OSPEDALE SAN RAFFAELE

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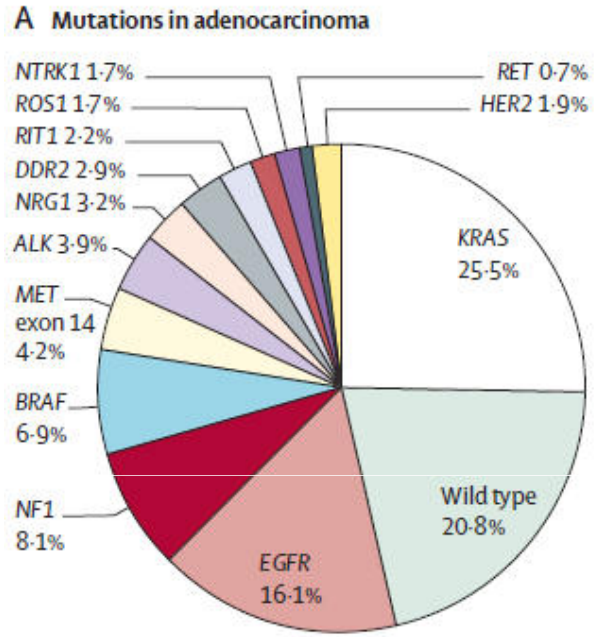
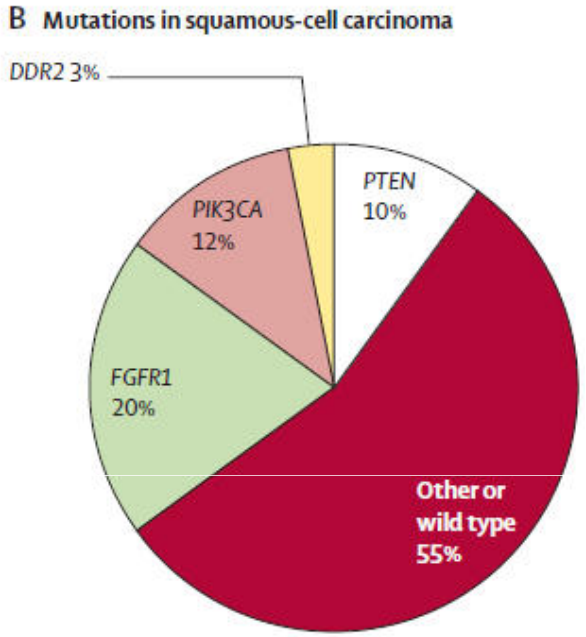
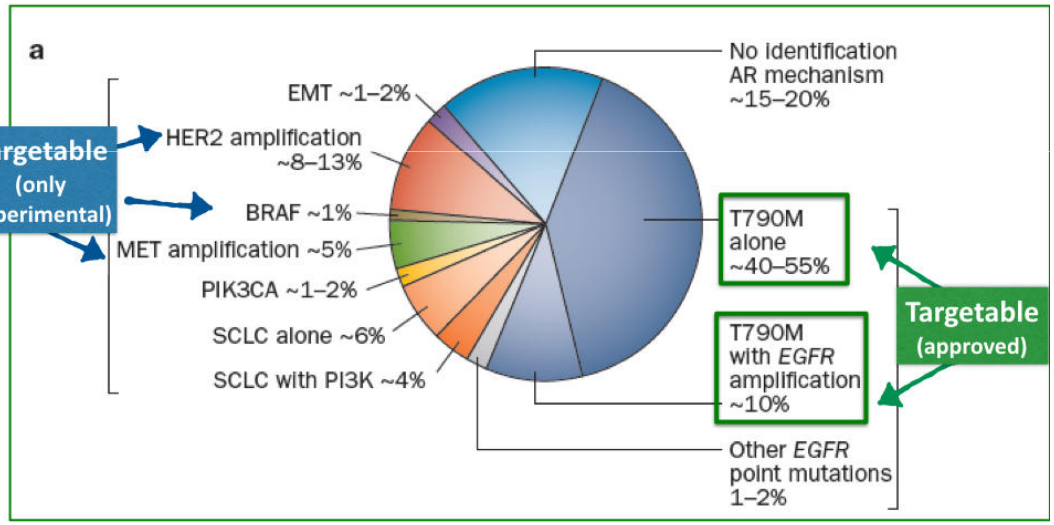
New options for oncogene addicted NSCLCs: options for acquired resistance

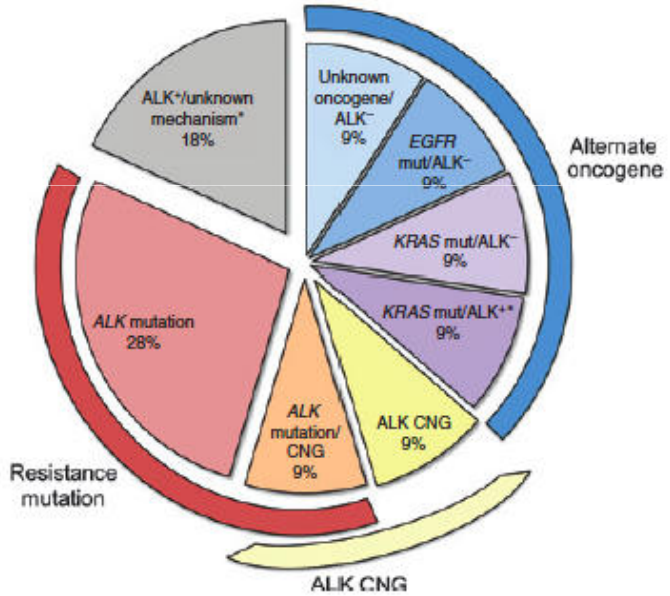




Rosell and Karachaliou; Lancet; 2016

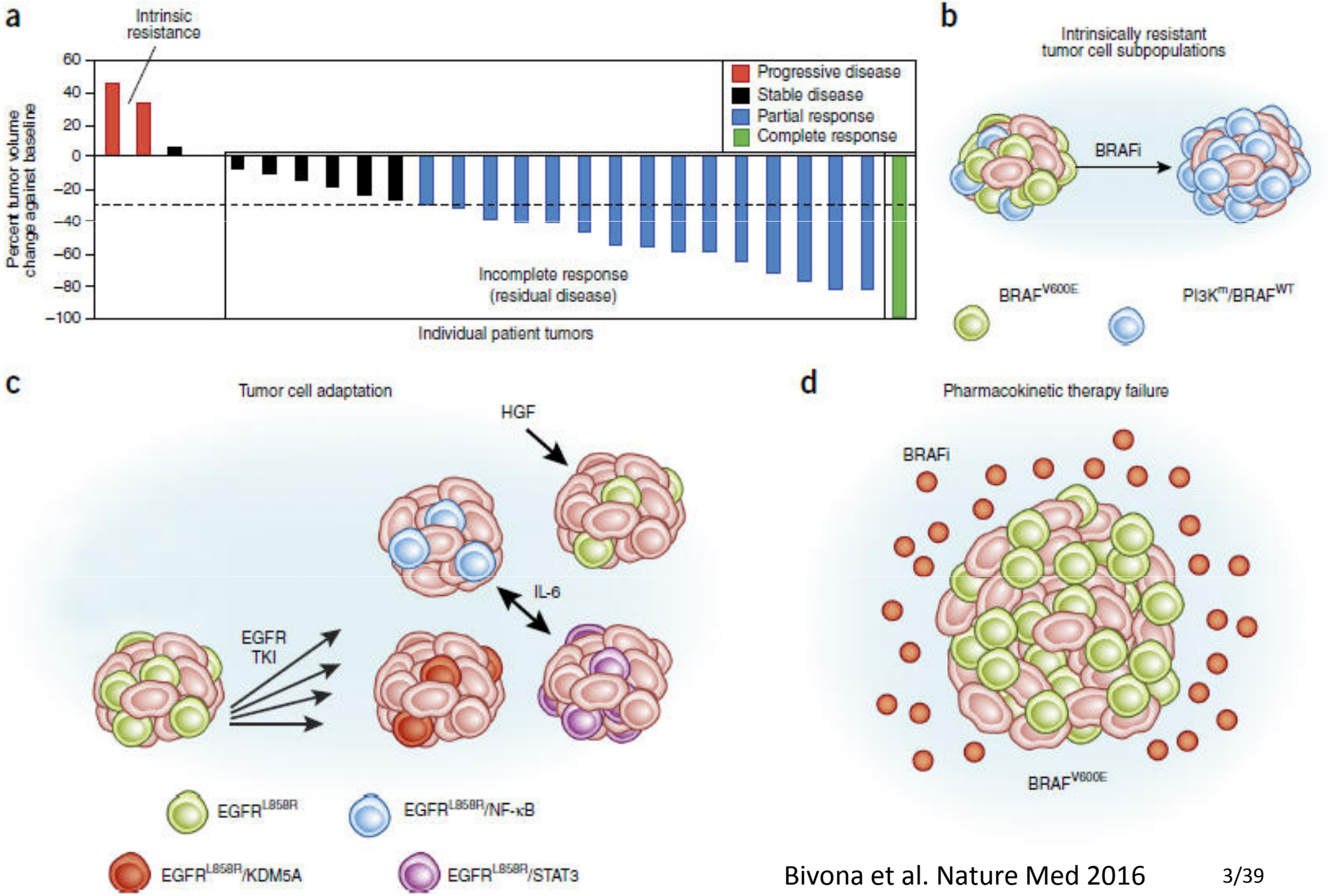


Camidge et al; Nat Rev Clin Oncol; 2014



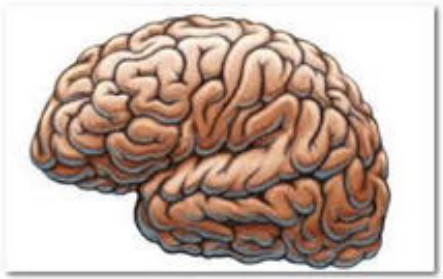
Doebele et al; Clin Canc Res; 2012

Which lesson from the use of targeted agents



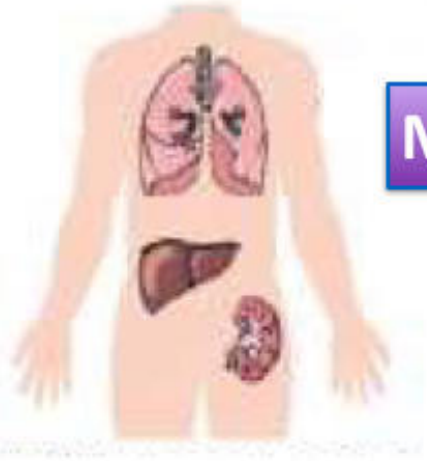
Bivona et al. Nature Med 2016

Patterns of progression in patients receiving targeting agents



Brain progression

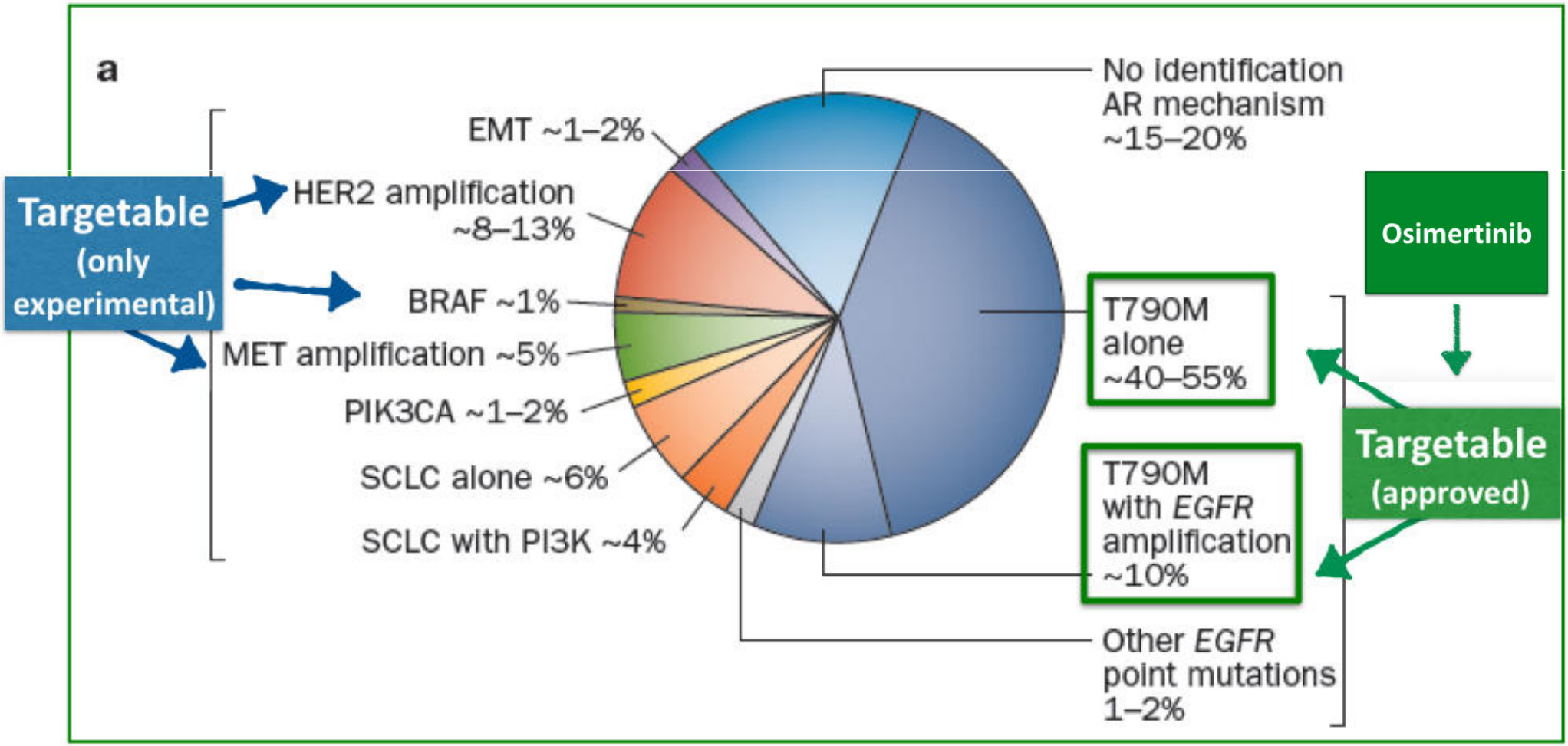
Oligoprogression



Multisite progression

EGFR mutated NSCLC

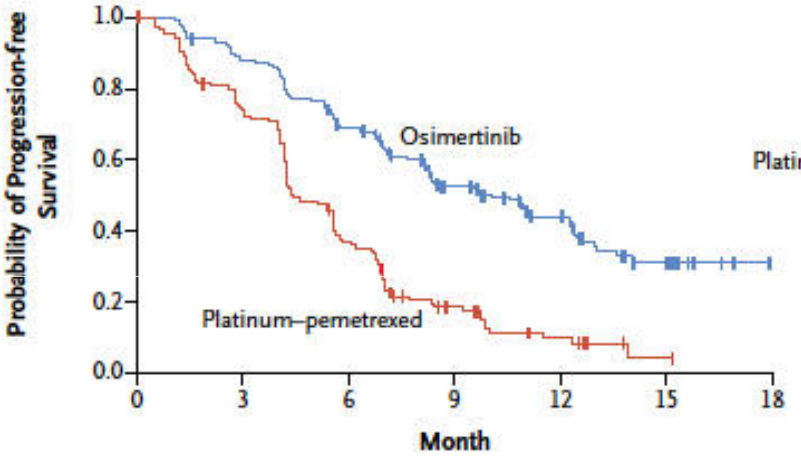
Heterogeneous acquired resistance mechanisms to EGFR-TKIs



Camidge et al, Nat Rev Clin Oncol 2014

Osimertinib - AURA3

A Patients in Intention-to-Treat Population

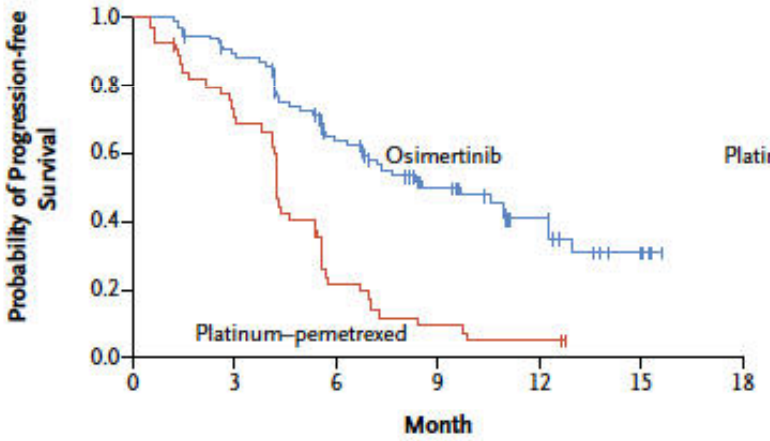


No. at Risk							
Osimertinib	279	240	162	88	50	13	0
Platinum–pemetrexed	140	93	44	17	7	1	0

	No. of Patients	Median Progression-free Survival *mo (95% CI)*
Osimertinib	279	10.1 (8.3–12.3)
Platinum–pemetrexed	140	4.4 (4.2–5.6)

Hazard ratio for disease progression or death, 0.30 (95% CI, 0.23–0.41)
P<0.001

B Patients with CNS Metastases



No. at Risk							
Osimertinib	93	80	46	27	14	4	0
Platinum–pemetrexed	51	32	9	4	2	0	0

	No. of Patients	Median Progression-free Survival *mo (95% CI)*
Osimertinib	93	8.5 (6.8–12.3)
Platinum–pemetrexed	51	4.2 (4.1–5.4)

Hazard ratio for disease progression or death, 0.32 (95% CI, 0.21–0.49)

Mok et al, NEJM 2016

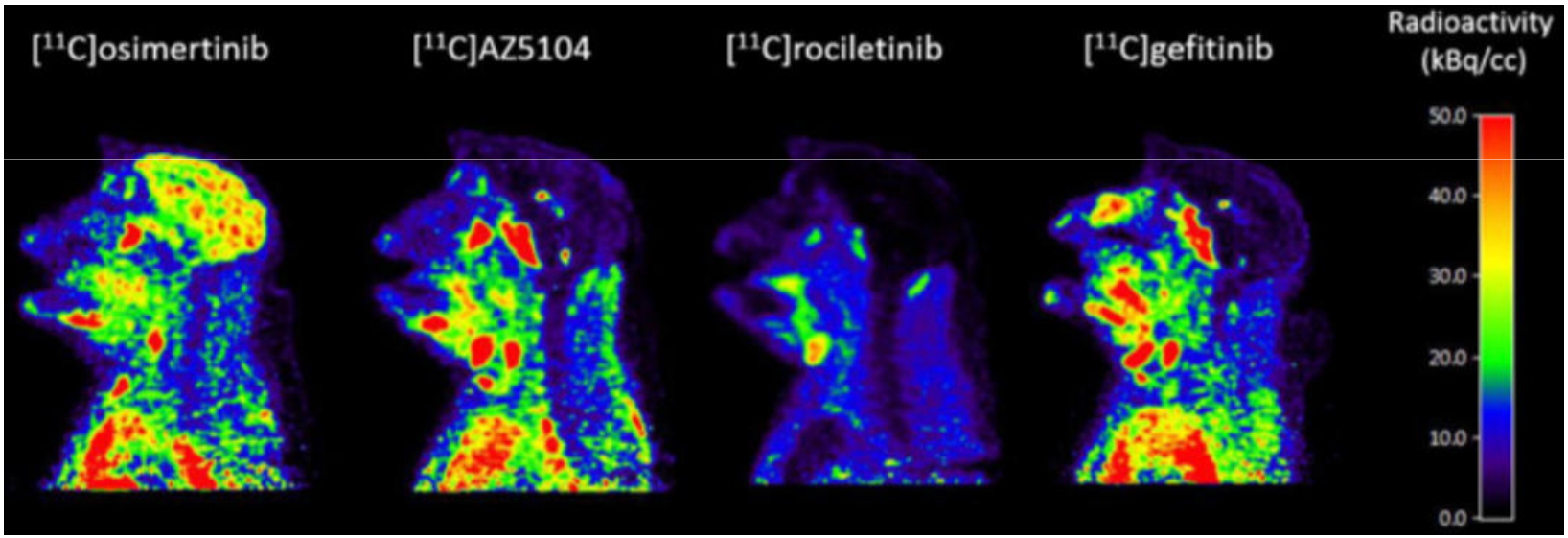
Osimertinib and brain metastases

30% of EGFR mutant patients develop brain lesions in the course of EGFR-TKis

Osimertinib and its metabolites AZ5104 and AZ7550 are substrates of Pgp and BCRP

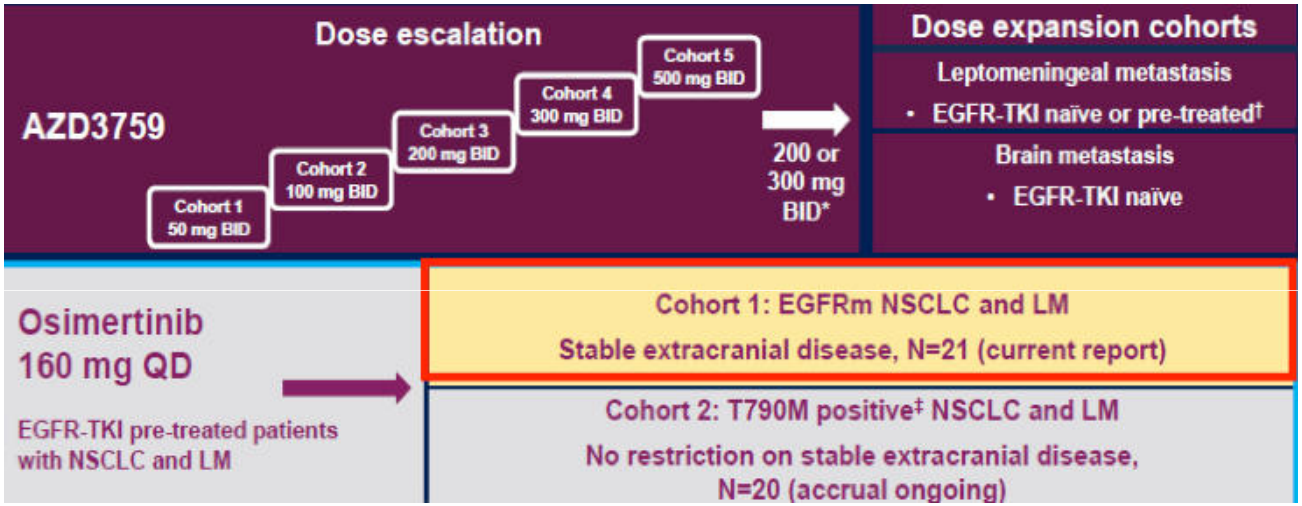
Table 2. Distribution to mouse brain of osimertinib, gefitinib, rociletinib, and afatinib following oral administration

	Osimertinib	Gefitinib	Rociletinib	Afatinib
Dose (mg/kg)	25	6.25	100	7.5
Plasma C_{max} (µmol/L)	0.82	0.82	3.32	0.14
Brain C_{max} (µmol/L)	2.78	0.17	BLQ	BLQ
Brain/plasma C_{max} ratio	3.41	0.21	<0.08	<0.36



Ballard et al, Clinical Cancer Research 2016

Osimertinib and leptomeningeal metastases - The BLOOM study



Efficacy assessments were conducted on 21 patients

- Seven patients had confirmed* radiological improvement
- Two patients had confirmed* CSF cytology clearance; no tumor cells were detected in two consecutive CSF samples
- Five patients had confirmed* improved neurological function

Best MRI imaging intracranial response, n (%)	N=21	
	Confirmed*	Unconfirmed
Responding	7 (33)	1 (5)
Stable disease	9 (43)	2 (10)
Early withdrawal	2 (10)	

Yang et al, ASCO 2016

How to identify the emerging mechanisms of acquired resistance - Analysis of circulating tumor DNA

Retrospective analysis of EGFR mutant patients enrolled in the AURA trial

Table 1. Sensitivity and Specificity of Plasma Genotyping Assays Compared With Tumor Genotype As a Reference Standard

Plasma Genotype (BEAMing)	Tumor Genotype (cobas, Central Laboratory)	
	Exon 19 del+ (n = 136)	Exon 19 del− (n = 80)
Exon 19 del+ (n = 114)	112 (82.3% sensitivity)	2
Exon 19 del− (n = 102)	24	78 (97.5% specificity)
	L858R+ (n = 73)	L858R− (n = 143)
L858R+ (n = 68)	63 (86.3% sensitivity)	5
L858R− (n = 148)	10	138 (96.5% specificity)
	T790M+ (n = 158)	T790M− (n = 58)
T790M+ (n = 129)	111 (70.3% sensitivity)	18
T790M− (n = 87)	47	40 (69.0% specificity)

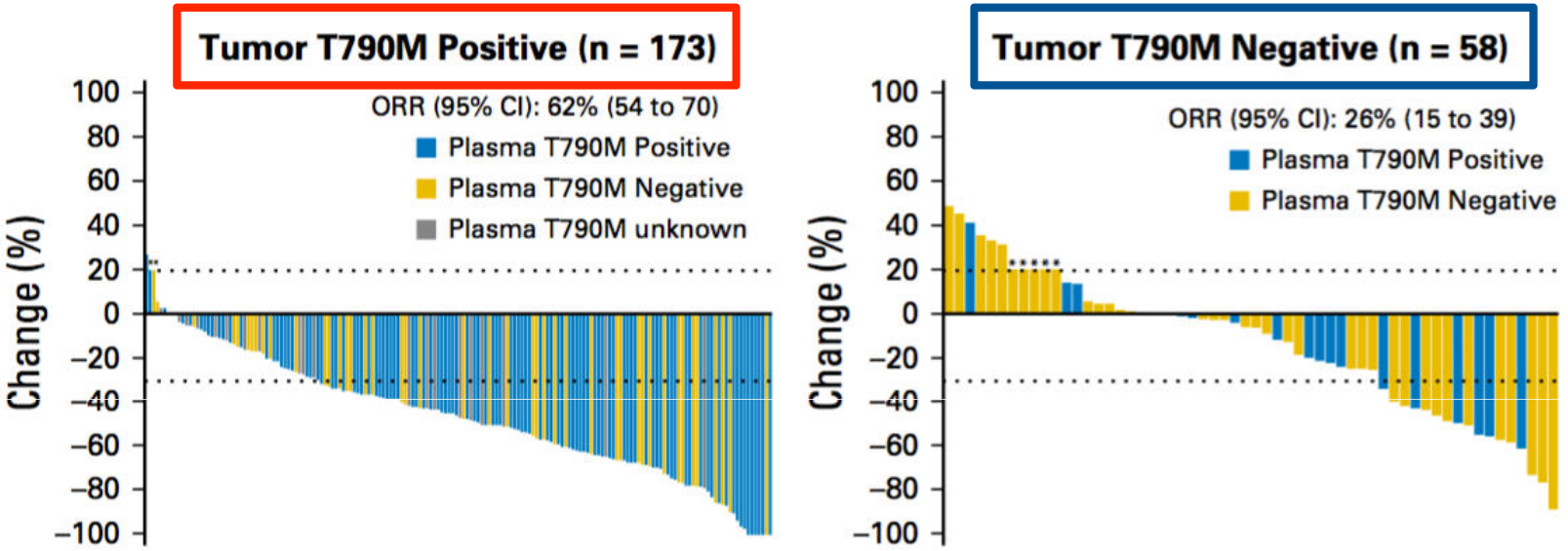
High specificity (~100%), good sensitivity (>80%) for EGFR sensitizing mutations

Good sensitivity and specificity for T790M (~70%)

20% false negative results for EGFR sensitizing mutations
30% false negative results for T790M

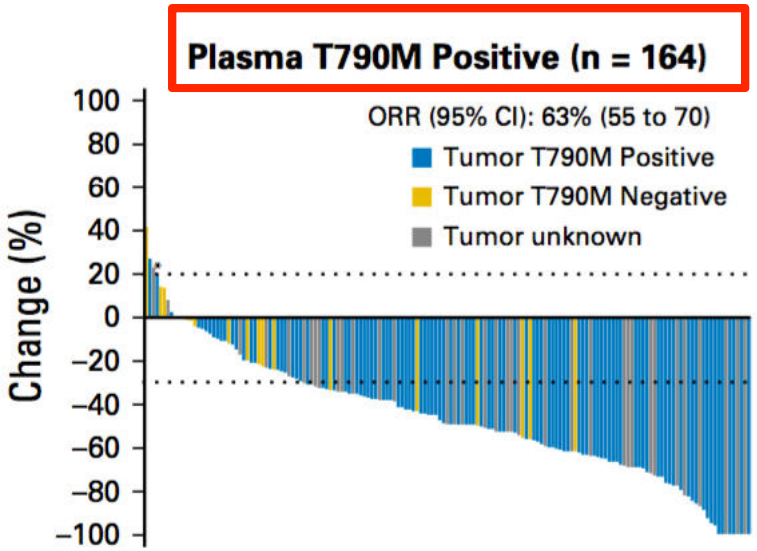
Oxnard et al, JCO 2016

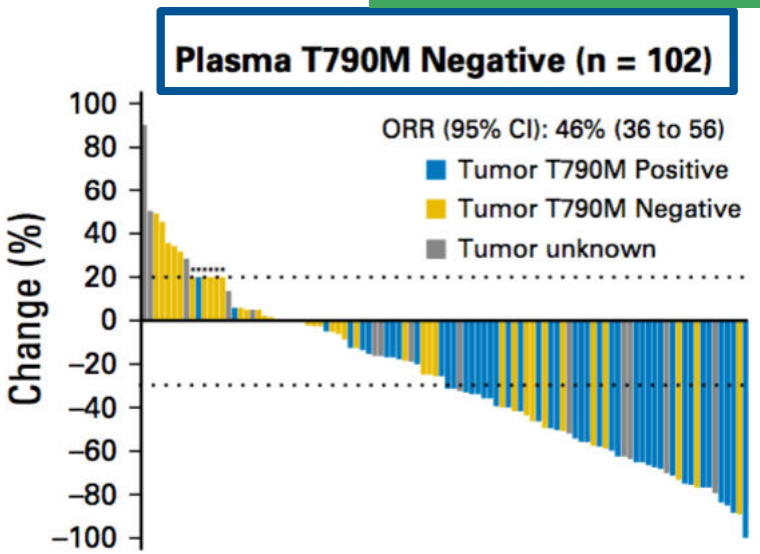
Response rate in T790M+ : tumor = plasma



	ORR (95% CI)	*P*
Tumor T790M Positive	62% (54 to 70)	< .001
Tumor T790M Negative	26% (15 to 39)	

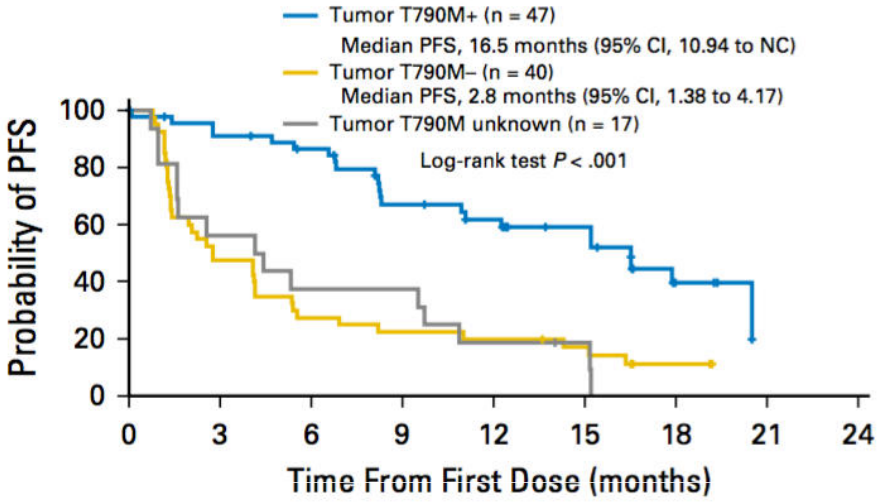
30% false negative results for T790M





	ORR (95% CI)	*P*
Plasma T790M Positive	63% (55 to 70)	.011
Plasma T790M Negative	46% (36 to 56)	

Oxnard et al, JCO 2016

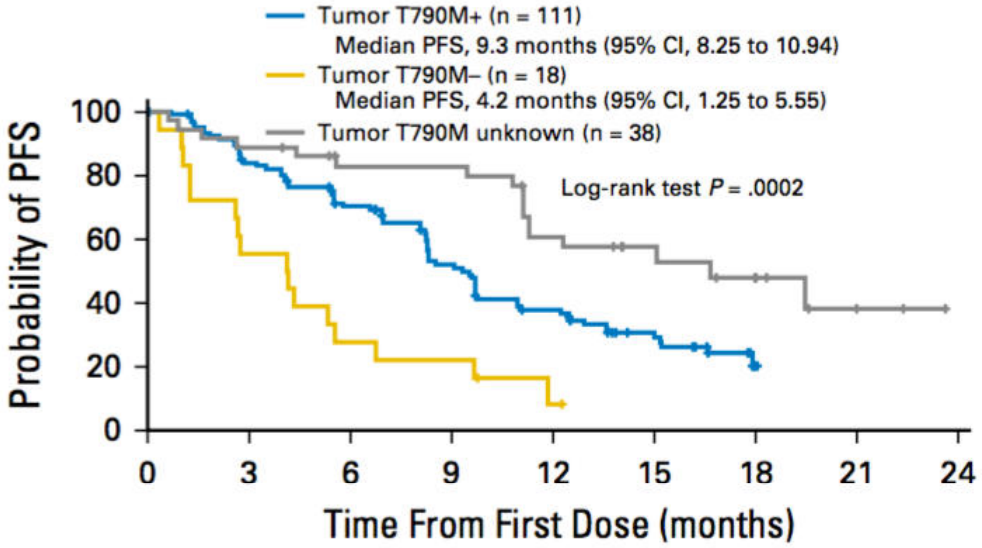


No. at risk

Tumor T790M+	47	41	37	27	23	17	4
Tumor T790M–	40	19	11	9	8	6	2
Tumor unknown	17	9	6	5	3	2	

Plasma T790M-

Plasma T790M+



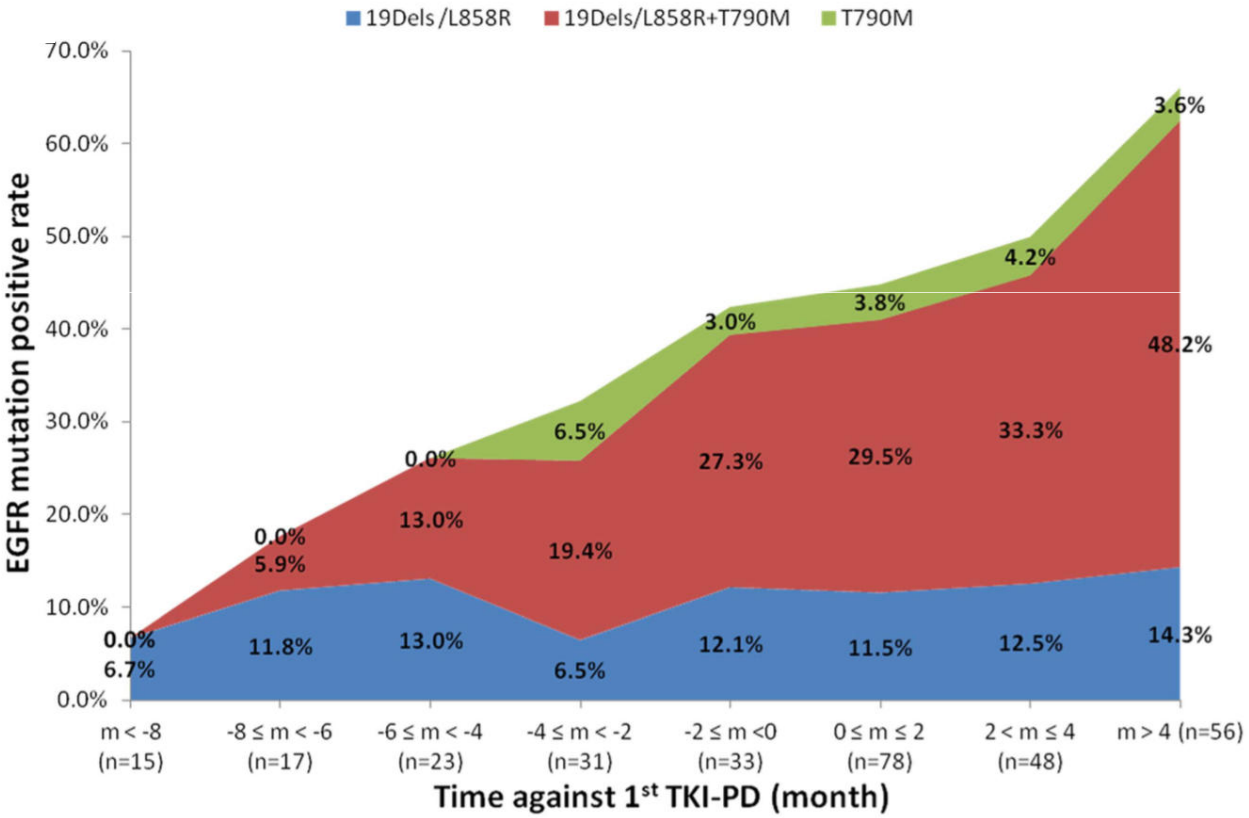
No. at risk

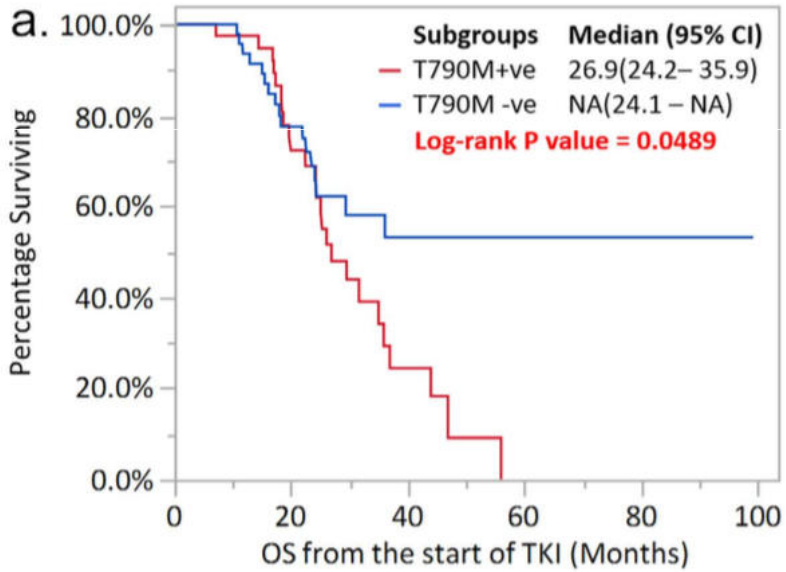
Tumor T790M+	111	88	70	48	33	19	1
Tumor T790M–	18	10	5	4	1		
Tumor unknown	38	32	27	26	19	12	8	2

Tumor positive, plasma negative (***16.5 months***) >
Tumor positive, plasma positive (***9.3 months***)>
Tumor negative, plasma positive (***4.2 months***)

Oxnard et al, JCO 2016

Longitudinal monitoring of EGFR sensitizing and T790M mutations





35 (46%) patients developed EGFR T790M, 16 (45%) of whom earlier than clinical progression (median time 2.2 months, increasing progressively from 6 months prior PD to 4 months beyond PD)

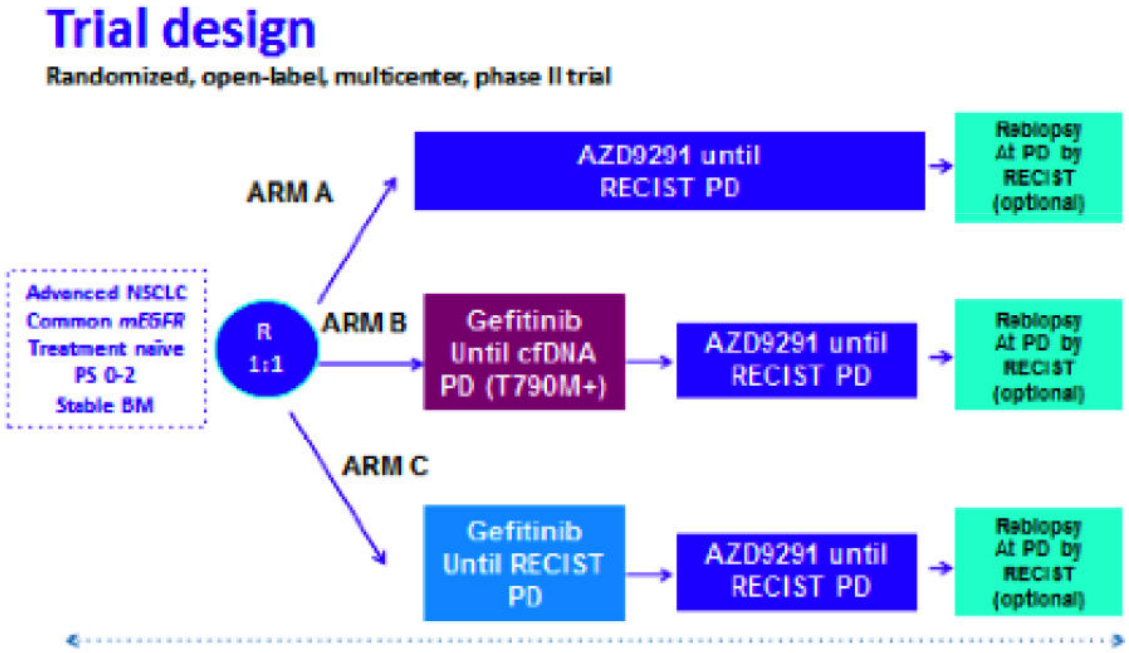
Zheng et al, Scientific Reports 2015

Longitudinal monitoring of EGFR sensitizing and T790M mutations

OPEN QUESTION

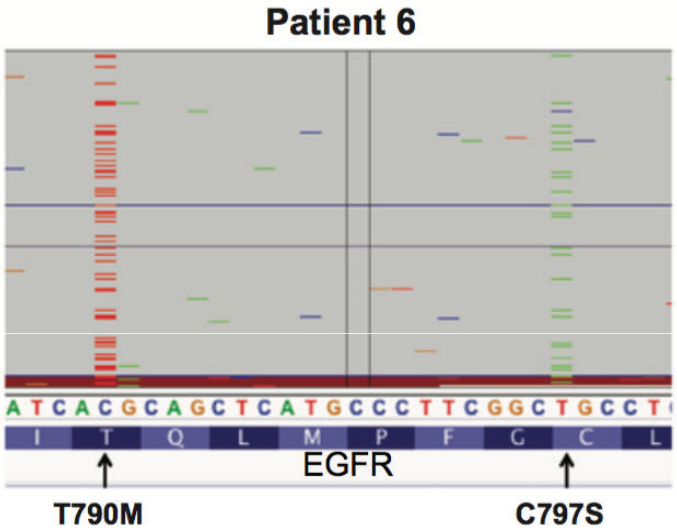
- How many patients develop EGFR T790M earlier than RECIST progression

- What is timing of EGFR T790M development and the clinical significance of early EGFR T790M detection?



Apple trial ongoing

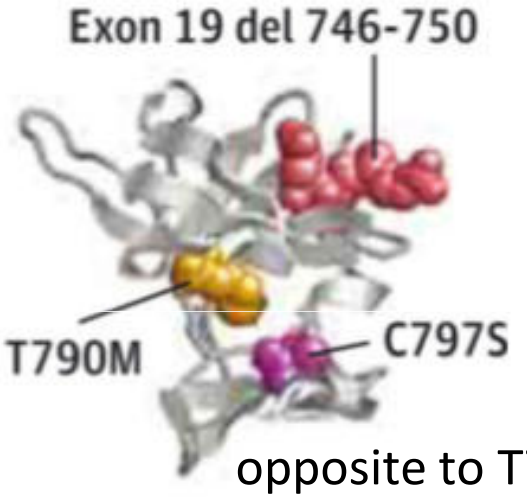
Acquired resistance mechanisms to third generation EGFR-TKIs - C797S



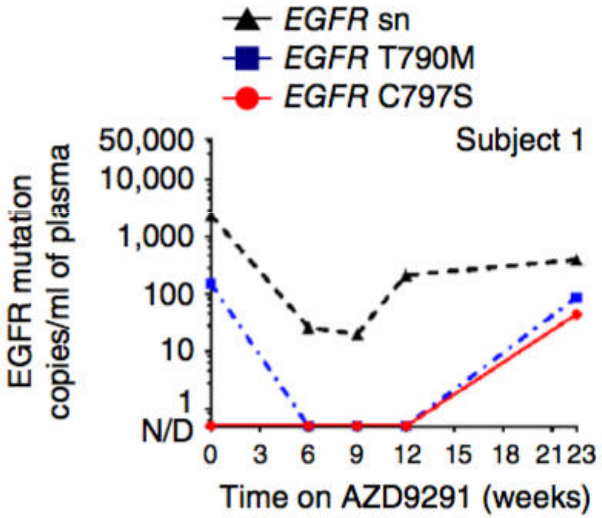
C797S in exon 20

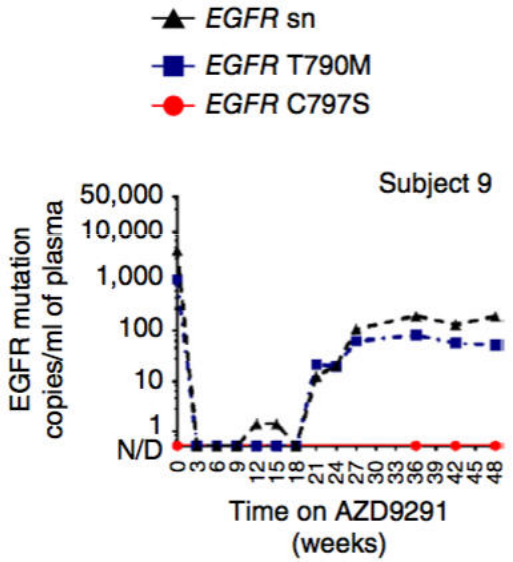
(abrogates the irreversible binding of third generation)

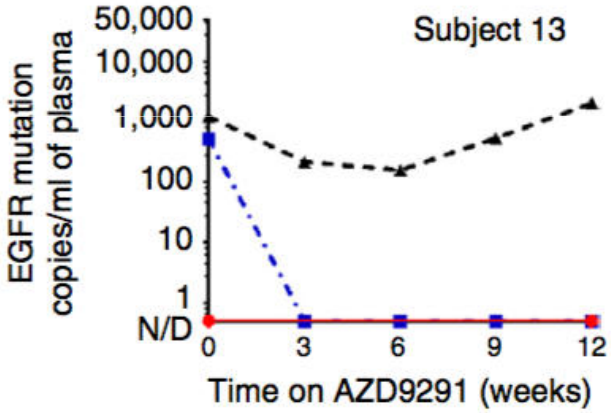
40% of cases



opposite to T790M
in the ATP binding pocket

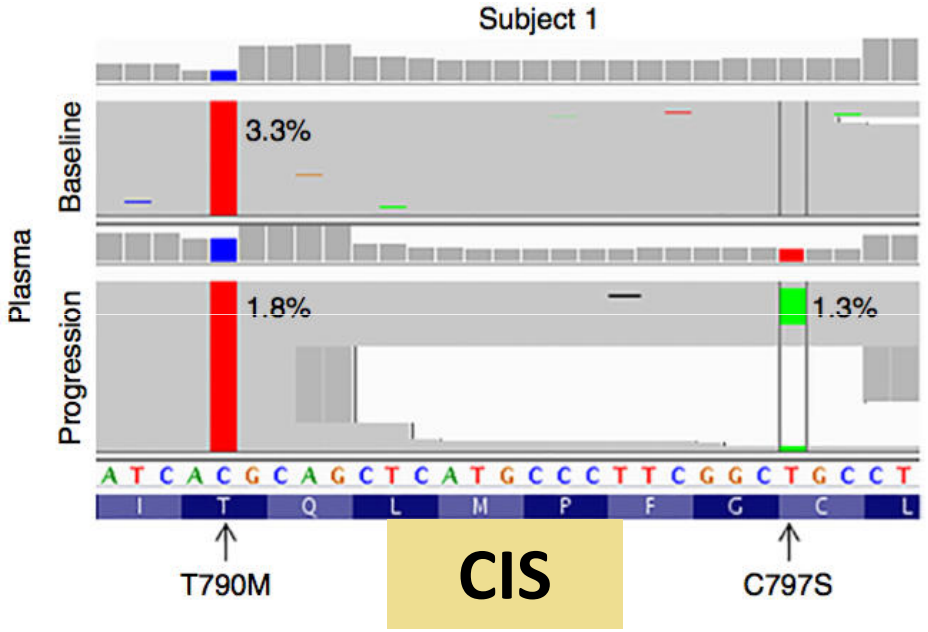






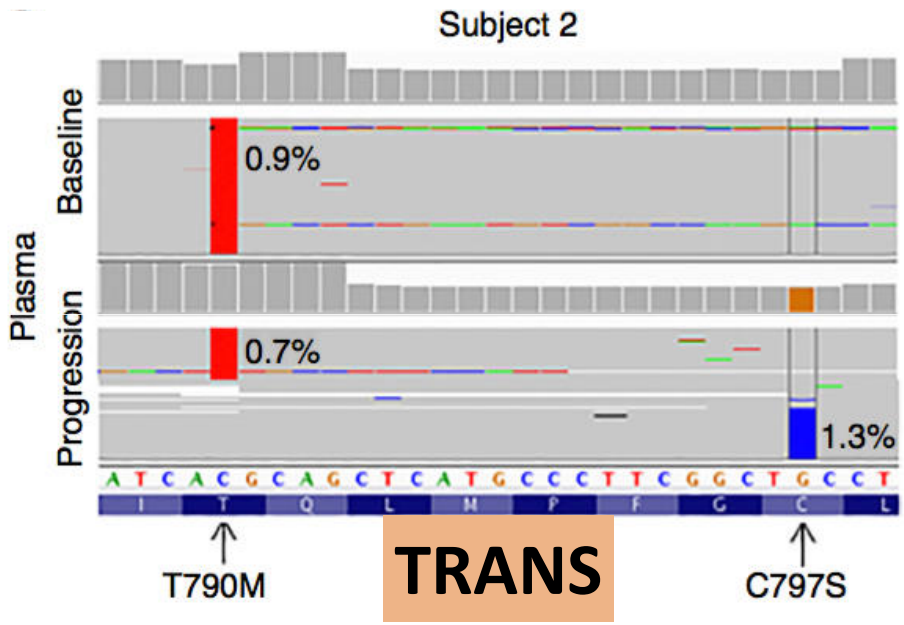
Ortiz-Charan et al; Clin Canc Res 2015. Thress et al; Nature Medicine 2015. Yu HA et al; JAMA 2015

Acquired resistance mechanisms to third generation EGFR-TKIs - C797S



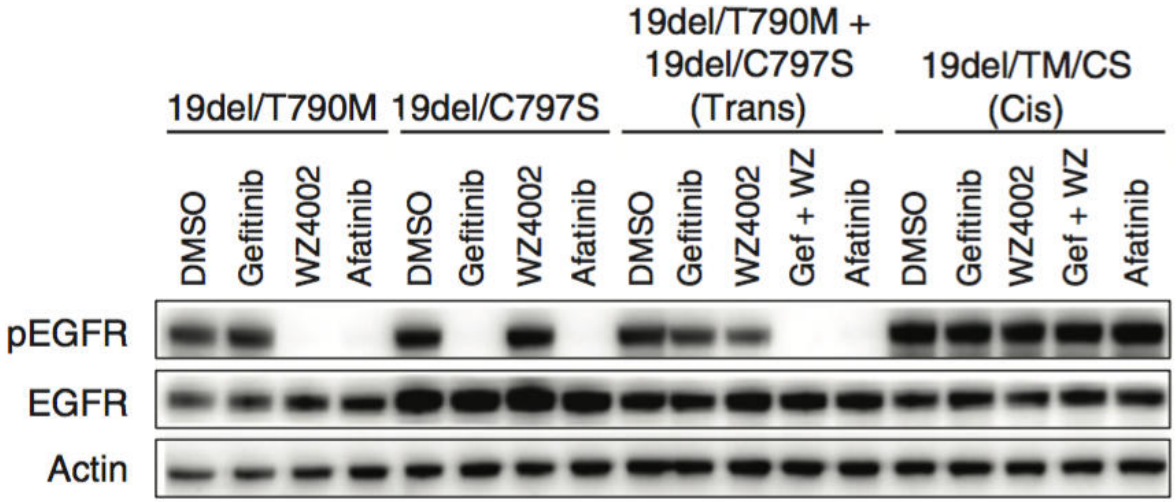
C797S coexists with T790M on the same alleles

Resistance to 3 generations EGFR-TKIs



C797S and T790M are on different alleles

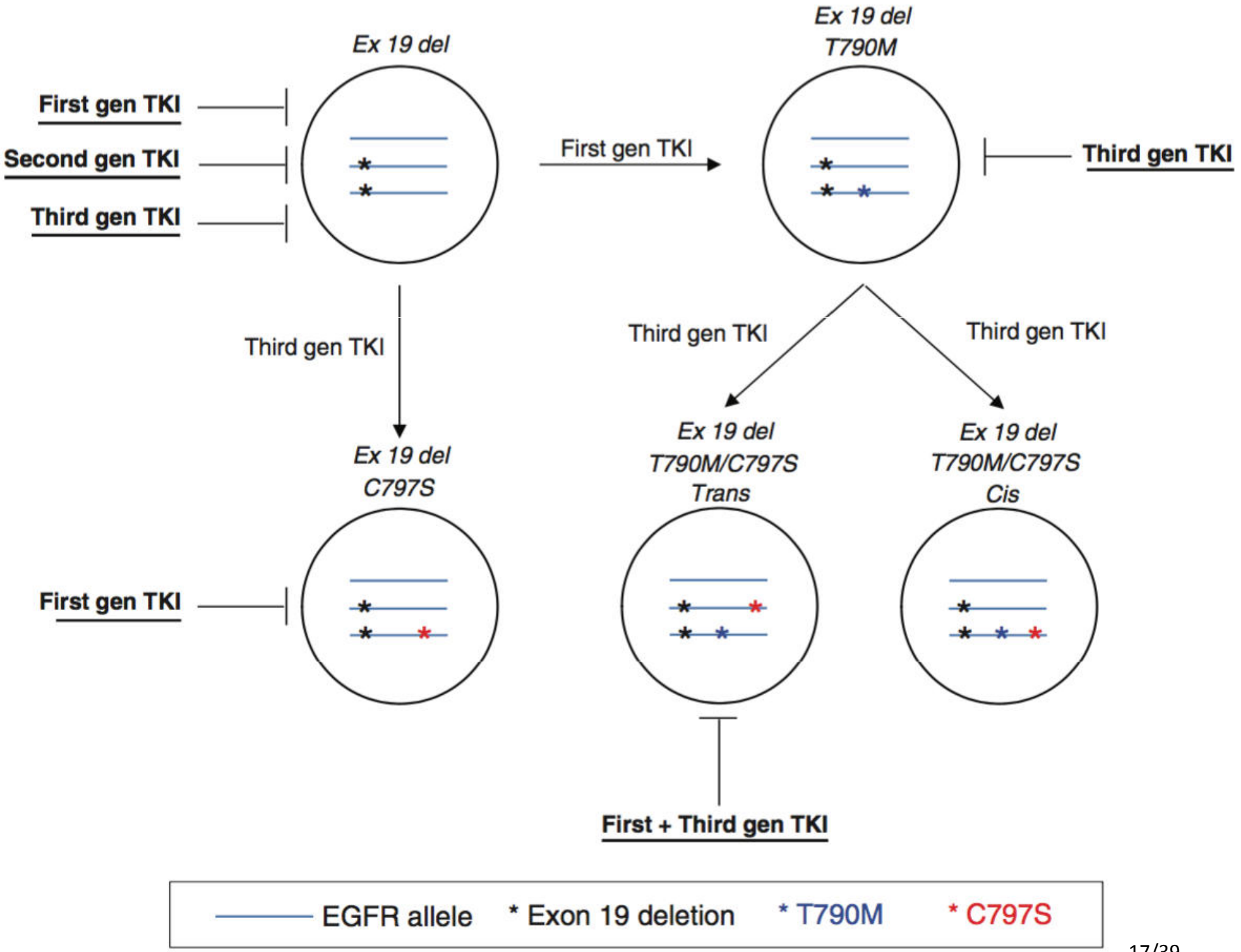
Sensitive to 1st/2nd generations EGFR-TKIs



Third generation require cyst for binding

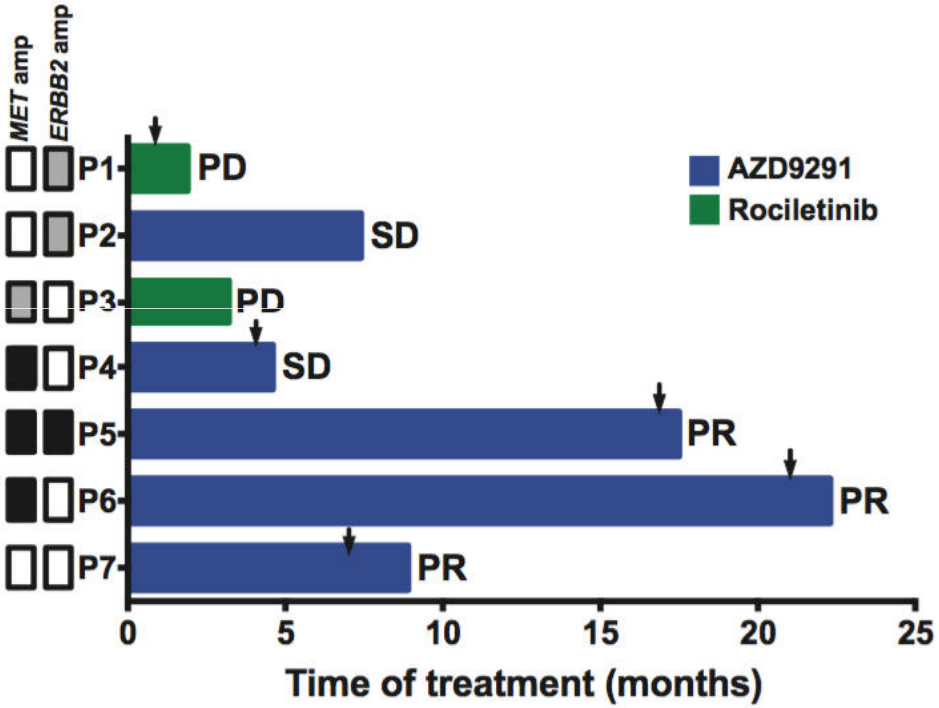
First and second generation do not bind cyst

Niederst et al; Clin Canc Res 2015



Niederst et al; Clin Canc Res 2015

Acquired resistance mechanisms to third generation EGFR-TKIs - MET and ERBB2



At baseline

At progression

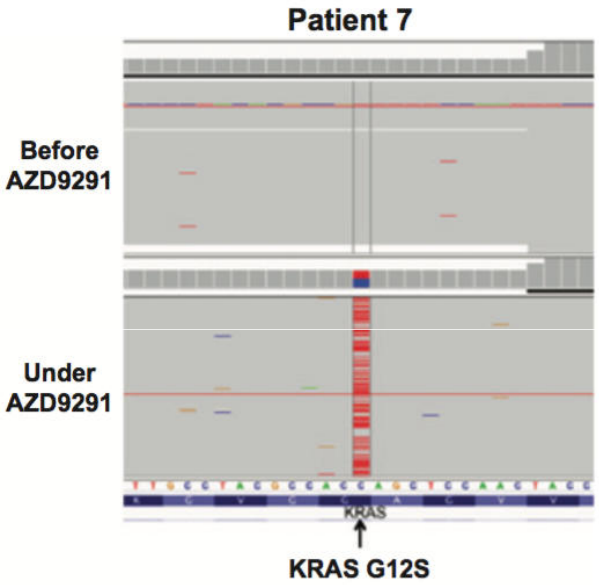
ERBB2 and **MET** amplification decrease sensitivity to third generation EGFR-TKIs

Ongoing studies:

- savolitinib + osimertinib (TATTON trial)

Ortiz-Charan et al; Clin Canc Res 2015

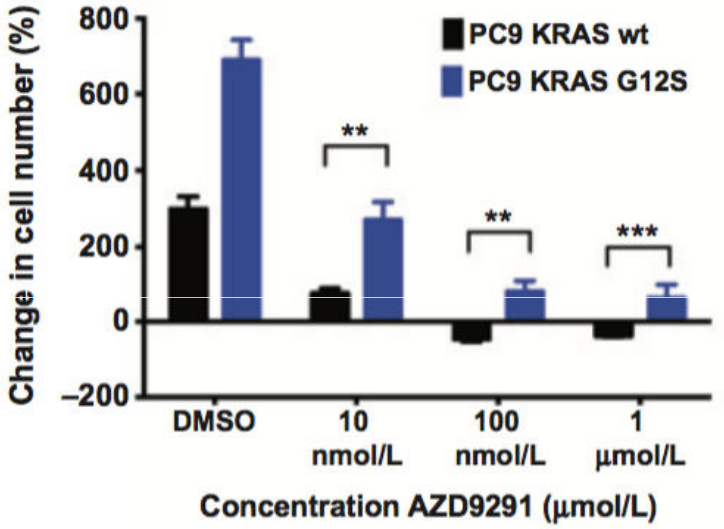
Acquired resistance mechanisms to third generation EGFR-TKIs - KRAS

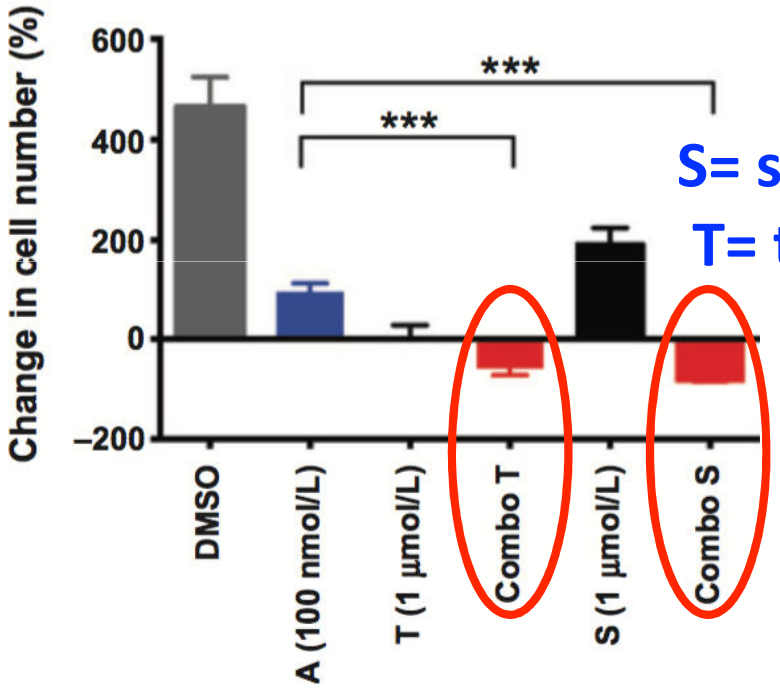


KRAS G12S in tissue

T790M - in tissue

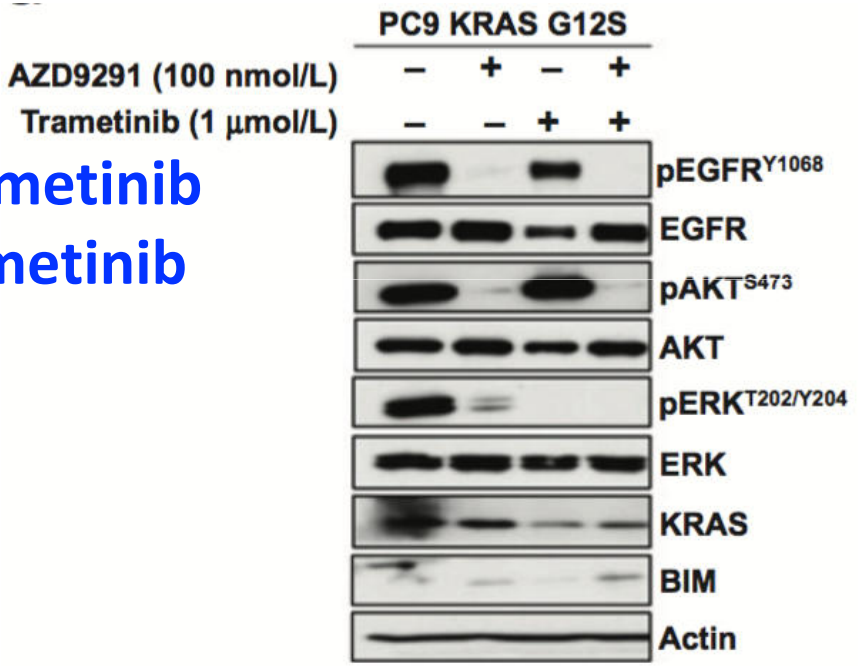
C797S + in blood





S= selumetinib

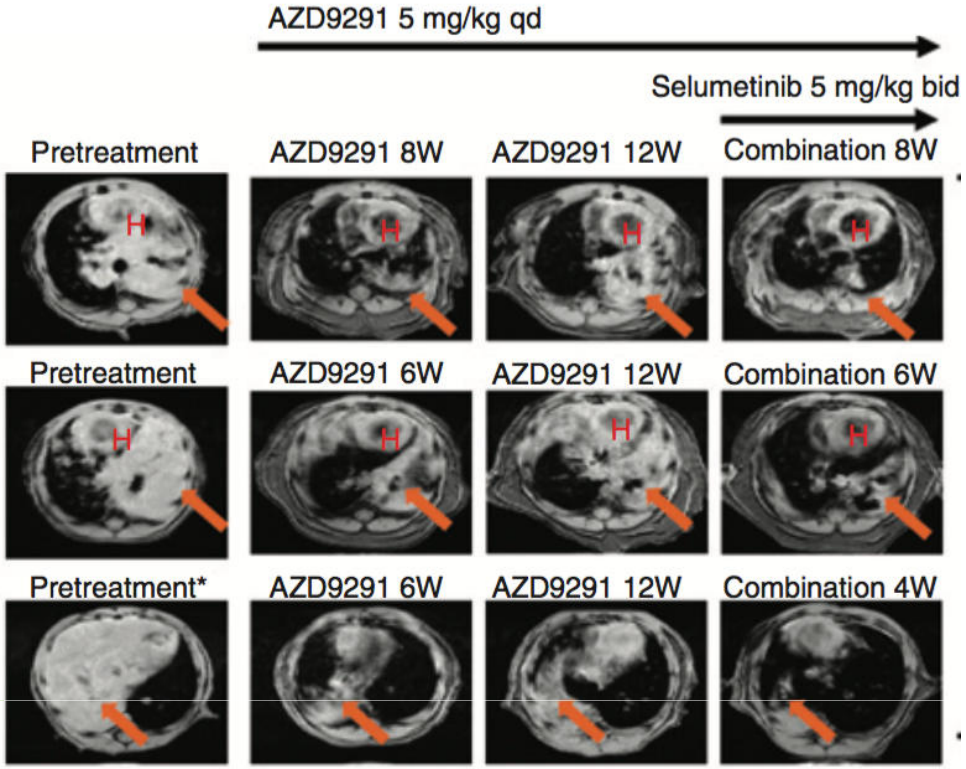
T= trametinib



Ortiz-Charan et al; Clin Canc Res 2015. Thress et al; Nature Medicine 2015

Acquired resistance mechanisms to third generation EGFR-TKIs - KRAS

Cell population	Genetic alterations detected within resistant populations	Selumetinib (MEK1/2)
PC9		6.95 (±2.5)
PC9 GR_1	EGFR T790M/KRAS gain (5.43-fold)	7.24 (±3.2)
PC9 GR_2	NRAS E63K	0.62 (±0.3)
PC9 GR_3	EGFR T790M	6.2 (±3.6)
PC9 GR_4	EGFR T790M	6.2 (±3.6)
PC9 GR_5	EGFR T790M	7.32 (±2.3)
PC9 GR_6	EGFR T790M	8.77 (±1.5)
PC9 GR_7	EGFR T790M	7.44 (±2.6)
PC9 GR_8	EGFR T790M/KRAS gain (7.06-fold)	3.7 (±0.99)
PC9 AR_1	KRAS gain (24.6-fold)	2.7 (±0.23)
PC9 AR_4	EGFR T790M	1.63 (±1.1)
PC9 AR_6	NRAS gain (4.23-fold)	0.89 (±0.6)
PC9 WZR_1	NRAS Q61K	0.23 (±0.04)
PC9 WZR_3	KRAS gain (2.64-fold)	0.22 (±0.1)
PC9 AZDR_1	NRAS gain (2.5-fold)/MAPK1 gain/CRKL gain	0.25 (±0.06)
PC9 AZDR_2	NRAS G12V	1.4 (±0.9)
PC9 AZDR_3	MAPK1 gain/CRKL gain	2.38 (±0.9)
PC9 AZDR_4	ND	0.19 (±0.1)
PC9 AZDR_5	NRAS E63K	0.17 (±0.05)
PC9 AZDR_6	NRAS E63K	0.11 (±0.03)
PC9 AZDR_7	NRAS G12R	0.14 (±0.03)
PC9 GR_1_AZDR_1	EGFR T790M/KRAS gain (6.23-fold)	3.6 (±0.7)
PC9 GR_1_AZDR_2	KRAS gain (5.66-fold)	6.7 (±1.4)
PC9 GR_1_AZDR_3	EGFR T790M/KRAS gain (4.44-fold)	3.4 (±0.5)
PC9 GR_1_AZDR_4	EGFR T790M/KRAS gain (5.46-fold)	3.6 (±2.6)
PC9 GR_6_AZDR_1	ND	0.28 (±0.2)
PC9 GR_6_AZDR_2	NRAS gain (2.4-fold)	0.54 (±0.3)
PC9 GR_6_AZDR_3	NRAS gain (3.68-fold)	0.13 (±0.06)
PC9 GR_6_AZDR_4	ND	0.73 (±0.5)
NCI-H1975	EGFR T790M	4.94 (±3)
NCI-H1975 AZDR_1	EGFR T790M	0.024 (±0.003)
NCI-H1975 AZDR_2	EGFR T790M	0.15 (±0.1)
NCI-H1975 AZDR_3	EGFR T790M	>10
NCI-H1975 AZDR_4	EGFR T790M/NRAS Q61K	5.46 (±3.7)



Ongoing studies:

- selumetinib + osimertinib (TATTON trial)

Eberlein et al; Canc Res 2015. Tricker et al; Cancer Discovery 2015

OSIMERTINIB + DURVALUMAB (Tatton trial)

Part A

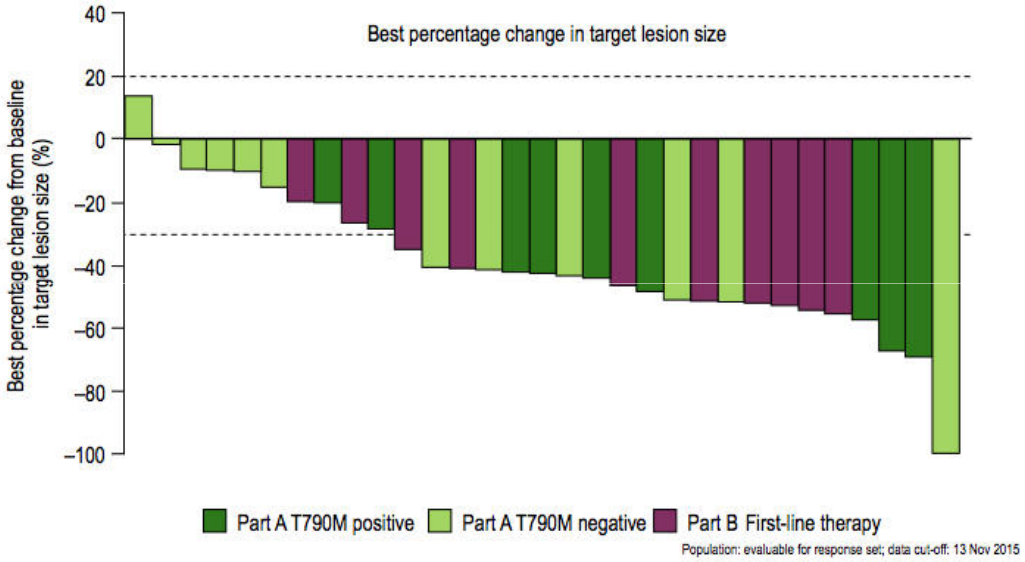
- EGFR mutated pre treated NSCLC patients
- No contraindication to immunotherapy
- No history of ILD

Dose escalation

Part B

- EGFR mutated naive NSCLC patients
- No contraindication to immunotherapy
- No history of ILD

Dose expansion



Increased percentage of ILD

Part A	6/23 (26%)
Dose 1: Osimertinib 80 mg QD / durvalumab 3 mg/kg Q2W	2/10 (20%)
Dose 2: Osimertinib 80 mg QD / durvalumab 10 mg/kg Q2W	4/13 (31%)
Part B: Osimertinib 80 mg QD / durvalumab 10 mg/kg Q2W	7*/11 (64%)
Part A and Part B	13/34 (38%; 95% CI 18, 52)†

†5 events were Grade 3/4 and there were no fatalities; most cases were managed using steroids

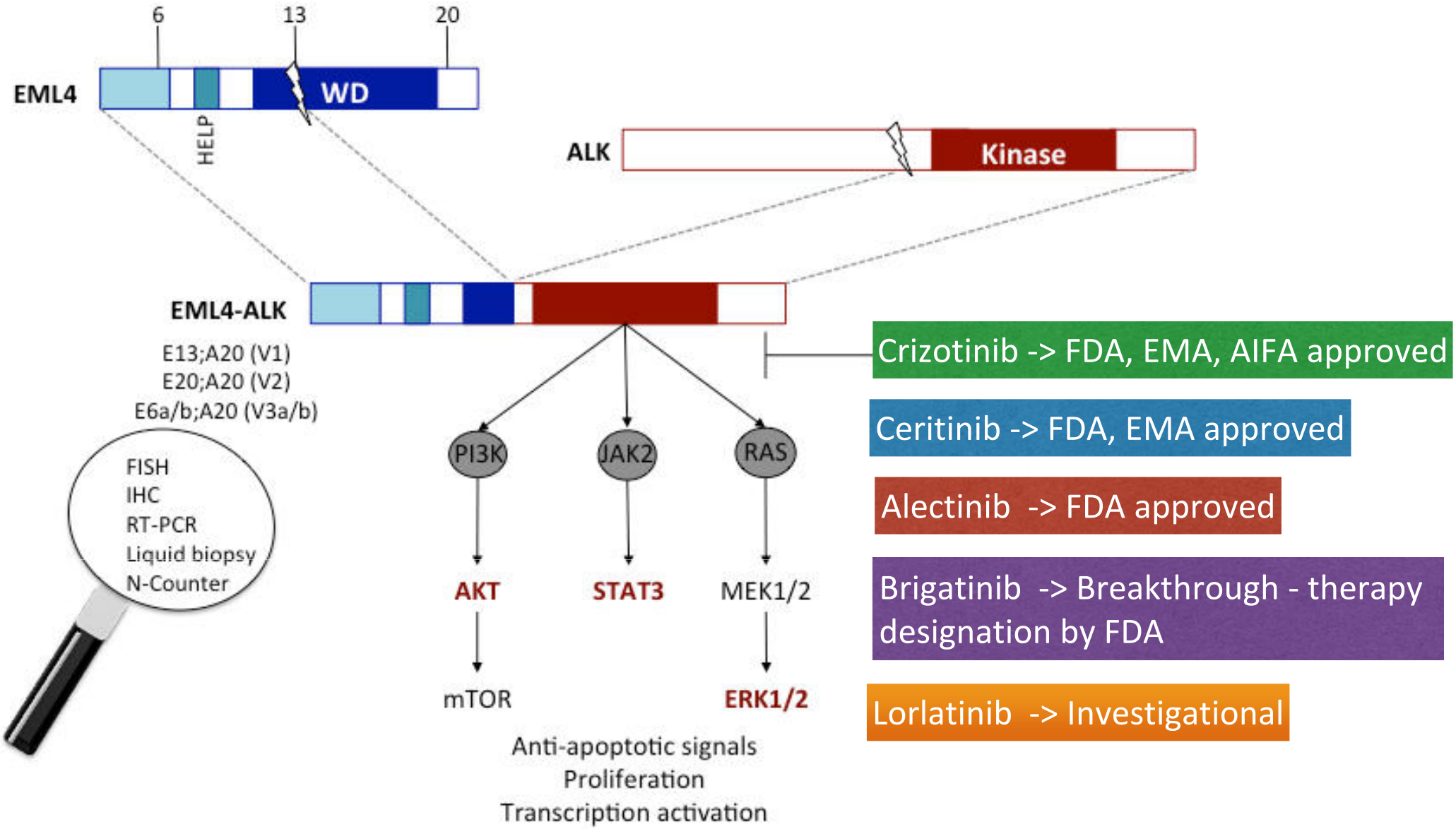
Entire osimertinib clinical programme (Phase I and II)	
Osimertinib monotherapy	35/1207 (3%)
Durvalumab monotherapy	23/1149 (2%)

23 patients in PART A (12 PR, 9 SD)

11 patients in PART B (8 PR, 2 SD)

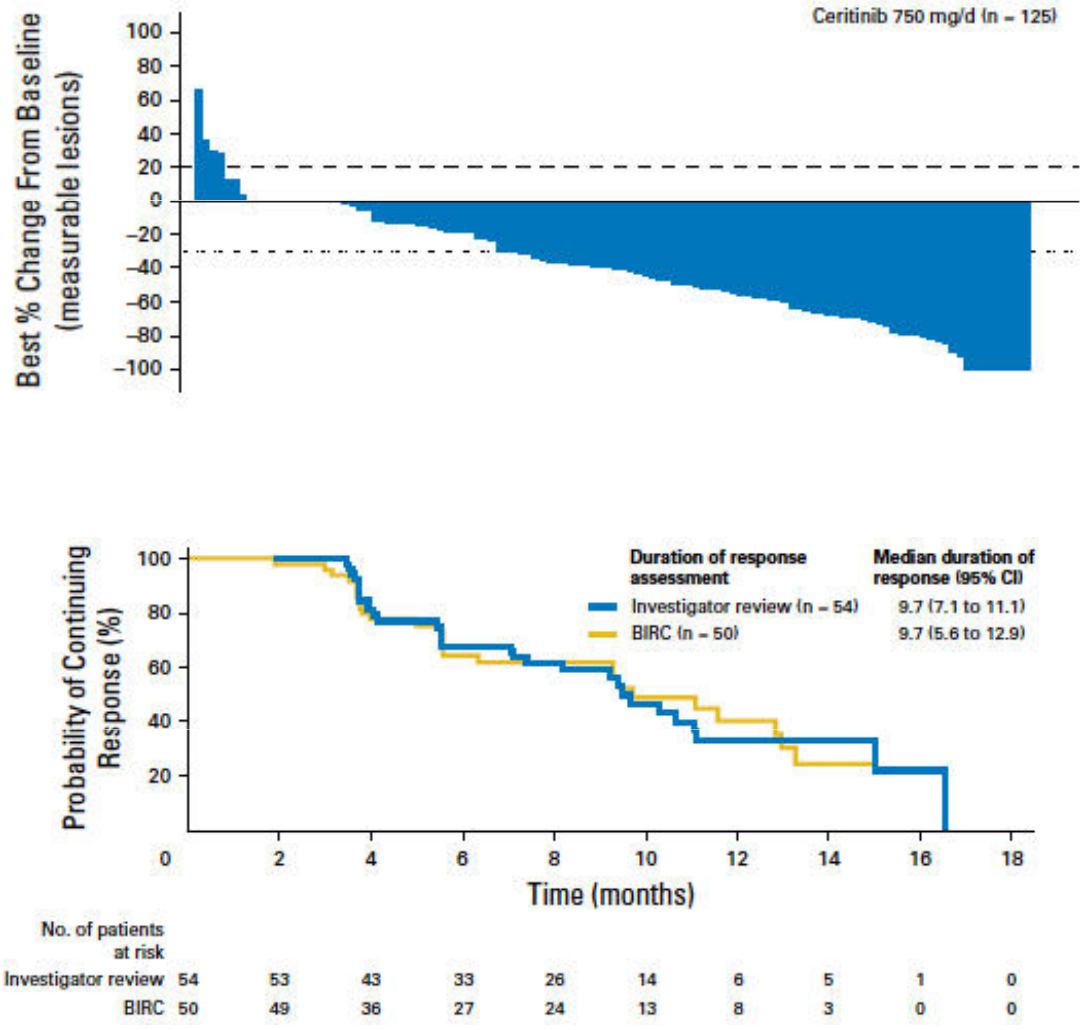
Ahn et al; ELCC 2016

EML4-ALK traslocated NSCLC

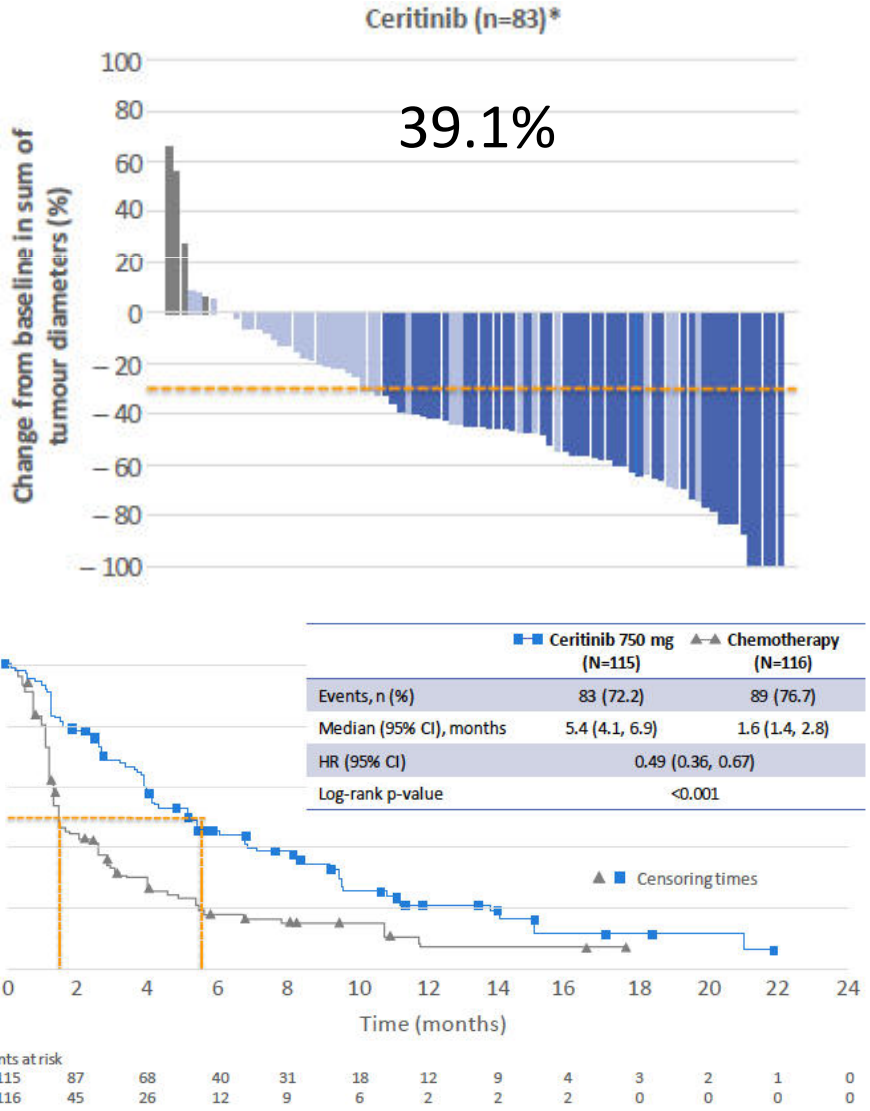


Ceritinib - crizotinib pretreated

ASCEND-2-Phase II



ASCEND-5- Phase III

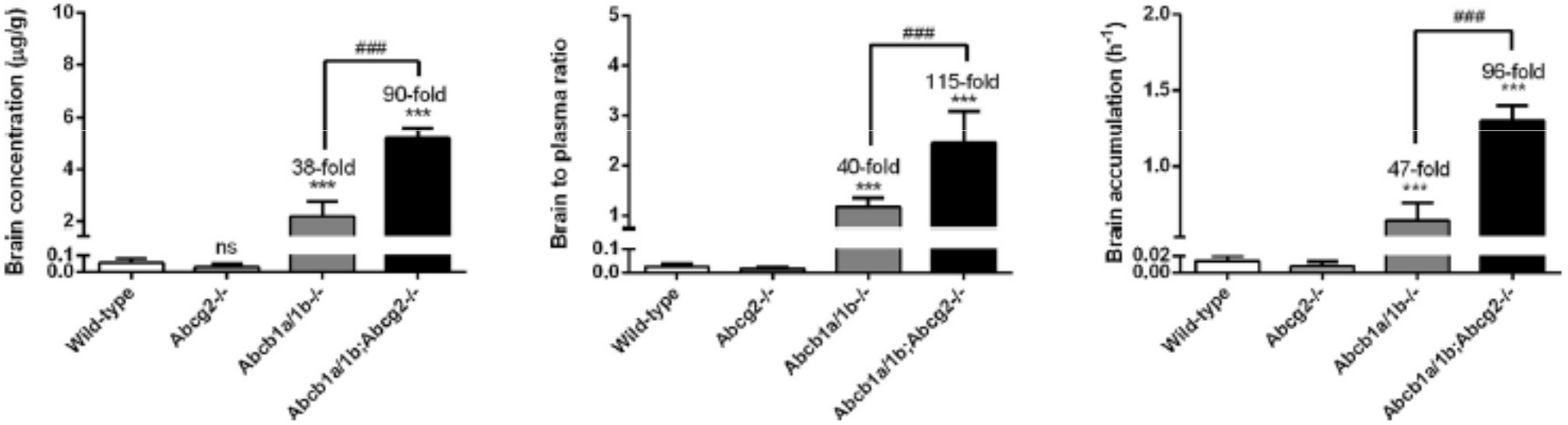


Crinò et al; JCO; 2016

Scagliotti et al; ESMO; 2016

Ceritinib and the blood brain barrier

Ceritinib is a good substrate of hABCB1 and hABCG2 at the level of the blood brain barrier in mice



The expression of hABCG2 relative to hABCB1 in the human BBB is 4.3-fold higher than the expression ratio of mAbcg2 and mAbcb1a in the mouse

The lipophilicity of ceritinib maybe allows the molecule to diffuse through the BBB at a significant rate.

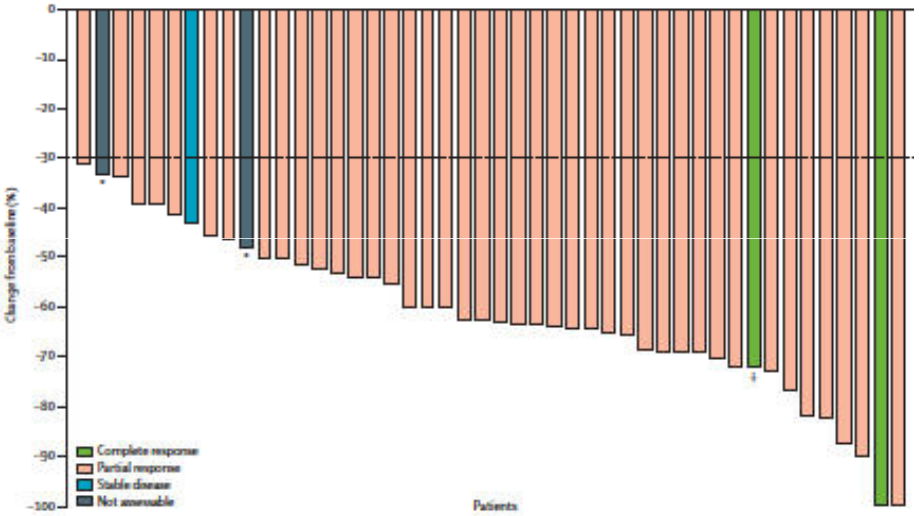
Objective intracranial responses in 45.0% (95%CI, 23.1% - 68.5%) ASCEND-2

Kort et al; Pharmacological Rese; 2015

Alectinib

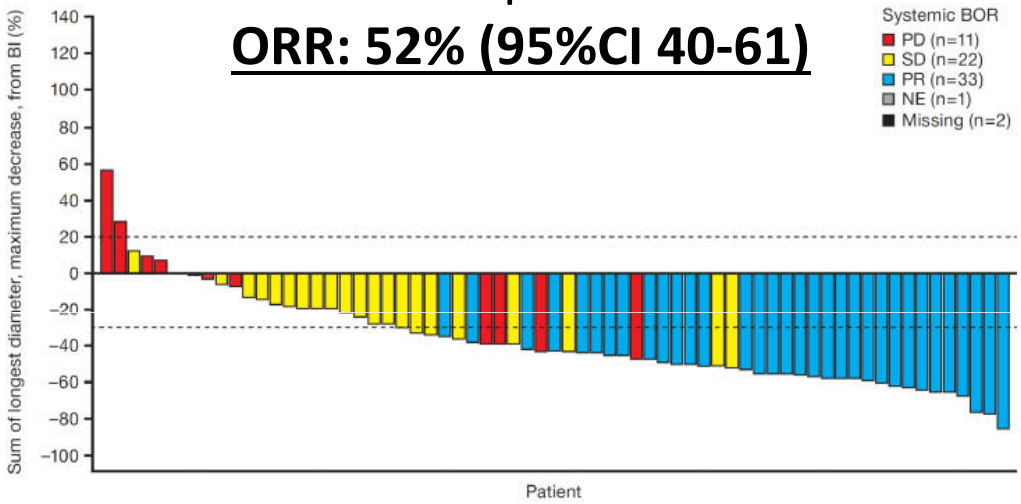
Crizotinib naive

ORR: 93.5% (95%CI 82-98)

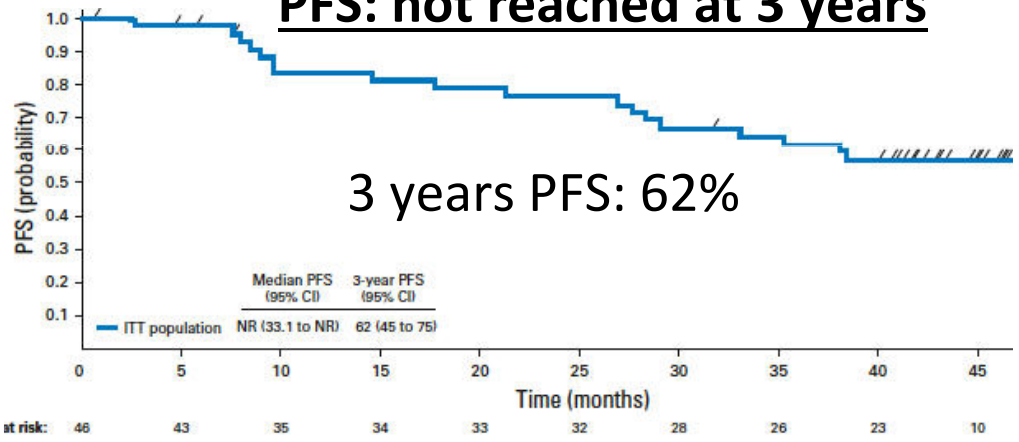


Crizotinib pre-treated

ORR: 52% (95%CI 40-61)



PFS: not reached at 3 years

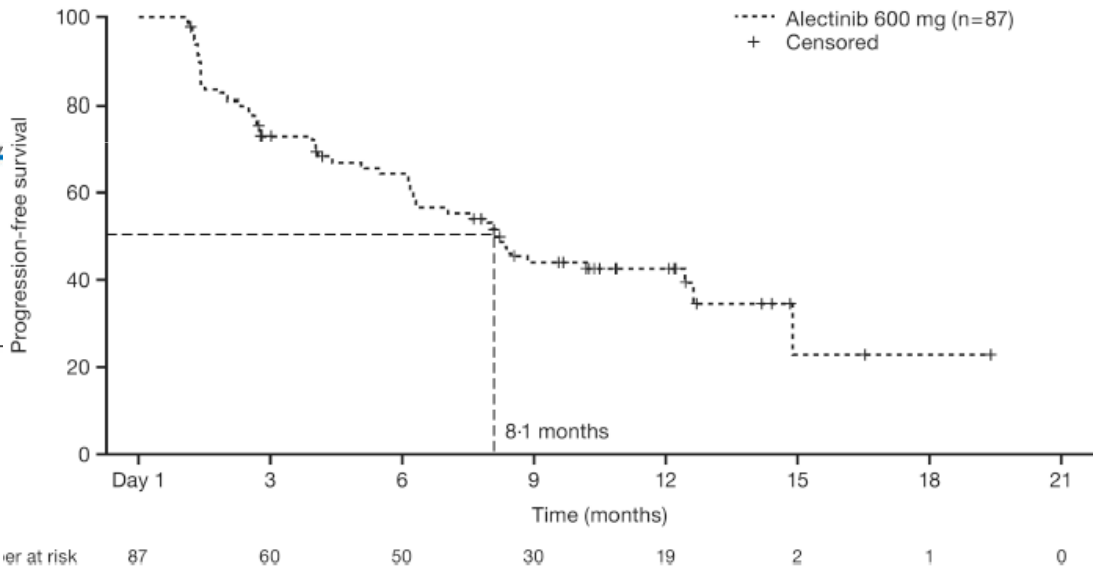


3 years PFS: 62%

Seto et al; Lancet Oncol; 2013

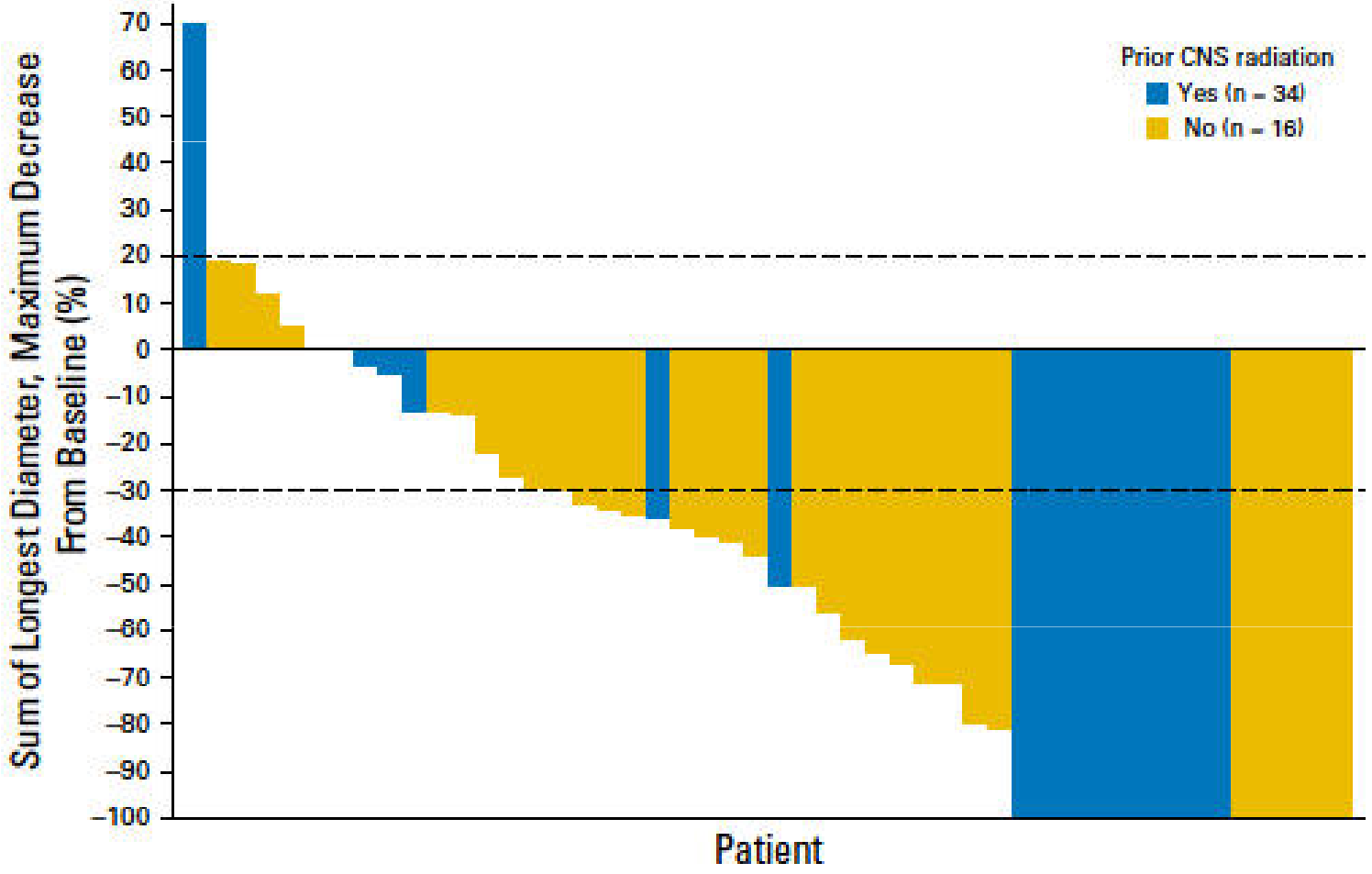
Tamara et al; JCO; 2017

PFS: 8.1 m (95% CI 6.2-12.6)



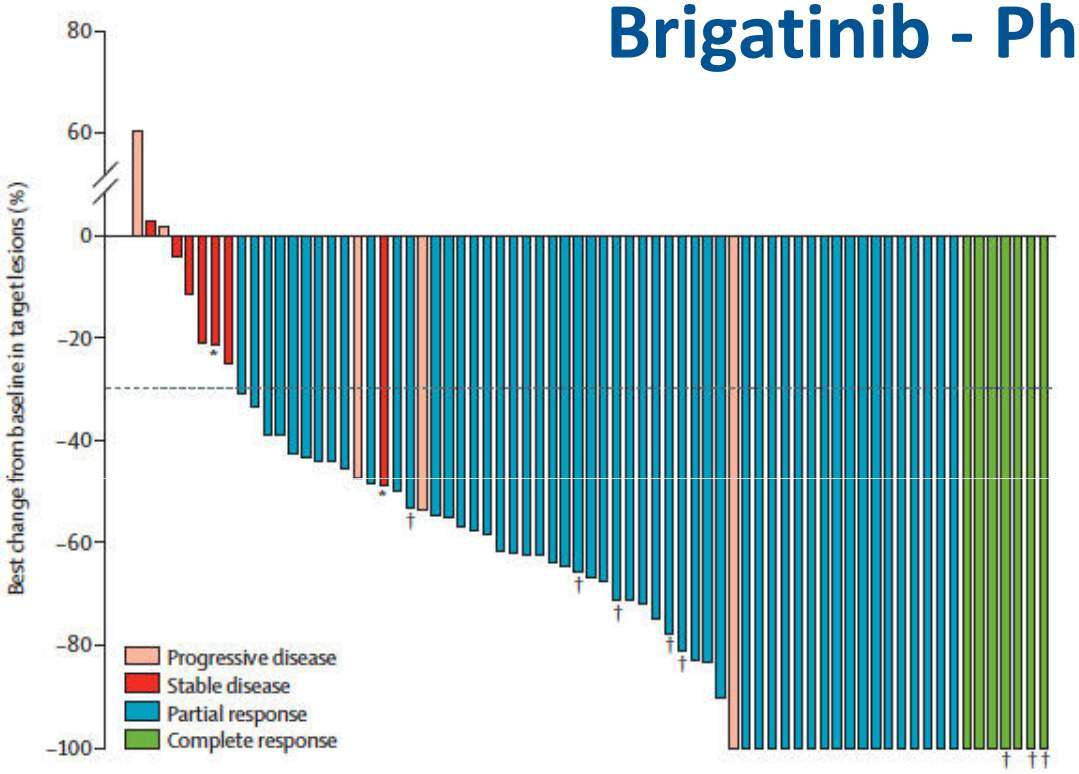
Shaw et al; Lancet Oncol; 2016

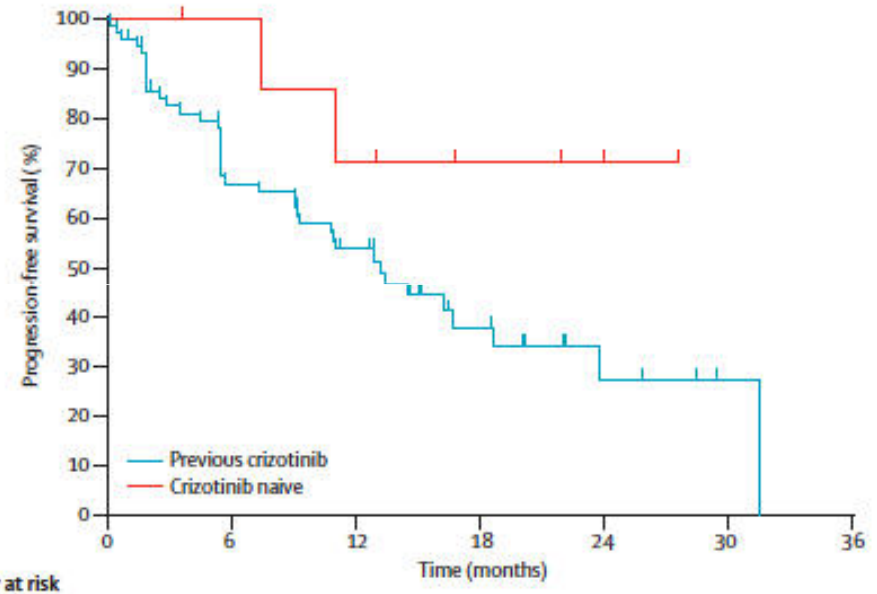
Alectinib activity on brain lesions - Pooled analysis of phase II NP28761 and NP28673 studies



Gadgeel et al; JCO; 2016

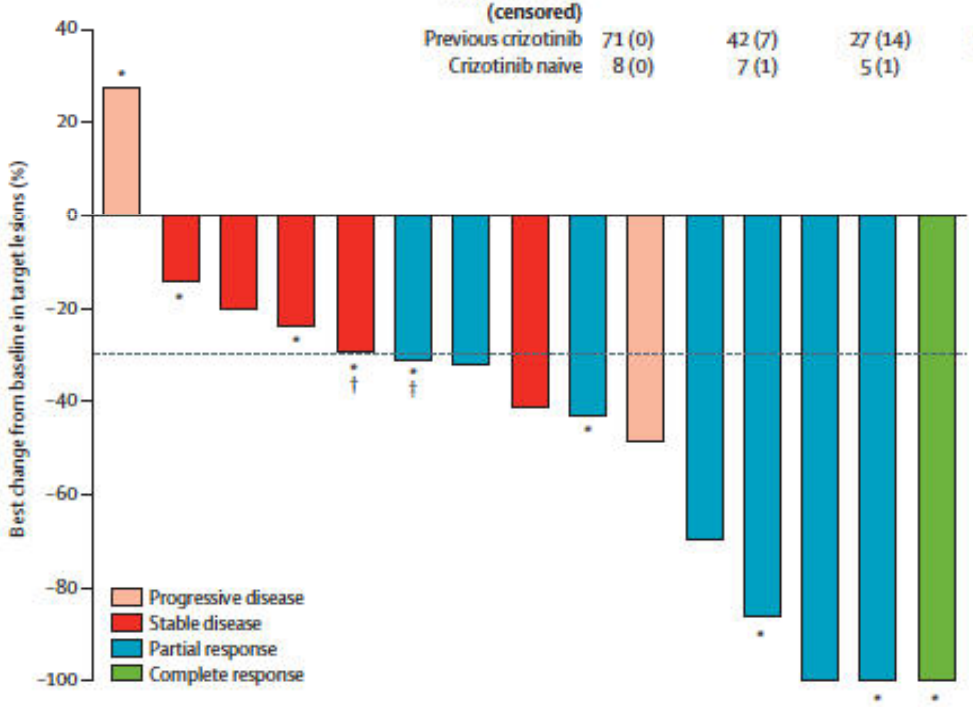
Brigatinib - Phase I/II trial





Number at risk (censored)							
Previous crizotinib	71 (0)	42 (7)	27 (14)	11 (24)	4 (29)	1 (32)	0 (32)
Crizotinib naive	8 (0)	7 (1)	5 (1)	3 (3)	1 (5)	0 (6)	0 (6)

Pulmonary toxicity

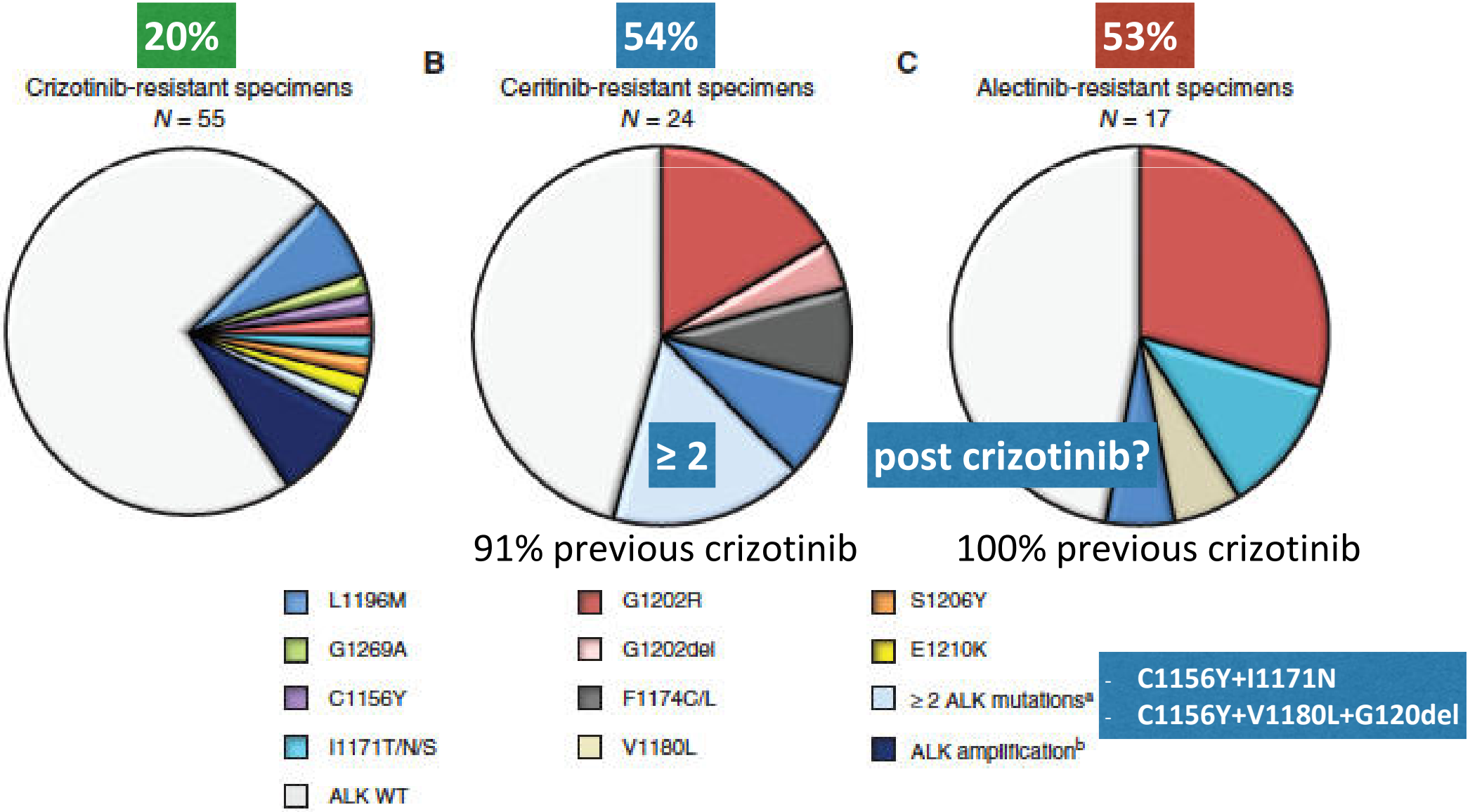


CNS activity:

RR 50%

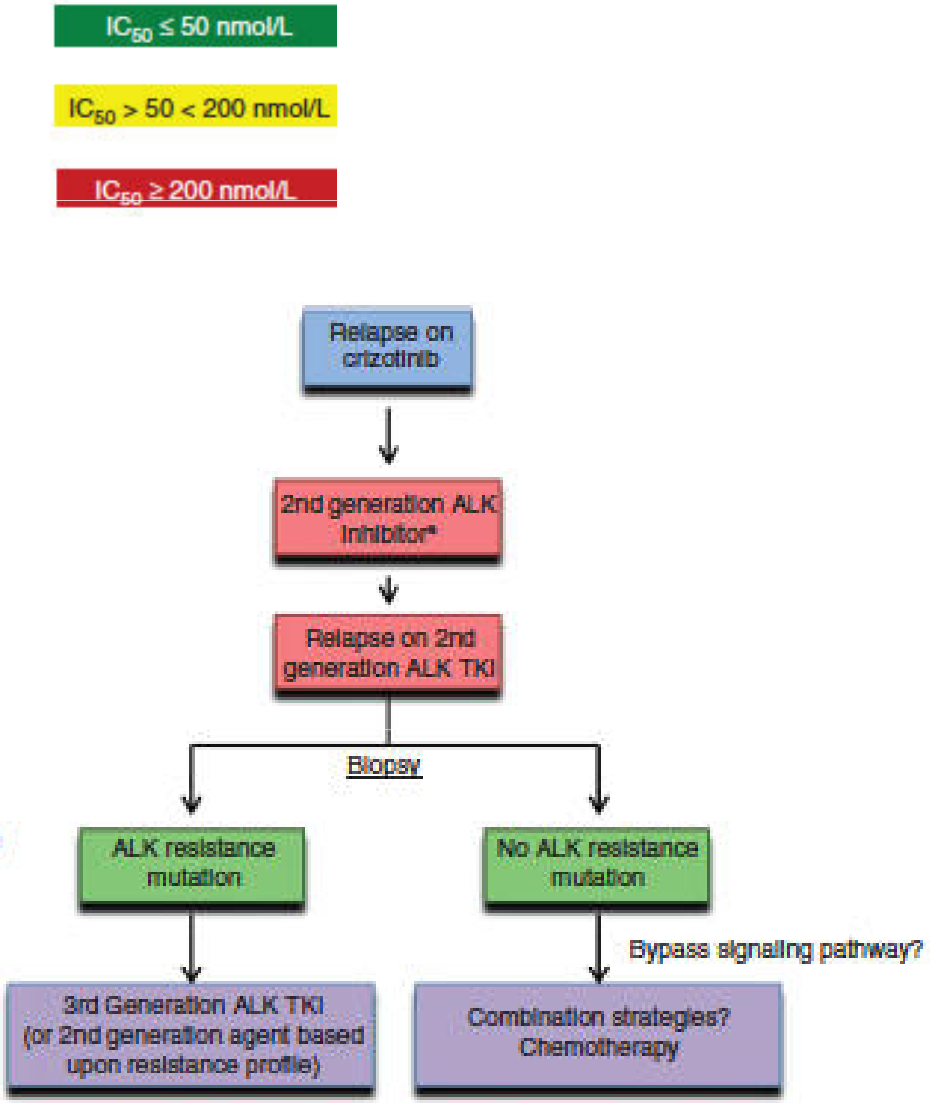
Gettinger et al; Lancet Oncol; 2016

Acquired resistance mechanisms to ALK inhibitors - ALK secondary mutations



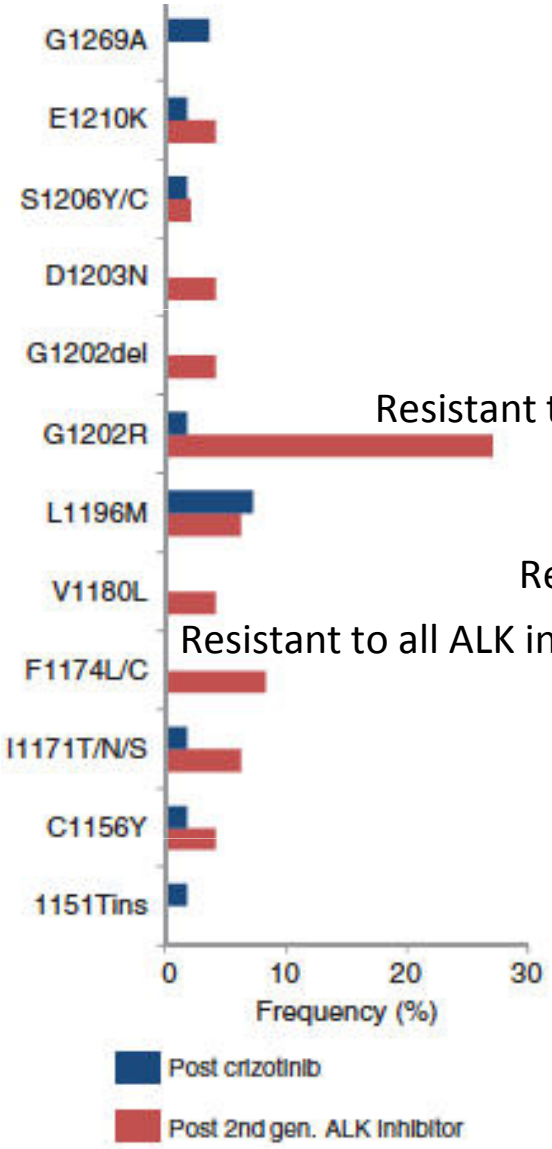
Gainor et al; Cancer Discovery; 2016

Mutation status	Crizotinib	Ceritinib	Alectinib	Brigatinib	Lorlatinib
	Cellular ALK phosphorylation mean IC_{50} (nmol/L)				
Parental Ba/F3	763.9	885.7	890.1	2774.0	11293.8
EML4–ALK V1	38.6	4.9	11.4	10.7	2.3
EML4–ALK C1156Y	61.9	5.3	11.6	4.5	4.6
EML4–ALK I1171N	130.1	8.2	397.7	26.1	49.0
EML4–ALK I1171S	94.1	3.8	177.0	17.8	30.4
EML4–ALK I1171T	51.4	1.7	33.6[a]	6.1	11.5
EML4–ALK F1174C	115.0	38.0[a]	27.0	18.0	8.0
EML4–ALK L1196M	339.0	9.3	117.6	26.5	34.0
EML4–ALK L1198F	0.4	196.2	42.3	13.9	14.8
EML4–ALK G1202R	381.6	124.4	706.6	129.5	49.9
EML4–ALK G1202del	58.4	50.1	58.8	95.8	5.2
EML4–ALK D1203N	116.3	35.3	27.9	34.6	11.1
EML4–ALK E1210K	42.8	5.8	31.6	24.0	1.7
EML4–ALK G1269A	117.0	0.4	25.0	ND	10.0
EML4–ALK D1203N+F1174C	338.8	237.8	75.1	123.4	69.8
EML4–ALK D1203N+E1210K	153.0	97.8	82.8	136.0	26.6



Gainor et al; Cancer Discovery; 2016

Significantly higher frequency of ALK secondary mutations with ALK second generation inhibitors



Resistant to 1st and 2nd generation

Resistant to 1st generation

Resistant to all ALK inhibitors, except lorlatinib

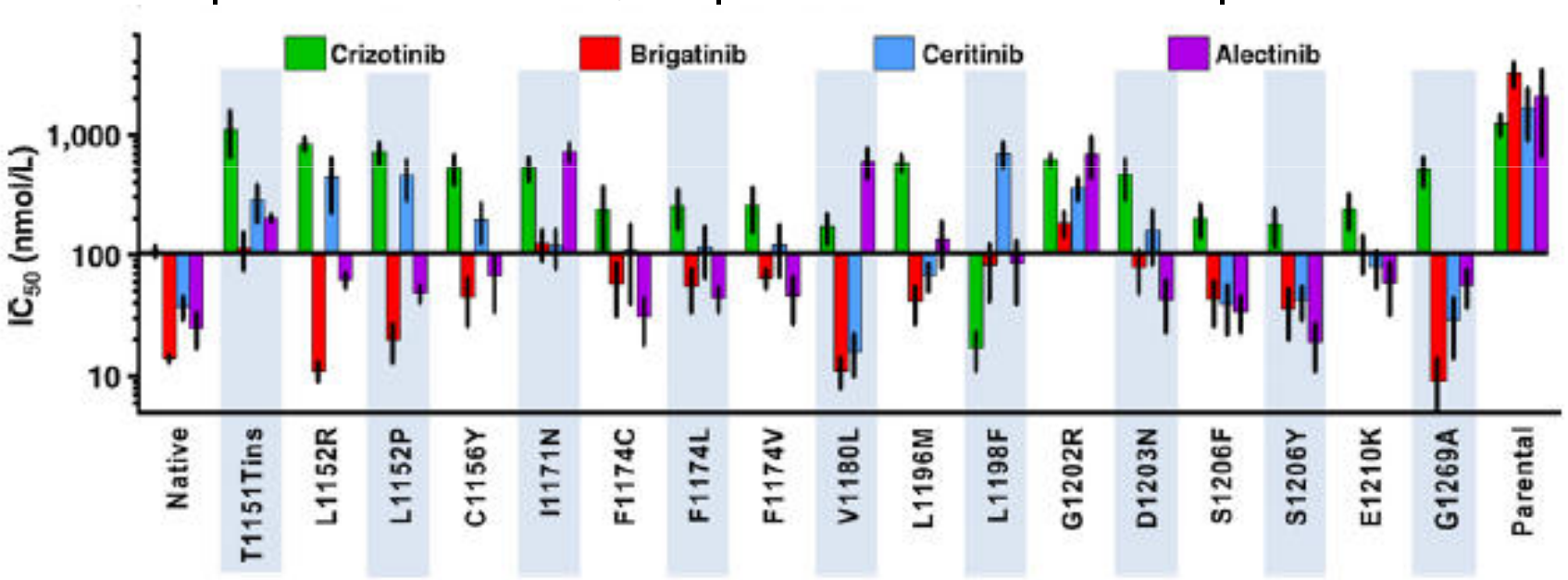
After crizotinib and alectinib

ALK resistance mutations[a]	Crizotinib (*N* = 55)	Ceritinib (*N* = 24)	Alectinib (*N* = 17)	Brigatinib (*N* = 7)
1151Tins	2%	0%	0%	0%
C1156Y	2%	8%	0%	0%
I1171T/N/S	2%	4%	12%	0%
F1174L/C	0%	17%	0%	0%
V1180L	0%	4%	6%	0%
L1196M	7%	8%	6%	0%
G1202R	2%	21%	29%	43%
G1202del	0%	8%	0%	0%
D1203N	0%	4%	0%	14%
S1206Y/C	2%	0%	0%	14%
E1210K	2%	0%	0%	29%
G1269A	4%	0%	0%	0%
ALK mutations[b]	**20%**	**54%**	**53%**	**71%**

Gainor et al; Cancer Discovery; 2016

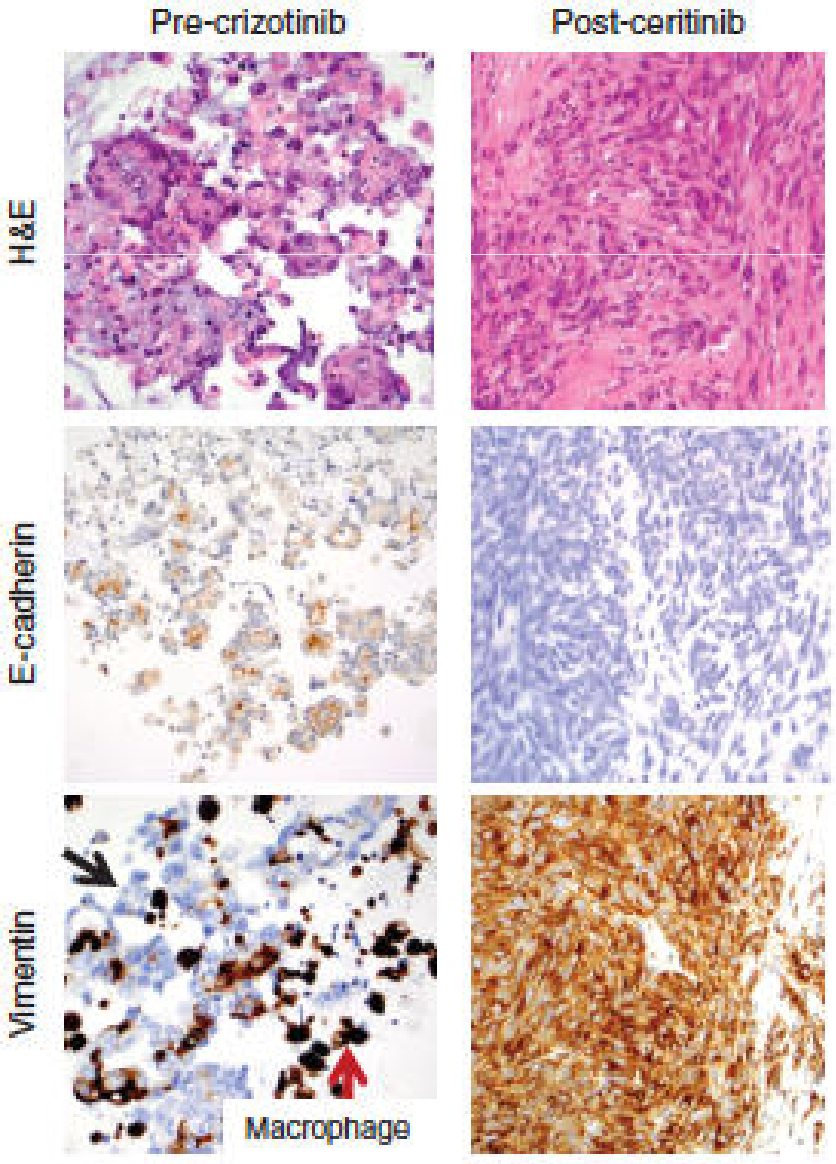
Up-front or sequential strategy?

	Ceritinib		Alectinib		Brigatinib	
	Crizotinib naive	**Crizotinib pretreated**	**Crizotinib naive**	**Crizotinib pretreated**	**Crizotinib naive**	**Crizotinib pretreated**
ORR	**79%** (ASCEND-4)	**56%** (ASCEND-1) **38.6%** (ASCEND-2)	**94%** (AF-001JP)	**52%** (NP28761)	**100%** (Phase I/II)	**74%** (Phase I/II)
PFS	**16.6 months** (12.6-27.2)	**6.9 months** **5.7 months** (5.4 - 7.6).	**NR at 3 years**	**8·1 months** (6·2–12·6)	**NR**	**11.2 months** (7·6–29·7)



Zhang et al; Clin Canc Res; 2016

Epithelial mesenchymal transition as an acquired resistance mechanism to 2nd generation ALK inhibitors



Assessments of EMT in ceritinib-resistant biopsies

Patient ID	ALK resistance mutation	Vimentin	E-cadherin
MGH023-2	ALK F1174C	**Positive**	**Negative**
MGH034-2	WT	**Positive**	**Negative**
MGH049-1	WT	Positive	Positive
MGH051-2	ALK G1202R	Positive	Positive
MGH061-1	WT	Negative	Positive
MGH065-2	ALK L1196M	**Positive**	**Negative**
MGH067-1	ALK L1196M	**Positive**	**Negative**
MGH084-1	ALK I1171N, C1156Y	Negative	Positive
MGH089-1	WT	Negative	Positive
MGH092-1	ALK G1202del	Negative	Positive
MGH902-1	WT	**Positive**	**Negative***
MGH908-1	WT	Negative	Positive

*Partial loss

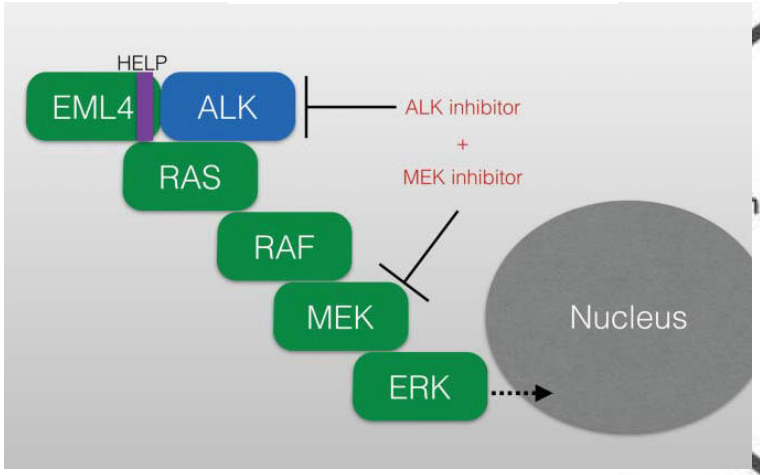
Gainor et al; Cancer Discovery; 2016

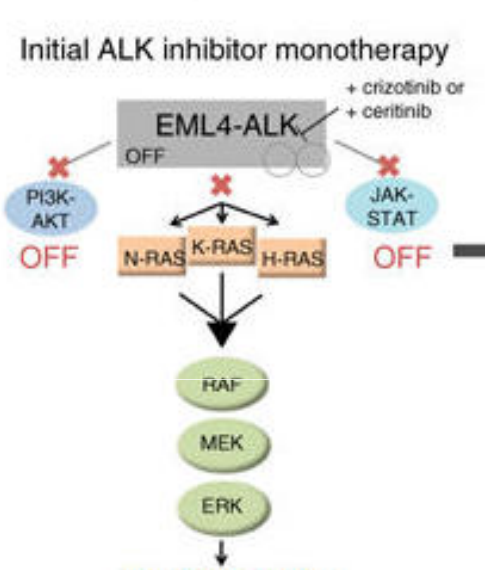
Small cell lung cancer transformation

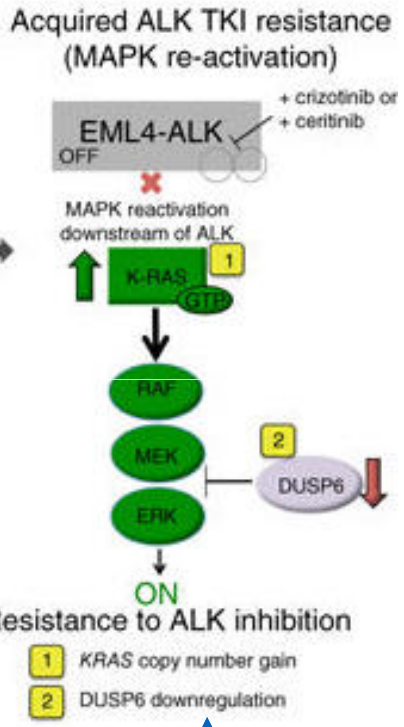
Case	Smoking status	Prior ALK inhibitor	Duration of previous ALKi Rx prior to biopsy	IHC	ALK rearrangement detected in transformed SCLC	Treatment after SCLC diagnosis	Reference
1	Ex-smoker	crizotinib	SD x 2 months	TTF-1, synaptophysin, CD56, ALK	NR	Not reported	Cha, JTO 2016
2	Never-smoker	crizotinib	PR x 6 months	synaptophysin, CD56, ALK	Present (FISH)	alectinib (with partial response)	Caumont, Lung Cancer 2016
3	Never-smoker	alectinib	3 months (prior 7 months of response on crizotnib)	Alk, TTF-1, CD56, Synaptophysin	Present (IHC)	alectinib/ irinotecan intercalating	Fujita, JTO 2016
4	Never-smoker	alectinib	8 months PR (PR x 2 years on crizotinib)	None	Present (IHC and FISH)	Cisplatin/ Irinotecan	Miyamoto, JpJCO 2016
5	Not-reported	alectinib	PR on alectinib x 13 months (PR x 6 months on crizotinib)	CD56, synaptophysin	Present (IHC and FISH); no acquired mutation and no ALK amplification	None	Takegawa, Ann Oncol 2016
6	Never-smoker	ceritinib	PR x 7 months (PR x 7 monts on crizotinib)	symaptophysin. chromogranin	Present (IHC, RNA sequencing, FISH)	cisplatin/VP16 then CAV	Levacq, Lung Cancer 2016

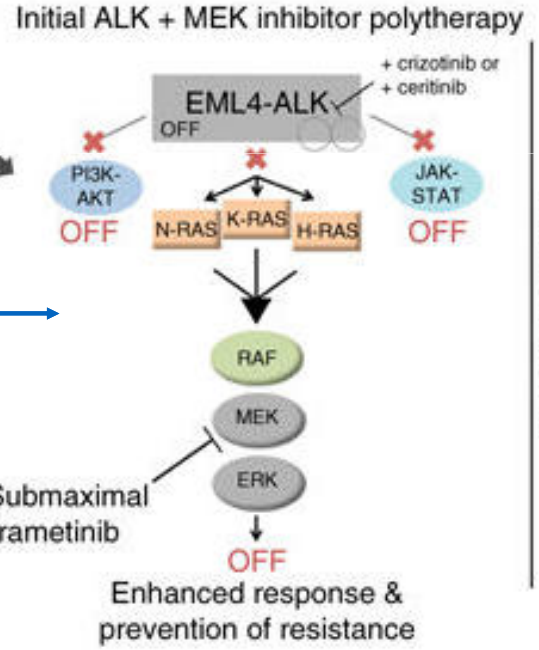
The KRAS activation

- The HELP domain of EML4 contains 50% of hydrophobic residue —> membrane localization and RAS-MAPK activation
- ALK inhibition does not fully abrogate MAPK activation









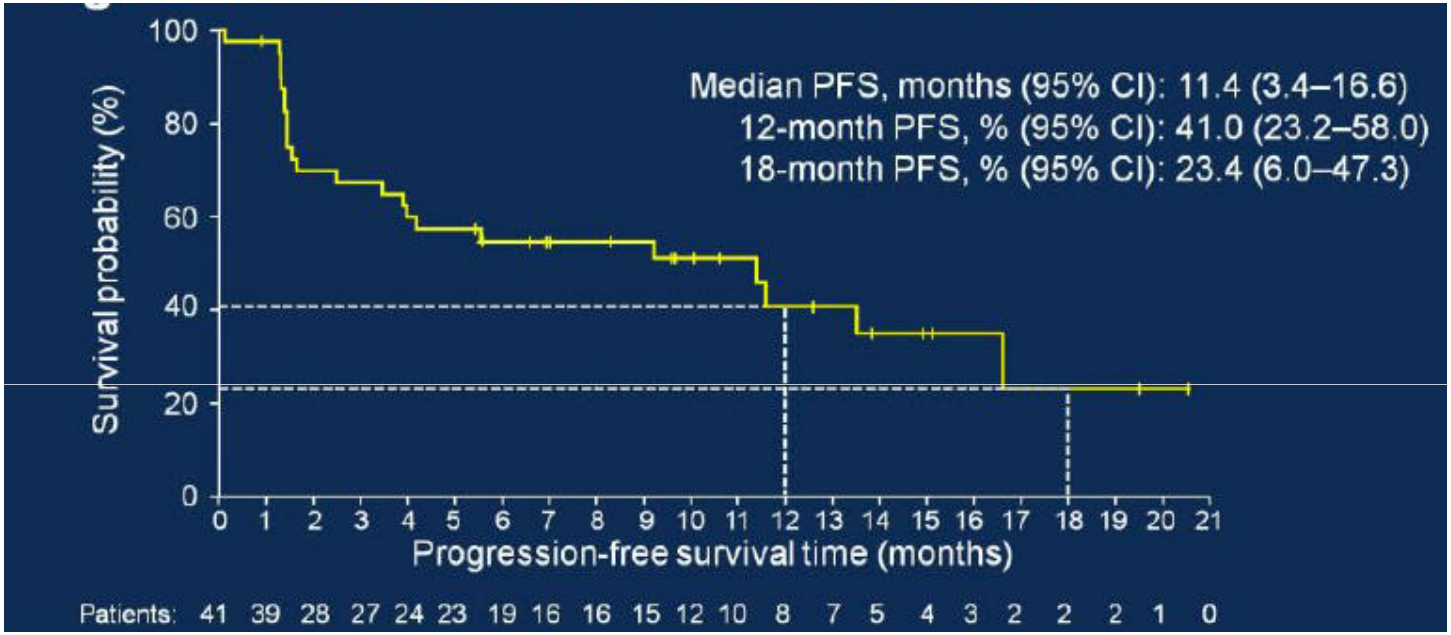
Durable response

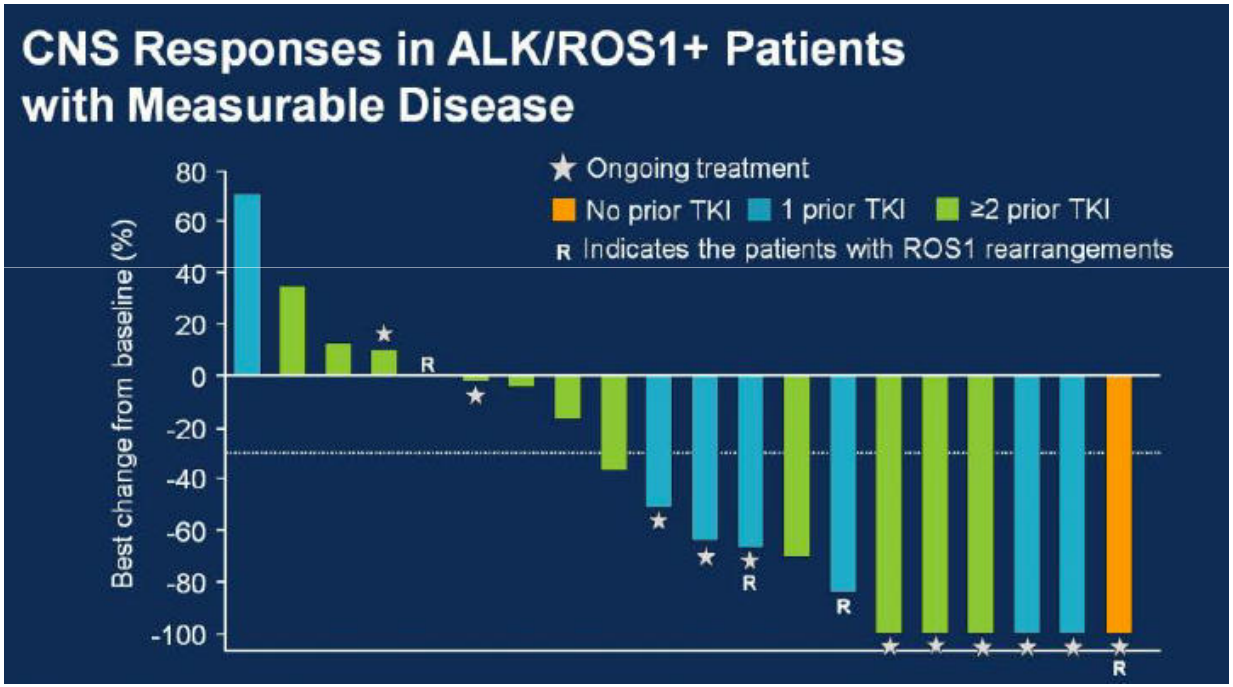
- Dual MAPK and ALK signaling inhibition

- Focal amplification of KRAS in 20% of patients with acquired resistance to the ALK inhibitor
- Downregulation of DUSP6 (that inhibits MAPK activation) in patients resistant to crizotinib

Hrustanovic et al; Nat Med 2015

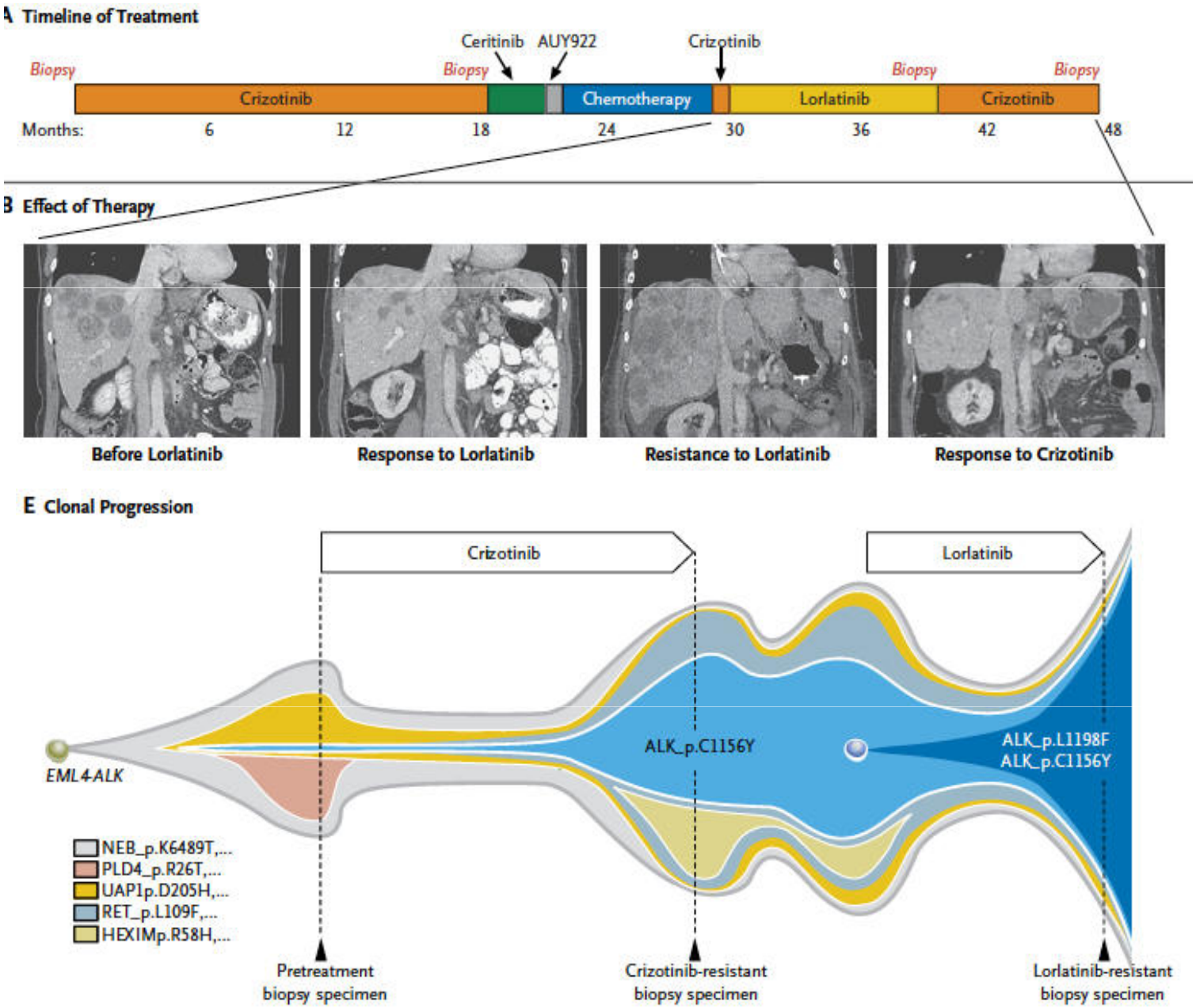
Lorlatinib - Phase I





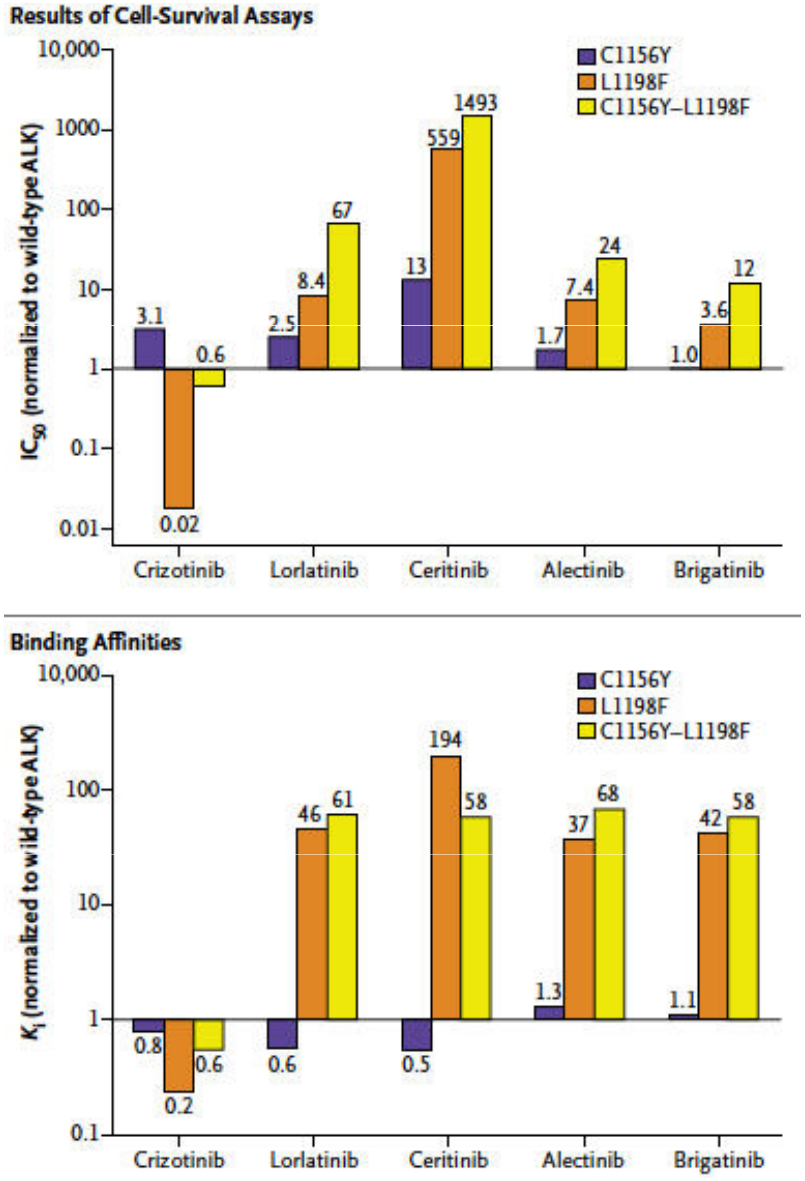
Solomon et al; ASCO 2016

Lorlatinib - mechanisms of resistance



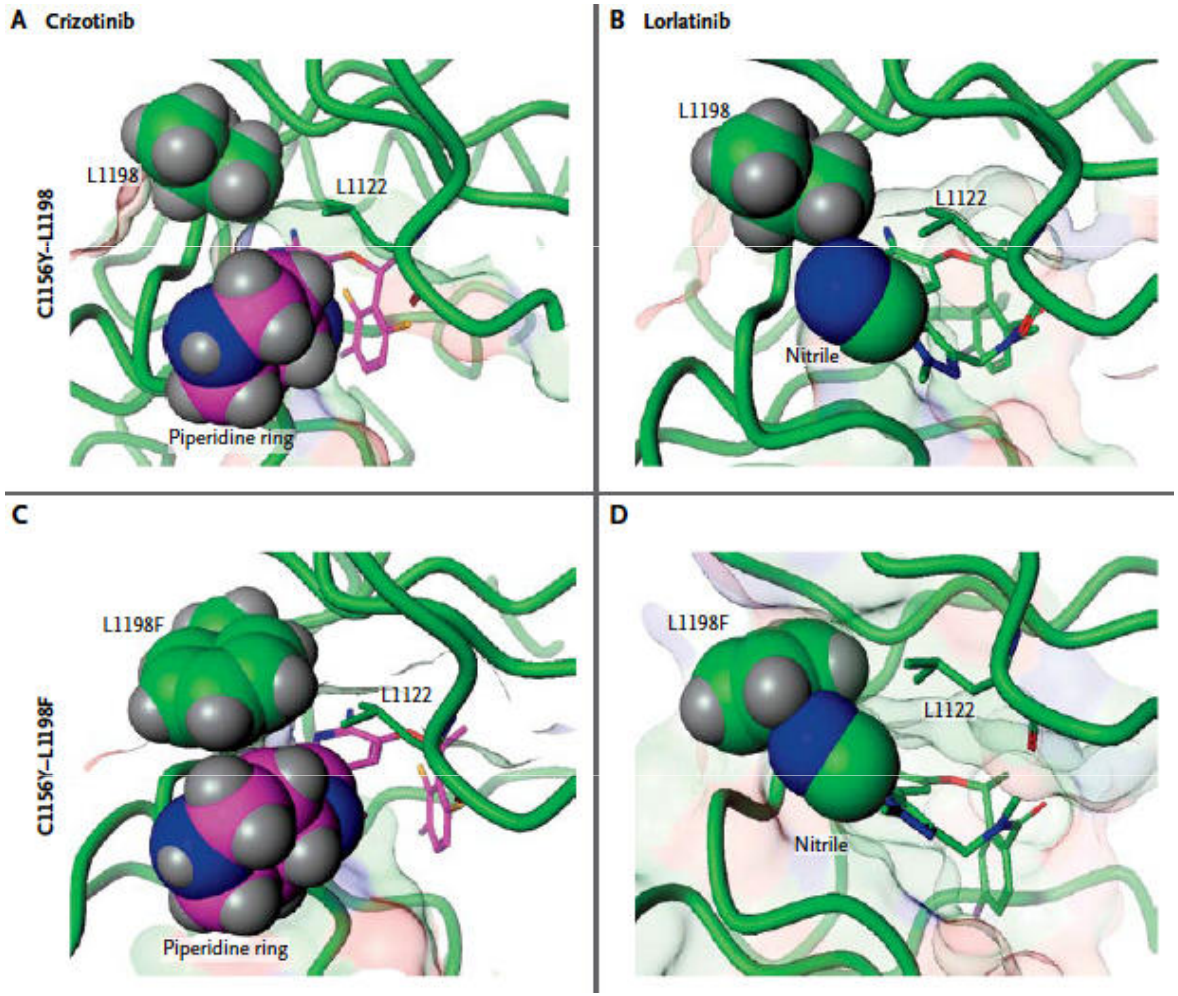
Shaw et al; NEJM 2016

Lorlatinib - mechanisms of resistance



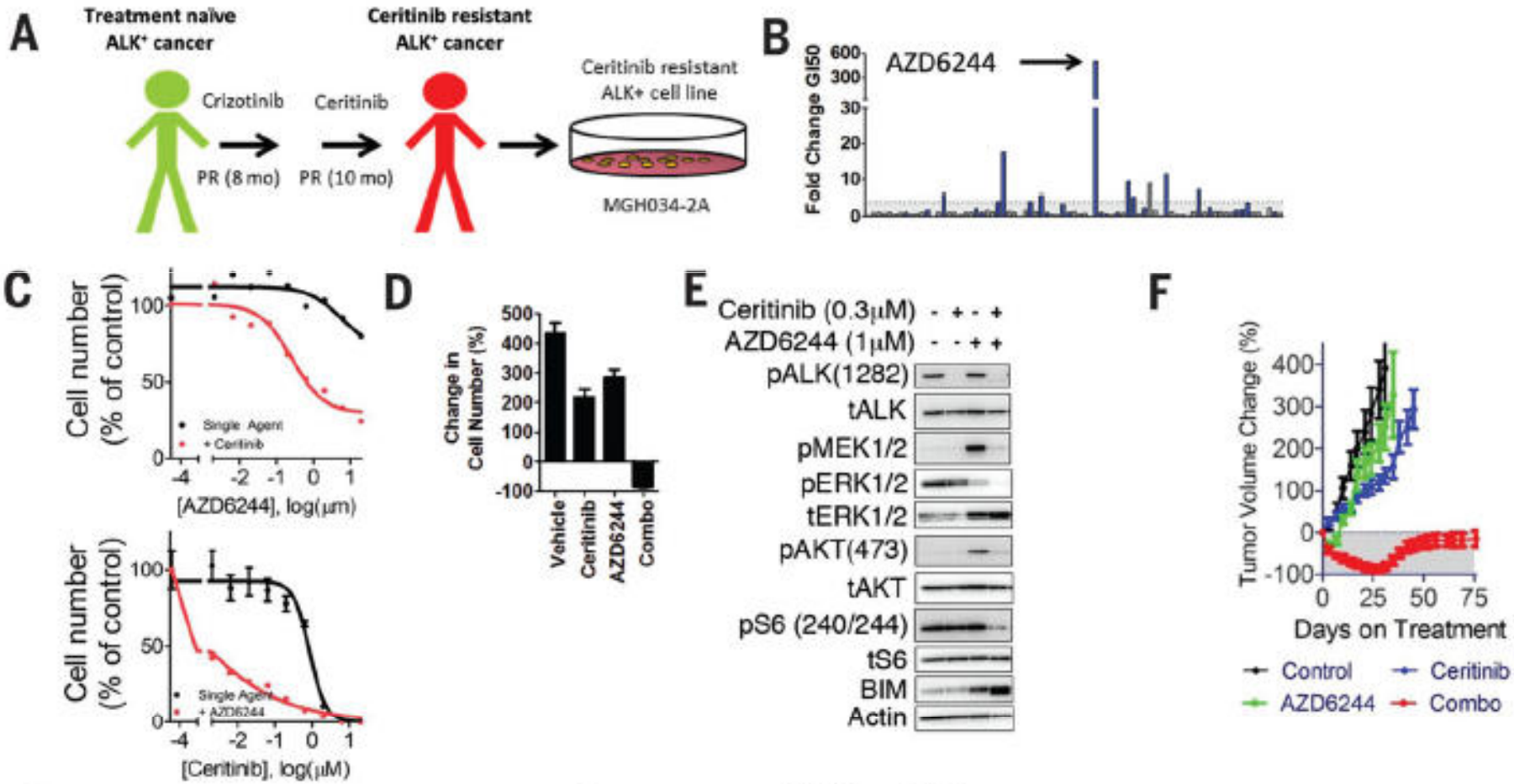
Shaw et al; NEJM 2016

The cysteine at position 1156 (**C1156**) is located far from the inhibitor binding site



L1198 resides near the ATP-binding site, and substitution of leucine with the larger phenylalanine leads to a steric clash with lorlatinib. Crizotinib binding to L1198 is more favorable —> in double mutant the enhanced crizotinib binding to L1198 offsets the increased kinase activity of C1156

Precision oncology: combination strategy based on patient´s derived models



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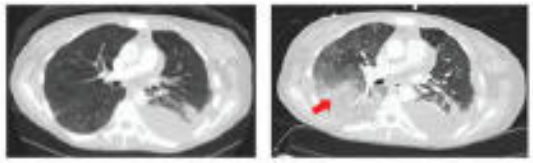

Response Progression

H

Site	MAP2K1 K57N	PIK3CA H1047R
Lung - RLL1	0.22	0.00
Lung - RLL2	0.20	0.00
Lung - RLL3	0.27	0.00
Lung - RLL4	0.22	0.00
Lung - Left apex	0.22	0.00
Lung - LUL1	0.00	0.00
Lung - LUL2	0.29	0.00
Lung - LUL3	0.24	0.00
Liver - Medial	0.00	0.28
Liver - Lateral	0.00	0.00
Liver - Posterior	0.00	0.00
Heart (Normal)	0.00	0.00

Crystal et al; Science 2014


INIZIA
UNA NUOVA
GRANDE
STAGIONE.