



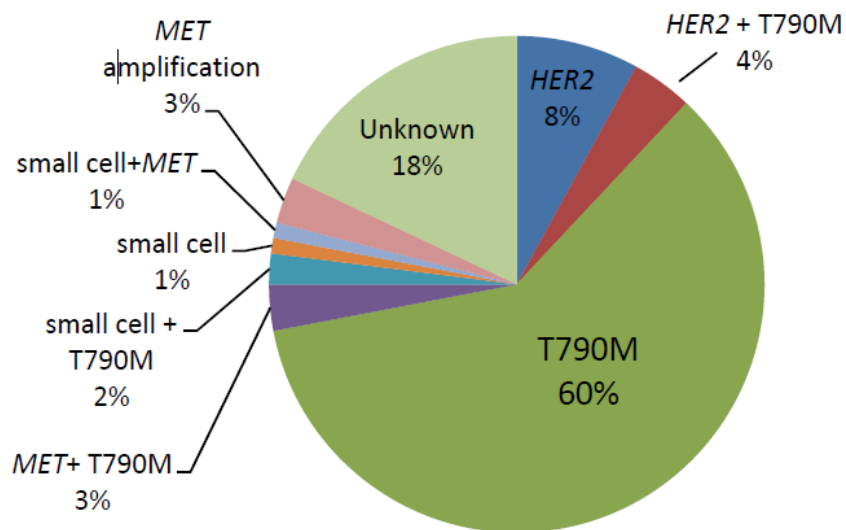
**Serial monitoring of *EGFR* mutations in plasma and evaluation of *EGFR* mutation status in matched tissue and plasma from NSCLC patients treated with CO-1686**

November 7, 2013

CNAPS VIII

# CO-1686 Overview

*T790M is dominant cause of acquired resistance to initial TKI therapy in EGFR<sup>mut</sup> NSCLC*



- CO-1686 is a novel, oral, selective covalent inhibitor of *EGFR* mutations in NSCLC

- Inhibits key activating and T790M resistance mutations
- Spares wild-type receptor signaling
- Promising activity in an ongoing Ph 1/2 clinical trial

Source: Yu et al CCR (2013)

# An *EGFR* blood test would be advantageous but needs high sensitivity/specificity for use in clinical practice

- Why blood?
  - May capture tumor heterogeneity
  - Is non-invasive
  - Enables serial monitoring of circulating tumor DNA (ctDNA) kinetics and emergence of resistance
  - Biopsies often provide inadequate/insufficient material for molecular analysis
- ***Key questions to be addressed***
  - Are current EGFR testing platforms sensitive and specific enough to inform NSCLC patient management?
  - What clinical characteristics impact the sensitivity of *EGFR* mutation detection in plasma?

## Methodology: Matched tissue and plasma were evaluated using the cobas EGFR test

- Matched tumor and plasma was obtained from NSCLC patients in ongoing Ph 1/2 trial of CO-1686 and an observational study
  - 97 evaluable Stage IIIB/IV patients; most received prior EGFR TKI
  - 10 Ph 1/2 patients not evaluable because of low/no tumor content
  - Plasma and tissue collections matched in time
  - Biopsies included CNB, FNA, bronchoscopic biopsies, and PE
- Baseline plasma and tissue evaluated with cobas EGFR mutation test
  - Qualitative test that uses allele-specific PCR (Taqman chemistry)
  - Input is ctDNA
  - Detects 41 mutations including T790M, L858R, ex19 deletions
- Baseline plasma from 30 patients was additionally tested by BEAMing for comparison with cobas results

# cobas test results in matched tissue and plasma displayed good sensitivity and very good specificity for plasma test

		Tissue		Tissue	
		Activating Mutations		T790M	
		positive	negative	positive	negative
Plasma	positive	57	0	21	2
	negative	21	23	13	61
total		78*	23	34	63

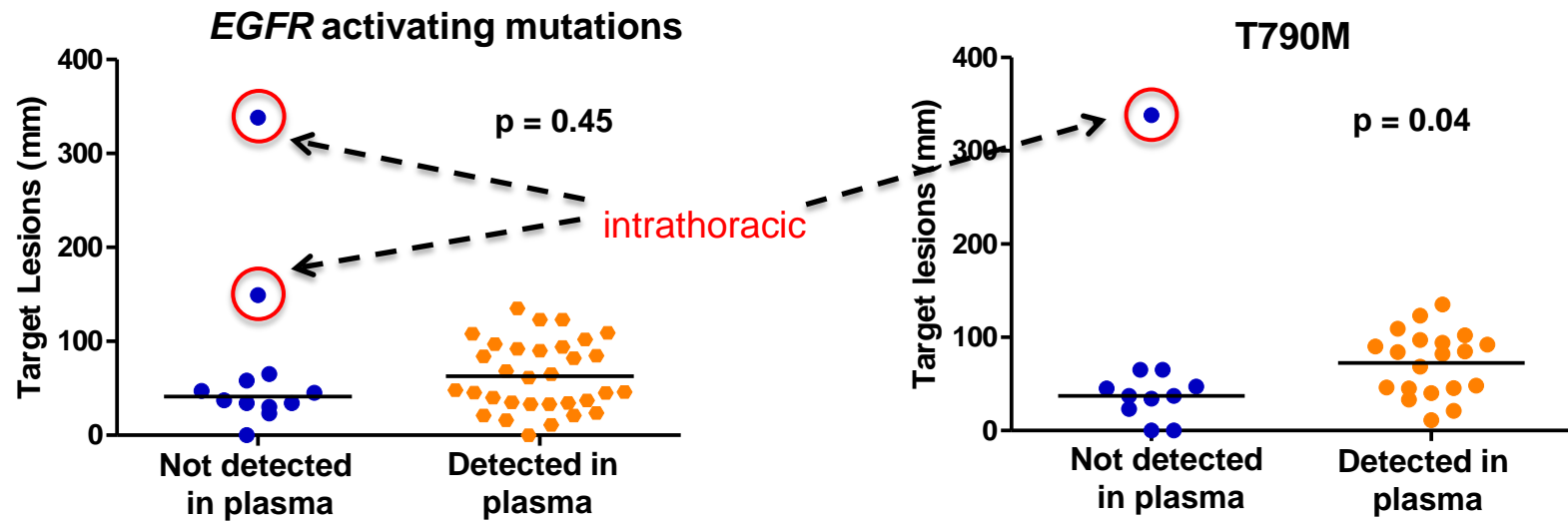
	<b>Activating</b>	<b>T790M</b>
<b>Plasma Sensitivity (tissue as reference)</b>	<b>73%</b>	<b>62%</b>
<b>Plasma Specificity (tissue as reference)</b>	<b>100%</b>	<b>97%</b>

- Two T790M plasma+/tumor- patients were confirmed plasma-positive by BEAMing
  - May reflect tumor heterogeneity and highlights potential advantages of plasma
- Plasma-/tumor+ patients may be plasma-neg due to biology (low/no ctEGFRmut)

\*4 patients had compound activating mutations in tissue (n = 97 patients)

# Tumor burden is a weak predictor of ability to detect *EGFR* mutations in plasma of NSCLC patients

- Tumor burden has been found to be associated with ctDNA levels<sup>1</sup> but may not capture differences in tumor biology (e.g. vascularization, immune infiltration)
  - While *EGFR* plasma test sensitivity was generally better in patients with higher tumor burden, there were notable exceptions



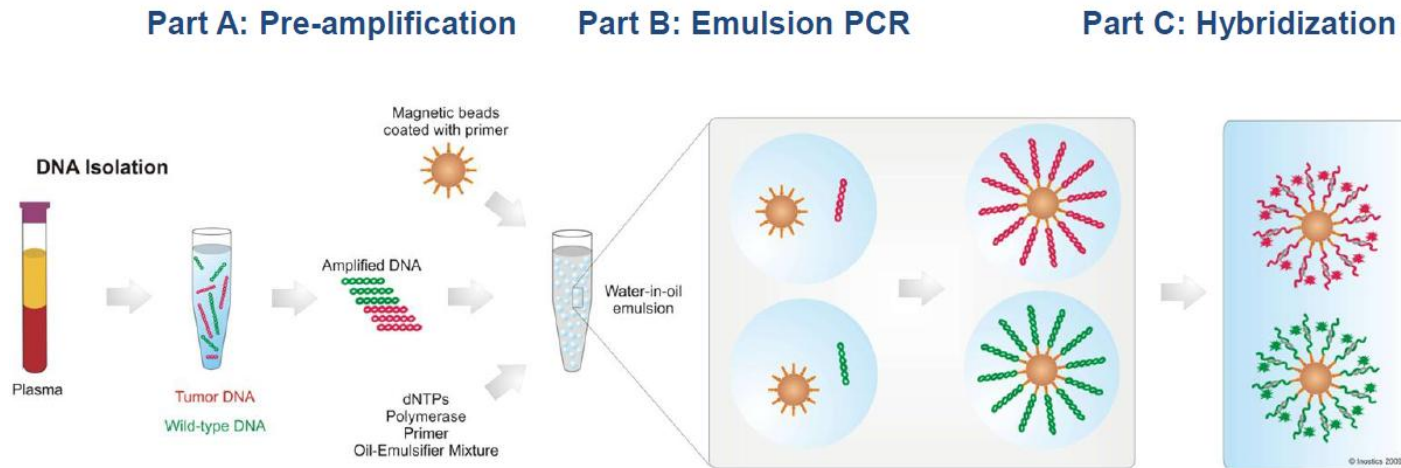
<sup>1</sup> Bidard Sci Transl Med 2013

## Mutations are more readily detected in plasma of patients with extrathoracic metastases (M1b) vs. intrathoracic (M1a/M0) disease

Mutation	Disease classification	Mutation detected in tissue	Subset with mutation in plasma	Percentage	P value
Activating Mutations	M1a/M0	24	12	50%	<0.001
	M1b	49	43	88%	
T790M	M1a/M0	13	2	15%	<0.001
	M1b	21	19	90%	

- M0/M1a/M1b status known for 73 patients

# cobas EGFR plasma test results were compared to sensitive BEAMing test



Dressman *et al.* PNAS, 2003  
Diehl *et al.* PNAS, 2005

- Baseline plasma samples from subset of 30 patients were tested by BEAMing and compared with cobas results
  - Serial monitoring using BEAMing is also ongoing
- BEAMing is droplet digital PCR followed by flow cytometry
  - 0.02% sensitivity



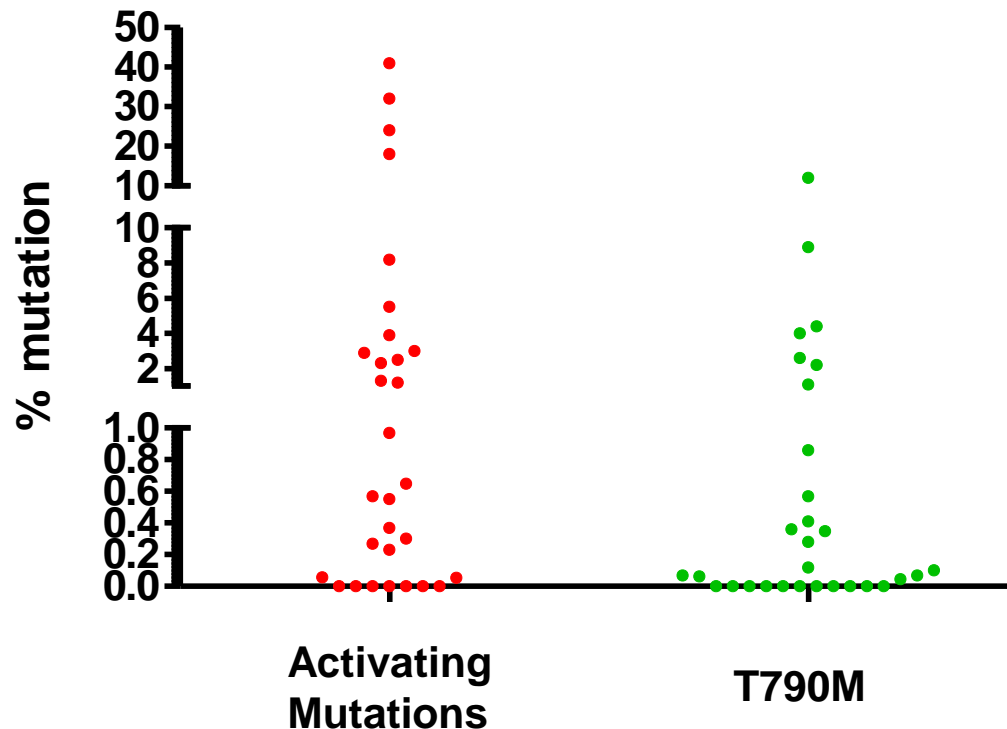
# Strong overall agreement observed between cobas and BEAMing *EGFR* plasma tests

Plasma Results		BEAMing			
		Activating Mutations		T790M	
		positive	negative	positive	negative
cobas	positive	21	2	16	1
	negative	1	6	3	10

- Overall agreement between platforms (n = 30)
  - 87% for T790M
  - 90% for activating mutations
  - Discordance occurred only at very low allele frequencies (<0.3%)
- Cobas/BEAMing had similar plasma sensitivity wrt tissue (n = 27)

	<u>Activating Mutations</u>	<u>T790M</u>
cobas	74%	70%
BEAMing	78%	70%

## Low median plasma *EGFR* allele fraction supports need for highly sensitive detection methods in this NSCLC patient population



### *EGFR* allelic fraction in plasma by BEAMing

#### **T790M**

median = 0.41%

range = 0.046 – 12%

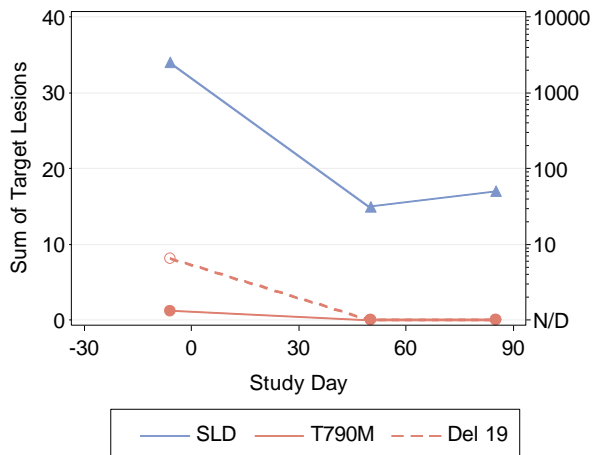
#### **Activating Mutations**

median = 1.3%

range = 0.053 – 41%

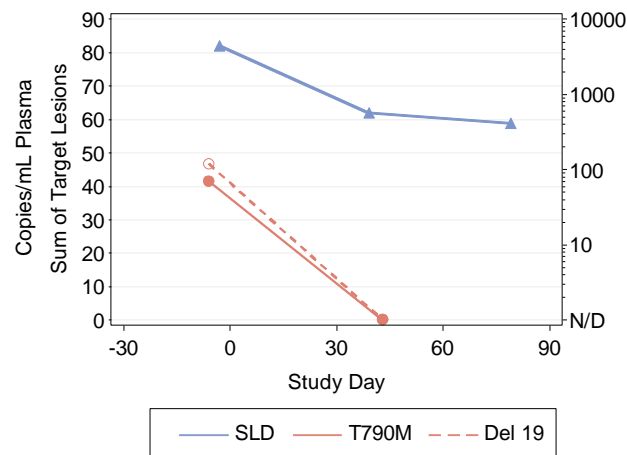
# Serial Monitoring: Initial drop in plasma *EGFR* seen in patients where clinical activity observed

900 mg BID



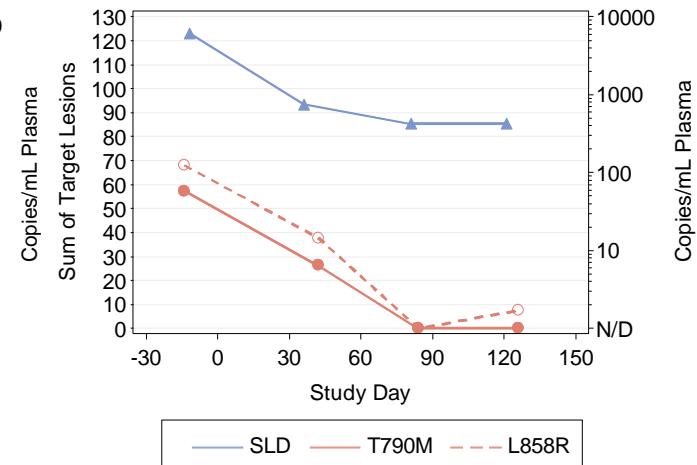
Base line **PR**

900 mg BID



Base line **SD**

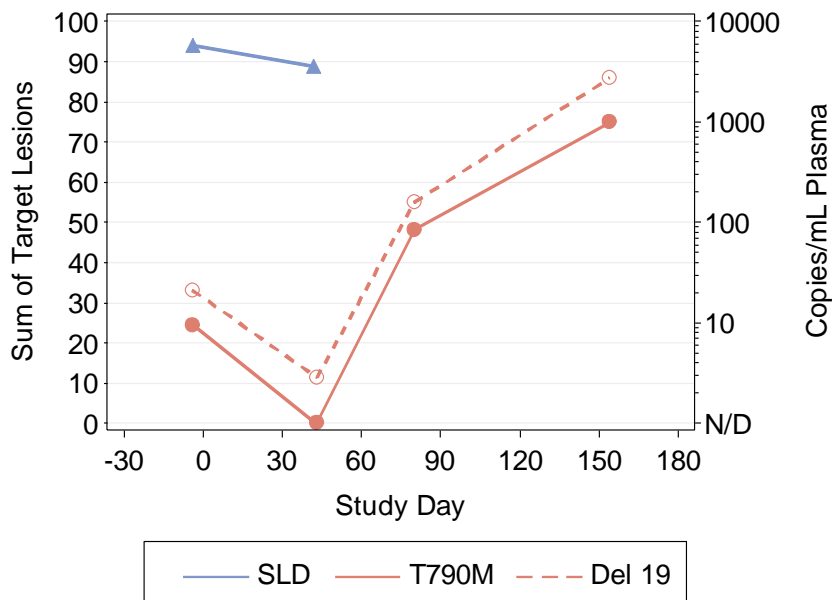
900 mg BID



Base line **PR**

- Serial monitoring ongoing for Ph 1/2 patients; data not yet mature for most (n = 25)
- Initial drop in plasma *EGFR* seen in 13/14 patients with SD or PR as best response
- CO-1686 inhibits both activating and T790M mutations
- 6 patients have undetectable plasma *EGFR* at baseline & during serial monitoring

# Serial monitoring will inform several key questions



Early cohort patient

- Does rate of initial decline in *EGFR* mutation levels correlate with response to CO-1686?
- When is re-emergence of mutant *EGFR* seen prior to clinical progression?
- Does the allele fraction of T790M relative to activating mutation correlate with response to CO-1686?

# Summary

- A high proportion of *EGFR* mutations identified in tissue were also detected in plasma using the cobas *EGFR* mutation test
- Mutations were more readily detectable in the plasma of patients with M1b rather than M1a disease
- Strong overall agreement observed between BEAMing and cobas plasma *EGFR* results
- Baseline T790M levels of <1% in many patients support use of sensitive technologies such as digital PCR or allele-specific PCR for mutation detection
- Serial monitoring of plasma ct*EGFR* kinetics is ongoing in the Ph 1/2 trial of CO-1686