=987)	175	146	Afatinib (1)	Erlotinib (130)	Erlotinib/OSI-906 (2)
			Dacomitinib (2)	Erlotinib/HCQ (2)	Gefitinib (1)
			Afatinib/Cetuximab (1)	Erlotinib/MM-121 (1)	Multiple lines* (6)
EGFR (other) (n=984)	35	23	AUY922 (1)	Erlotinib (14)	Neratinib/Temsirolimus (2)
			Dacomitinib (1)	Erlotinib/HCQ (1)	Multiple lines* (2)
			Erlotinib/MM-121 (1)	Erlotinib/OSI906 (1)	
HER2 (n=920)	23	11	Dacomitinib (5)	Neratinib/Temsirolimus (2)	
			Lapatinib/Trastuzumab/	STA-9090 (1)	
			Bevacizumab (1)	Trastuzumab (2)	
KRAS (n=981)	245	22	AUY922 (1)	Erlotinib/Tivantinib (3)	LY2835219 (2)
			AZD6244 (2)	Everolimus (2)	Ridaforalimus (1)
			Docetaxel/AZD6244 (1)	GDC0941/GDC0973 (3)	STA-9090 (3)
			Erlotinib/AZD6244 (2)	Imetelstat (1)	MEK inhibitor (1)
MEK1 (n=939)	2	0	N/A		
NRAS (n=940)	5	0	N/A		
PIK3CA (n=945)	7	0	N/A		
ALK (n=926)	80	52	Crizotinib (51)	Crizotinib/LDK378 (1)	
MET (n=833)	6	3	Crizotinib (3)		
Doubletons	27	15	ALK:Crizotinib (PF-02341066) (3) EGFR:Erlotinib (Tarceva) (10)	MET/HER2:Crizotinib/Daconitinib (1) PIK3CA:GSK2141795 (1)	

***Multiple lines of therapy include erlotinib, dacomitinib, erlotinib/BMS-936558.

eTable 3. Enrollment and genotyping frequencies by study site

Site	Enrollment	Total number with any or full genotyping	Any Genotyping number (%)	Full Genotyping number (%)
University of California, Los Angeles	104	50	8(16%)	42(84%)
University of Colorado, Denver	264	188	48(25%)	140(75%)
H. Lee Moffit Cancer Center	77	63	11(17%)	52(83%)
Emory University	71	42	4(10%)	38(90%)
Massachusetts General Hospital	118	85	25(29%)	60(70%)
Dana Farber Cancer Institute	322	184	25(14%)	159(86%)
Johns Hopkins Medical Institute	87	49	12(24%)	37(76%)
National Cancer Institute	16	13	13(100%)	0(0%)
Memorial Sloan Kettering Cancer Center	190	173	48(28%)	125(72%)
Pittsburgh Cancer Institute	20	9	1(11%)	8(88%)
Medical University of South Carolina	16	3	3(100%)	0(0%)
Vanderbilt-Ingram Cancer Center	60	47	41(87%)	6(13%)
University of Texas, Southwestern	53	40	11(27%)	29(73%)
MD Anderson Cancer Center	139	61	24(39%)	37(27%)
Totals	1537	1007	274	733
Median %			25%	73%
Range			10%-100%	0%-90%

eTable 4. Frequency of oncogenic drivers in all specimens in which a driver in a single gene was identified*

*To reconcile counts presented here with Table 2, all occurrences of each mutation, alone and/or in combination with other mutations, must be considered. For example, *EGFR* exon19 deletions appears with a count of 102 alone, plus an additional occurrence in combination with *G719A*, for a total count of 103, as presented in Table 2; the instances of exon 19 deletion with *T790M*, as presented in this table, is accounted for among the *EGFR* "other" mutations in Table 2, as explained in the footnote to that table that describes the categorization of *EGFR* mutation types.

Gene (n=cases tested)	**Amino Acid Mutant**	**Nucleotide Mutant**	**Frequency**
BRAF (n=951)	G469A	G469 BRAF_c.1406G.C	2
	G469V	G466 BRAF_c.1406G.T	1
	V600E	BRAF_c.1799T.A	14
	n/a	BRAF other	1
EGFR (n=987)	G719S	EGFR_c.2155G.A	1
	G719C plus S768I	EGFR_c.2155G.T, EGFR_c.2303G.T	3
	G719A	EGFR_c.2156G.C	2
	G719A plus L861Q	EGFR_c.2156G.C,EGFR_c.2582T.A	1
	G719A plus exon 19 del	EGFR_c.2156G.C,EGFR_exon.19.del	1
	T790M plus L858R p.L858R	EGFR_c.2369C.T,EGFR_c.2573T.G	5
	T790M plus exon 19 del	EGFR_c.2369C.T,EGFR_exon.19.del	3
	L858R	EGFR_c.2573T.G	64
	L861Q	EGFR_c.2582T.A	4
	exon 19 del	EGFR_exon.19.del	102
	exon 20 ins	EGFR_exon.20.ins	22
	n/a	EGFR other	1
HER2 (n=920)	exon 20 ins	ERBB2_ins.A775	23
MEK1 (n=939)	K57N	MEK1_c.171G.T	2
PIK3CA (n=945)	E545K	PIK3CA_c.1633G.A	5
	H1047R	PIK3CA_c.3140A.G	2
KRAS (n=981)	Q61R	KRAS_c.182A.G	1
	Q61L	KRAS_c.182A.T	1
	Q61H	KRAS_c.183A.C	5
	Q61H	KRAS_c.183A.T	5
	Q61H plus G13C	KRAS_c.183A.T,KRAS_c.37G.T	1
	G12S	KRAS_c.34G.A	7
	G12R	KRAS_c.34G.C	5
	G12C	KRAS_c.34G.T	98
	G12C plus G12V	KRAS_c.34G.T,KRAS_c.35G.T	2
	G12D	KRAS_c.35G.A	54
	G12D plus G13C	KRAS_c.35G.A,KRAS_c.37G.T	1
	G12A	KRAS_c.35G.C	12
	G12V	KRAS_c.35G.T	37
	G13C	KRAS_c.37G.T	7
	G13D	KRAS_c.38G.A	7
	G13V	KRAS_c.38G.T	1
	G 13R	KRAS_c.37G.C	1
NRAS (n=940)	Q61K	NRAS_c.181C.A	1
	Q61R	NRAS_c.182A.G	2
	Q61L	NRAS_c.182A.T	1
	G12R	NRAS_c.34G.C	1

eTable 5. Specific genetic aberrations in tumors with drivers in more than one gene – Any Genotyping group

	Gene 1	Gene 2	Frequency
Double oncogenic driver: *PIK3CA* and another mutation	PIK3CA E542K	BRAF V600E	1
	PIK3CA E545K	BRAF V600E	1
	PIK3CA E542K	EGFR L858R	1
	PIK3CA E542K	EGFR exon 19 del	1
	PIK3CA E542K	EGFR other	1
	PIK3CA H1047Y	EGFR L858R	1
	PIK3CA H1047R	EGFR exon 19 del	3
	PIK3CA H1047R	EGFR exon 20 ins	1
	PIK3CA H1047L	EGFR exon 19 del	1
	PIK3CA E545K	KRAS G12D	1
	PIK3CA H1047R	MEK1 K57N	1
Double oncogenic driver: *MET* amplification and another mutation	MET amplification	EGFR L858R	1
	MET amplification	EGFR exon 19 del	1
	MET amplification	HER2 exon 20 ins ERBB2_ins.A775	1
	MET amplification	KRAS G12C	2
	MET amplification	KRAS G12A	1
	MET amplification	KRAS G12V	1
Double oncogenic driver: *ALK* rearrangement and another mutation	ALK rearrangement	BRAF V600E	1
	ALK rearrangement	EGFR L858R	1
	ALK rearrangement	EGFR L861Q	1
	ALK rearrangement	EGFR exon 19 del	1
Double oncogenic driver: *ALK* rearrangement and *MET* amplification	MET amplification	ALK rearrangement	2
Double oncogenic driver: Two genes other than *PIK3CA*	AKT E17K	EGFR exon 19 del	1

eTable 6. Oncogenic drivers identified by cigarette smoking status*: Never, Current, Former

	Gene with mutational or structural change	Current Smokers Driver in indicated gene (n=73) Count (%)	Former Smokers Driver in indicated gene (n=589) Count (%)	Never Smokers Driver in indicated gene (n=341) Count (%)
Any gene(s)		45 (62%)	340 (58%)	238 (70%)
SINGLETONS**	*KRAS*	33 (45%)	198 (34%)	14 (4%)
	EGFR (sensitizing)***	4 (5%)	59 (10%)	114 (33%)
	exon19 del	4 (5%)	29 (5%)	70 (21%)
	L858R	0 (0%)	24 (4%	40 (12%)
	G719X	0 (0%)	3 (1%)	2 (1%)
	L861Q	0 (0%)	3 (1%)	2 (1%)
	ALK (rearrangement)	3 (4%)	26 (4%)	51 (15%)
	EGFR (other)****	0 (0%)	14 (2%)	21(6%)
	HER2	1 (1%)	6 (1%)	16 (5%)
	BRAF	2 (3%)	13 (2%)	3 (1%)
	V600E	1 (1%)	10 (2%)	3 (1%)
	Non-*V600E*	1 (1%)	3 (1%)	0 (0%)
	PIK3CA	0 (0%)	4 (1%)	3 (1%)
	MET (amplification)	1 (1%)	4 (1%)	1 (0%)
	NRAS	0 (0%)	5 (1%)	0 (0%)
	MEK1	0 (0%)	2 (0%)	0 (0%)
	AKT1	0 (0%)	0 (0%)	0 (0%)
	>1 gene (doubletons)	1 (1%)	11 (2%)	15 (4%)

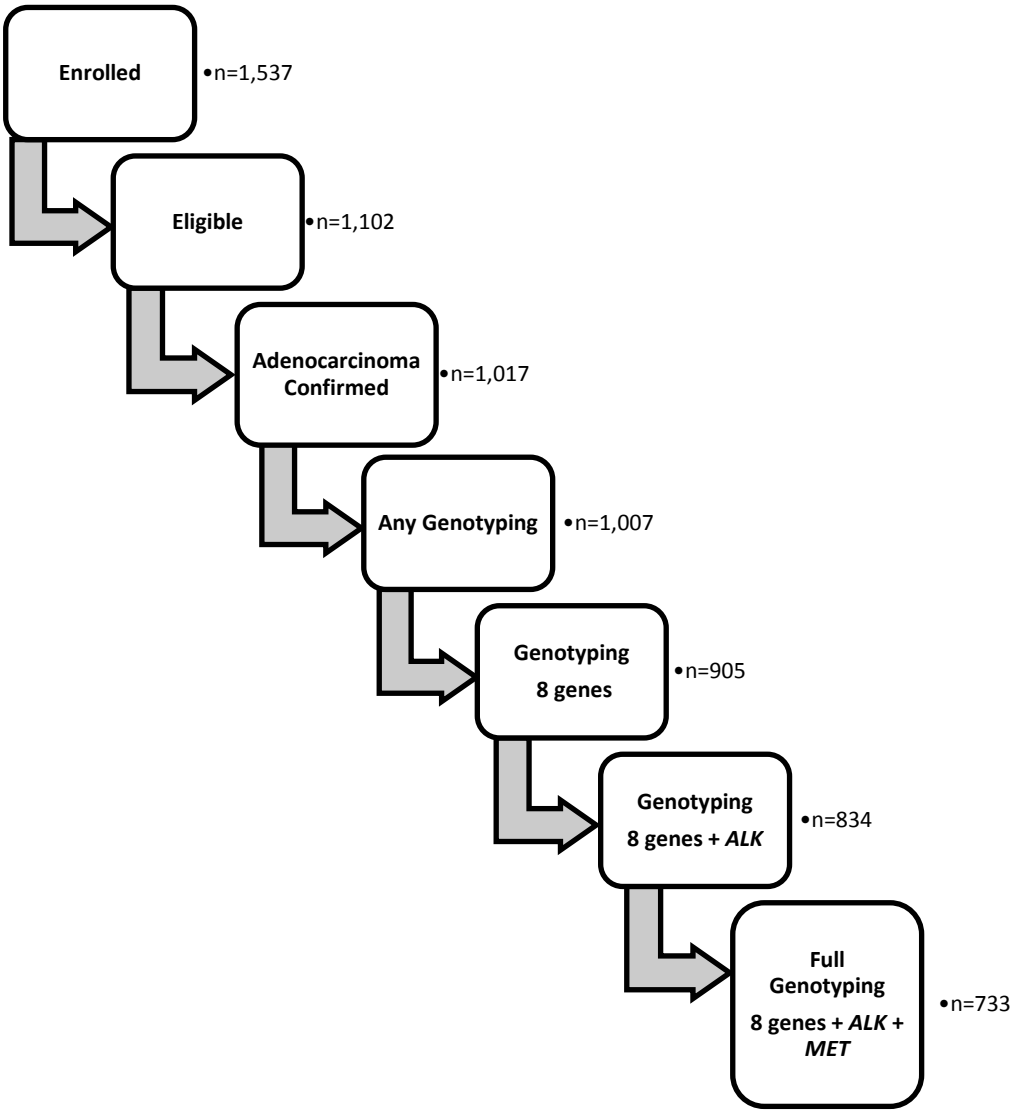
*Patient smoking status from Any Genotyping group; as not all cases in this group were tested for all genes, percentages therefore reflect rate of detection of mutation where non-detection is a combination of negative findings and no findings.

**Per-gene count (percent) of patients with mutations occurring in a single gene (Singletons) . Patients with oncogenic drivers in more than one gene (Doubletons) are included as their own category. For detailed information on mutations occurring within Doubletons, see eTable 5.

***The sum of counts for the four categories of *EGFR*-sensitizing mutations differs from the patient-level count of total number of patients with EGFR-sensitizing mutations due to two patients with the co-occurrence of two different sensitizing mutations in the same specimen.

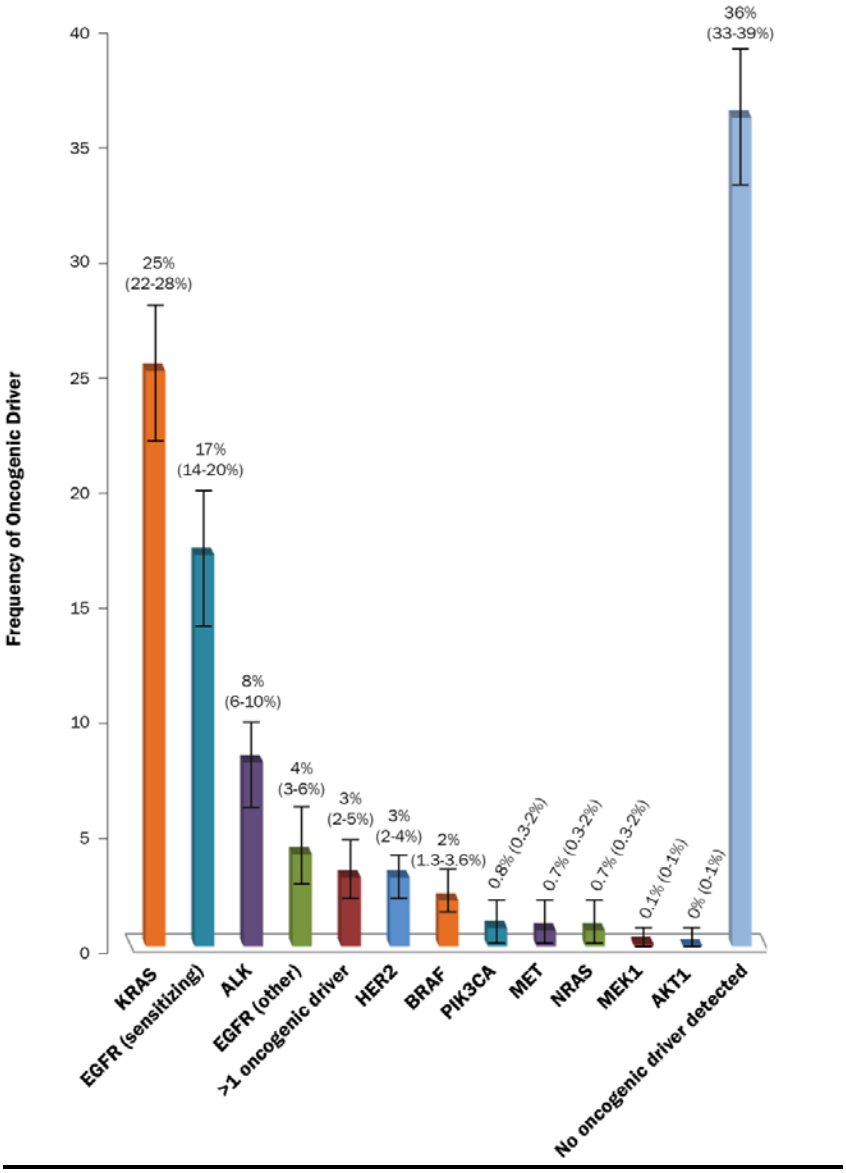
****Patients are counted in the *EGFR* (other) group based on a finding of any one or more mutations in EGFR other than exon 19 deletions, *L858R, G719X*, or *L861Q*, with or without co-occurrence in the same case of one of these sensitizing mutations. See eTable 5 for detailed count of co-occurring *EGFR* mutations.

eFigure 1. Status of All 1537 Patients Enrolled



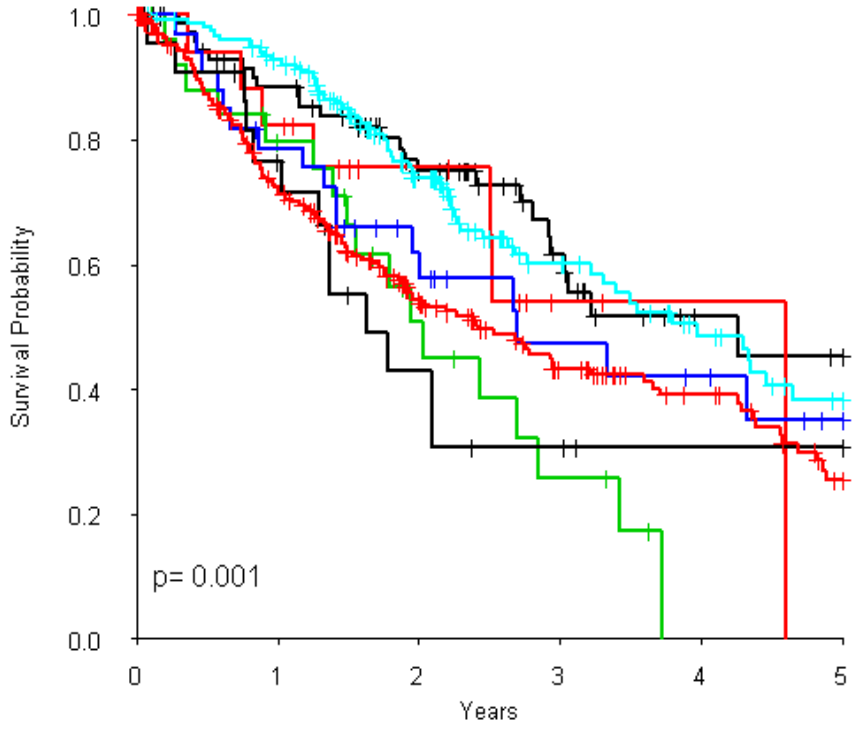
Patient flow from enrollment through screening, pathology review, and genotyping.

eFigure 2. Frequency of Oncogenic Drivers Detected in the 733 Patients Tested for All 10 Drivers



Frequency of oncogenic drivers detected in the 733 patients tested for all 10 drivers (Full Genotyping group)

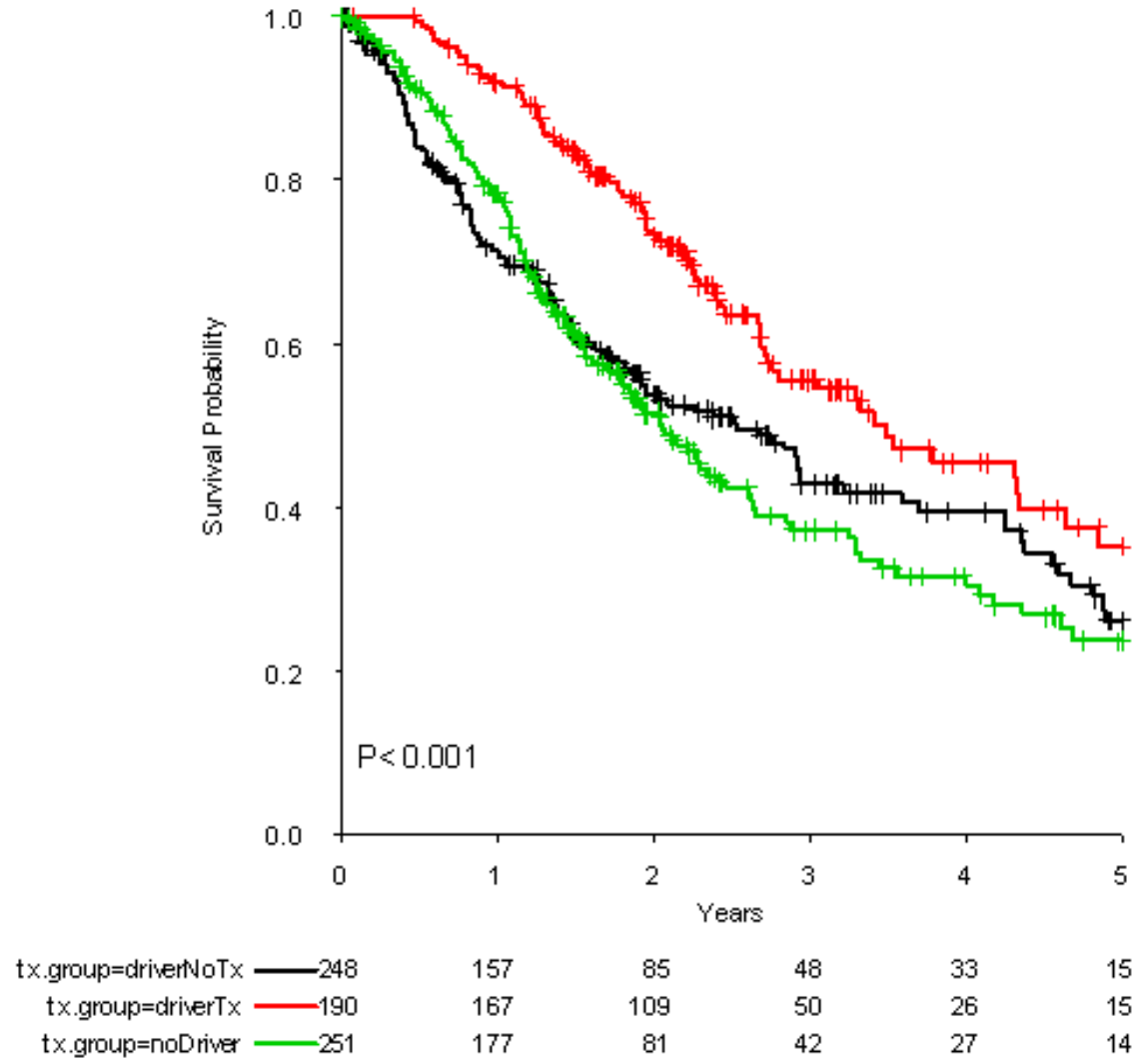
eFigure 3. Survival by Oncogenic Driver for the 7 Drivers Identified in at Least 10 Patients



driver=ALK	74	59	41	21	8	6
driver=BRAF	17	14	8	2	1	
driver=doubs	26	18	9	4		
driver=EGFR(o)	33	25	15	9	7	3
driver=EGFR(s)	158	137	80	41	26	16
driver=HER2	22	15	7	4	2	1
driver=KRAS	232	152	88	52	33	16

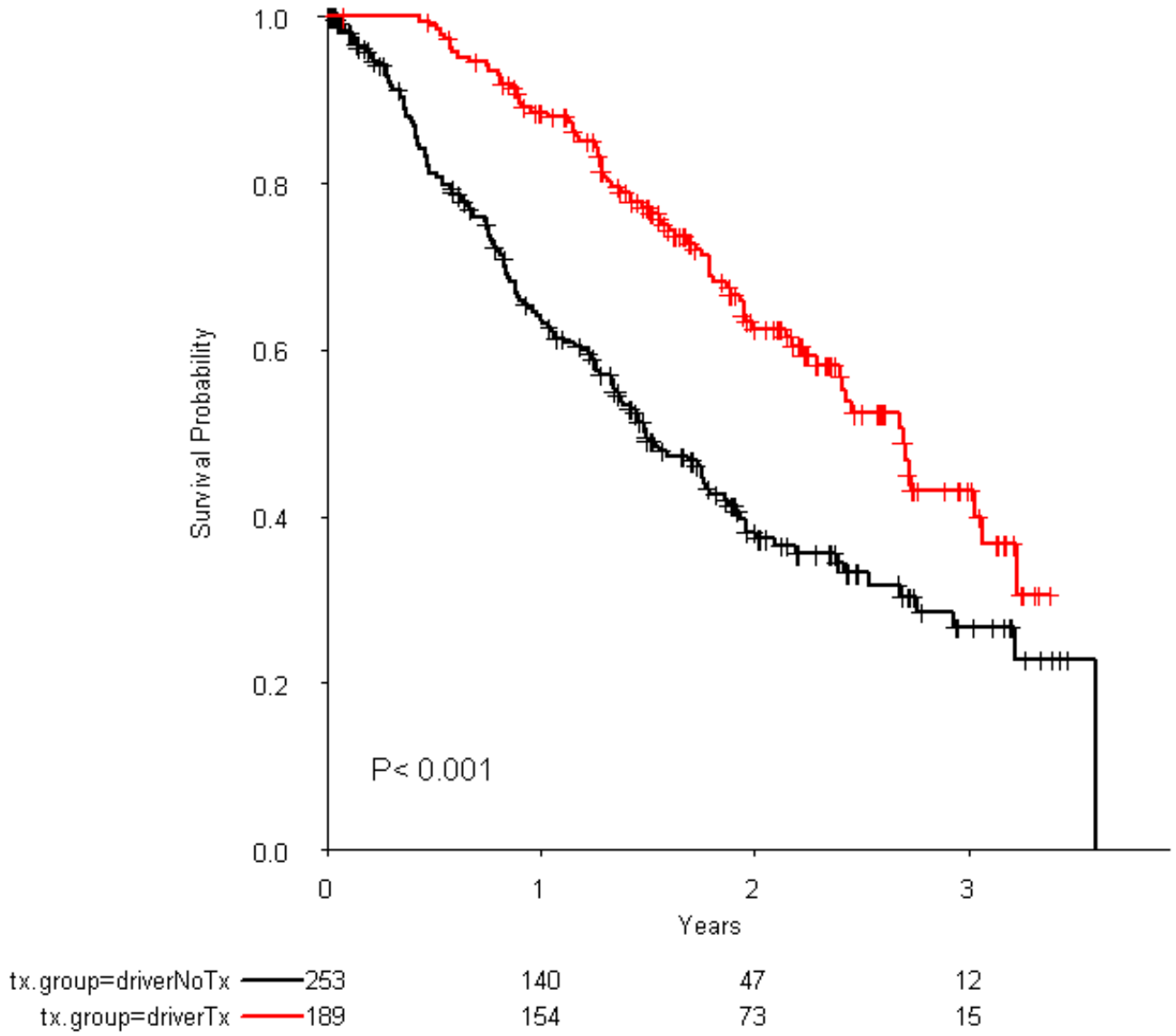
Comparison of survival among patients with an oncogenic driver detected, including all oncogenic driver genes with lesions detected in at least 10 patients. Median survival (95% CI): EGFR (sensitizing), 3.97 (3.21-4.64); EGFR (other), 2.70 (1.42-NA); ALK, 4.25 (2.92-NA); KRAS, 2.41 (1.87-3.21); doubletons, 2.03 (1.39-2.84); HER2, 1.63 (1.03-NA); BRAF, 4.59 (1.25-NA); p=0.001 (log-rank test). Vertical tick-marks indicate censoring events.

eFigure 4. Survival by Driver-Treatment Status in Patients With Full Genotyping (10 genes)



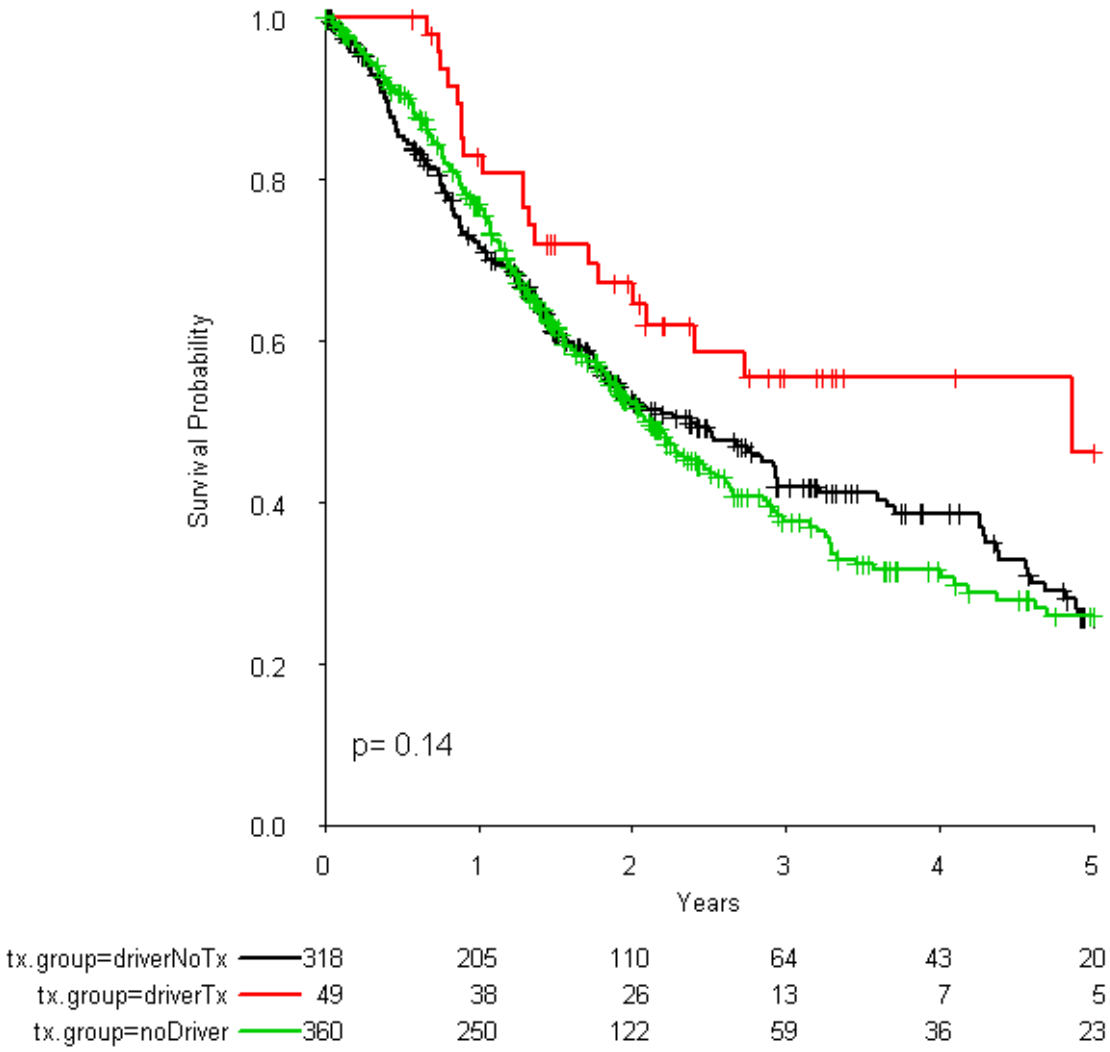
Among patients with Full Genotyping, comparison of those with a driver detected and treated with a targeted therapy, against those with a driver detected and not treated with a targeted therapy. Survival of patients with no oncogenic driver identified also included. Median survival (95% CI): driver, no targeted therapy, 2.53 (1.85-3.21); driver, targeted therapy, 3.49 (2.71-4.33); no oncogenic driver identified, 2.05 (1.70-2.46); p<0.001 (log-rank test). Vertical tick-marks indicate censoring events.

eFigure 5. Survival in Patients With Metastatic Cancer Diagnosed Within 6 Months Prior to Study Initiation



Among patients with metastatic disease diagnosed <=6 months prior to study initiation (September 1, 2009), comparison of those with a driver detected and treated with a targeted therapy, against those with a driver detected and not treated with a targeted therapy. Median survival (95%CI): driver, no targeted therapy, 1.49 (1.33-1.79); driver, targeted therapy, 2.69 (2.29-3.06); p<0.001 (log-range test). Vertical tick-marks indicate censoring events.

eFigure 6. Survival in Patients With Oncogenic Drivers Other Than *EGFR* or *ALK*



Comparison of survival of patients with an oncogenic driver other than EGFR or ALK and treated with a targeted therapy, against those with an oncogenic driver and no targeted therapy. Survival of patients with no oncogenic driver identified also displayed. Median survival (95% CI): driver, no targeted therapy, 2.38 (1.81-2.93); driver, targeted therapy, 4.85 (2.00-NA); no oncogenic driver identified, 2.08 (1.84-2.46); p=0.14 (log-rank test). Vertical tick-marks indicate censoring events.