Aparna Parikh MD
Massachusetts General Hospital Cancer Center
Harvard Medical School

“Liquid Biopsies” in gastrointestinal malignancies: Are we ready?

HARVARD MEDICAL SCHOOL

MASSACHUSETTS GENERAL HOSPITAL
CANCER CENTER

Disclosures

- Consultant/advisory board member for Puretech, Foundation Medicine
- Institutional research funding from Array, Eli Lily, Plexxikon, Guardant, BMS, MacroGenics, Novartis, Tesaro, Genentech

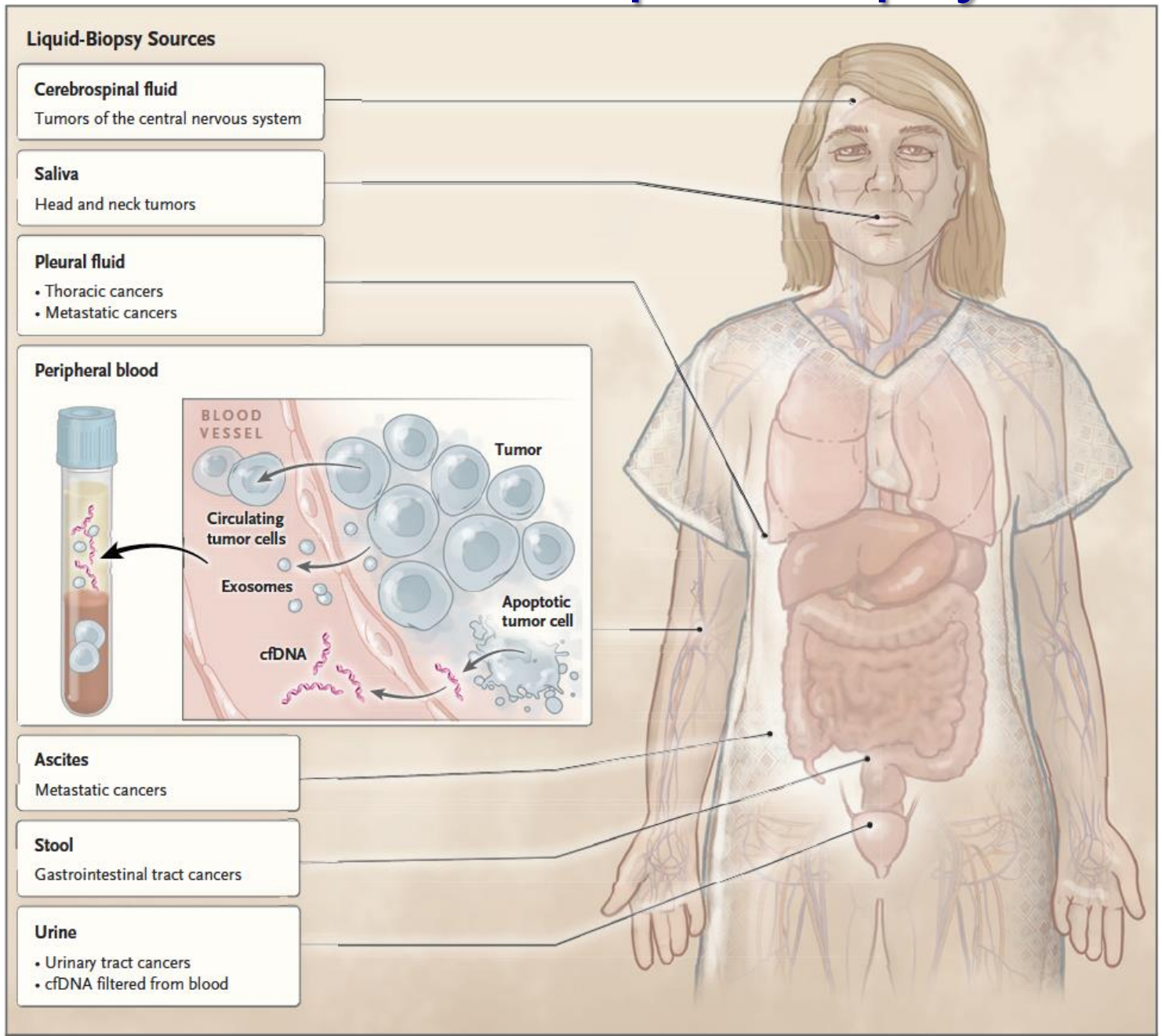
Applications of Liquid Biopsies

Resistance

Treatment Response

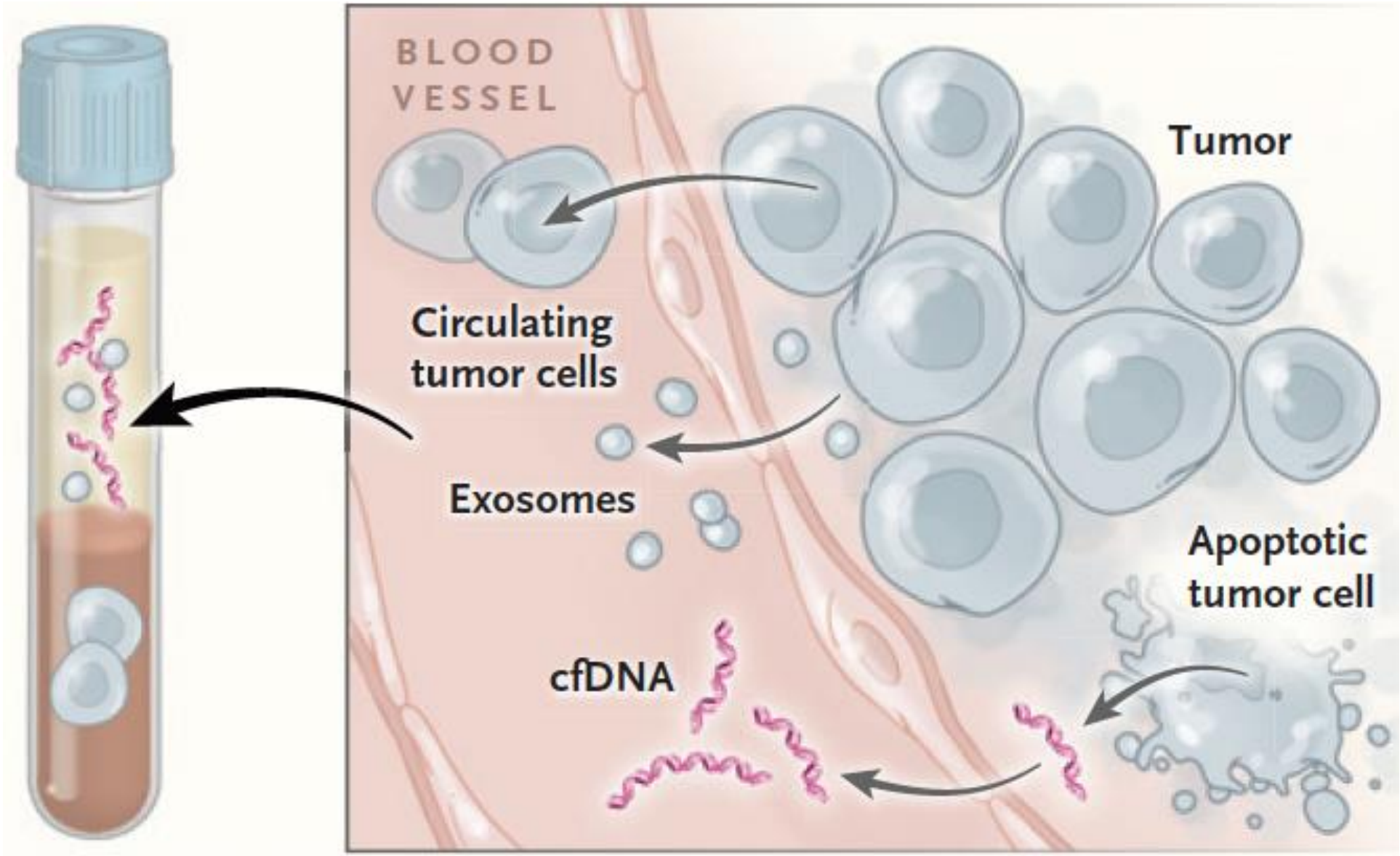
MRD

What is a “Liquid Biopsy”?



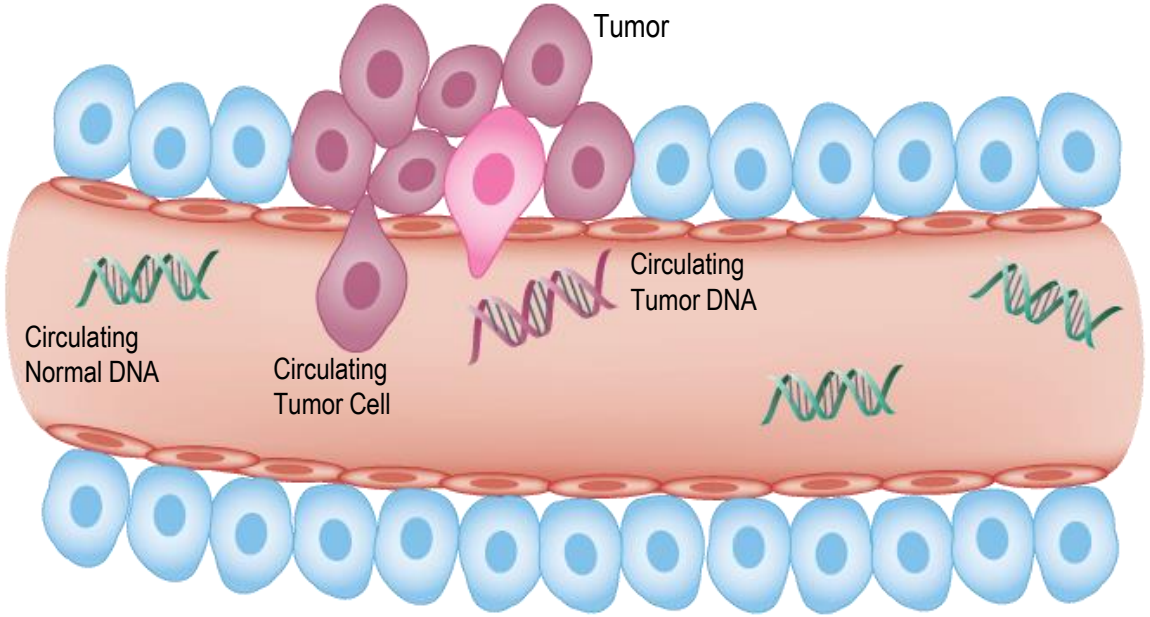
Corcoran and Chabner, NEJM 2018

Different forms of liquid biopsy



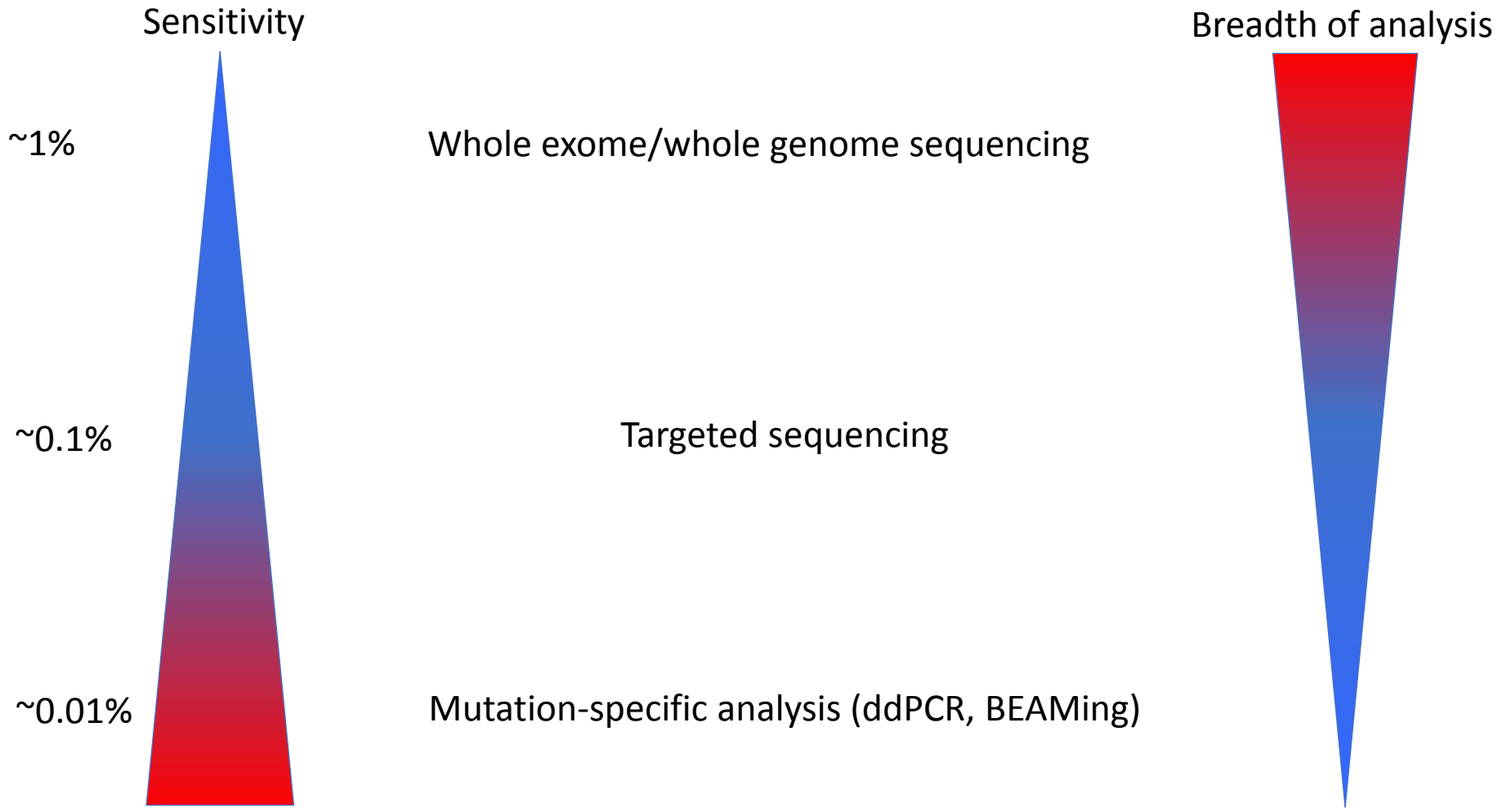
Corcoran and Chabner, NEJM 2018

Circulating tumor DNA (ctDNA)



- Cell-free DNA (cfDNA) is shed into the bloodstream by cells (normal or cancer) throughout the body
- Thus, only a fraction of cfDNA may be ctDNA (tumor-derived)

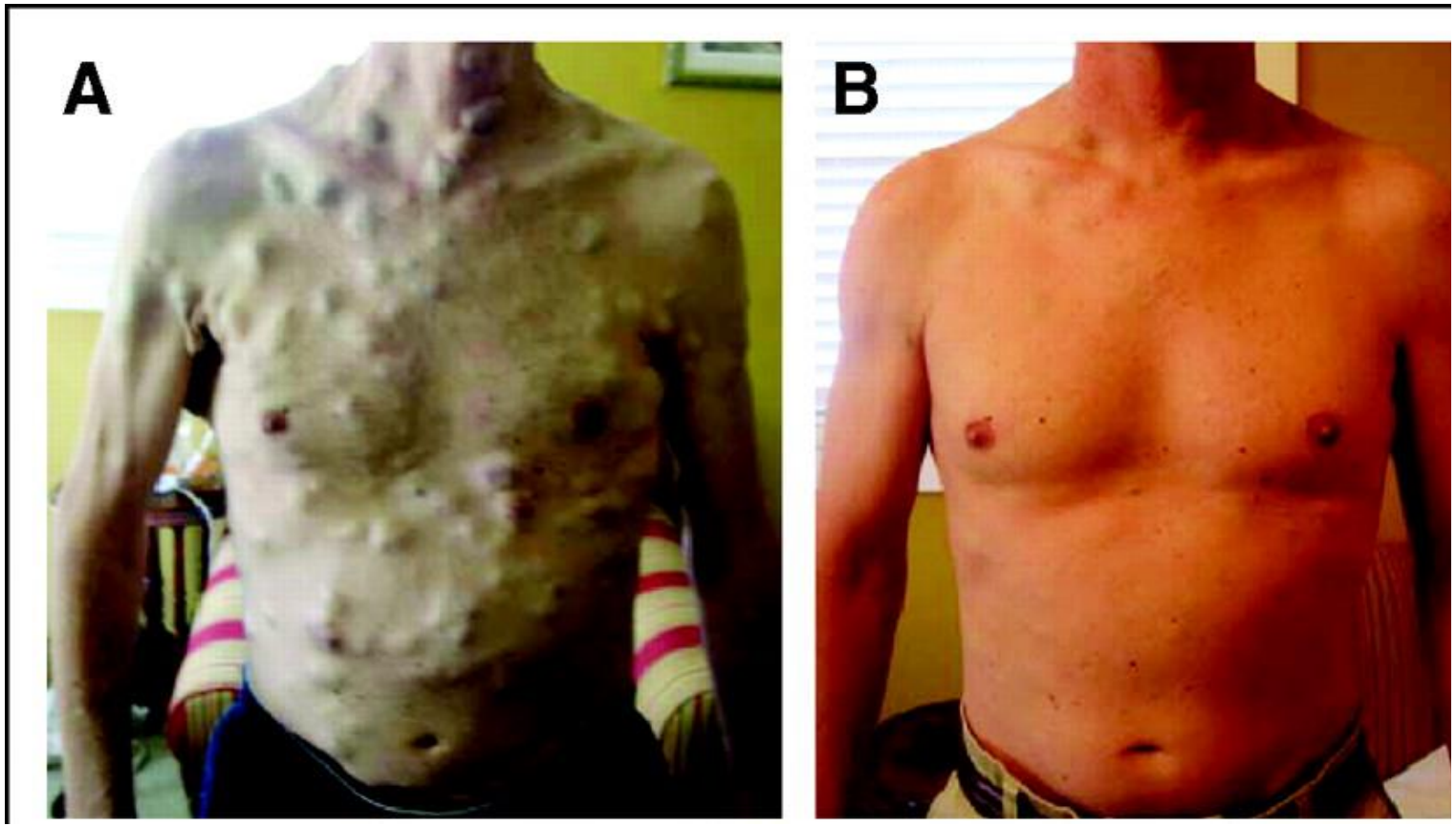
Methods for ctDNA analysis



Resistance

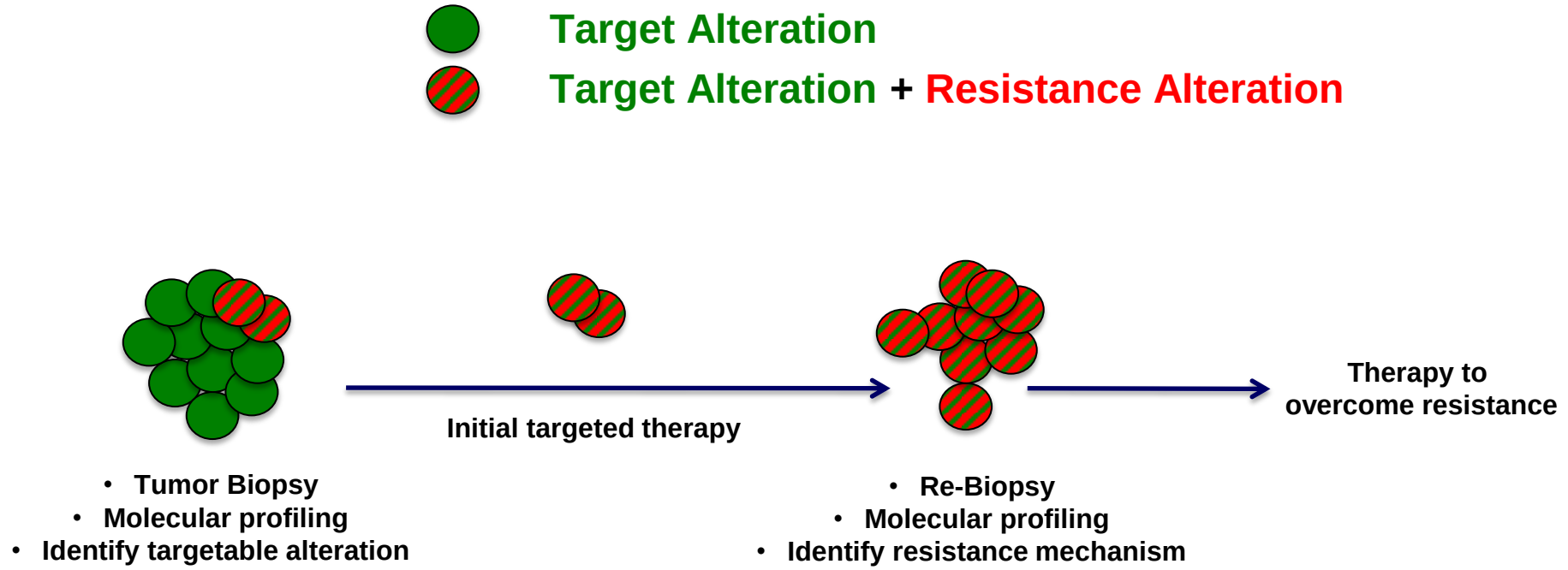
Secondary (acquired) Resistance

Pre-treatment **+ Vemurafenib 15 weeks**

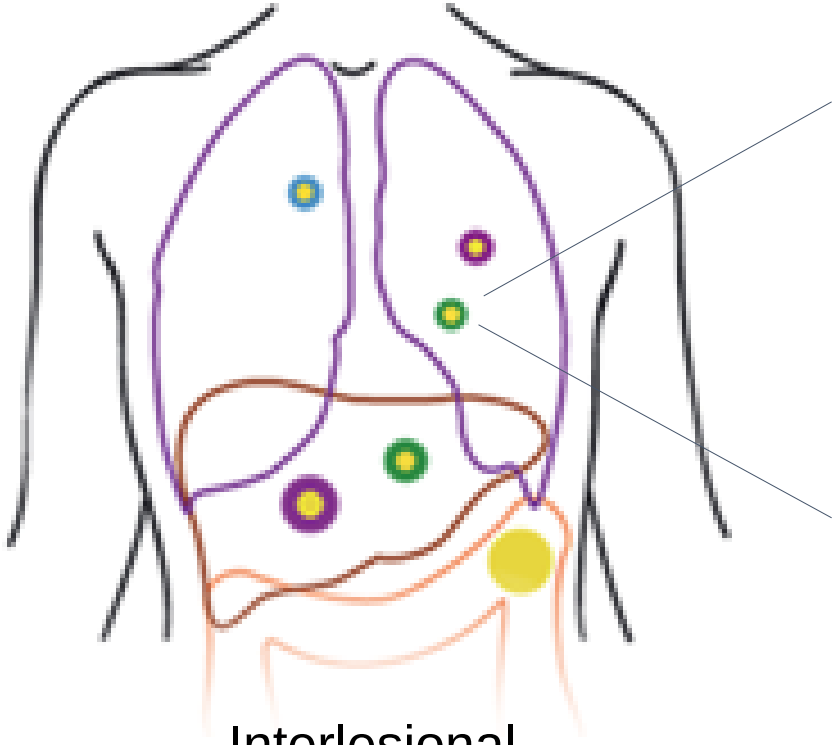


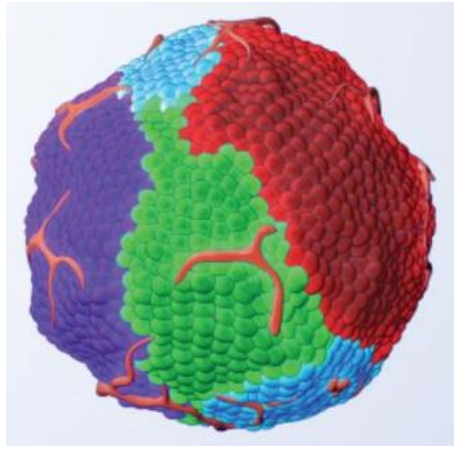
Wagle et al, JCO 2011

Overcoming acquired resistance to therapy



Tumor heterogeneity and acquired resistance





Interlesional

Heterogeneity between distinct metastatic lesions

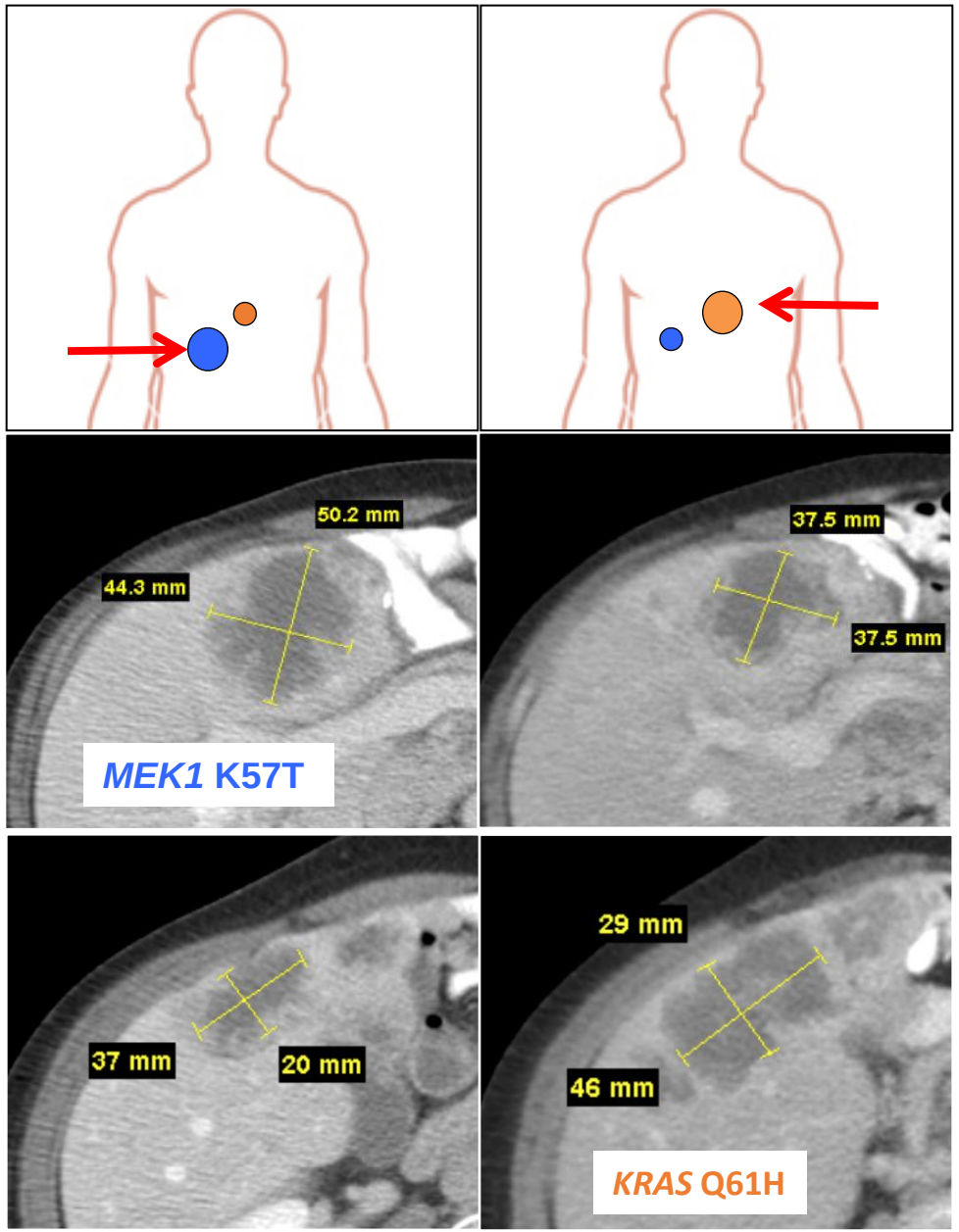
Intralesional

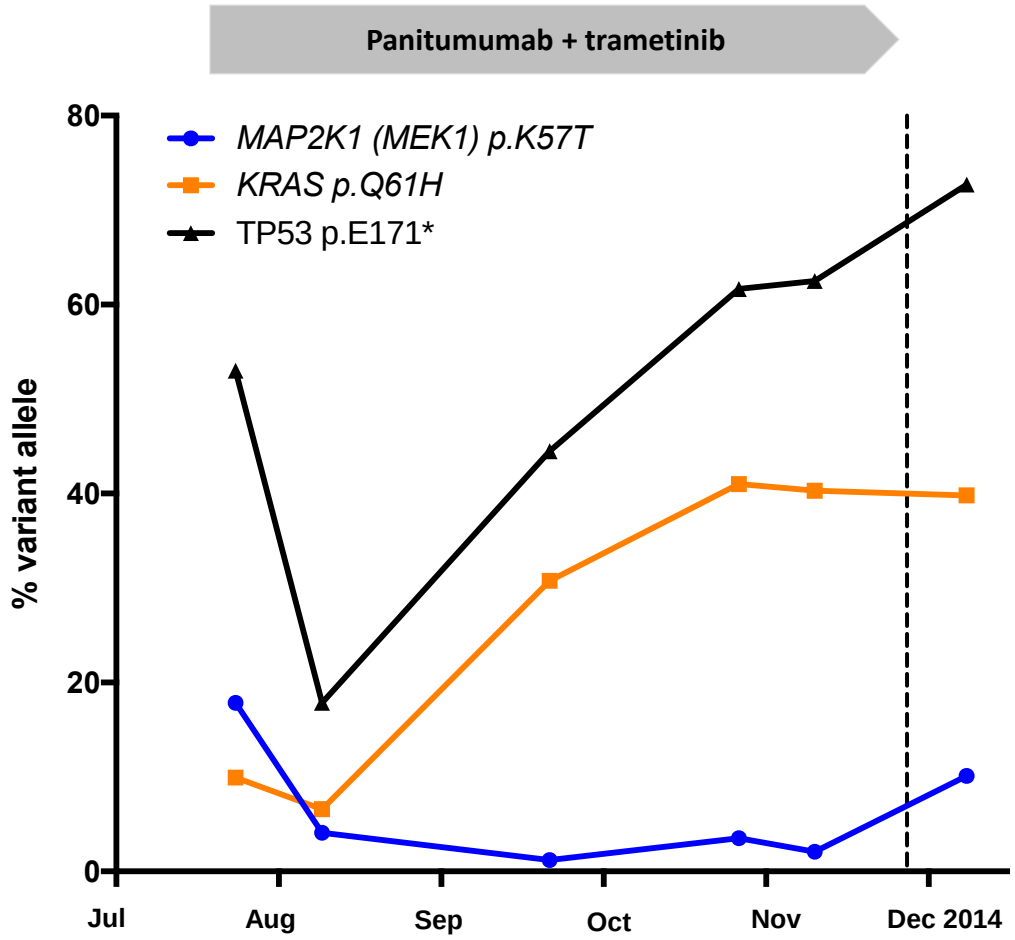
Heterogeneity within a single metastatic lesion

- A single needle biopsy may vastly underrepresent molecular heterogeneity
- Liquid biopsy may detect alterations in ctDNA shed by tumor cells throughout the body

Adapted from Bardelli, ASCO 2013; Misale, Cancer Discovery 2014

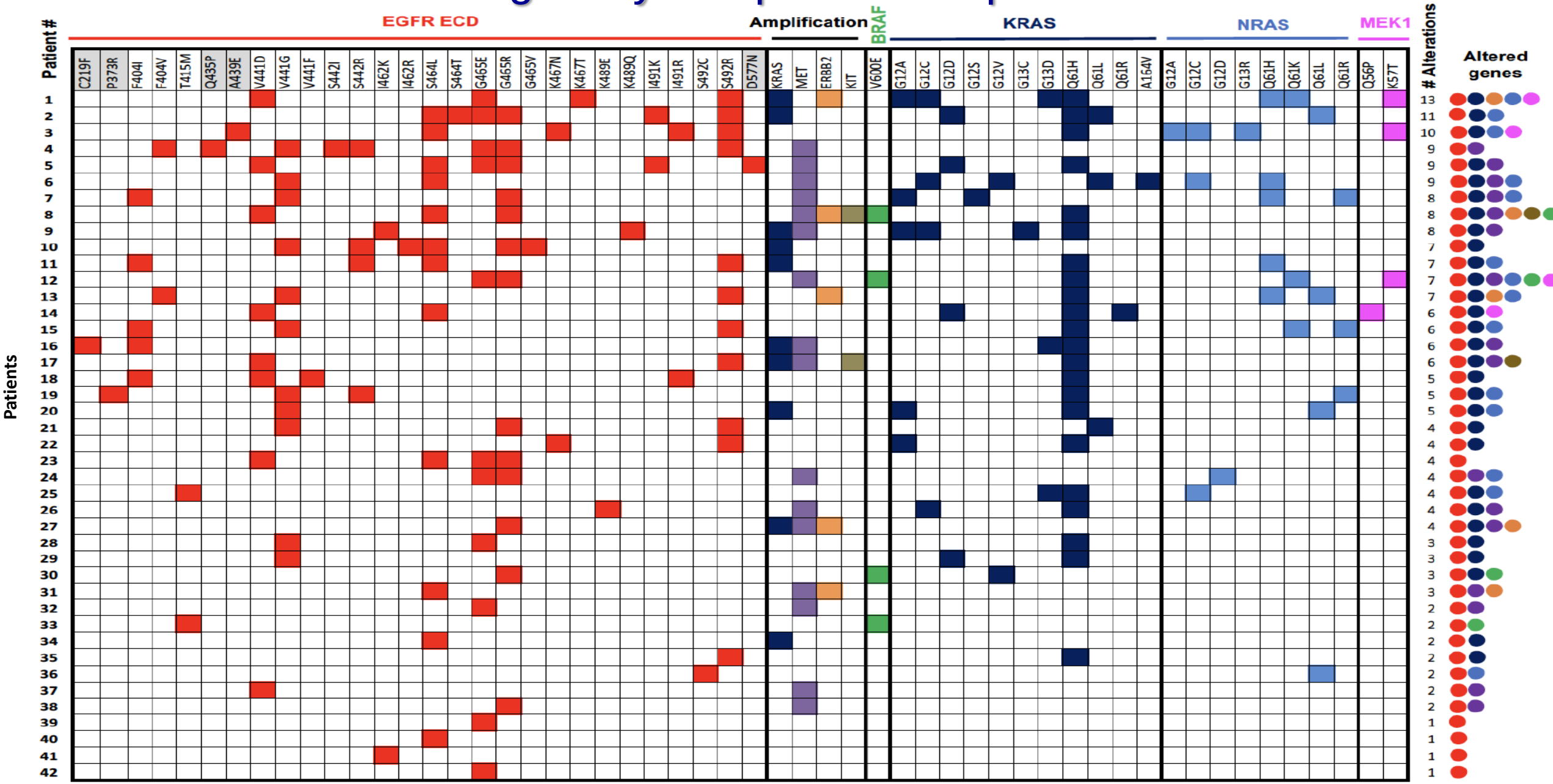
Are single tumor biopsies sufficient to guide therapy?





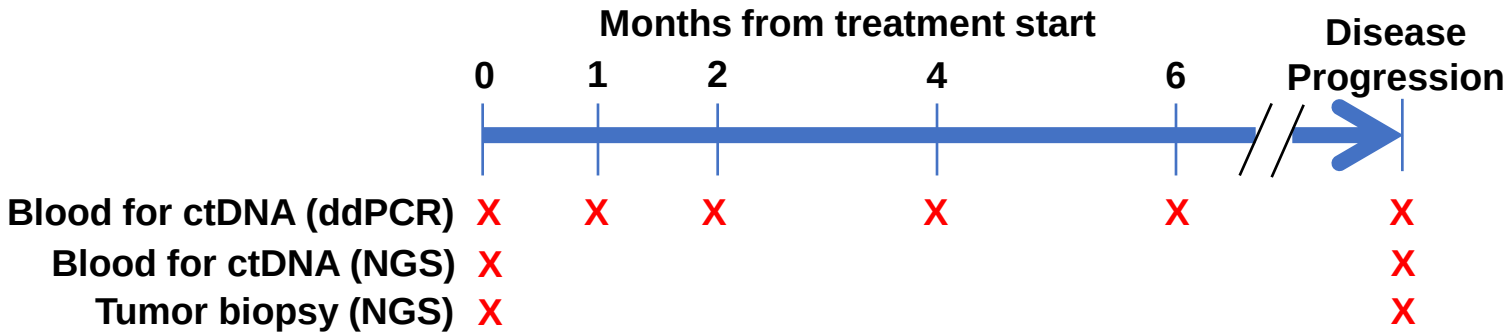
(Ryan Corcoran in collaboration with Larry Blaszkowsky, Alberto Bardelli)
Russo et al, Cancer Discovery 2016

Tumor heterogeneity complicates acquired resistance



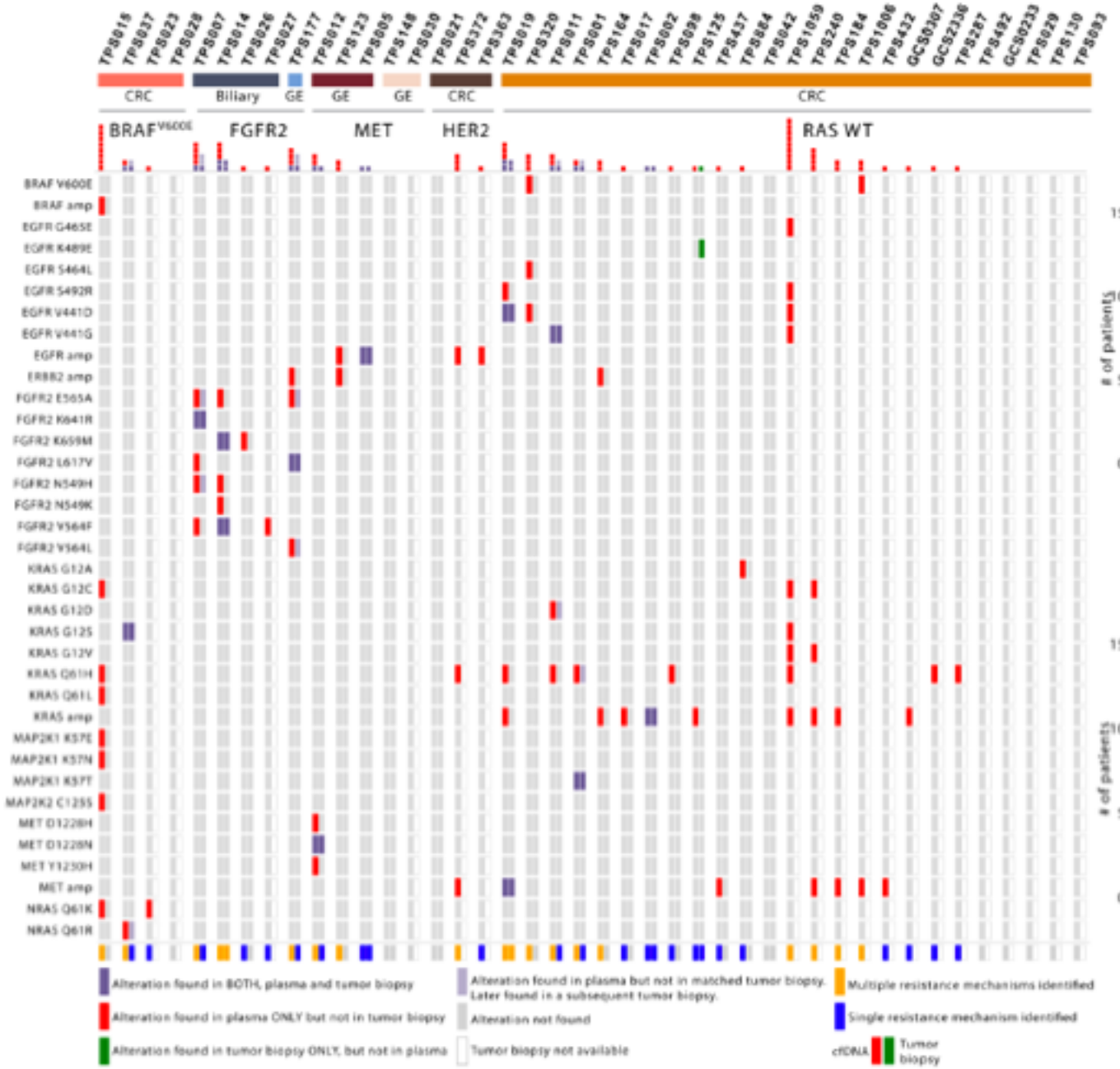
Strickler et al, Cancer Discovery 2018

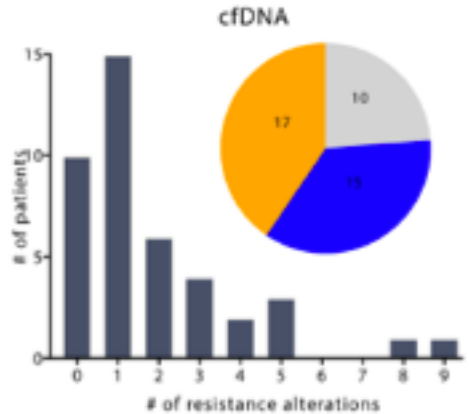
Systematic liquid biopsy collection during targeted therapy

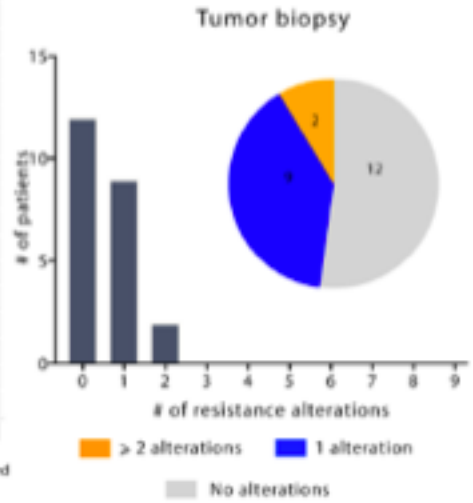


- Integrated, disease center-wide protocol for all targeted therapy patients
- One page consent “brochure” given to patients at first clinic visit
- Fully staffed by CRCs/technicians
- Activated by single page/email from MD to CRC at time of treatment start

Liquid biopsy to identify heterogeneous resistance mechanisms





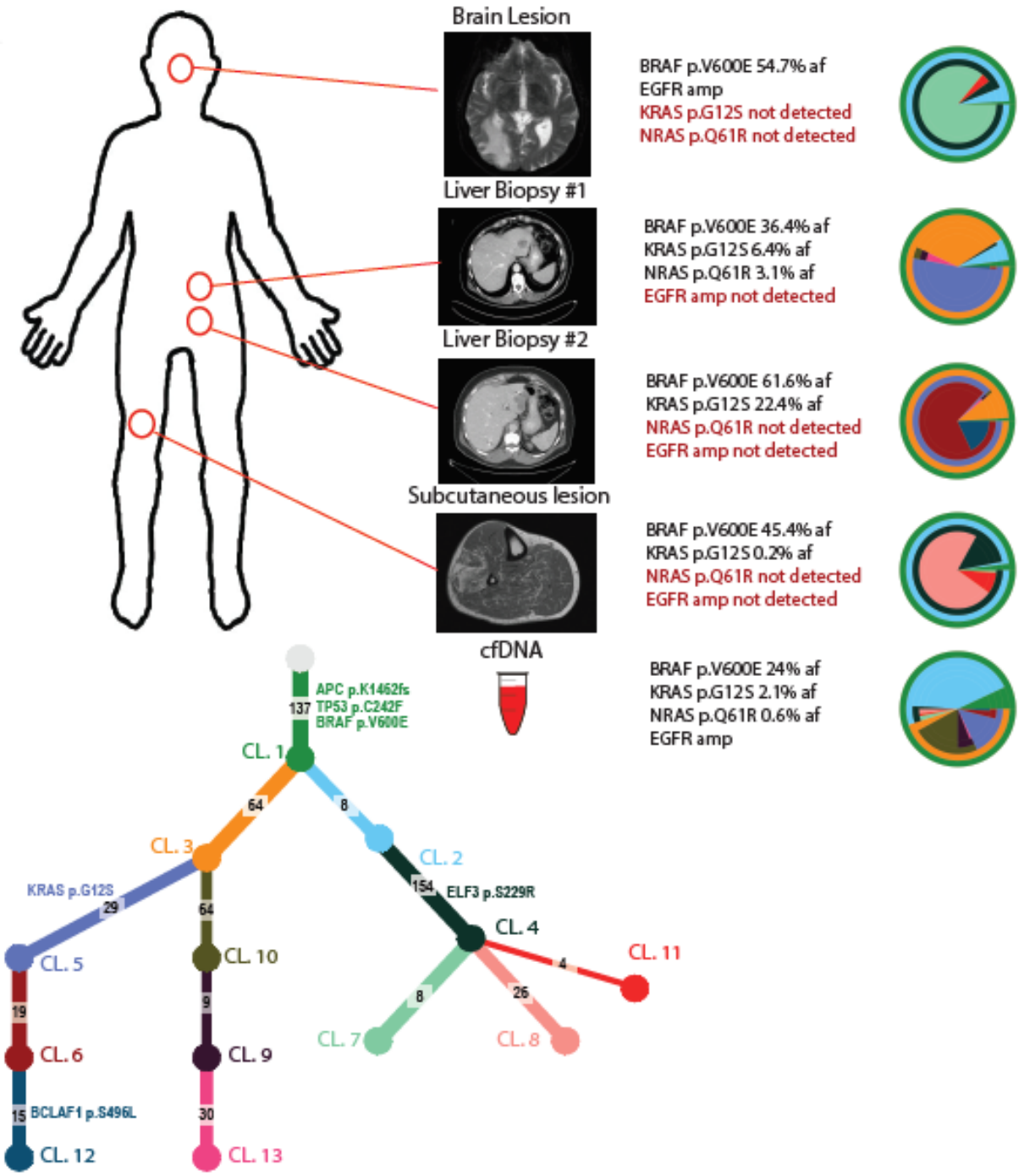


In 23 patients with matched tumor biopsies, ctDNA identified additional resistance mechanisms in 78% of cases

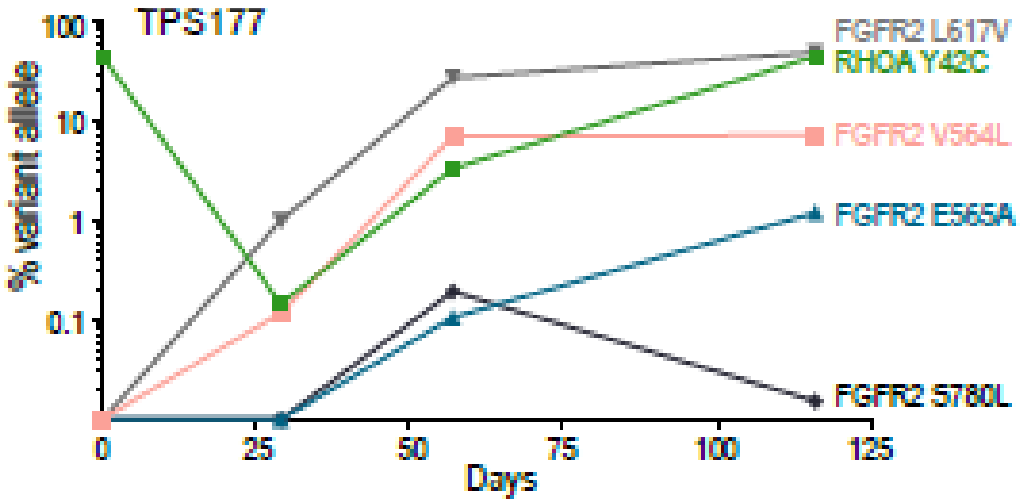
IN COLLABORATION WITH GADDY GETZ

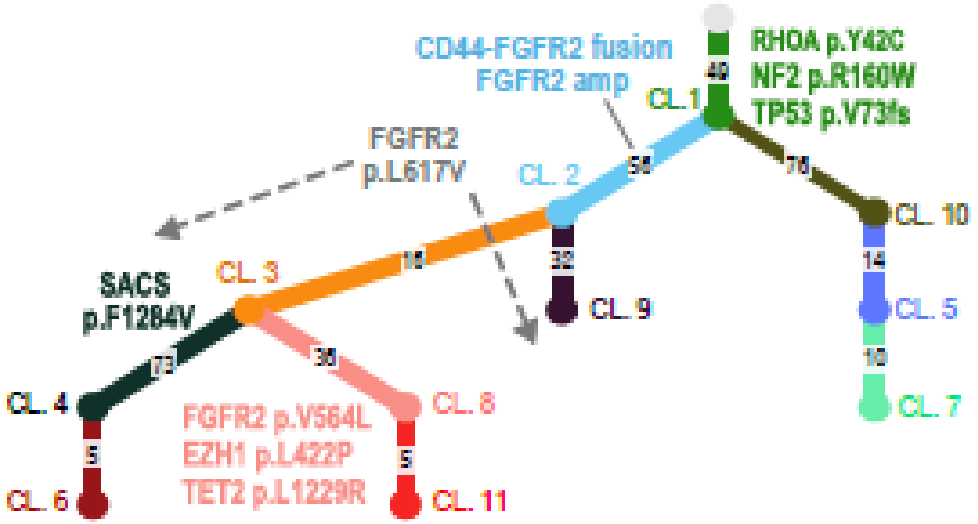
Parikh*, Leshchiner*, Elagina* et al, Nature Medicine 2019

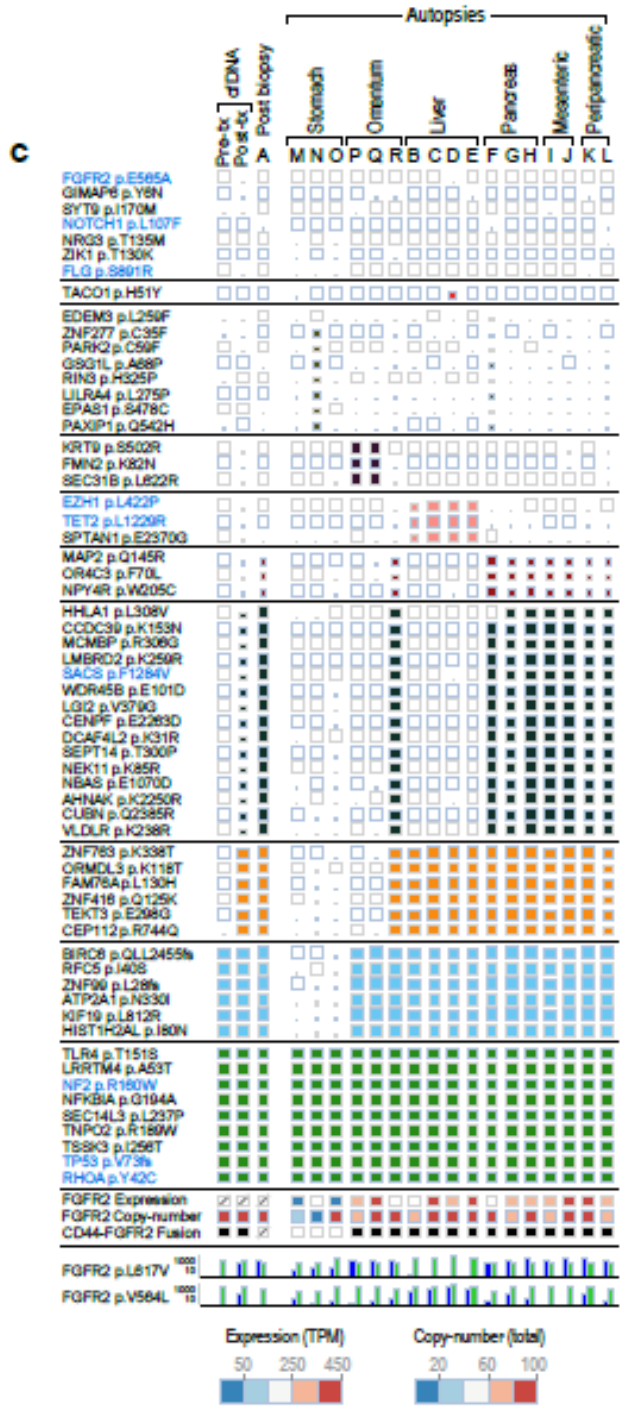
Comparison of multiple tumor biopsies versus liquid biopsies in a BRAF-mutant colorectal cancer patient



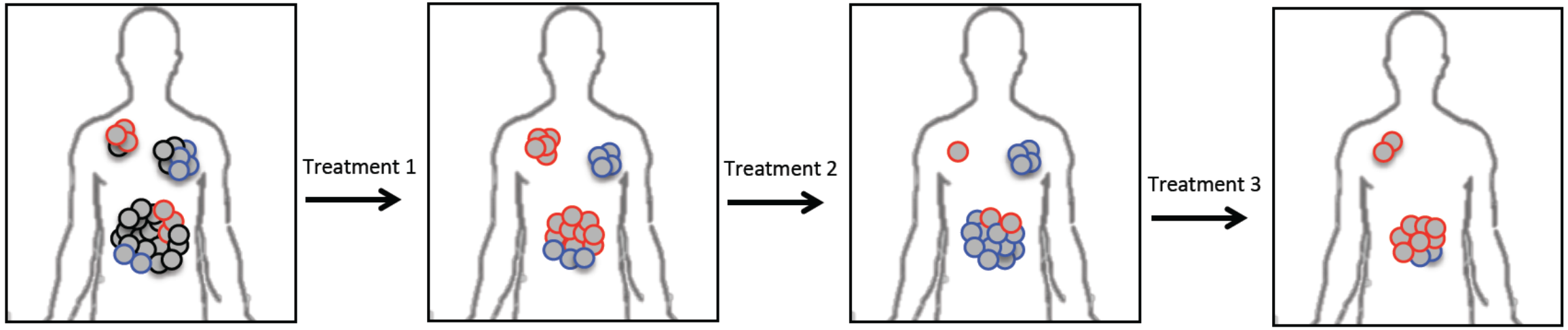
Serial liquid biopsy and autopsy in a patient with a FGFR-2 fusion positive gastric cancer

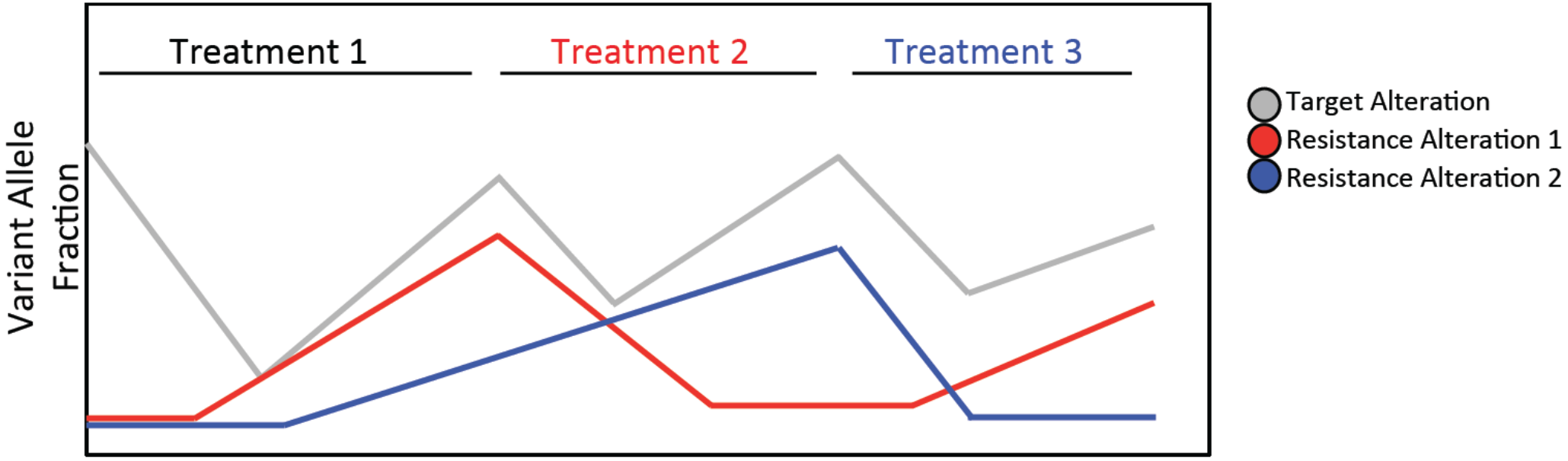




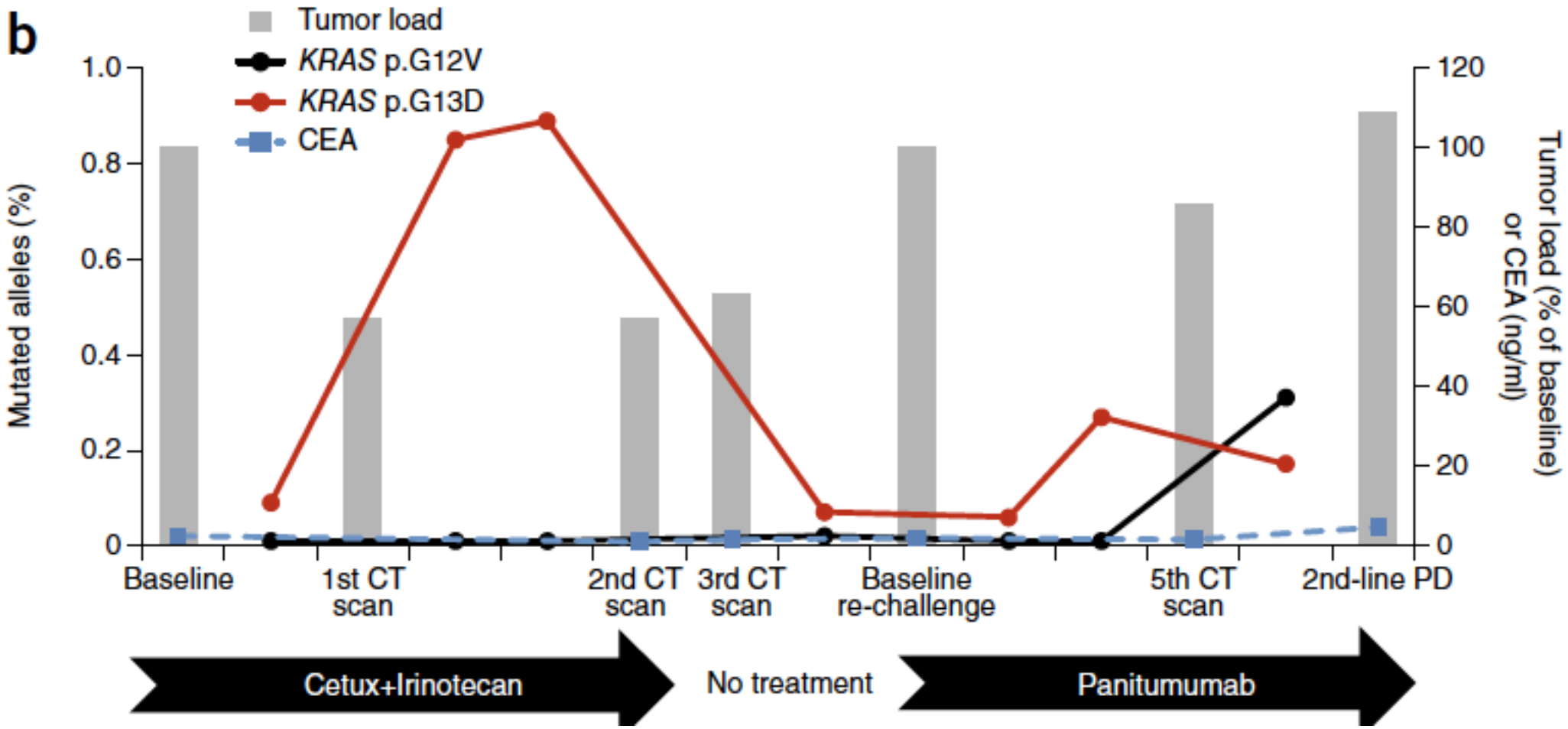

c
Autopsies
Pre-tx
Post-tx
cfDNA
Post biopsy
Stomach
Omentum
Liver
Pancreas
Mesenteric
Peripancreatic
A M N O P Q R B C D E F G H I J K L
FGFR2 p.E565A
GIMAP6 p.Y6N
SYT9 p.I170M
NOTCH1 p.L107F
NR0B3 p.T135M
ZIK1 p.T130K
FLG p.S891R
TACO1 p.H51Y
EDEM3 p.L259F
ZNF277 p.C35F
PARK2 p.C59F
GSG1L p.A88P
RIN3 p.H325P
ULRA4 p.L275P
EPAS1 p.S478C
PAXIP1 p.Q542H
KRT9 p.S502R
FMN2 p.K82N
SEC31B p.L822R
EZH1 p.L422P
TET2 p.L1229R
SPTAN1 p.E2370G
MAP2 p.Q145R
OR4C3 p.F70L
NPY4R p.W205C
HHLA1 p.L308V
CCDC39 p.K153N
MCMBP p.R306G
LMBRD2 p.K259R
SACS p.F1284V
WDR45B p.E101D
LGI2 p.V379G
CENPF p.E2283D
DCAF4L2 p.K31R
SEPT14 p.T300P
NEK11 p.K85R
NBAS p.E1070D
AHNAK p.K2250R
CUBN p.Q2385R
VLDLR p.K238R
ZNF763 p.K338T
ORMDL3 p.K118T
FAM76A p.L130H
ZNF416 p.Q125K
TEKT3 p.E298G
CEP112 p.R744Q
BIRC6 p.QLL2455fs
RFC5 p.I40S
ZNF99 p.L28fs
ATP2A1 p.N330I
KIF19 p.L812R
HIST1H2AL p.I80N
TLR4 p.T151S
LRRTM4 p.A53T
NF2 p.R160W
NFKBIA p.G194A
SEC14L3 p.L237P
TNPO2 p.R189W
TSSK3 p.I256T
TP53 p.V73fs
RHOA p.Y42C
FGFR2 Expression
FGFR2 Copy-number
CD44-FGFR2 Fusion
FGFR2 p.L617V
FGFR2 p.V564L
Expression (TPM)
50 250 450
Copy-number (total)
20 60 100
d
Liver
Stomach
Pancreas
Post Biopsy (paraaortic node)
Peripancreatic nodes
Mesenteric nodes
Omentum nodes
cfDNA pretx
cfDNA posttx
Post Biopsy (A)
Autopsy B
Autopsy C
Autopsy D
Autopsy E
Autopsy F
Autopsy G
Autopsy H
Autopsy I
Autopsy J
Autopsy K
Autopsy L
Autopsy M
Autopsy N
Autopsy O
Autopsy P
Autopsy Q
Autopsy R

Real-time adaptation of therapy guided by ctDNA





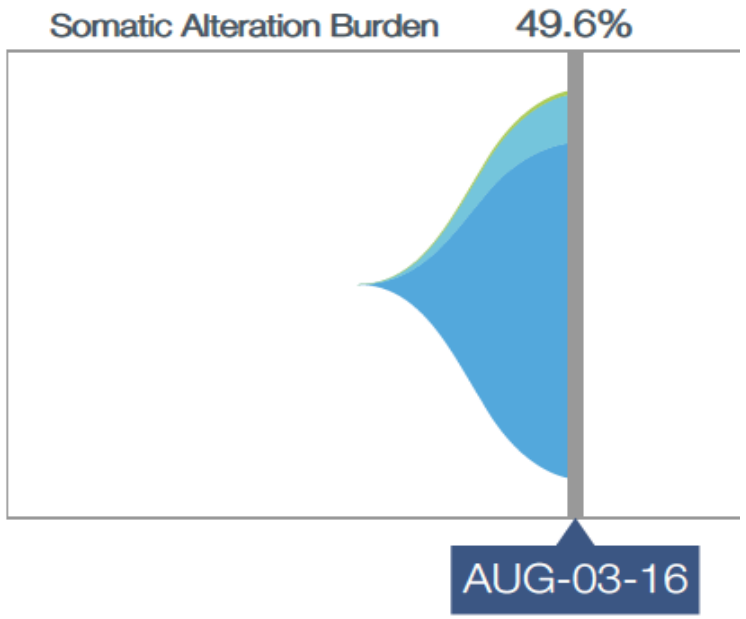
Real-time adaptation of therapy guided by ctDNA



Siravegna et al, Nature Medicine 2015

“Re-challenge” with anti-EGFR therapy in CRC

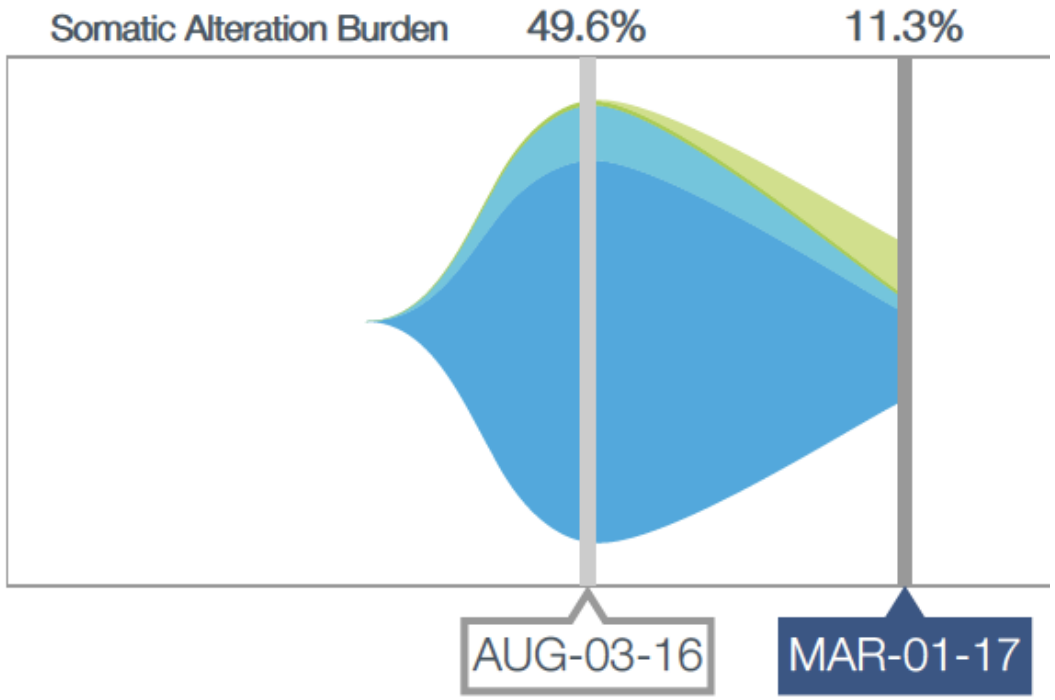
- 54M w metastatic CRC. 1st line FOLFOX bevacizumab for 15 months.
- Molecular testing revealed RAS wild type
- Received irinotecan + cetuximab for 16 months



TP53 R282W 49.6%
KRAS amp 2+
HER2 amp 1+

Case: “Re-challenge” with anti-EGFR therapy in CRC

- Received local ablation, followed by 3 months of Trifluridine/tipiracil

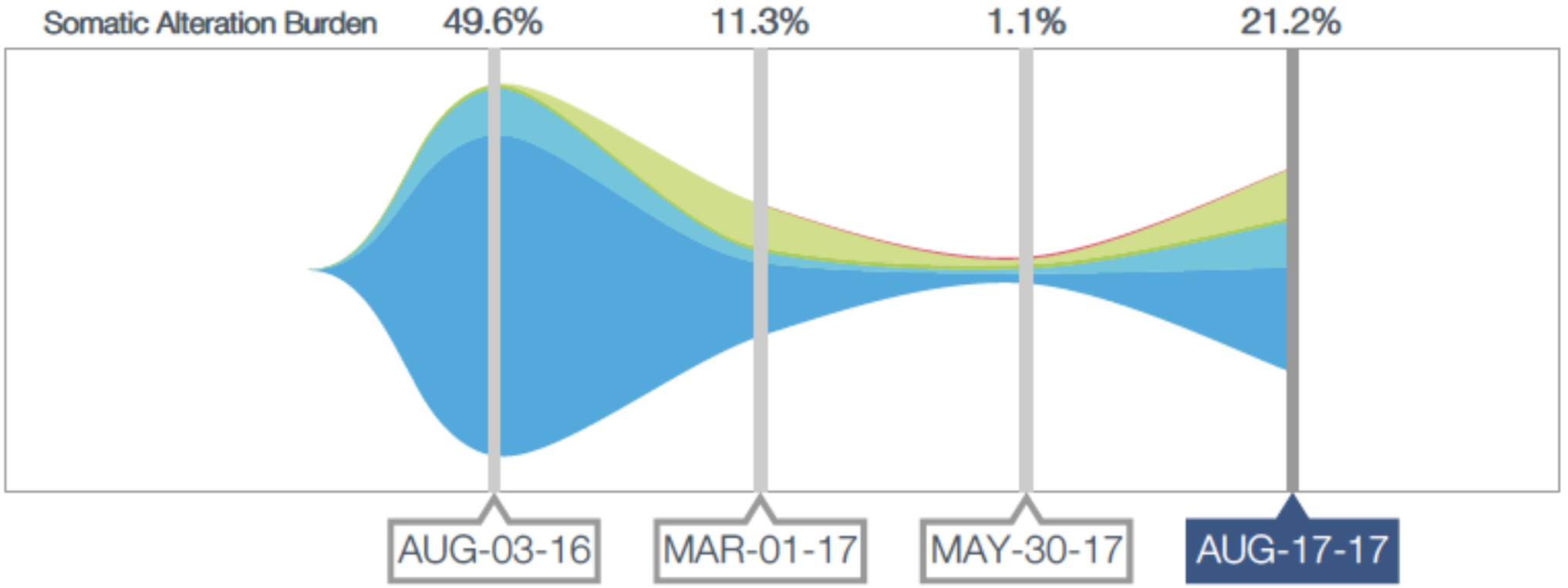


TP53 R282W 49.6%
KRAS amp 2+
HER2 amp 1+

TP53 R282W 11.3%
No KRAS amp
No HER2 amp

Case: “Re-challenge” with anti-EGFR therapy in CRC

- Received irinotecan + panitumumab. Response lasting ~6 months.



TP53 R282W 49.6%
KRAS amp 2+
HER2 amp 1+

TP53 R282W 11.3%
No KRAS amp
No HER2 amp

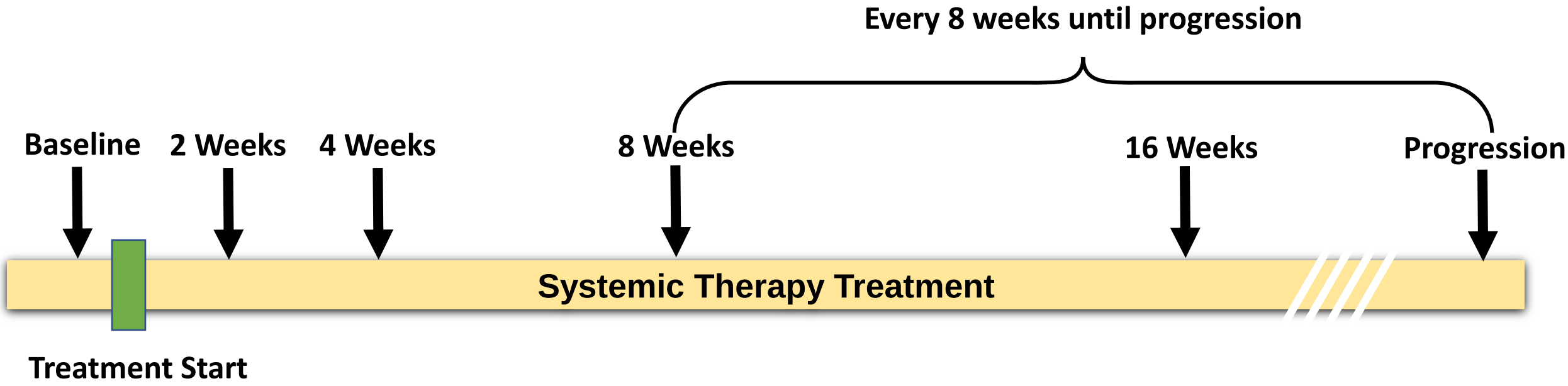
TP53 R282W 21.2%
No KRAS amp
No HER2 amp

Conclusions

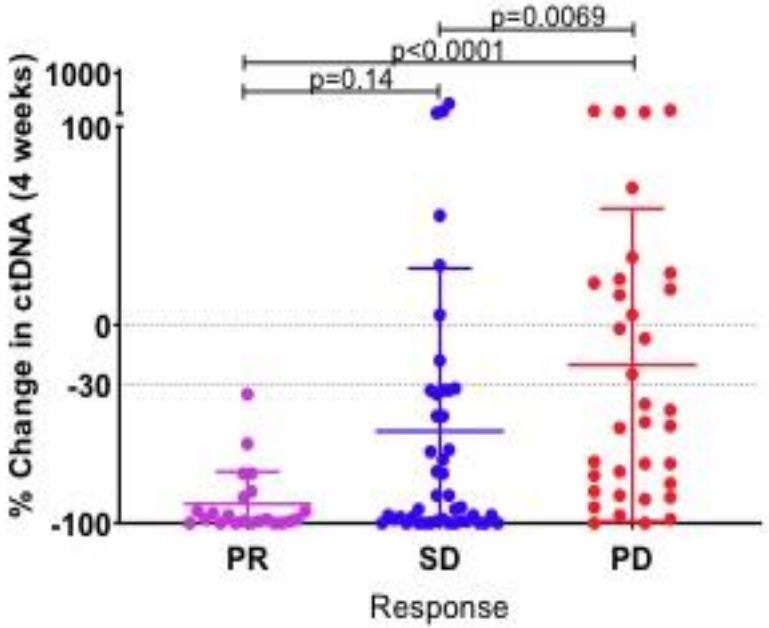
- Molecular profiling through ctDNA can be used to guide treatment decisions, particularly when inadequate tissue is available
- Diverse mechanisms can promote resistance in different patients AND in the same patient
- Single-lesion biopsies may not capture heterogeneity of resistance, missing alterations that might drive treatment failure
- Integrating liquid biopsy into clinical-decision making may be key to developing strategies to overcome heterogeneity of resistance
- Liquid biopsy may offer the ability to monitor emergence of resistance mechanisms in real-time and adjust therapy accordingly

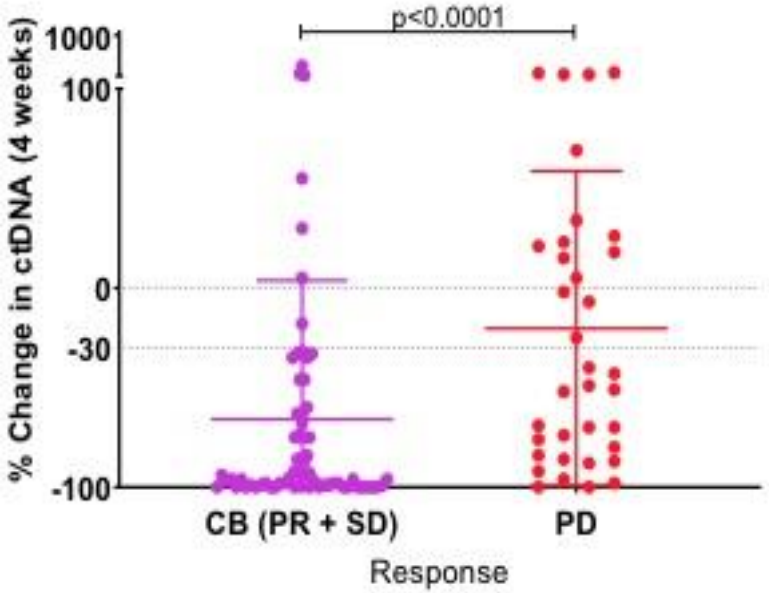
Treatment Response

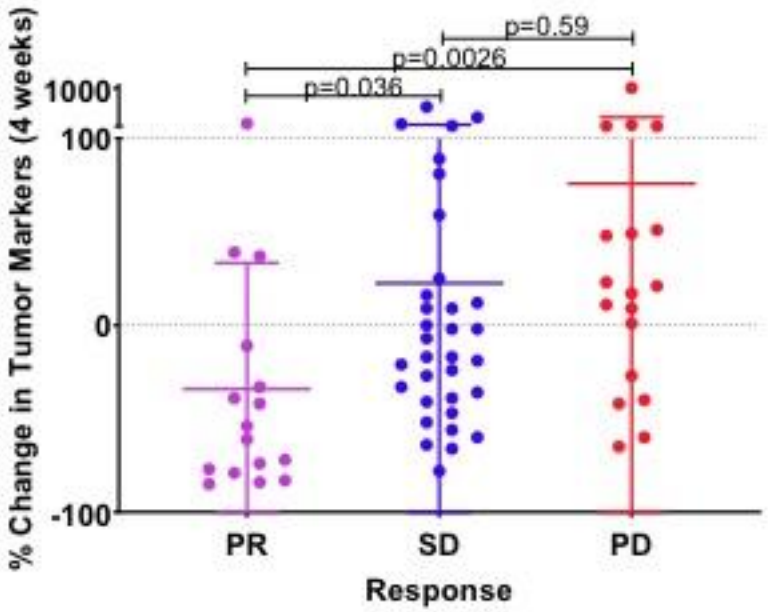
Serial ctDNA to predict treatment response

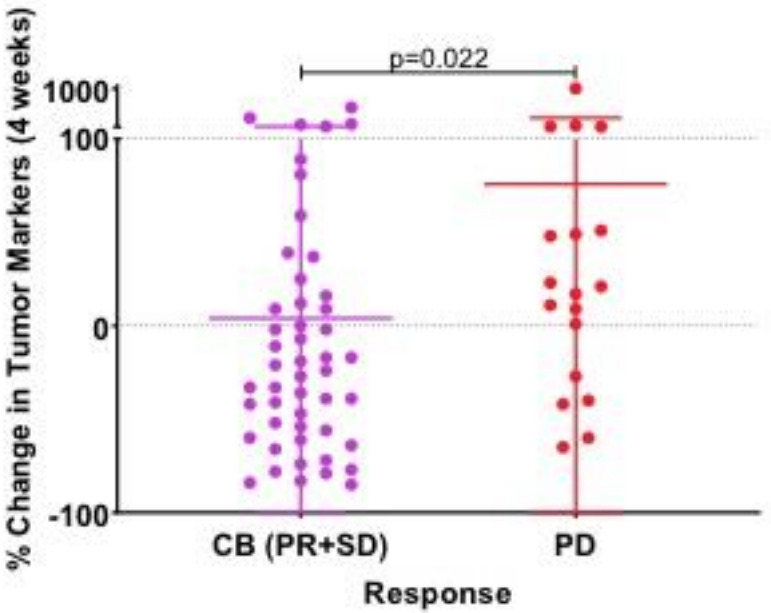


Change in ctDNA and Tumor Markers at 4 weeks

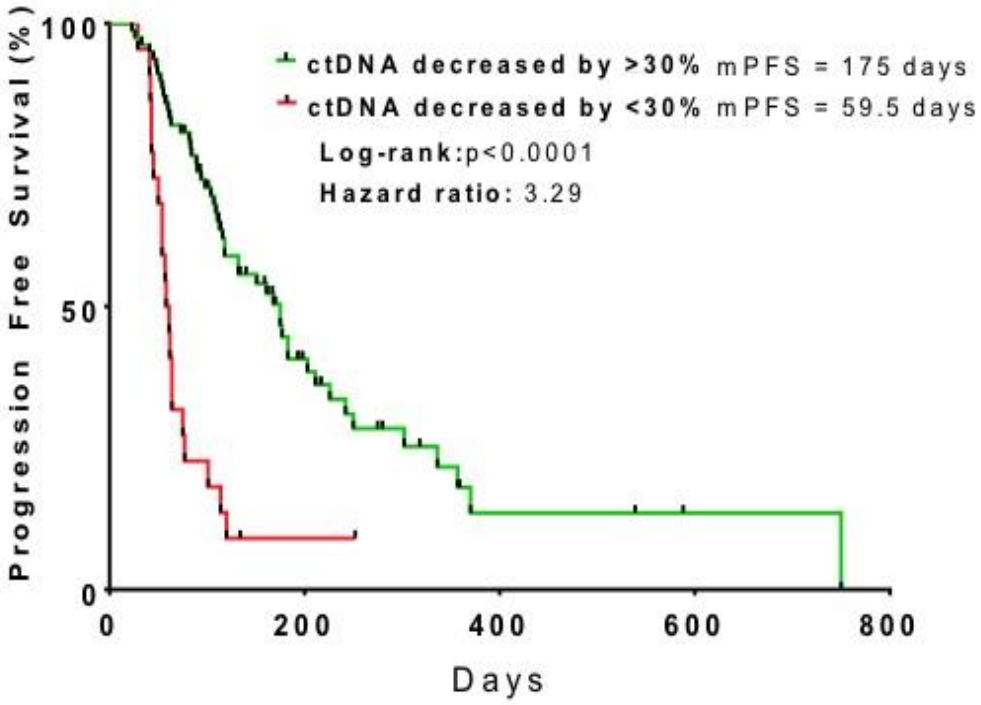


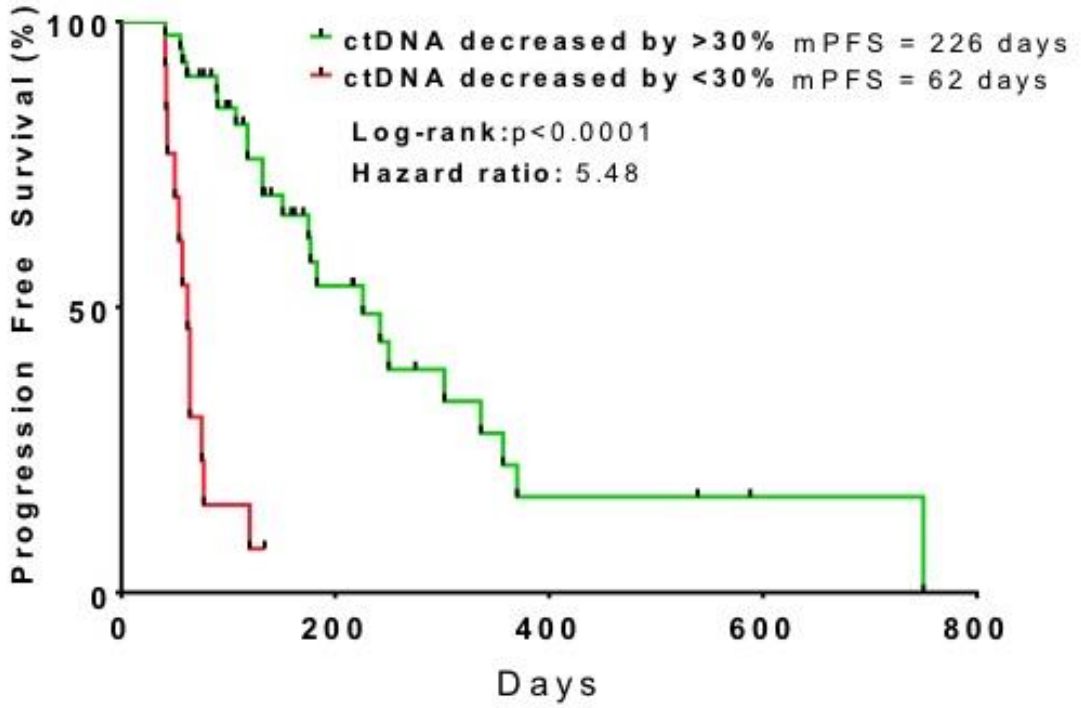




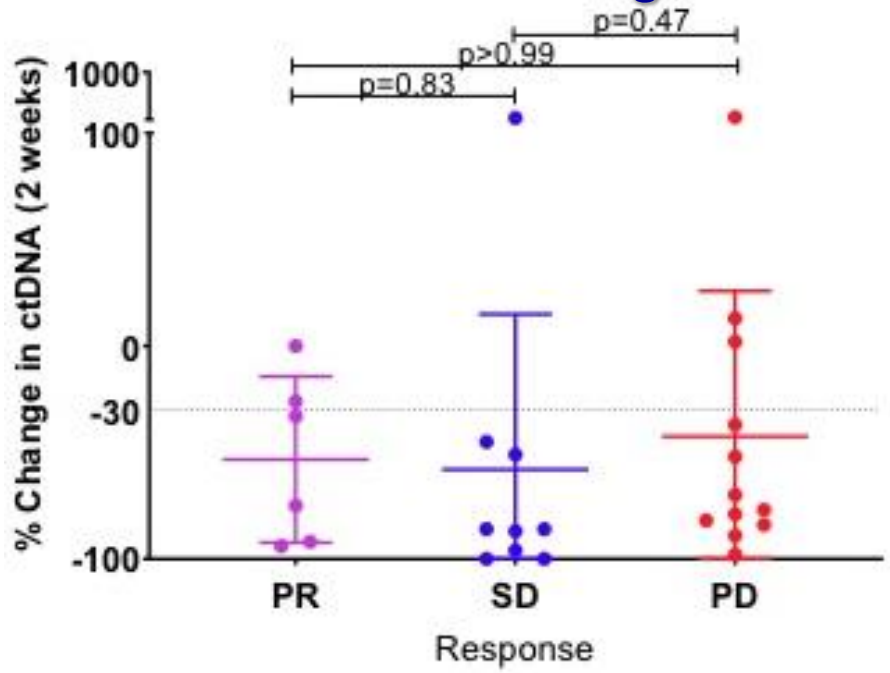


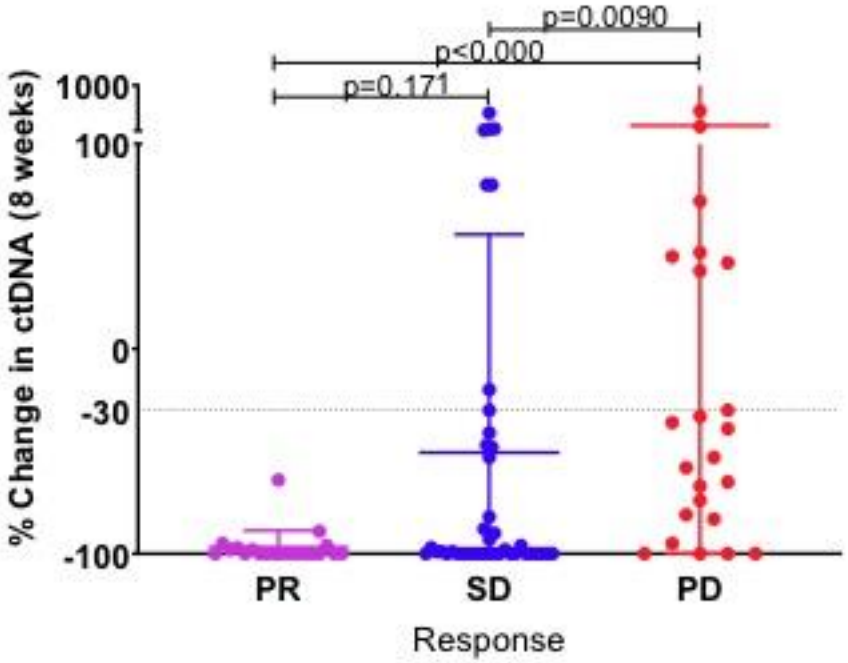
Progression-Free Survival by % Change in ctDNA

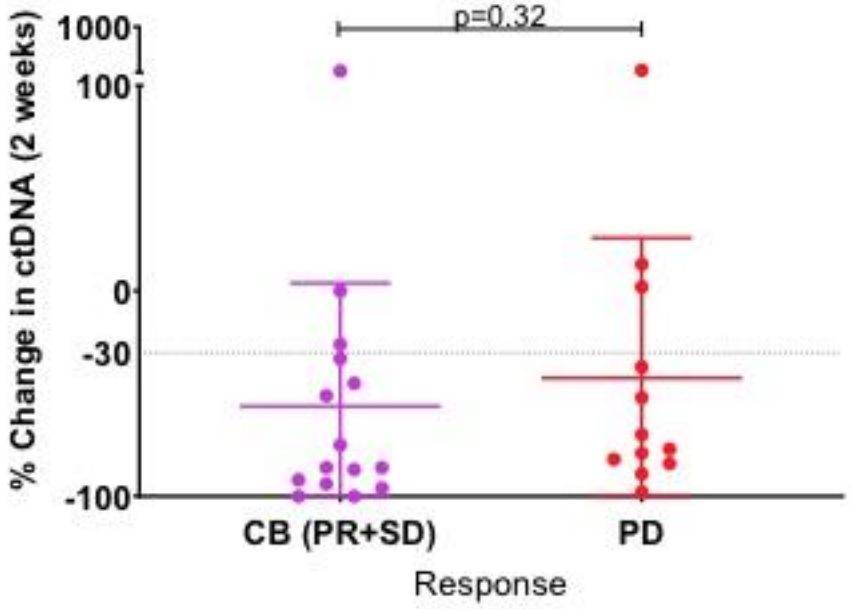


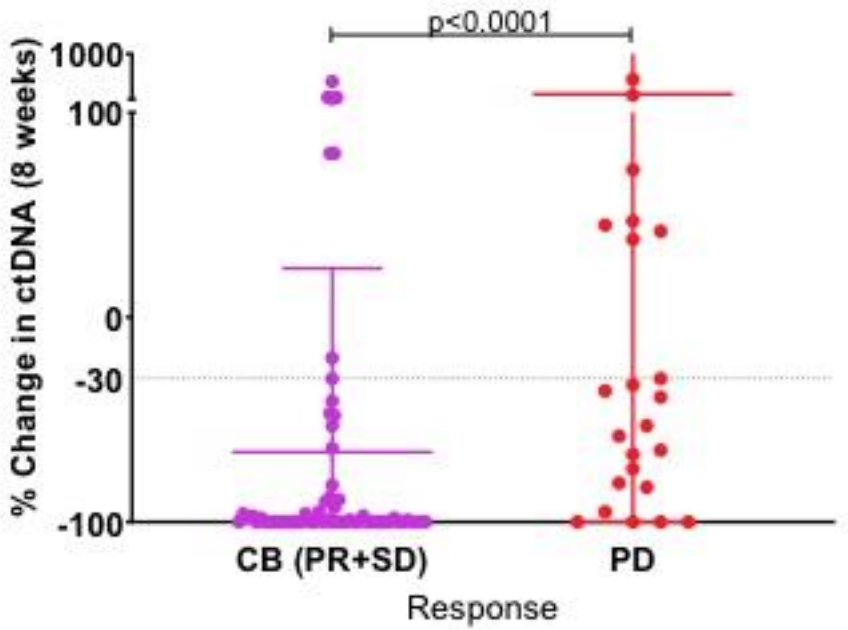


Change in ctDNA and tumor markers over time

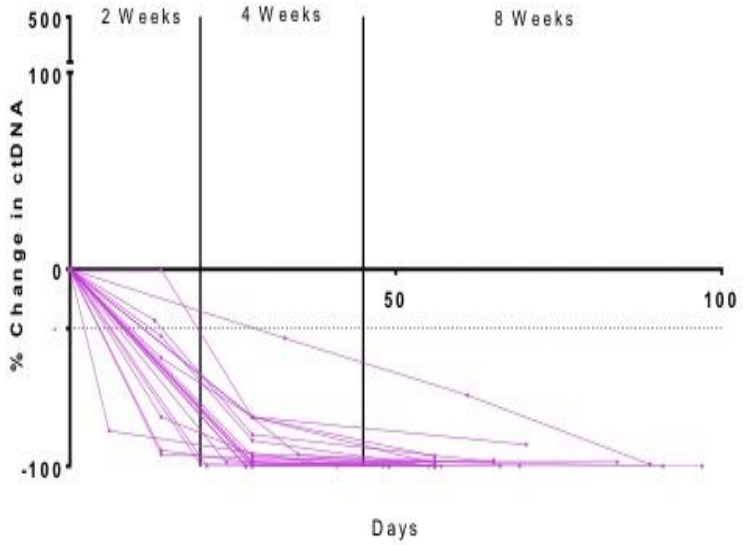


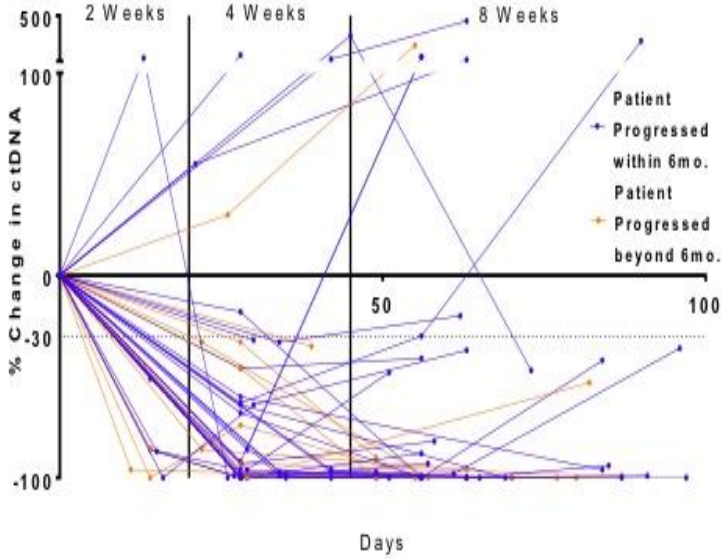


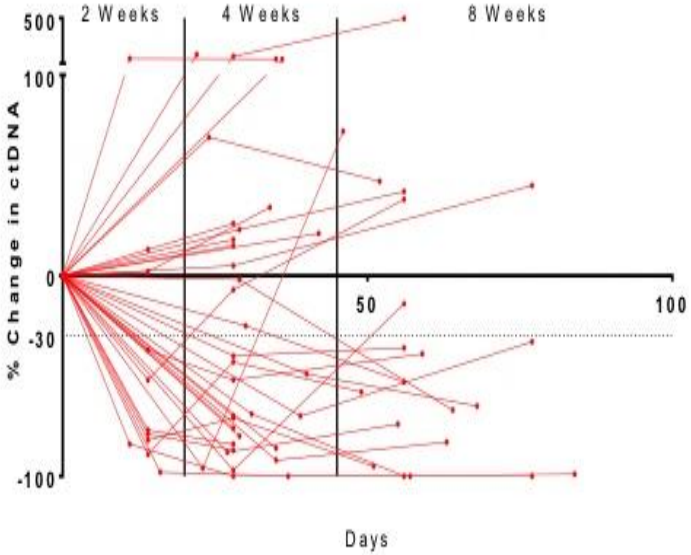




Longitudinal ctDNA changes during the first 100 days of therapy



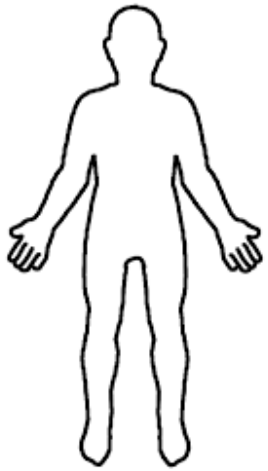




MRD

Residual disease: The Problem

Stage III CRC:
All patients get adjuvant chemo
>50% cured by surgery alone



Curative Intent Surgery

Negative

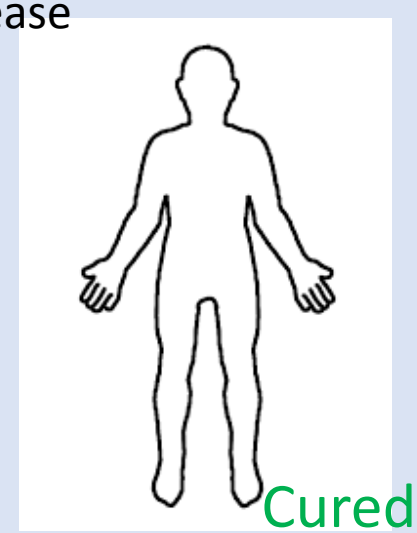
ctDNA

Positive

Stage II CRC:
SOC is NO adjuvant chemo
10-15% of patients recur

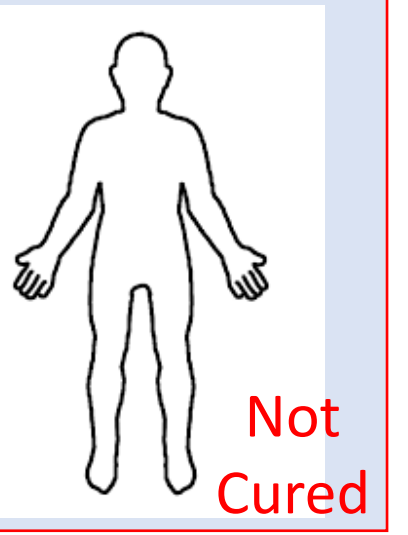
Minimal Residual Disease

None



Cured

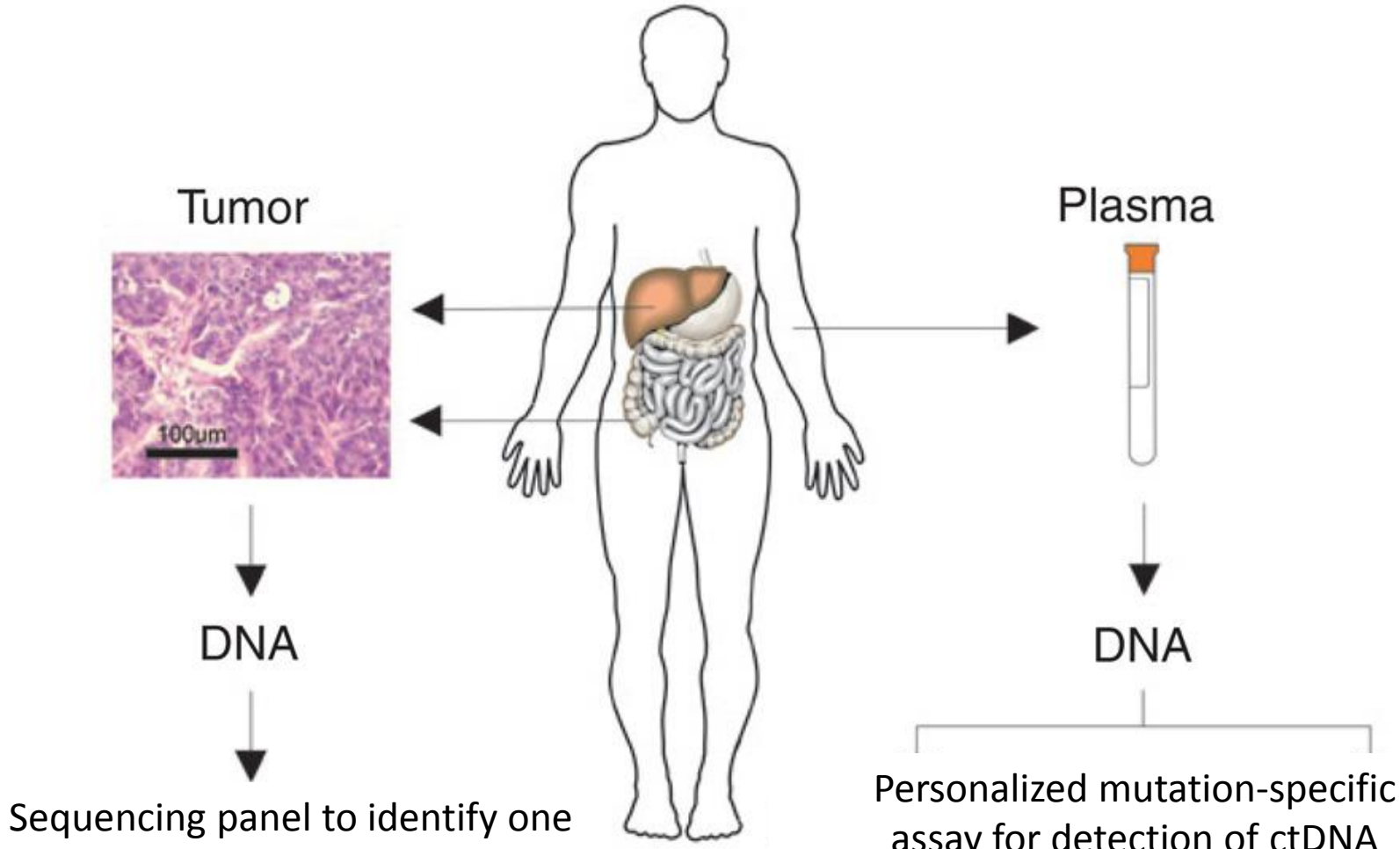
Present



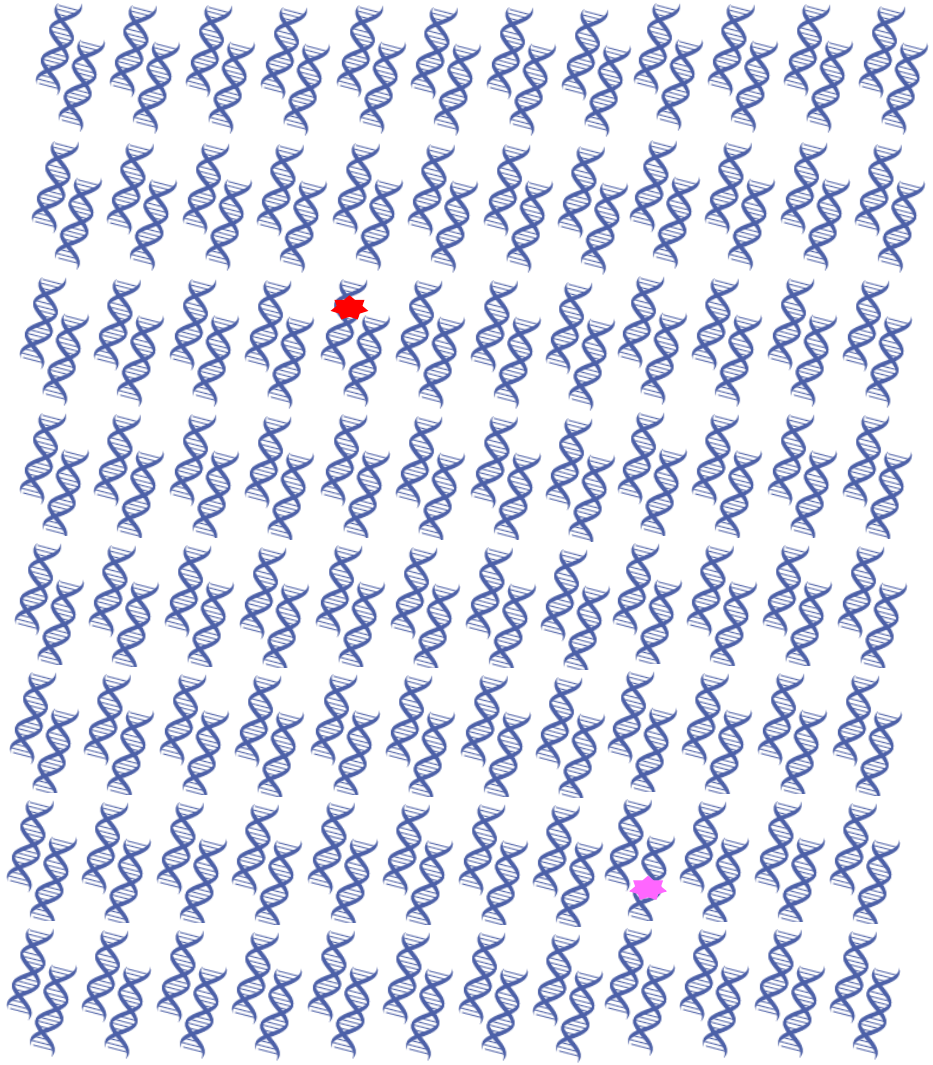
Not Cured

We have no way to determine who is cured and who will recur

Strategy for sensitive and specific detection of residual disease



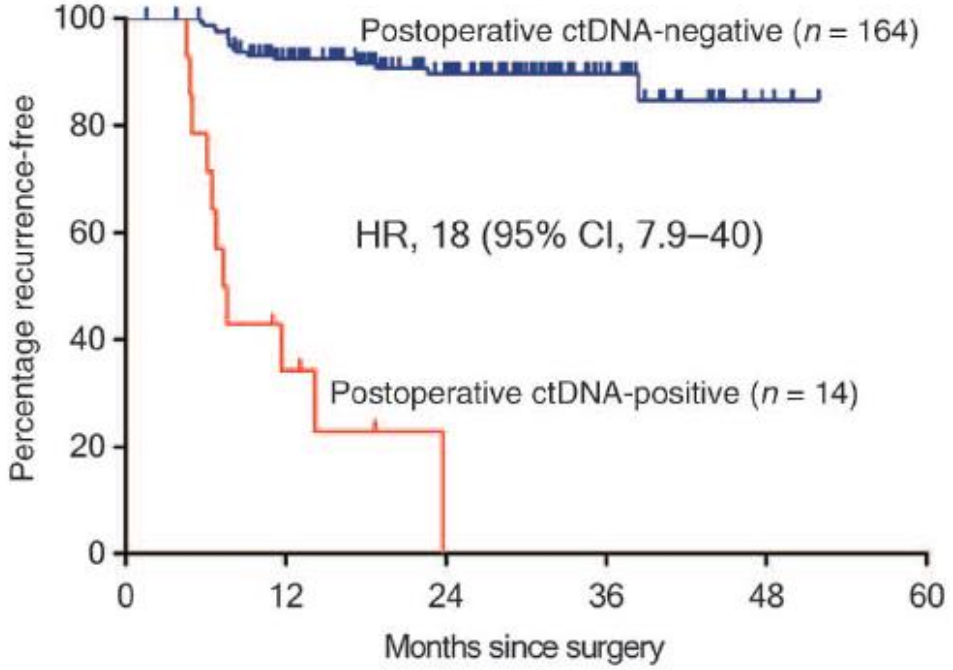
ctDNA detection requires high sensitivity and specificity



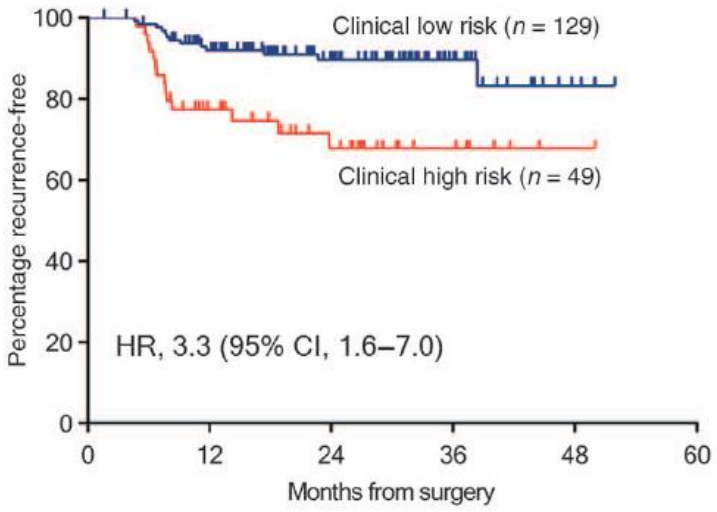
- SENSITIVITY: Trace amounts of ctDNA present in a background of normal cfDNA (~0.01%)
- SPECIFICITY: Need to have high confidence in detection methods to base treatment decisions on a positive result

Prediction of relapse in stage II CRC

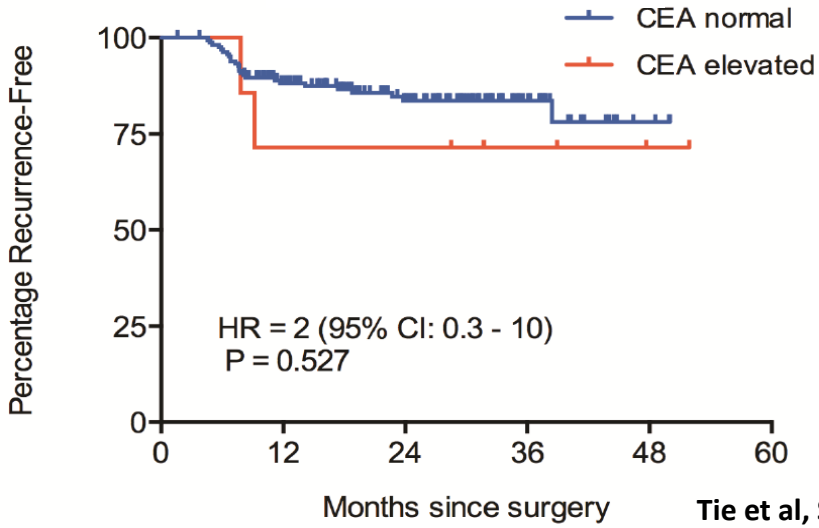
Post-operative ctDNA



Clinical Risk Factors

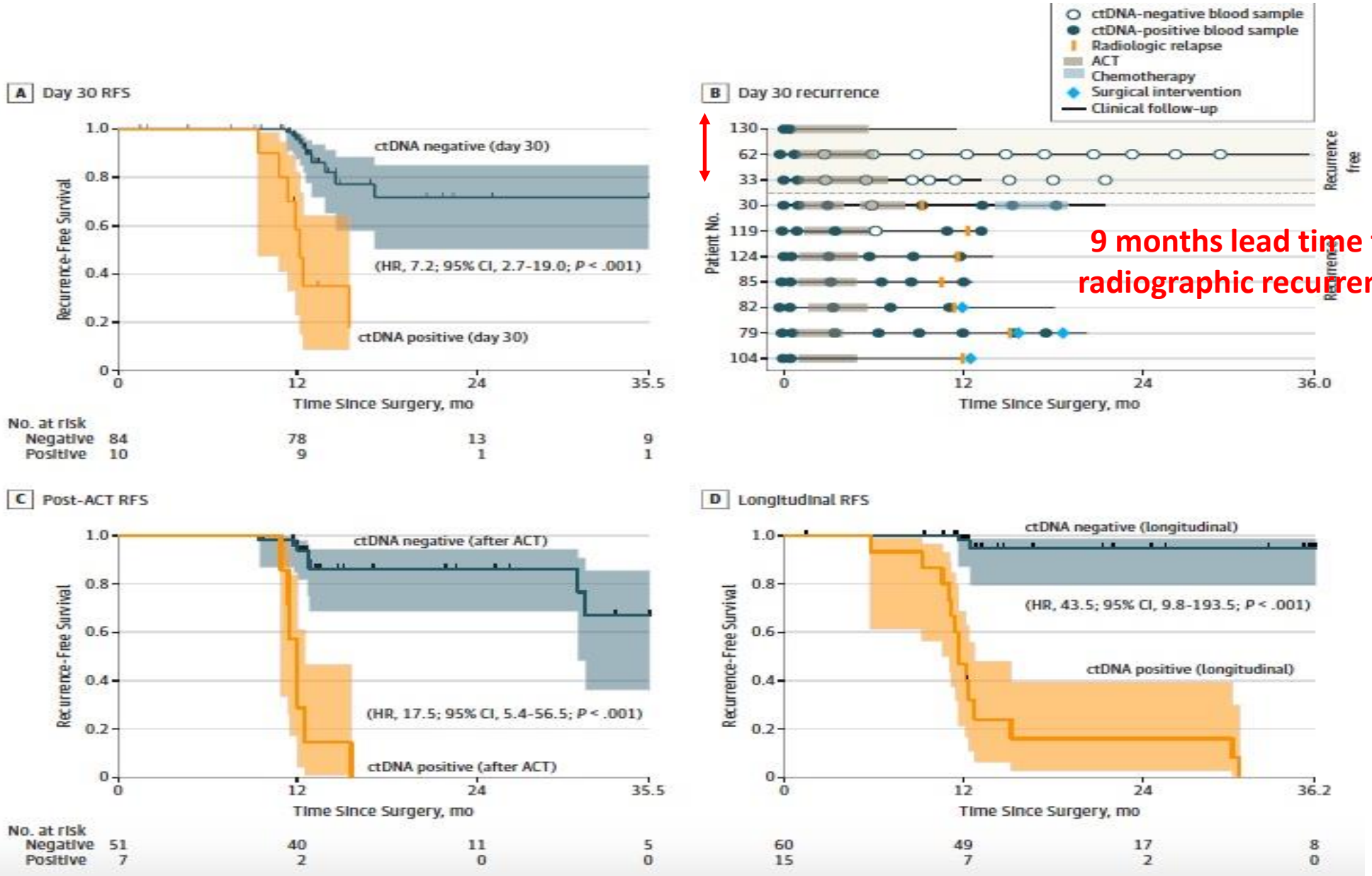


Post-operative CEA



Tie et al, STM 2016

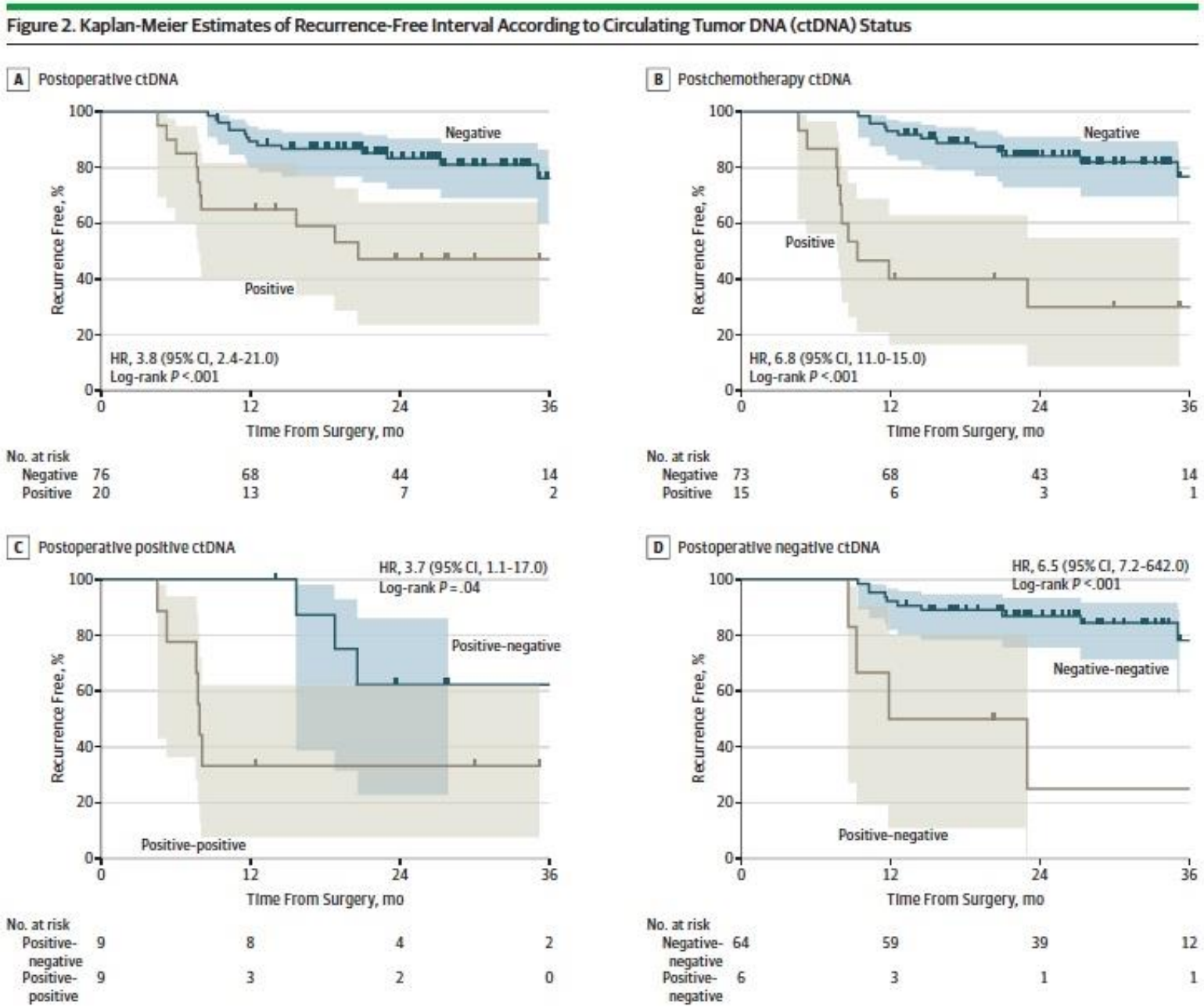
Prognostic but can it be a predictive biomarker???



Reinert et al
JAMA Onc 2019

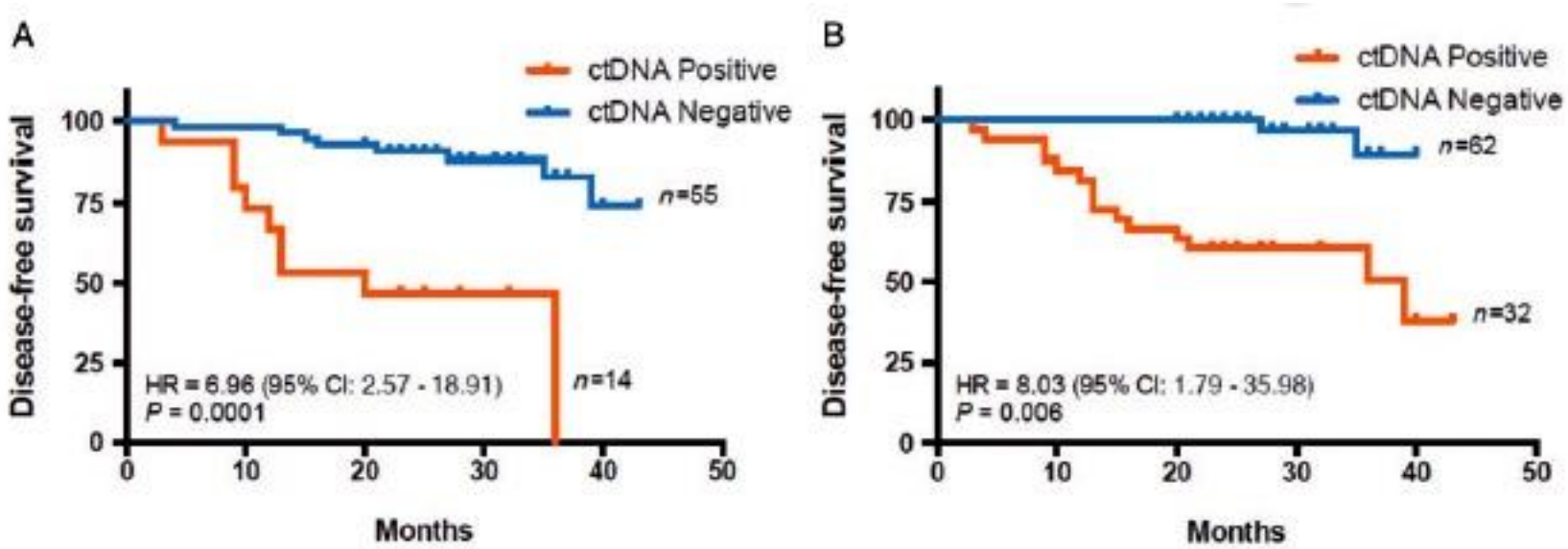
Efficacy of adjuvant chemotherapy

- ctDNA detectable in 20 of 96 (21%) post surgery
  - inferior recurrence-free survival
  - 47% vs 76% despite adjuvant
- ctDNA detectable in 15 of 88 (17%) post chemo
  - 3-year RFI
  - 30% when ctDNA was detectable after chemotherapy and 77% when ctDNA was undetectable



Serial tracking improves sensitivity

47% detectable with 1 post op draw

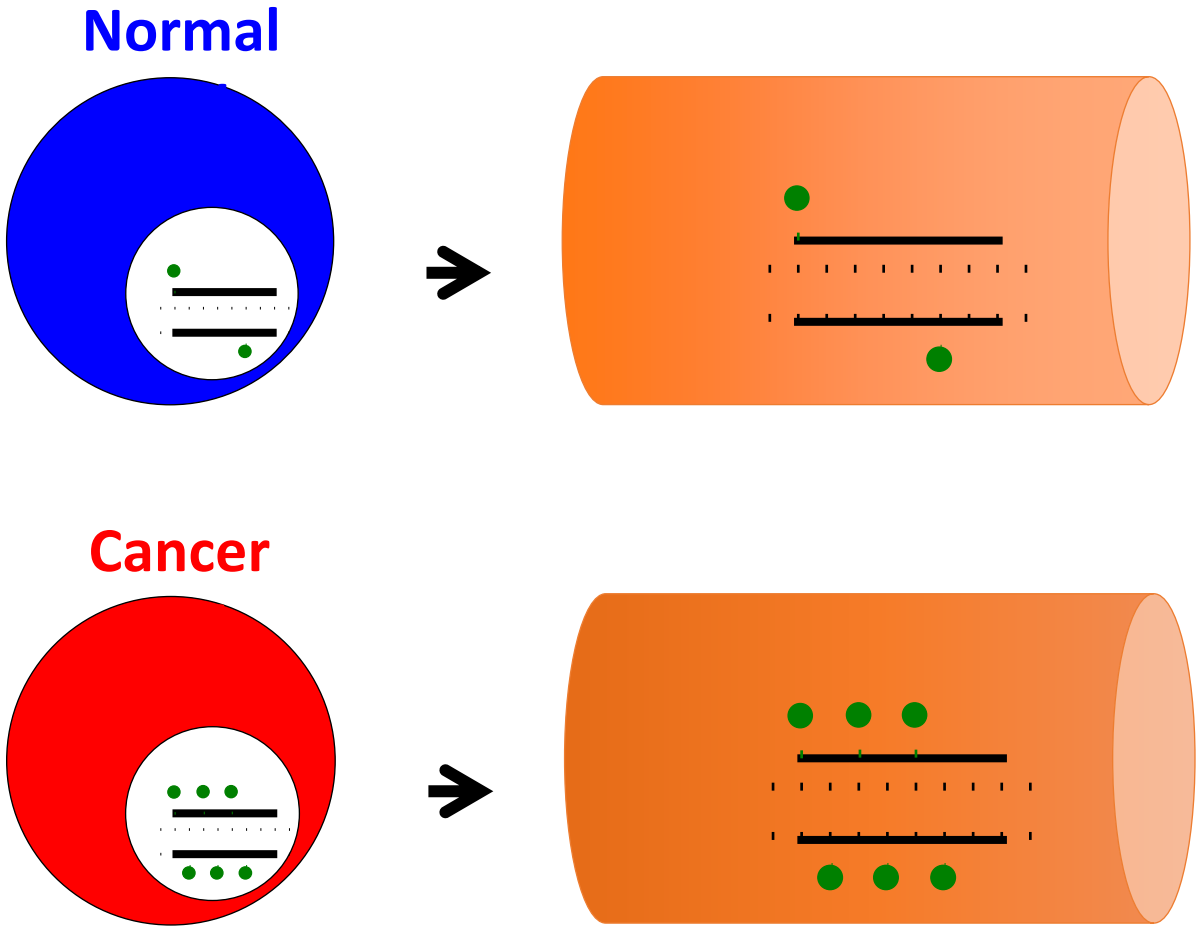


82% detectable with serial draws

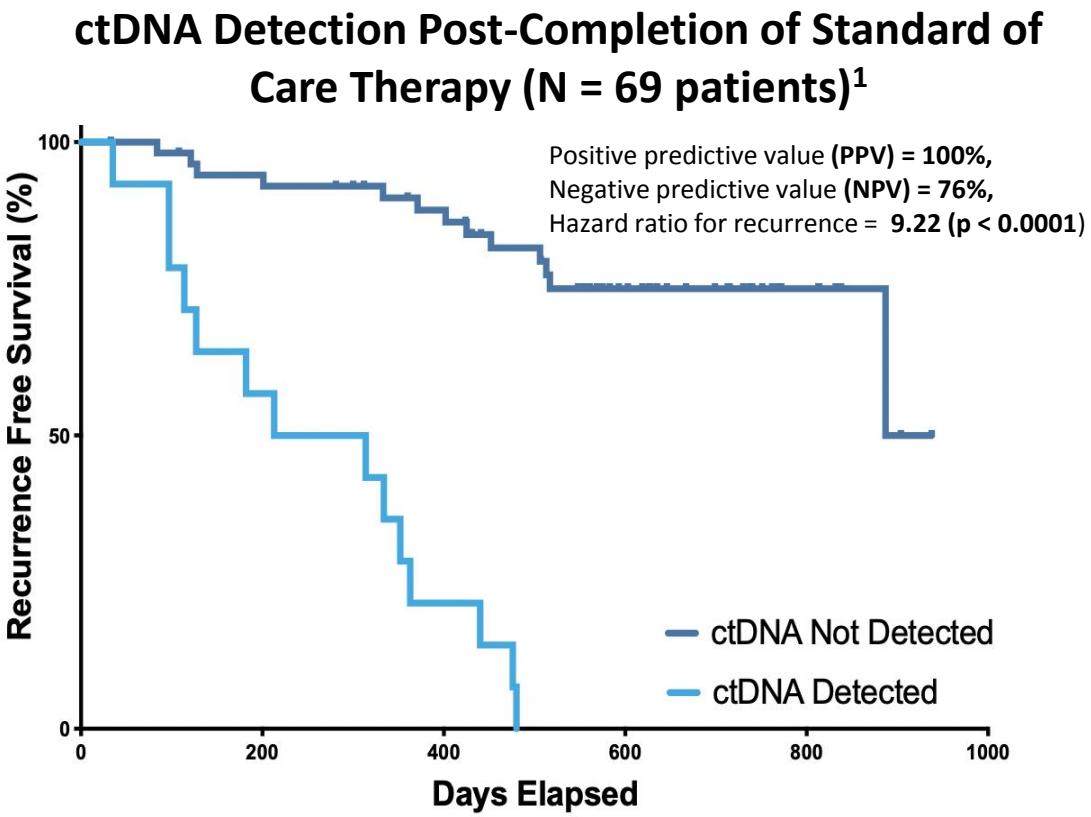
35% improvement

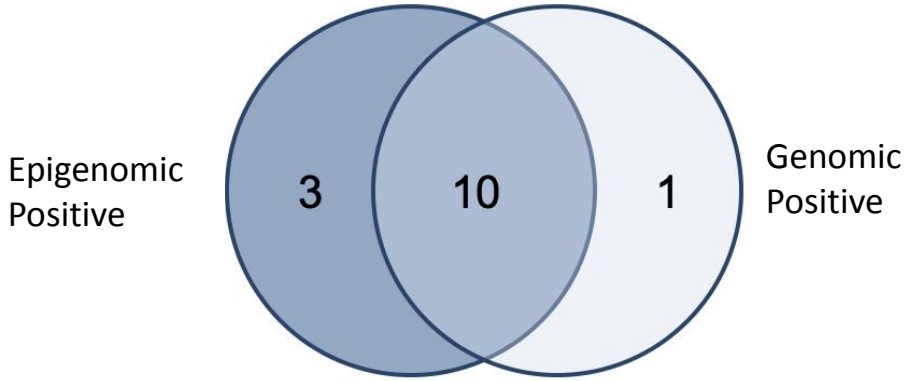
Integrated genomic and epigenomic plasma for recurrence prediction

Tumor-specific methylation patterns as a potential biomarker



Integrated assay has high PPV for CRC recurrence





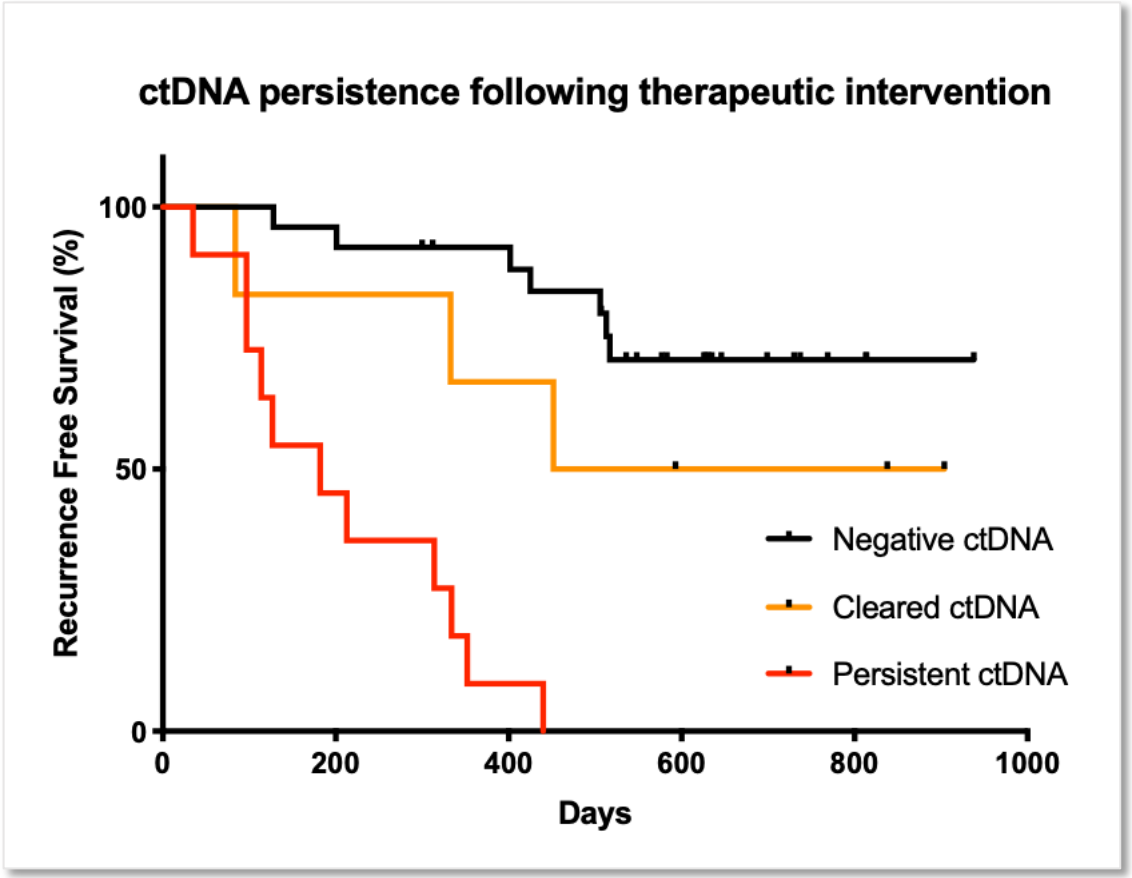
Assay Performance by Analysis		
	Genomic (N)	Integrated Genomic and Epigenomic (N)
PPV (N of patients with ctDNA detected who recurred)	100% (11 / 11)	100% (14 / 14)
NPV (N of patients with ctDNA not detected who were recurrence free)	72% (42 / 58)	76% (42 / 55)
Sensitivity for recurrence within one year of surgery	56% (9 / 16)	69% (11 / 16)
Specificity for recurrence within one year of surgery	96% (51 / 53)	94% (50 / 53)

[2]Parikh, et al (2019). JCO. 37 (suppl abstr 3602)

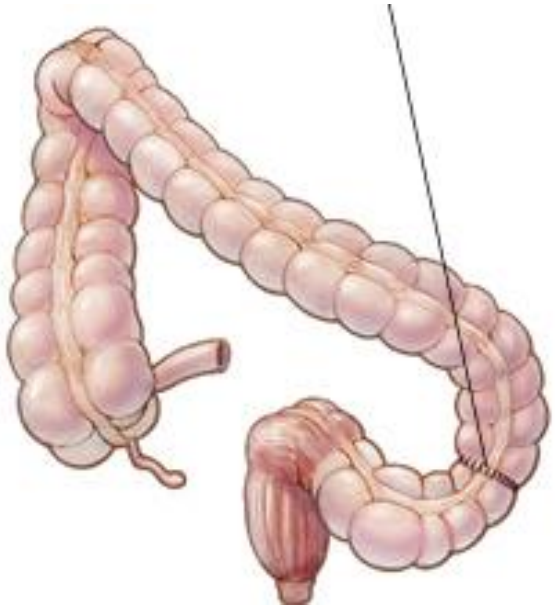
Parikh (Corcoran). 2019. ESMO World GI Congress. Abstract #O-016.

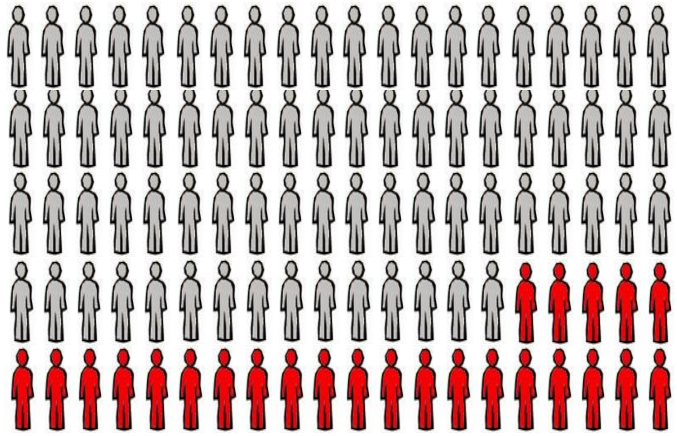
Patients with persistent ctDNA were significantly more likely to experience disease recurrence in a shorter time period

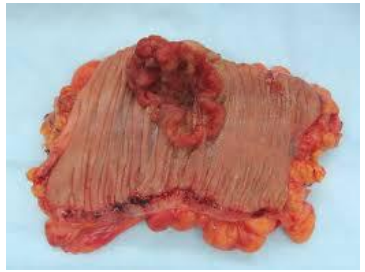
	Recurred	Recurrence Free	Median Time to Recurrence (days)
Persistent ctDNA	11	0	182
Cleared ctDNA	3	3	333
Negative ctDNA	7	19	NR* (median follow-up: 580 days)

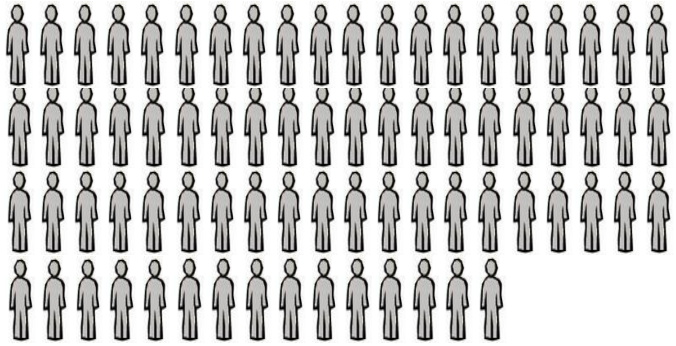


Detecting residual disease with circulating tumor DNA

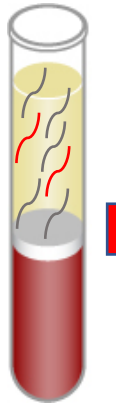


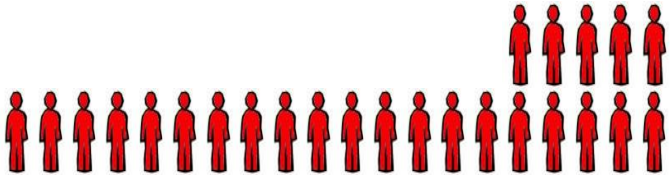






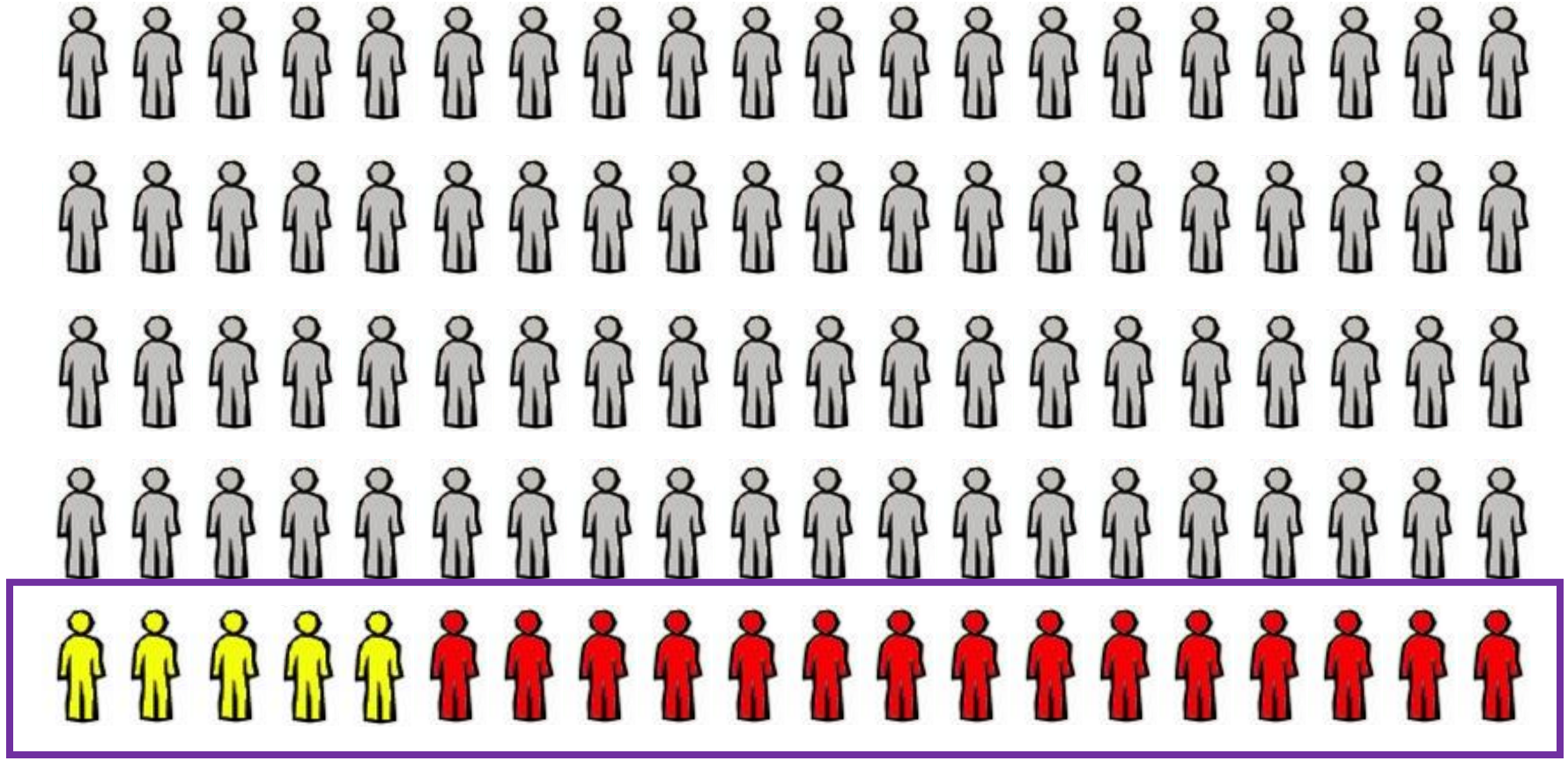
Observation only





Additional therapy

Potential to transform adjuvant therapy

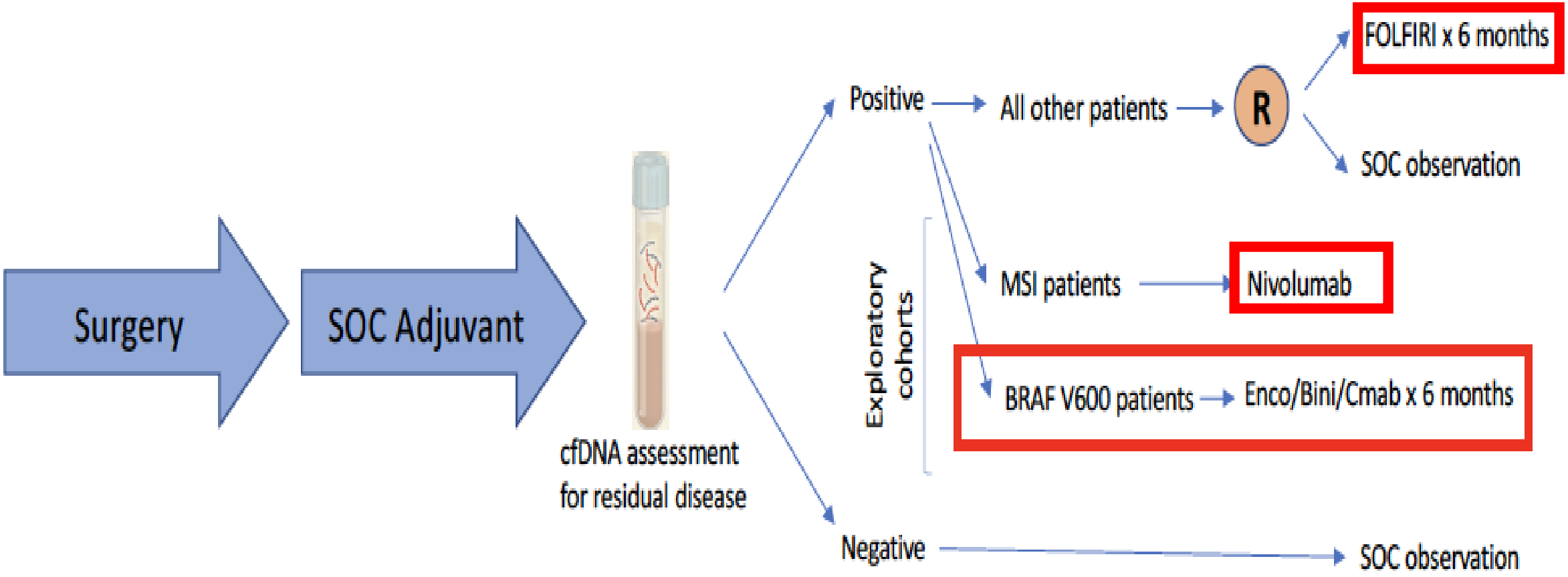


Residual disease, tumor recurrence

Cured with surgery alone

Cured with adjuvant therapy

SU2C CRC Dream Team: Early treatment of occult metastatic disease following standard adjuvant therapy in stage III CRC



STAND UP TO CANCER

MASSACHUSETTS GENERAL HOSPITAL
CANCER CENTER

Acknowledgements

*First and foremost the patients and their families

MGH Center for GI Cancers

- ***Ryan Corcoran
- Ted Hong
- David Ting
- David Ryan
- Larry Blaszkowsky
- Jeff Clark
- Lipika Goyal
- Jill Allen
- Ryan Nipp
- Bruce Giantonio
- Colin Weekes
- Jennifer Wo
- Sam Klepmner

MGH Center for Melanoma

- Genevieve Boland

MGH Pathology

- John Iafrate
- Dora Dias-Santagata
- Joe Lennerz
- Nicholas Jessop

MGH GI Research Assistant Team

- Emily Van Seventer GI Program Director
- Madeline Fish
- Katie Kanter

Other Mentors

- Alan Venook, UCSF
- ***Bruce Chabner

Funding

- NIH/NCI DF/HCC GI SPORE
- ACS
- ASCO CDA
- SU2C Colorectal Dream Team Award
- Gateway Foundation