Molecular Tumor Board

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University of Wisconsin- Madison

Disclosures

M.E.B. declares the following:

- Strata Oncology **(Medical Advisory Board)**
- Abbvie, Arcus, Apollomics, Elevation Oncology, Genentech, Puma, Loxo Oncology, Seagen **(Research funding)**
- Novartis, Strata Oncology **(Consultant)**

Many vendors

Genomics (somatic)

Tempus
Strata Oncology
Foundation Medicine
Caris
Guardant

Expression (somatic)

Exact Sciences/Genomic Health
Agendia
Prosigna
BioTheranostics

Germline

Myriad
Prevention Genetics
Color Genetics
Ambry Genetics

M. Burkard, unpublished

Many vendors

Genomics (somatic)

Tempus
Strata Oncology
Foundation Medicine
Caris
Guardant

M. Burkard, unpublished

Topics

1. How Next-Generation Sequencing (NGS) tests work
2. Differences in commercial NGS tests
3. What to do?
4. What's next?

Topics

1. **How Next-Generation Sequencing (NGS) tests work**
2. Differences in commercial NGS tests
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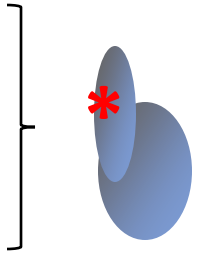
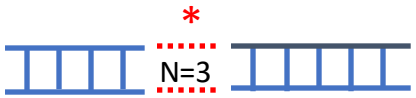
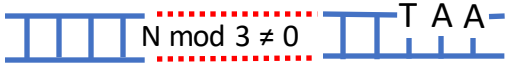
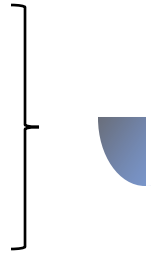
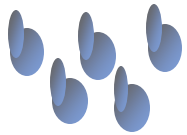
Types of alterations

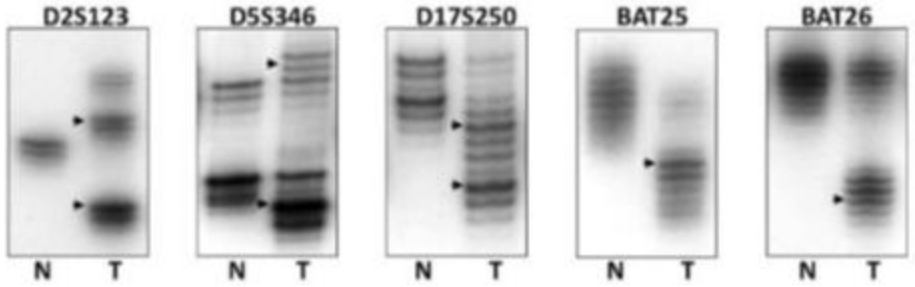
Type	Example	DNA/chromosome	Protein	Function
Point mutation (missense)	ERBB2 S310F			Protein with aberrant function
Inframe deletion	EGFR L747_T451del			
Frameshift	CDH1 F317fs*39			Nonfunctional protein or No protein (*nonsense-mediated decay*)
Nonsense/Stop	CREBBP Q792*			
Amplification	ERBB2 amplification			Excess protein
Deletion	CDKN2A del or loss			Absent protein

M. Burkard, unpublished

MSI and tumor mutational burden

MSI

-changes in length in repetitive DNA (microsatellites)



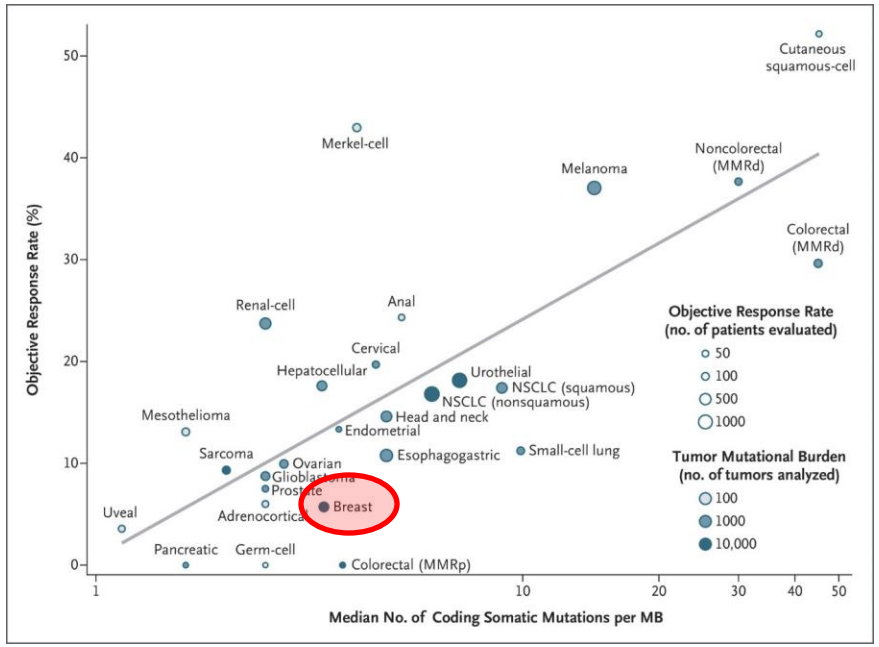
Boland and Goel, Gastroenterology 2010

-caused by mutations:

MLH1, MSH2, MSH6, PMS2

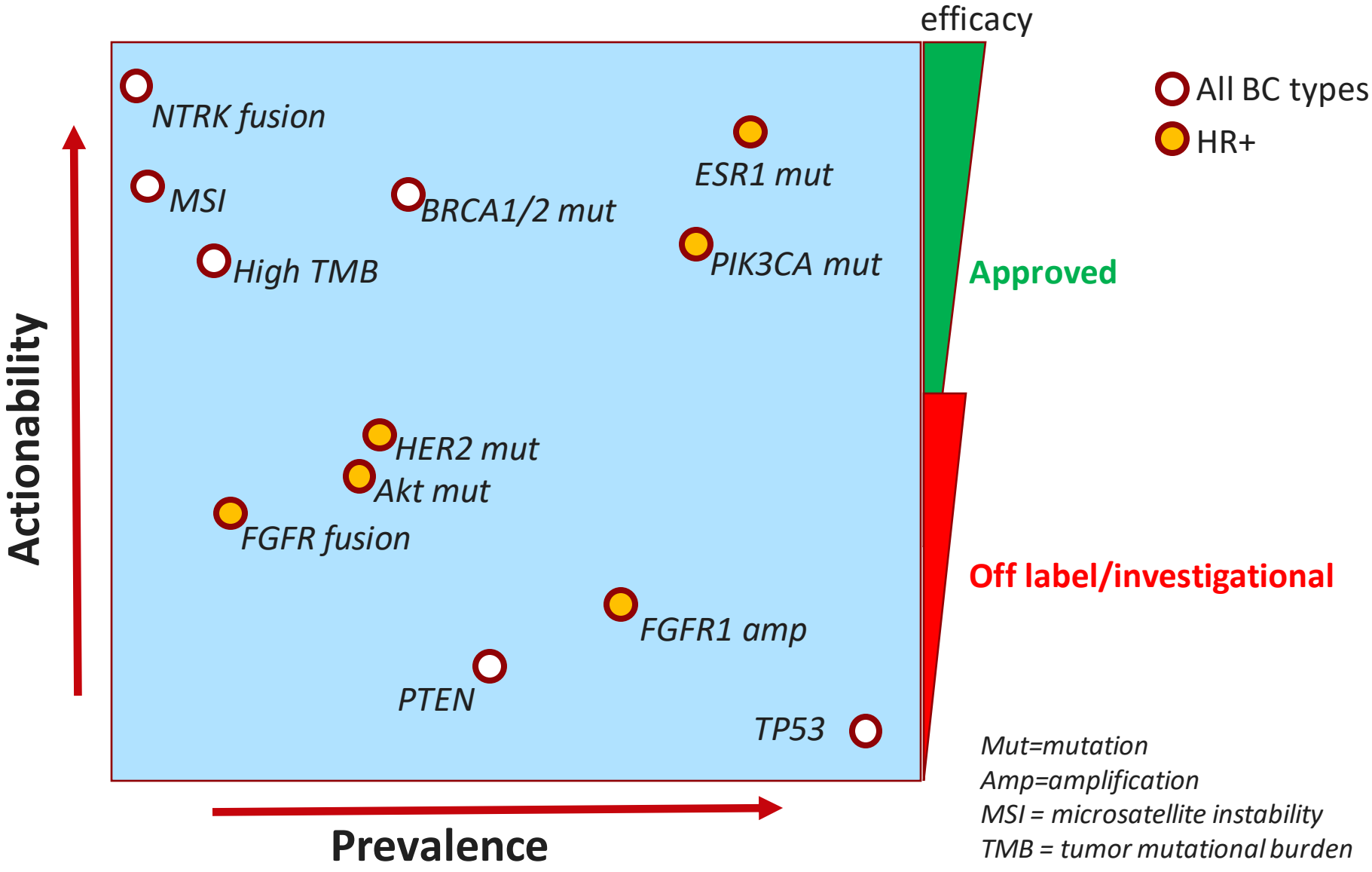
TMB

-number of non-synonymous mutations per Megabase



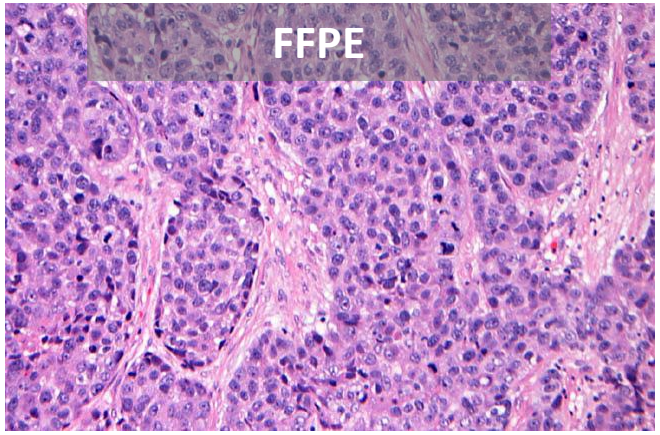
Yarchoan, Hopkins & Jaffee, NEJM 2017

Druggable targets by NGS



Mut=mutation
Amp=amplification
MSI = microsatellite instability
TMB = tumor mutational burden

M. Burkard, unpublished



plasma

DNA
+/- RNA
extraction

200ng

Panel/
WES

capture

WGS

Sequencer

Reads

ATGGCATTGCAA
TGGCATTGCAATTTG
AGATGGTATTG
GATGGCATTGCAA
GCATTGCAATTTGAC
ATGGCATTGCAATTT
AGATGGTATTGCAATTTG

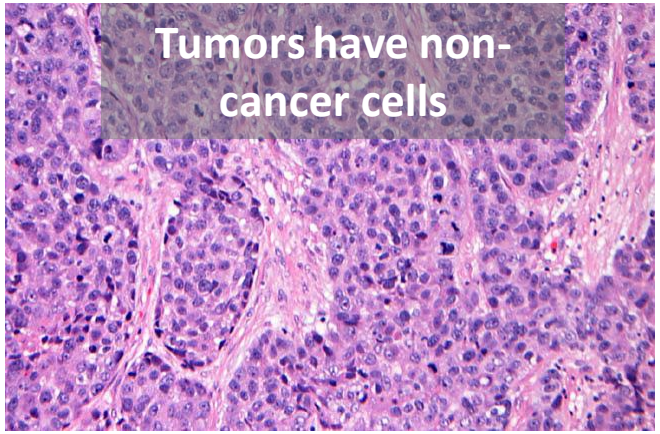
M. Burkard, unpublished

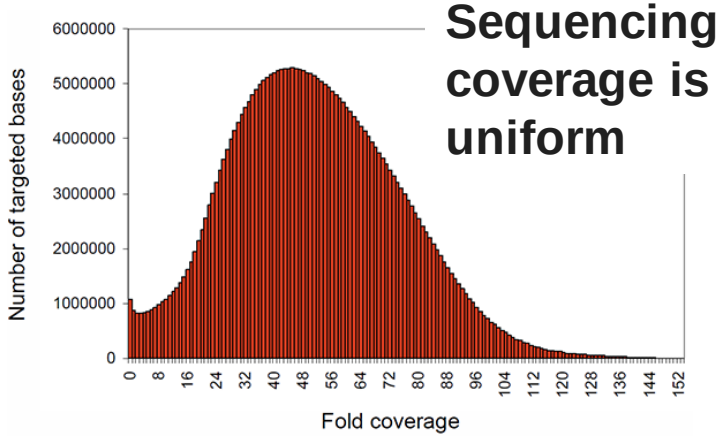
Topics

1. How Next-Generation Sequencing (NGS) tests work
2. **Differences in commercial NGS tests**
3. What to do?
4. What's next?

Difference 1:
breadth and depth of sequencing

Depth: why it matters



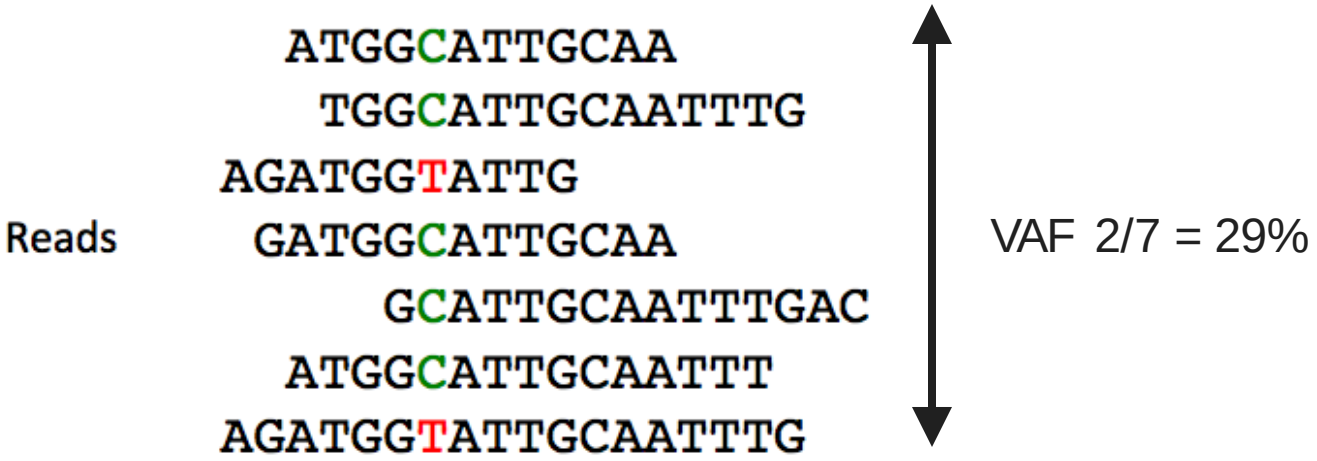


Sequencing coverage is non-uniform

Ng et al. Nature 461, 272, 2009. Targeted capture and massively parallel sequencing of 12 human exomes

M. Burkard, unpublished

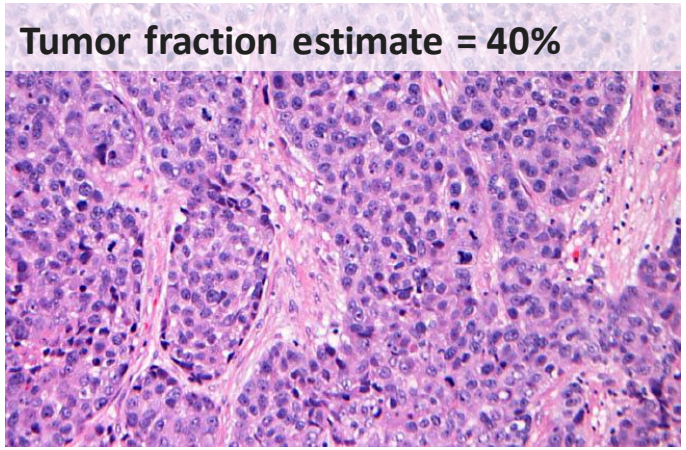
Variant allele frequency



Ng et al. Nature 461, 272, 2009. Targeted capture and massively parallel sequencing of 12 human exomes

M. Burkard, unpublished

Variant allele frequency (VAF)

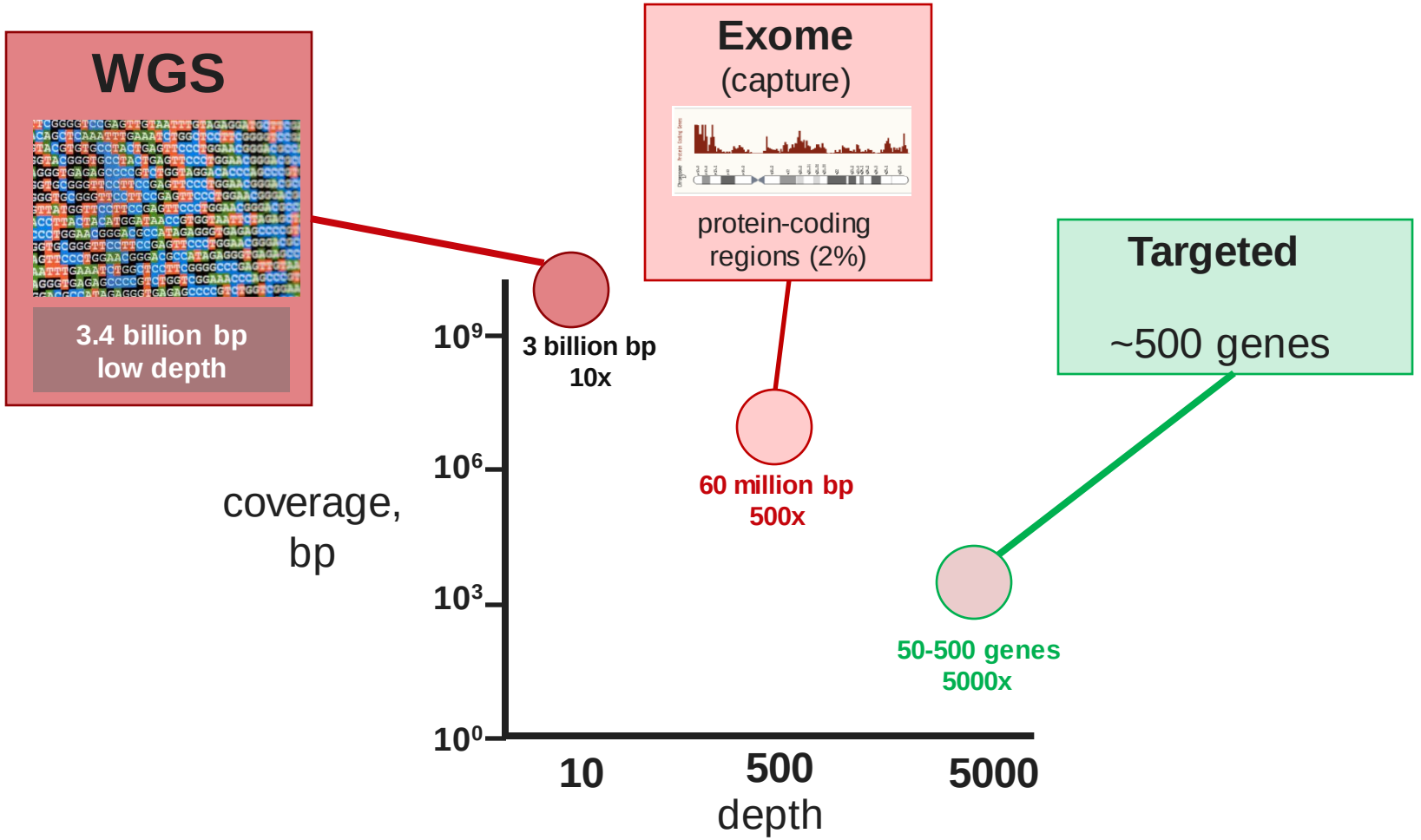


VAF result	Interpretation*	Example
0.2	tumor, heterozygous	*PIK3CA*
0.25	tumor, heterozygous + LOH	*TP53*
0.62	germline, heterozygous (LOH)	*BRCA1*

**take as 'hint' with a large grain of salt*

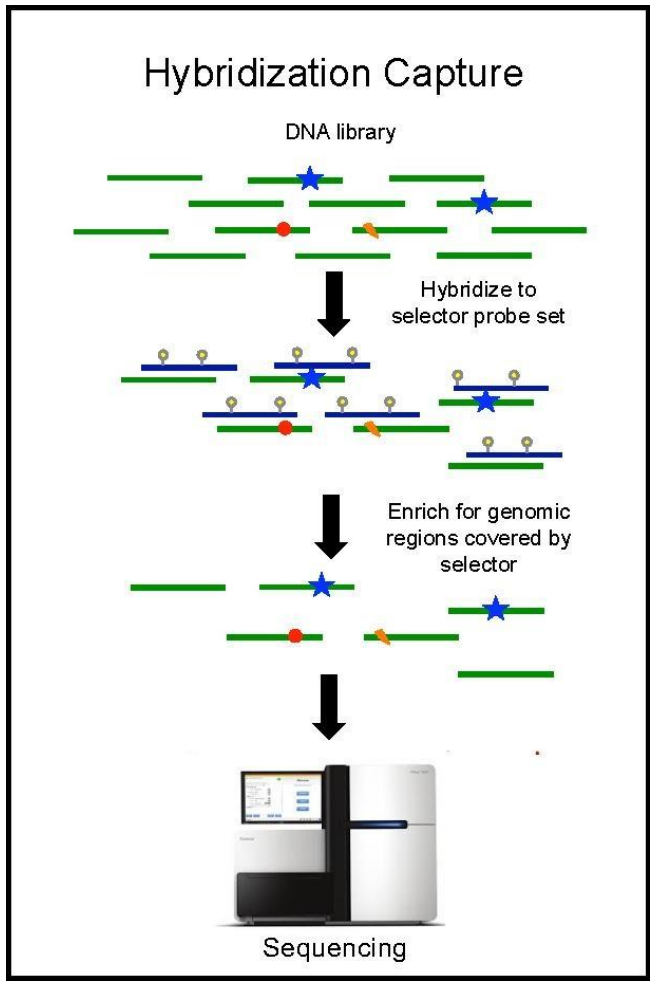
M. Burkard, unpublished

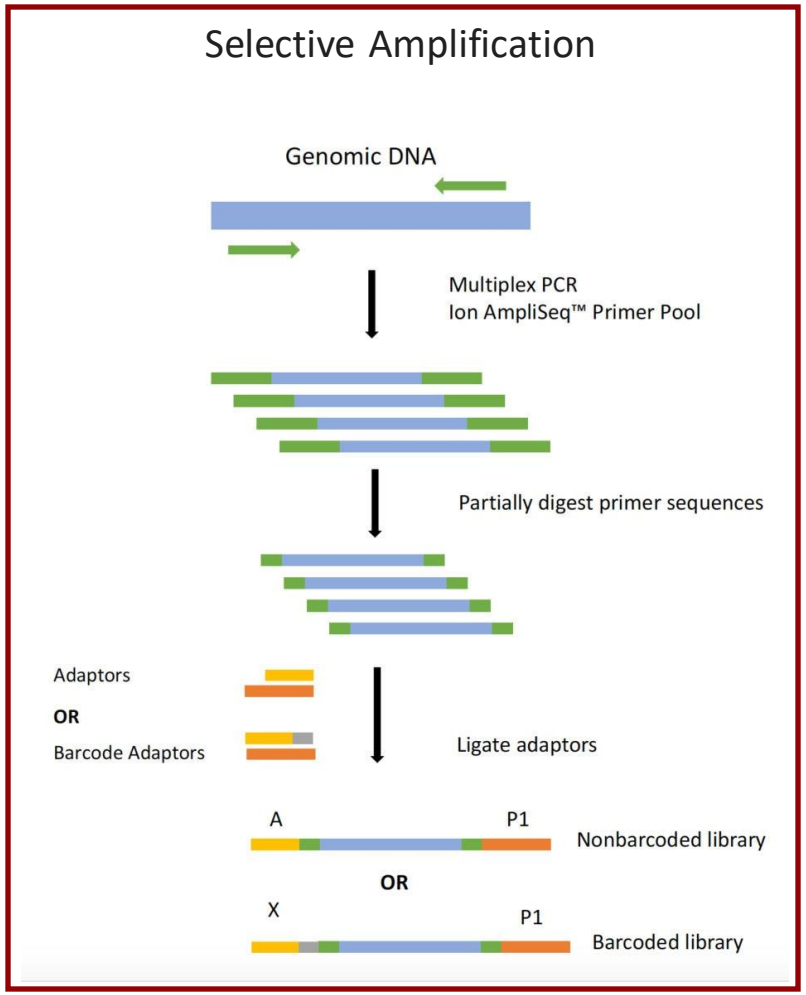
Genomic analyses



M. Burkard, unpublished

Selecting important parts of genome

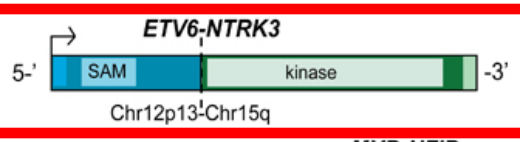
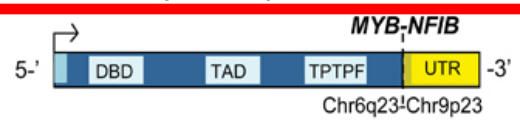
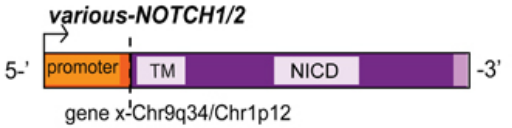
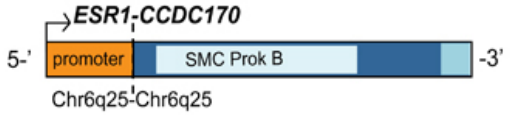
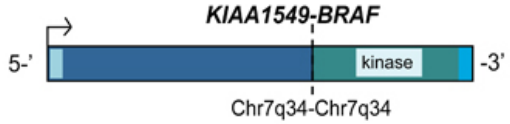
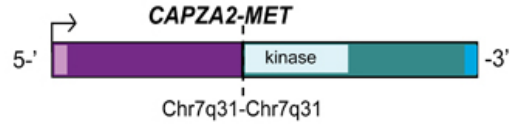
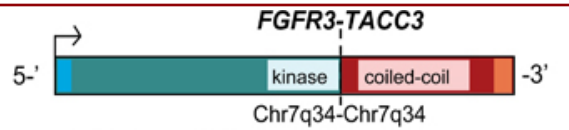
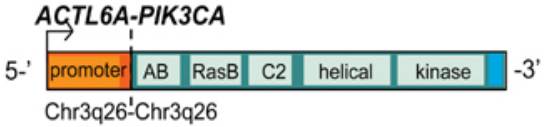
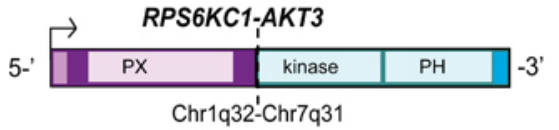
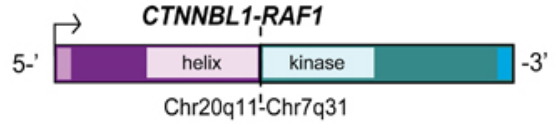
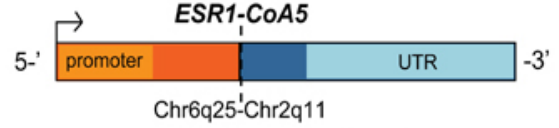
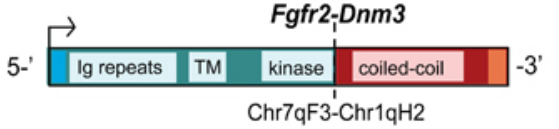
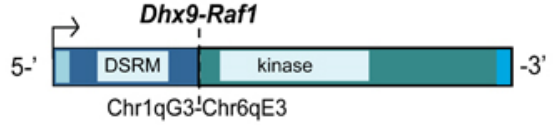




M. Burkard, unpublished

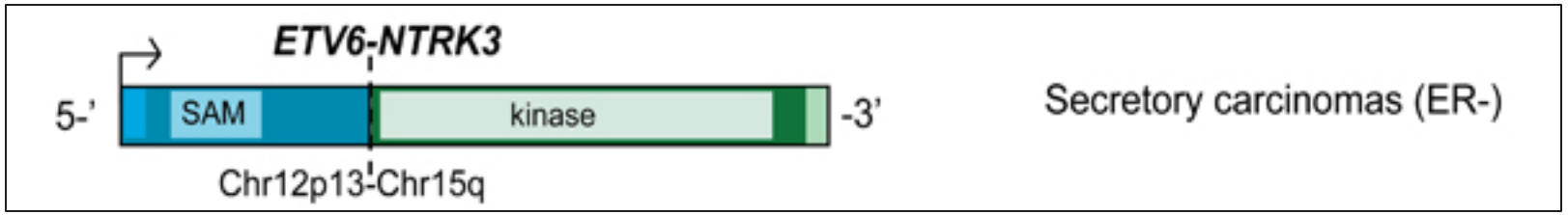
Difference 2:
Detecting Fusions

Potentially druggable fusions in breast cancer

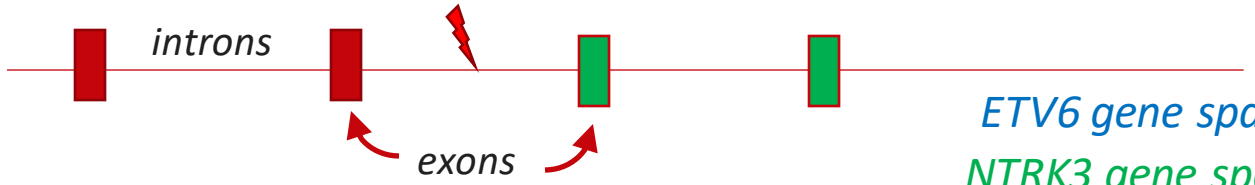
Fusion gene structure	Breast Cancer subtype	Reference
	Secretory carcinomas (ER-)	Tognon et al (5)
	Adenoid-cystic carcinomas (ER-)	Persson et al (5)
	ER+/ER-	Robinson et al (3)
	ER-	Robinson et al (3)
	ER+/Luminal B	Veeraraghavan et al (6)
	Metastatic	Ross et al (7)
	ER+	Yoshihara et al (8)
	Triple negative breast cancer	Shaver et al
	ER+ advanced disease	Matiseek et al (1)
	ER+ advanced disease	Matiseek et al (1)
	ER+ advanced disease	Matiseek et al (1)
	ER+ advanced disease	Matiseek et al (1)
	*Tp53/Brca1* GEMM	Liu et al (2)
	*Tp53/Brca1* GEMM	Liu et al (2)

Natrajan et al. Cancer Discov 8: 272, 2018

Finding fusions



DNA



ETV6 gene spans 245,000 nucleotides

NTRK3 gene spans 397,000 nucleotides

mRNA

Exons are spliced

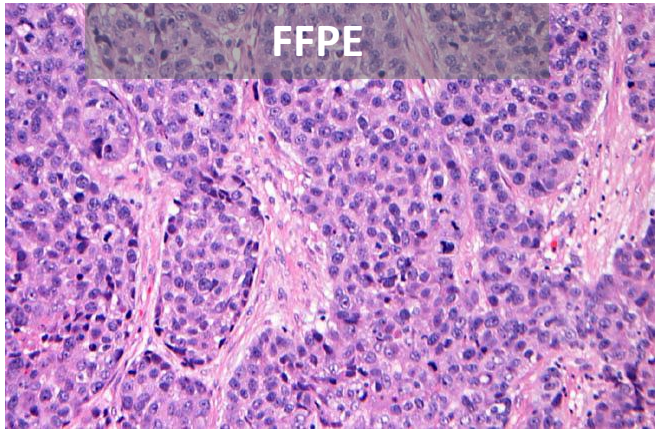
ETV6 mRNA 1,356 nucleotides

NTRK3 mRNA 2,517 nucleotides

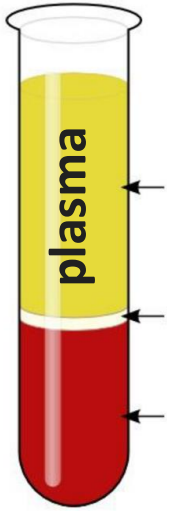
Modified from: Natrajan et al. Cancer Discov 8: 272, 2018

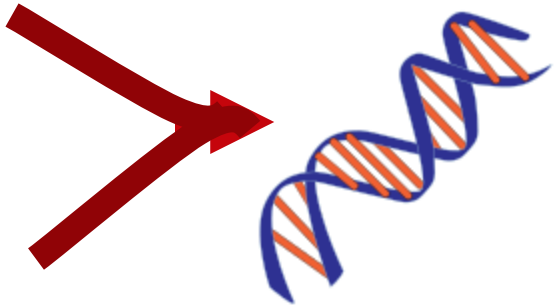
Difference 3:
tissue vs. ctDNA

FFPE Advantages:
Sensitive
Less likely to be impacted by CHIP
Amplifications are more reliable



ctDNA Advantages:
Samples all sites of disease
Easy to obtain
Fast turnaround
Companion diagnostic





DNA
+/- RNA
extraction

M. Burkard, unpublished

Topics

1. How Next-Generation Sequencing (NGS) tests work
2. Differences in commercial NGS tests
3. **What to do?**
4. What's next?

How to implement NGS into routine clinical practice

1. **Decide what to do**

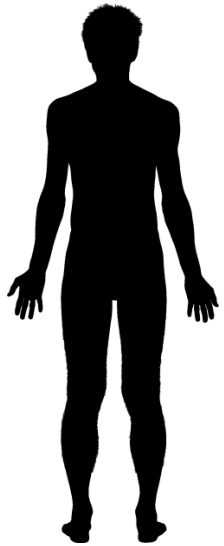
2. **What do you want to accomplish?**

Help some patients?	- any multigene panel and/or ctDNA
Clinical trials?	- any multigene panel
Make discoveries?	- whole genome panels or new technologies

3. **Make it efficient!**

UWCCC/WON Molecular Tumor Board

Committee of physicians, genomicists, bioinformatics, pharmacists, cancer biology researchers



Submit pathology or genomics analysis

Recommendations

- Clinical trial
- Off-label treatment
- Standard treatment (not targeted)

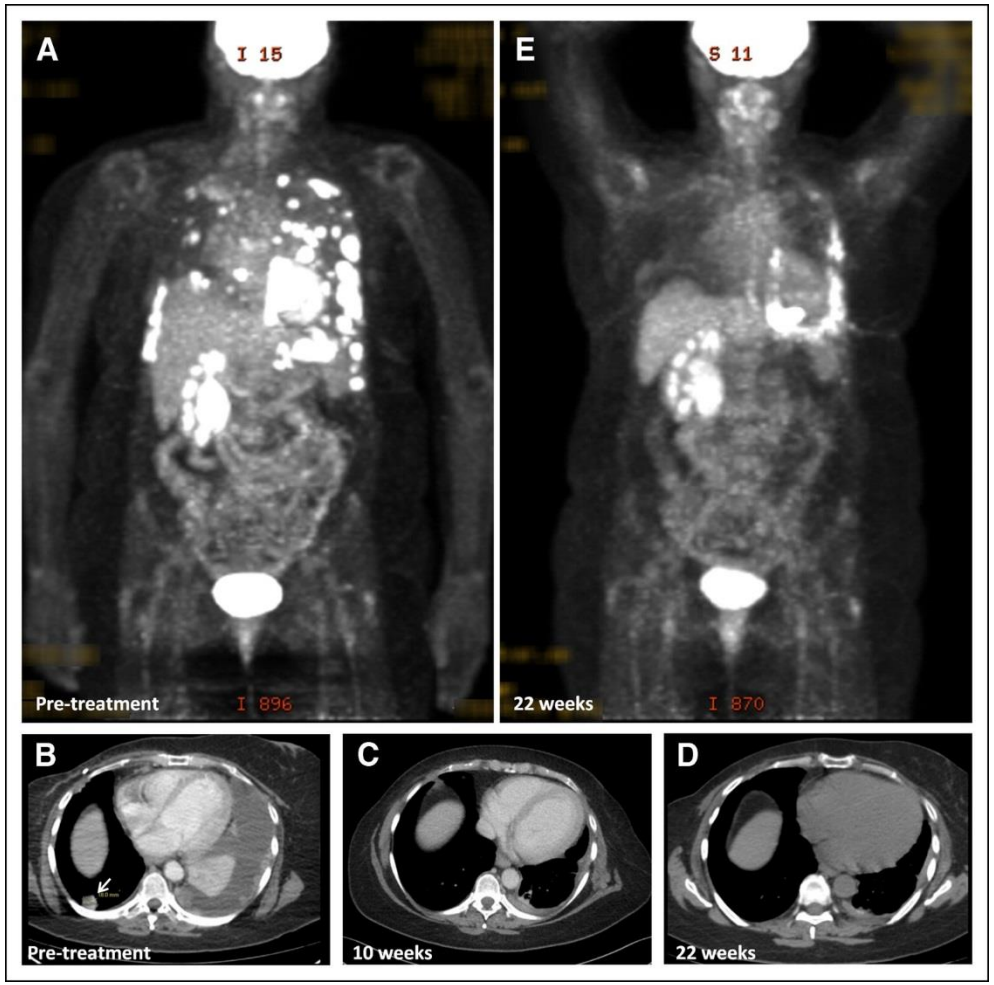
In return:

- Submit clinical outcome data
- Enrolled in registry protocol

Burkard et al. JCO Precision Oncology 2017

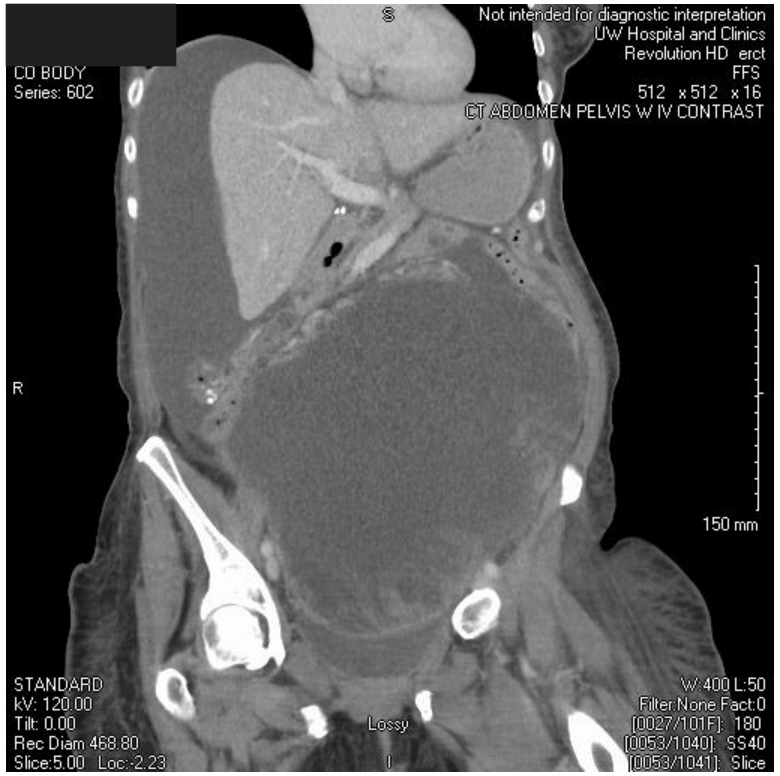
Oncogene amplification

Crizotinib for MET amplification

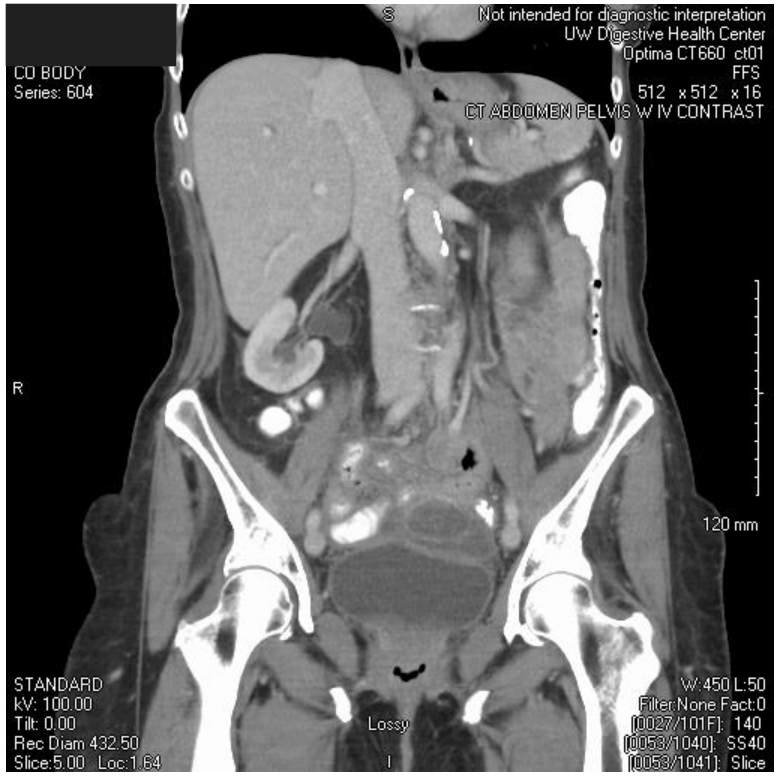


Parsons et al. JCO Precision Oncology 2017

TRK fusion in GIST



6/15/17

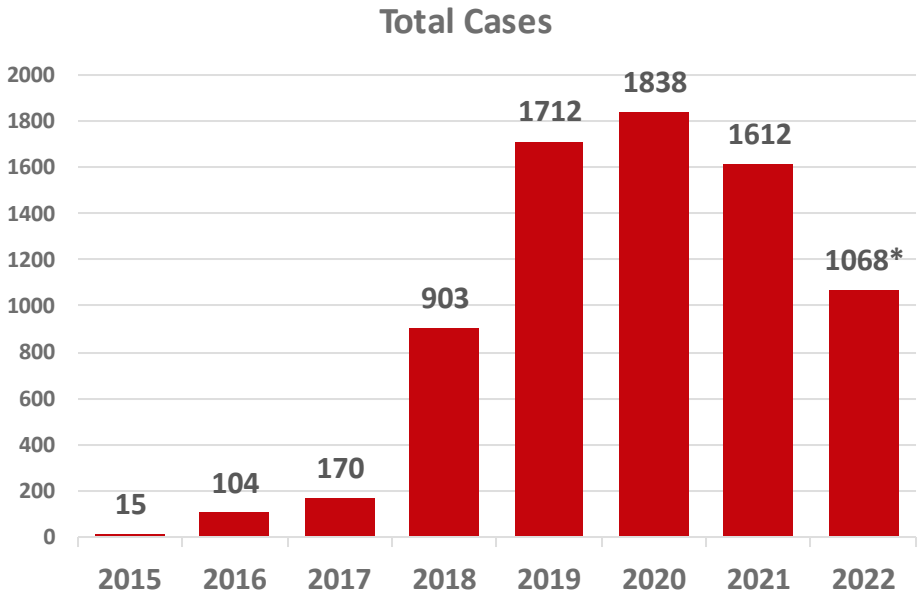


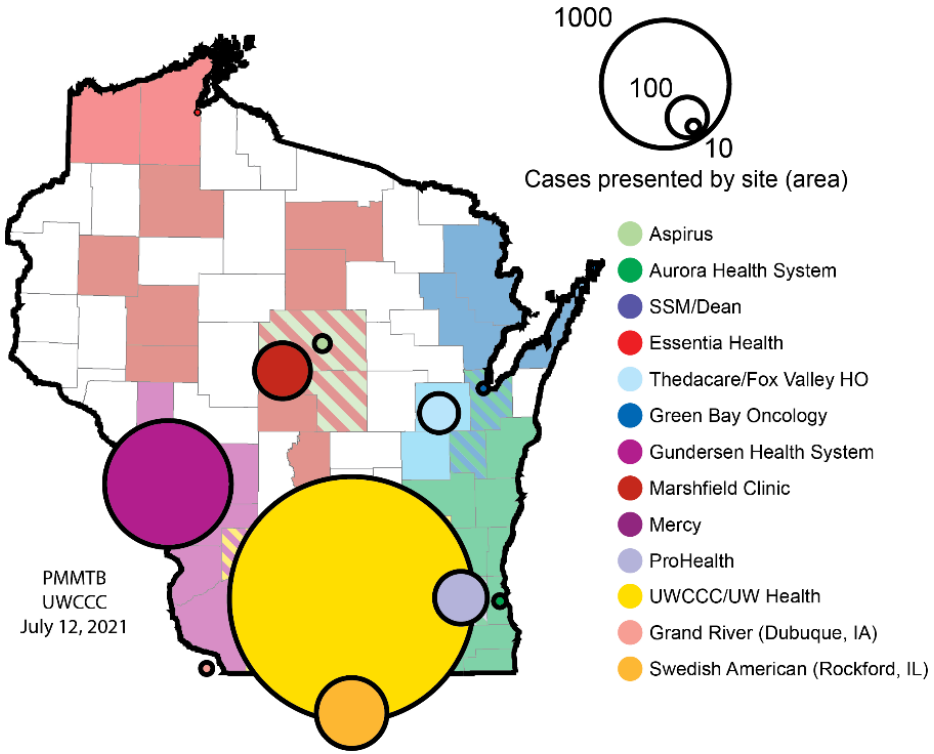
2/05/18

N Uboha, unpublished

MTB activity to date

7,422 cases





Updated from Burkard et al. JCO Precision Oncology 2017

Clinical Trials Navigation Office

Sarah Kotila, RN, BSN
Clinical Trials Navigation Manager:

Karen Arkin, RN, BSN
Clinical Trial
Nurse Navigator

Katie Browen, RN, BSN
Clinical Trial
Nurse Navigator

- Provide general trial information
- Communicate with patients and physicians
- Conduct preliminary screenings for studies
- General education about clinical research
- Outreach to outside clinics and hospitals
- Work with external and internal referrals
- Connect the right people

Topics

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Future opportunities in cancer genomics

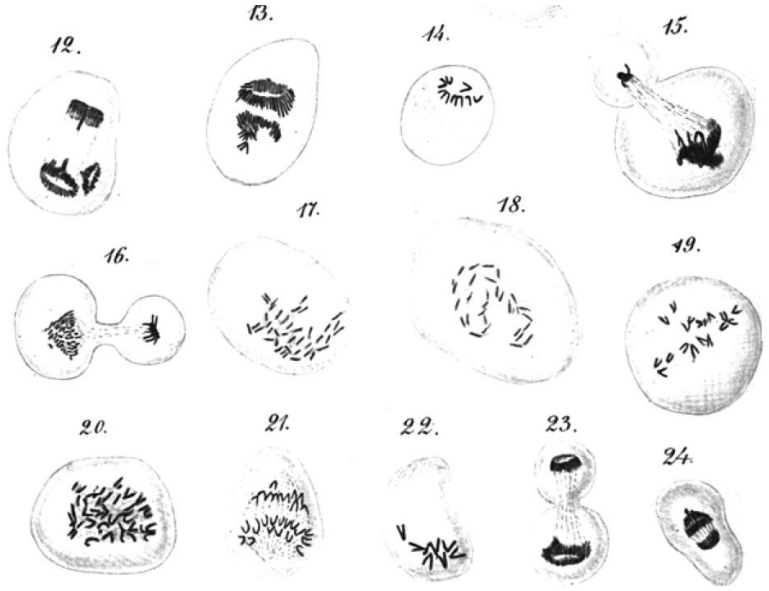
Things we find now:

- Point mutations
- Small indels
- Fusions
- Common drivers

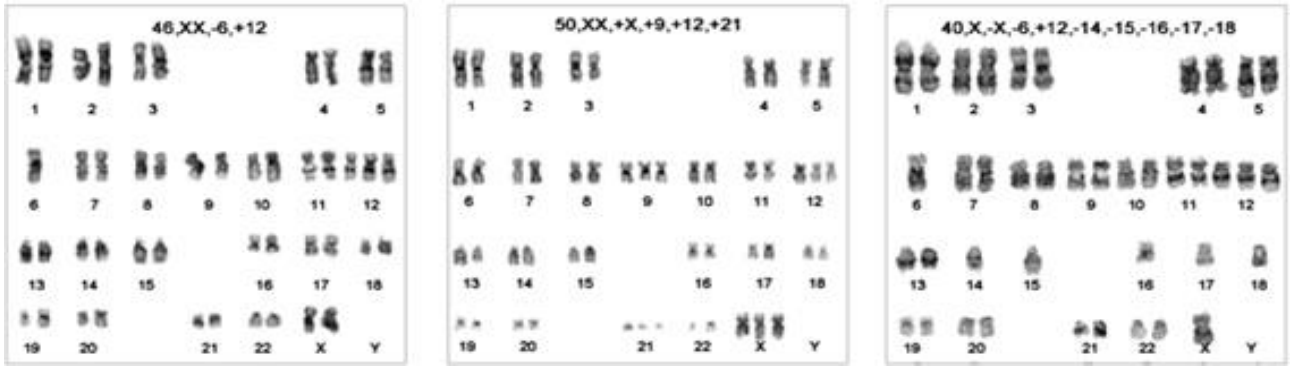
Things we are missing:

- Large-scale genomic alterations
- Centromere variation (T2T)
- Cell-cell variation
- Drivers restricted to rare populations

Chromosomal Instability



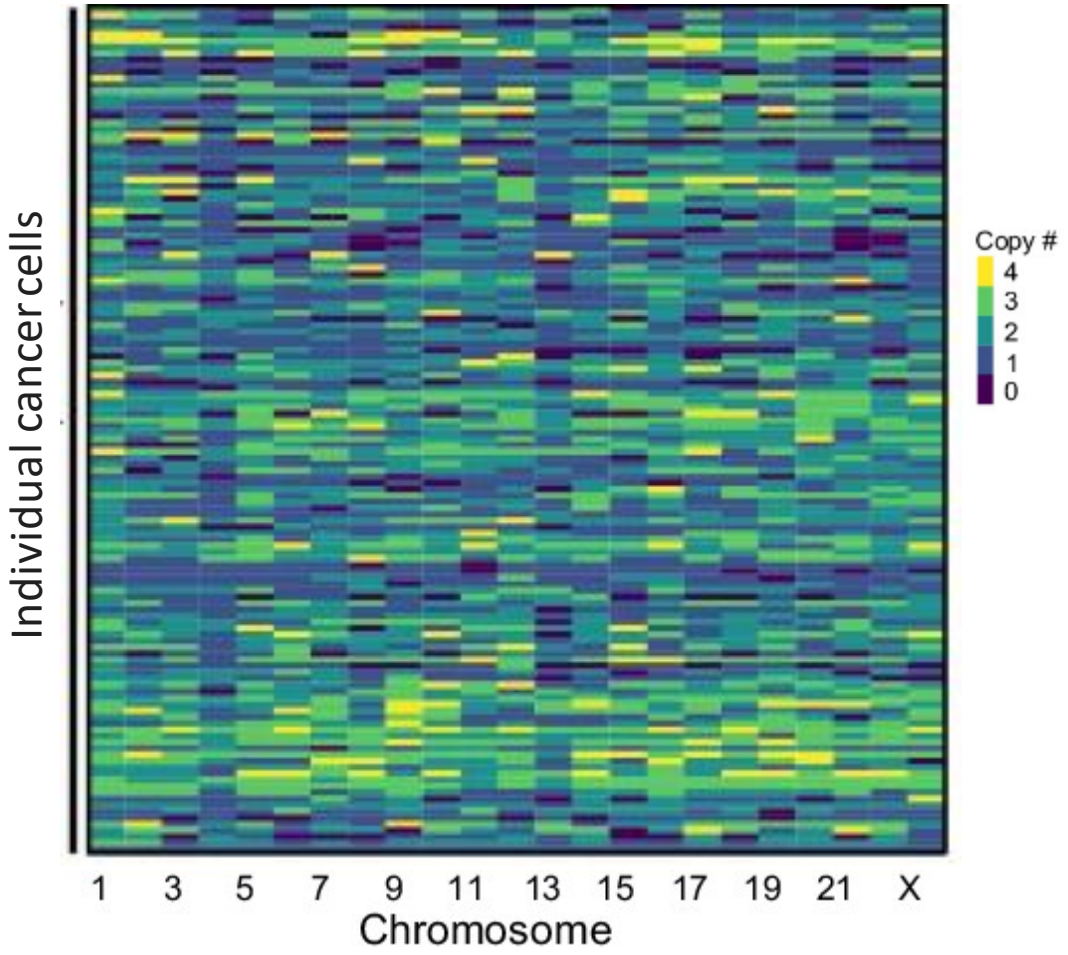
Von Hansemann D (1890) Arch Pathol Anat Physiol Klin Medicin 119: 299-326.

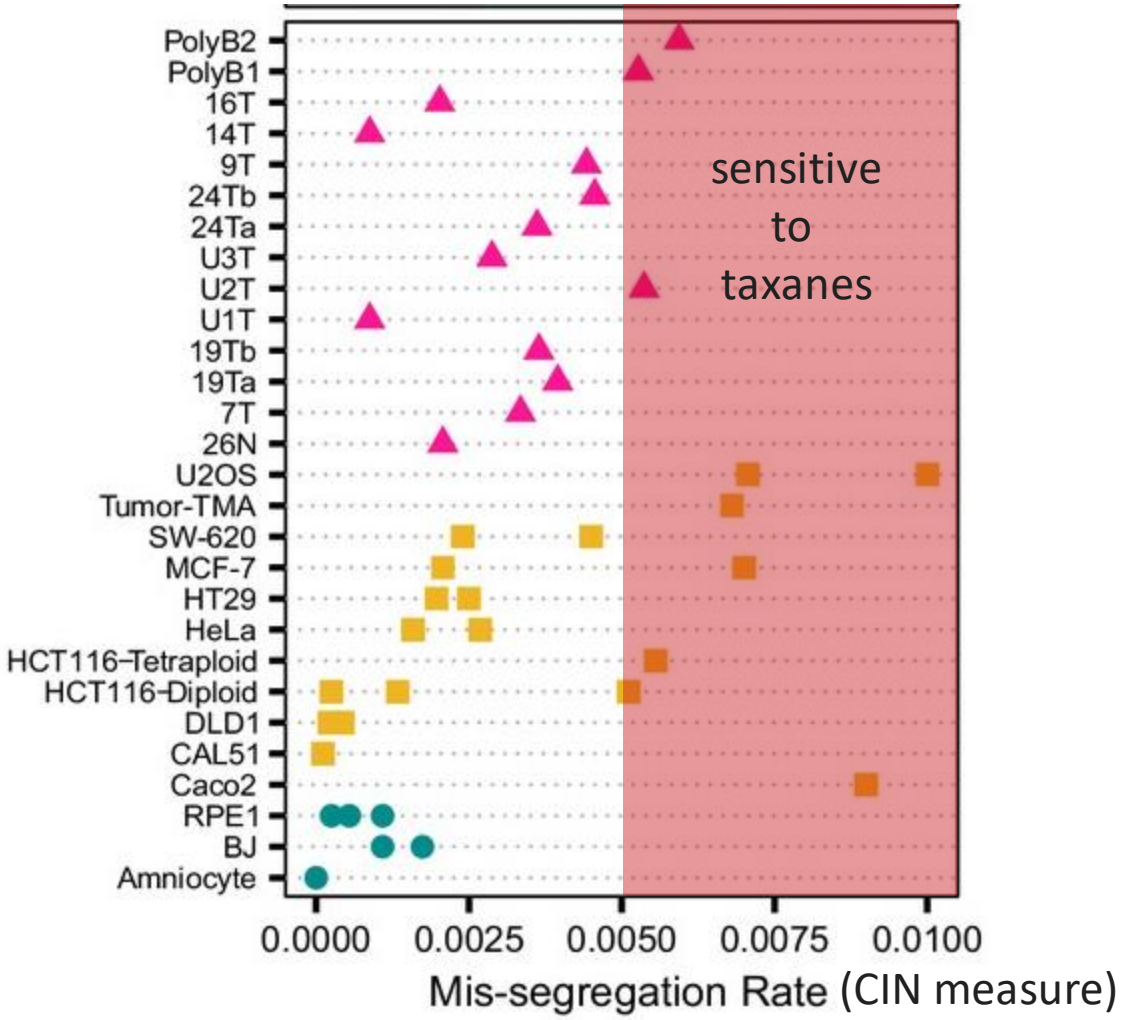


Hanks et al. (2004) Nature Genet 36: 1159-1161

Chromosomal Instability

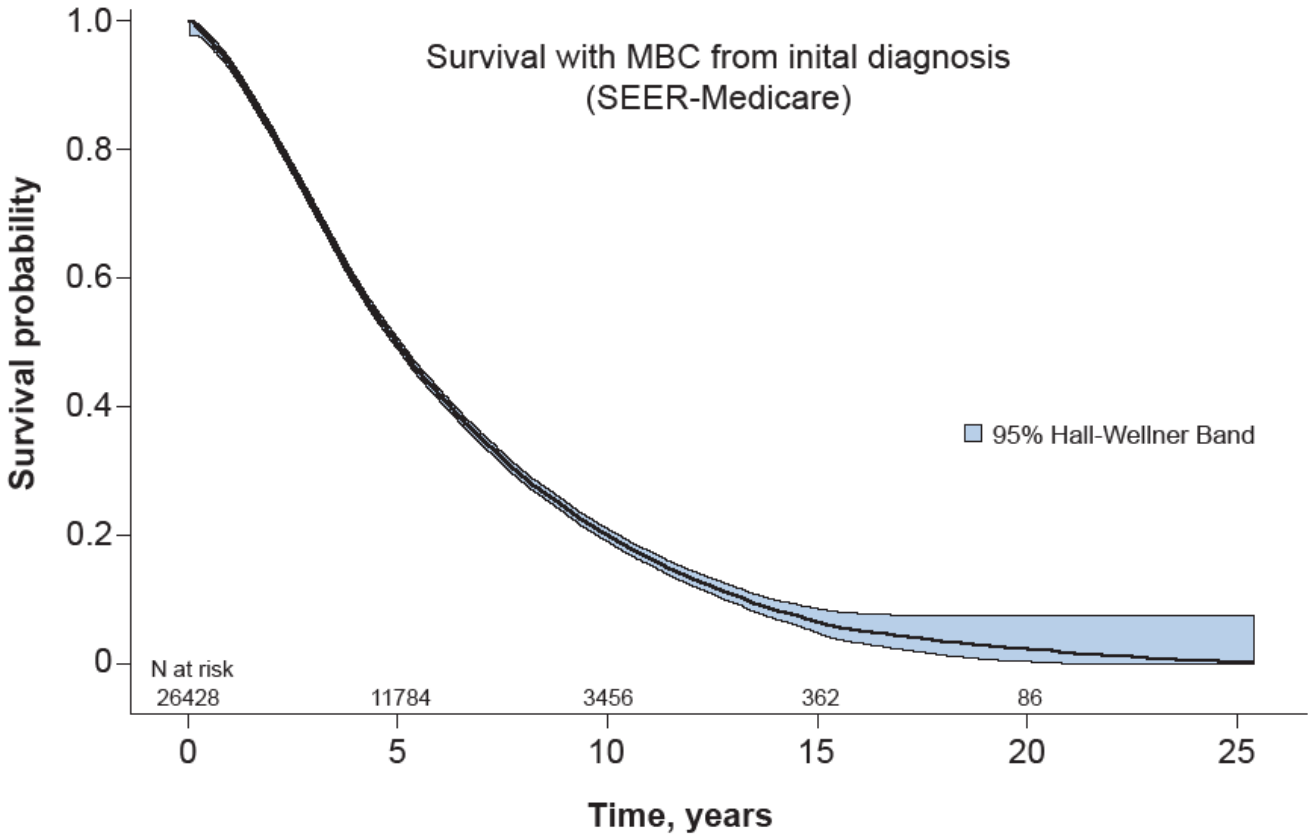
Single-cell DNA sequencing CNV by chromosome

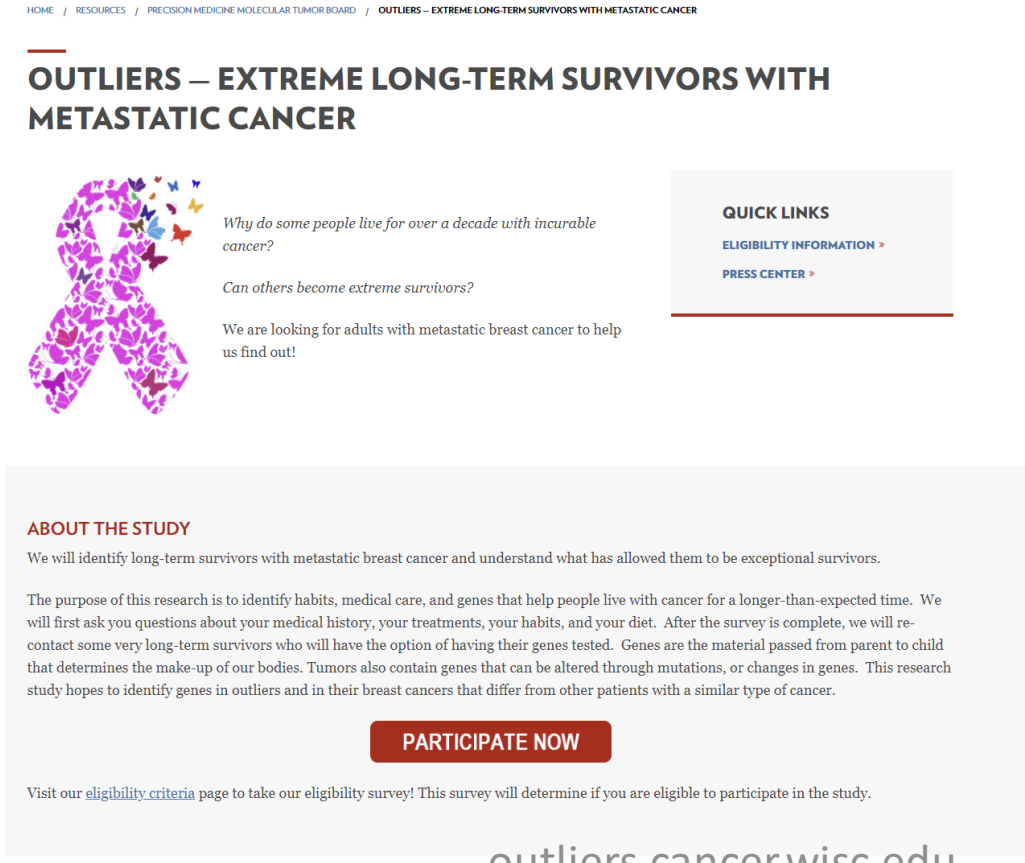




Lynch et al. eLife 11:e69799, *2022*
Sci Trans Med 13(610):eabd4811, 2011

Long term survival with MBC >20 years is uncommon





outliers.cancer.wisc.edu

Gilbert and Rocque, unpublished

Conclusions

- Clinical NGS is ready for prime time with multiple approved therapies for:

  ESR1, PIK3CA, BRCA1/2, PALB2, MSI/high TMB, and NTRK fusions

  (and many studies for other alterations)

- All MBC should have NGS, preferably at diagnosis

- MTBs can help adjudicate and prioritize rare mutations and coordinate trial enrollment in a catchment area

- NGS is a tool and more discoveries are at hand

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Effcansah.com

Thank you.