DNA sequencing of human tumor: Next generation pathology

ASCLC-Idaho

April 12. 2019

Matthew Burtelow MD PhD

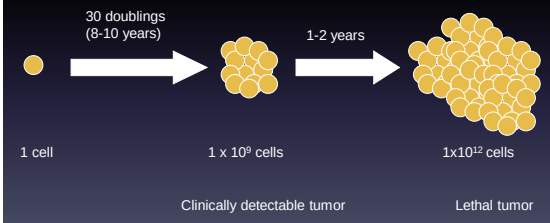
Lecture Outline: Part 1

1. Brief review of the fundamentals of cancer biology and taxonomy of cancer
2. Review of modern pathology histology laboratory
3. Next generation DNA sequencing and deep tumor sequencing analysis

Fundamentals of cancer biology and taxonomy of cancer

Definition of cancer

Uncontrolled proliferation of cells

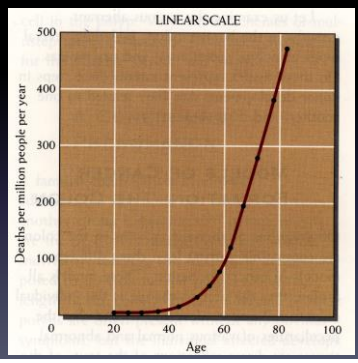


1 cell — 1 x 10^9 cells — $1x10^{12}$ cells

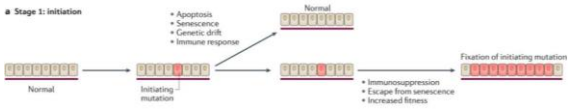
Clinically detectable tumor — Lethal tumor

Virtually all cancers are initiated by a single mutation

A single mutation is not enough to cause cancer

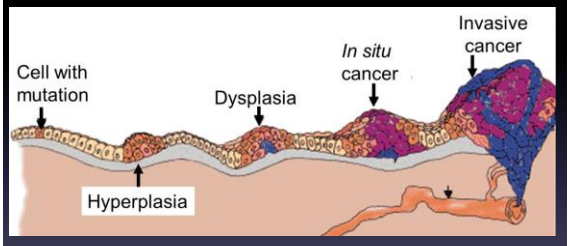


Most cancer cells derive from a single abnormal cell

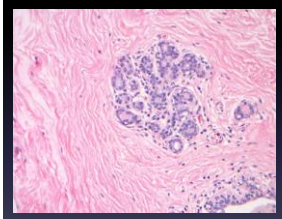


Makohon-Moore A, Iacobuzio-Donahue CA. Pancreatic cancer biology and genetics from an evolutionary perspective. Nat Rev Cancer. 2016 Sep;16(9):553-65. doi: 10.1038/nrc.2016.66. Epub 2016 Jul 22. Review. PMID: 27444064

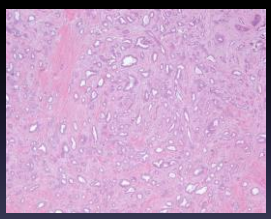
Cancers develop in slow stages from mildly aberrant cells



Cancer cells differ according to the cell type from which they derive



Normal breast



Breast cancer

Cancers arise in different tissues with different frequencies

Published in final edited form as:
Science. 2015 January 2; 347(6217): 78–81. doi:10.1126/science.1260825.

Variation in cancer risk among tissues can be explained by the number of stem cell divisions

Cristian Tomasetti[1,*] and **Bert Vogelstein**[2,*]

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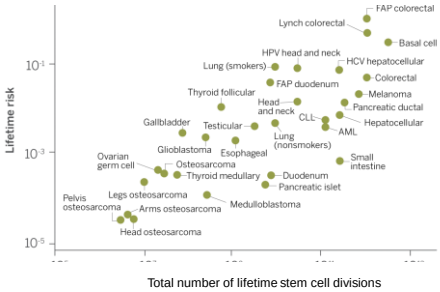
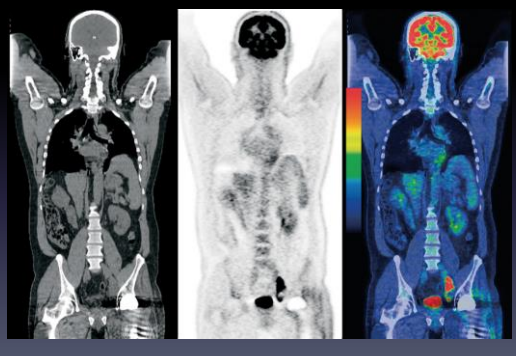
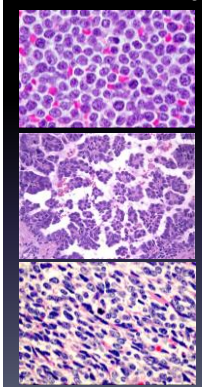
Lifetime total of stem cell divisions versus cancer risk in a tissue



Taxonomy of neoplasia: Anatomic site and cell of origin



Taxonomy of neoplasia: Cell of origin



hematolymphoid

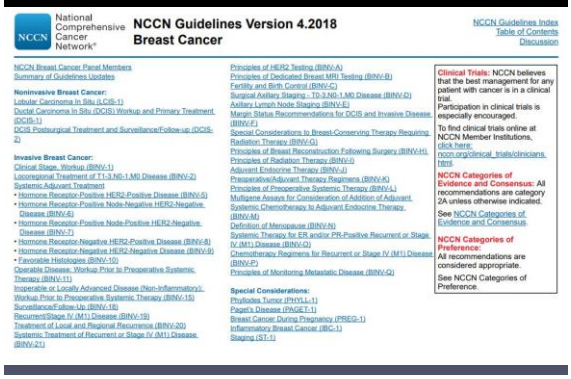
carcinoma

sarcoma

National Comprehensive Cancer Network®

NCCN GUIDELINES FOR TREATMENT OF CANCER BY SITE

NCCN Guidelines for breast cancer



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NCCN Guidelines Version 4.2018
Breast Cancer

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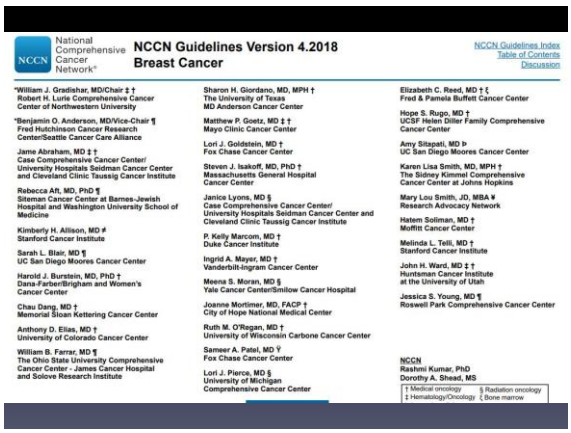
Clinical Trials: NCCN believes that the best management for any patient with cancer is in a clinical trial.
Participation in clinical trials is especially encouraged.
To find clinical trials online at NCCN Member Institutions, click here: nccn.org/clinical_trials/clinicians.html

NCCN Categories of Evidence and Consensus: All recommendations are category 2A unless otherwise indicated.
See NCCN Categories of Evidence and Consensus.

NCCN Categories of Preference: All recommendations are considered appropriate.
See NCCN Categories of Preference.



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Janice Lyons, MD §
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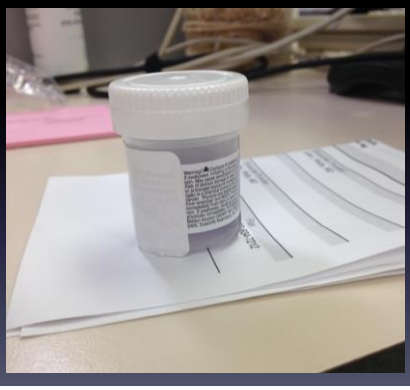
NCCN
Rashmi Kumar, PhD
Dorothy A. Shead, MS

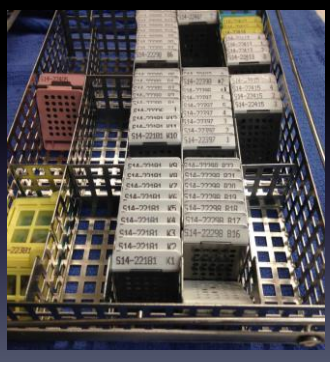
† Medical oncology ‡ Hematology/Oncology § Radiation oncology ξ Bone marrow

The modern pathology tissue processing laboratory

Formalin specimen container

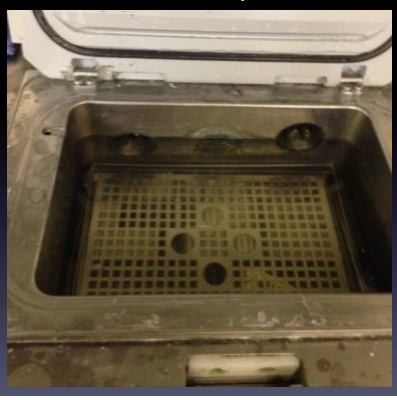


Tissue cassettes in a rack

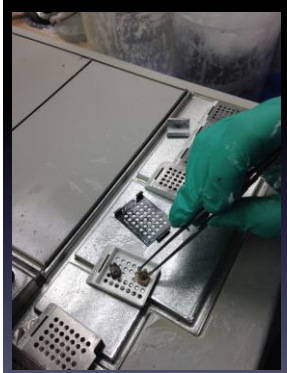


Tissue processor

Inside of the tissue processor



Paraffin embedding



Microtome

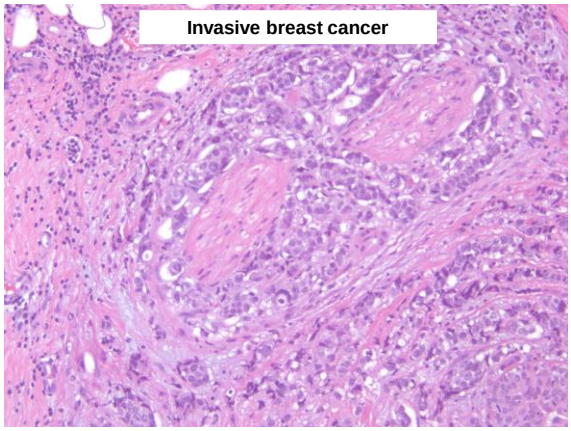
Paraffin tissue block

Tissue slide transfer

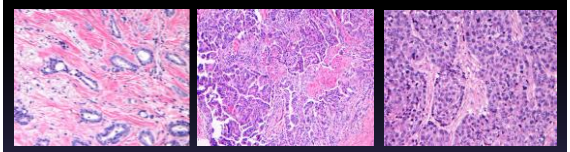
Automated slide stainer

H&E stained glass slides

Microscopic analysis of slides

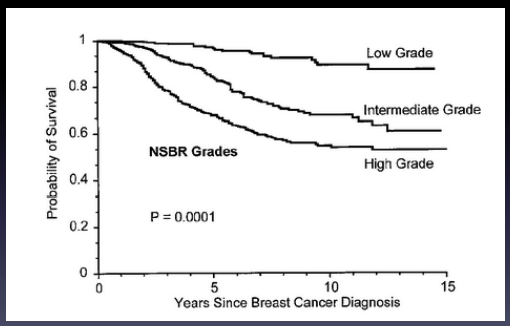


Cancer histologic grade



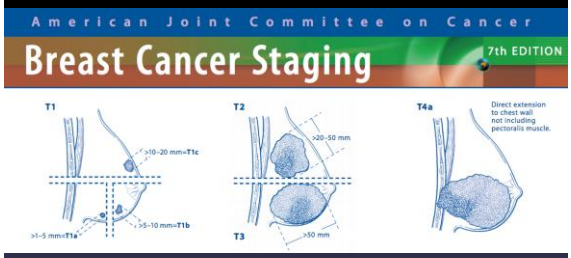
Low grade (1) Intermediate grade (2) High grade (3)

Breast cancer grade and overall survival



Mod Pathology ; 13 (7), 2000 730-735

Breast cancer stage, TNM system

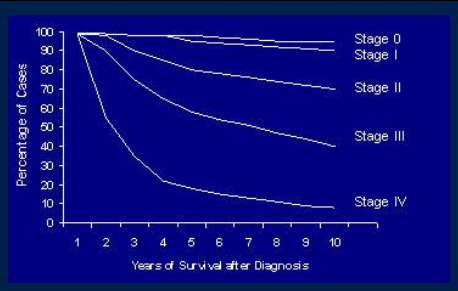


T = Tumor
N = Lymph nodes
M = Metastasis

Breast cancer stage, AJCC grouping

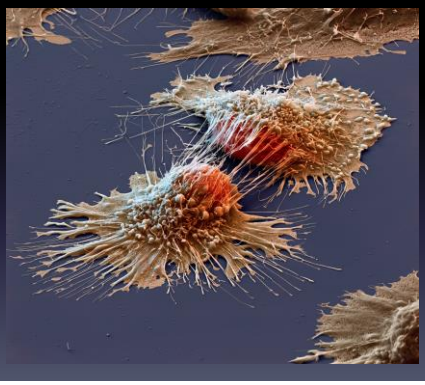
ANATOMIC STAGE/PROGNOSTIC GROUPS			
Stage 0	Tis	N0	M0
Stage IA	T1*	N0	M0
Stage IB	T0	N1mi	M0
	T1*	N1mi	M0
Stage IIA	T0	N1**	M0
	T1*	N1**	M0
	T2	N0	M0
Stage IIB	T2	N1	M0
	T3	N0	M0
Stage IIIA	T0	N2	M0
	T1*	N2	M0
	T2	N2	M0
	T3	N1	M0
	T3	N2	M0
Stage IIIB	T4	N0	M0
	T4	N1	M0
	T4	N2	M0
Stage IIIC	Any T	N3	M0
Stage IV	Any T	Any N	M1

Breast cancer stage and overall survival



Bland KI, et al. *Cancer*. 1998;83:1262-1273.

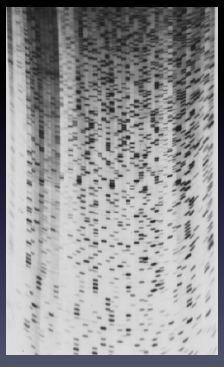
Genetic lesions driving tumors

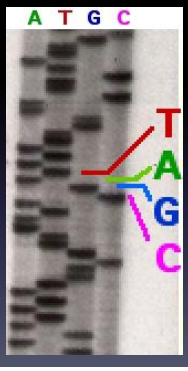


"New directions in science are launched by new tools much more often than by new concepts or ideas"

Freeman Dyson,
Imagined Worlds

Really old DNA sequencing technology: chain-terminator





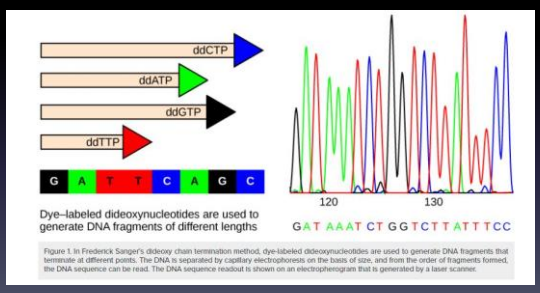
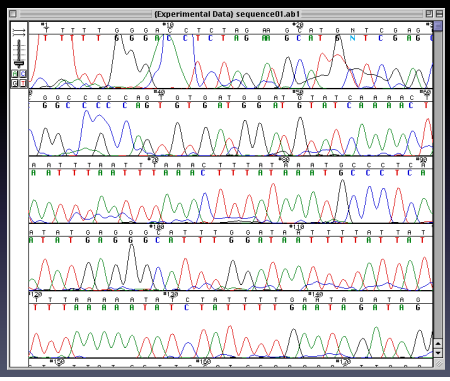
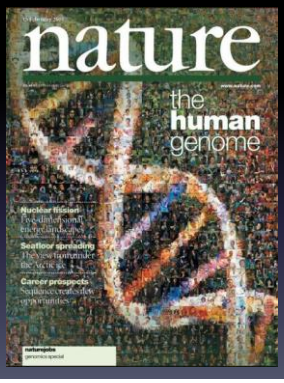


Figure 1. In Frederick Sanger's dideoxy chain termination method, dye-labeled dideoxynucleotides are used to generate DNA fragments that terminate at different points. The DNA is separated by capillary electrophoresis on the basis of size, and from the order of fragments formed, the DNA sequence can be read. The DNA sequence readout is shown on an electropherogram that is generated by a laser scanner.

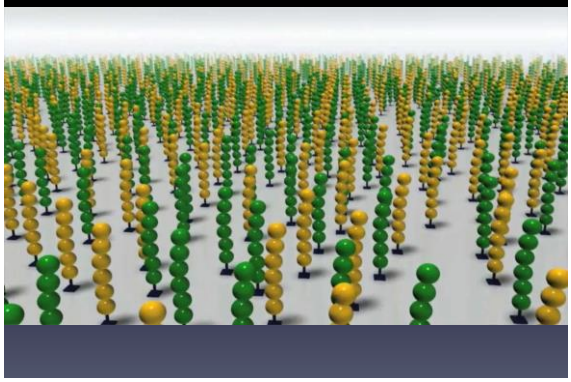
Sanger DNA sequencing method



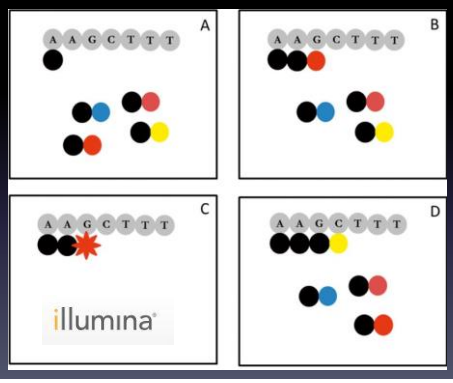
February 2001: First draft of the human genome completed



Next Generation DNA sequencing (NGS)

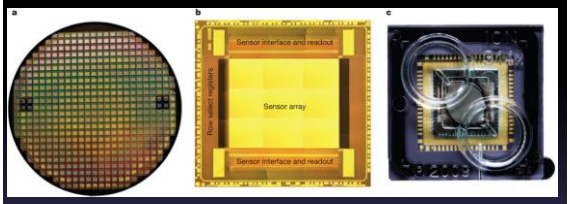


Sequencing by synthesis

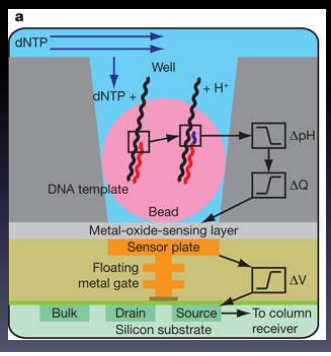


Illumina MiSeq Instrument

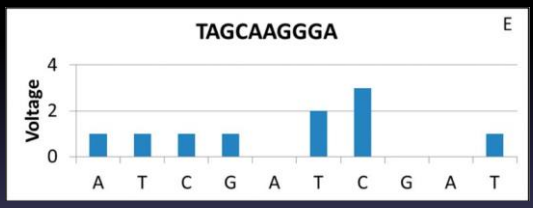
Ion torrent: Semiconductor chip based DNA sequencing



Ion torrent: semiconductor chip based DNA sequencing

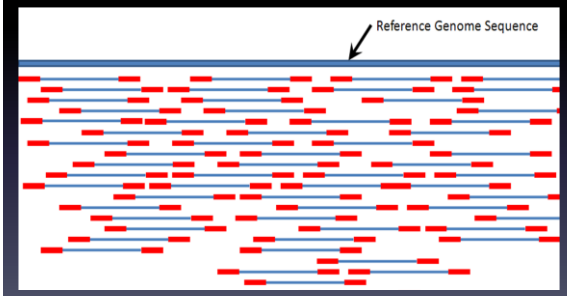


Sequencing by voltage



ion torrent
by life technologies

Assembly of next generation DNA sequencing data



Foundation Medicine Assay for Cancer

FOUNDATIONONE® CDx

- For all solid tumor cancers
- Performed on tumor tissue
- May help identify targeted therapy, immunotherapy, and clinical trial options
- Includes more than 300 genes and biomarkers
- Can include PD-L1 if ordered by your physician

LEARN MORE →

FOUNDATIONONE® LIQUID

- Also for all solid tumor cancers
- Performed on blood
- This is called a liquid biopsy
- May help identify targeted therapy, immunotherapy, and clinical trial options
- Includes more than 70 genes and biomarkers

LEARN MORE →

FOUNDATIONONE® HEME

- For blood cancers like leukemia and lymphoma, and for sarcomas
- Performed on blood, bone marrow, or tissue
- May help identify targeted therapy, immunotherapy, and clinical trial options
- Includes more than 400 genes and biomarkers

324 cancer associated genes interrogated by the FoundationOne CDx assay

Current Gene List[2]

Genes with full coding exonic regions included in FoundationOne CDx for the detection of substitutions, insertion-deletions (indels), and copy-number alterations (CNAs).

ABL1	ACVR1B	AKT1	AKT2	AKT3	ALK	ALOX12B	AMER1 (FAM123B)
APC	AR	ARAF	ARID1A	ARID1B	ASXL1	ATM	ATR
ATRX	AURKA	AURKB	AXIN1	AXL	BAP1	BARD1	BCL2
BCL2L1	BCL2L2	BCL6	BCOR	BCORL1	BRAF	BRCA1	BRCA2
BRD4	BRIP1	BTG1	BTG2	BTK	CTNNB1 (CHN1)	CALR	CARD11
CASP8	CBFB	CBL	CCND1	CCND2	CCND3	CCNE1	CD22
CD274 (PD-L1)	CD70	CDH1A	CDH1B	CDC73	CDH1	CDK12	CDK4
CDK6	CDKN1A	CDKN1B	CDKN2A	CDKN2B	CDKN2C	CEBPA	CHEK1
CHEK2	CIC	CREBBP	CRKL	CSF1R	CSF3R	CTCF	CTNNA1
CTNNB1	CUL3	CUL4A	CXCR4	CYP17A1	DAXX	DDR1	DDR2
DIS3	DNMT3A	DOT1L	EED	EGFR	EP300	EPHA3	EPHB1
EPHB4	ERBB2	ERBB3	ERBB4	ERCC4	ERG	ESR1	EZH2
FAMSEC	FANCA	FANCC	FANCG	FAS	FBXW7	FGF10	FGF12
FGF14	FGF19	FGF23	FGF3	FGF4	FGF6	FGFR1	FGFR2
FGFR3	FGFR4	FH	FLCN	FLT1	FLT3	FOXL2	FUBP1
GABRA6	GATA3	GATA4	GATA6	GID4 (C17orf39)	GNA11	GNAQ	GNAS
GRM3	GSK3B	H3F3A	HDAC1	HGF	HNF1A	HRAS	HSD3B1
IDH1	IDH2	IGF1R	IKBKE	IKZF1	INPPL1	IRF2	IRF4
IRS2	JAK1	JAK2	JAK3	JUN	KDM5A	KDM5C	KDM6A
KDR	KEAP1	KEL	KIT	KLHL6	KMT2A (MLL)	KMT2D (MLL2)	KRAS
LTK	LYN	MAF	MAP2K1 (MEK1)	MAP2K2 (MEK2)	MAP2K4	MAP3K1	MAP3K13
MAPK1	MCL1	MDM2	MDM4	MED12	MEF2B	MEN1	MERTK
MET	MKNK1	MLH1	MPL	MRE11A	MSH2	MSH3	MSH6
MST1R	MTAP	MTOR	MUTYH	MYC	MYCL (MYCL1)	MYCN	MYD88
NBN	NF1	NF2	NFE2L2	NFKBIA	NKX2-1	NOTCH1	NOTCH2
NPM1	NRAS	NTRK1	NTRK2	NTRK3	P2RY8	PAK1	PAK7
PARP1	PARP2	PARP3	PAX5	PBRM1	PDCD1 (PD-1)	PDCD1LG2 (PD-L2)	PDGFRA
PDGFRB	PDK1	PIK3C2B	PIK3C2G	PIK3CA	PIK3CB	PIK3R1	PIM1
PMS2	POLD1	POLE	PPARG	PPP2R1A	PPP2R2A	PRDM1	PRKC1
PTCH1	PTEN	PTPN11	PTPRD	QKI	RAC1	RAD21	RAD50
RAD51B	RAD51C	RAD51D	RAD52	RAD54L	RAF1	RARA	RB1
RBM10	REL	RET	RICTOR	RNF43	ROS1	RPTOR	SDHA
SDHB	SDHC	SDHD	SETD2	SF3B1	SGK1	SMAD2	SMAD4
SMARCA4	SMARCB1	SMO	SNCAIP	SOCS1	SOX2	SOX9	SPEN
SPOP	SRC	STAG2	STAT3	STK11	SUFU	SYK	TBX3
TEK	TET2	TGFBR2	TIPARP	TNFAIP3	TNFRSF14	TP53	TSC1
TSC2	TYRO3	U2AF1	VEGFA	VHL	WHSC1 (NSD2)	WHSC1L1	WT1
XPO1	XRCC2	ZNF217	ZNF703				

Foundation Medicine Assay for Cancer

FOUNDATIONONE® TECHNICAL INFORMATION	BASE SUBSTITUTIONS[1]	INSERTIONS AND DELETIONS[1]	COPY NUMBER ALTERATIONS – AMPLIFICATIONS[1]	COPY NUMBER ALTERATIONS – DELETIONS[1]	REARRANGEMENTS[1,2]
Sensitivity	>99% (MAF ≥5%)	>97% (MAF ≥5%; 1-40bp)	>99% (CNA ≥8; ≥30% tumour nuclei)	>97% (homozygous deletions; ≥30% tumour nuclei)	>90%[4] >99% for ALK fusion (95% CI 88%-100%)[3] (≥20% tumour nuclei)
Specificity (PPV)	>99%	>99%	>99%	>99%	>99%[4]
Concordance MSI	97%[1,6]				
Concordance TMB	>90%[1,4,5]				
Typical median depth of coverage	500[1]				
Sample requirements	≥40 µm tissue, of which a minimum of 20% is of malignant origin,[1] on 10 unstained slides or in an FFPE block. Needle biopsy is also acceptable.[10]				
Turn-around time	14-day average[4]				

Foundation Medicine Assay for Cancer

FOUNDATIONONE®

Patient Name | Report Date | Diagnosis: Lung adenocarcinoma

Date of Birth	Not Given	Client	Cancer Center	Specimen Received	Not Given
Gender	Female	Ordering Physician	Doctor, Denise	Specimen Site	Lymph Node
FMI Case #	SRF000005	Additional Recipient	Not Given	Date of Collection	Not Given
Medical Record #		FMI Client #	-1	Specimen Type	Block
Specimen ID	Not Given	Pathologist	Not Given		

ABOUT THE TEST:
FoundationOne™ is a next-generation sequencing (NGS) based assay which identifies genomic alterations within hundreds of cancer-related genes.

PATIENT RESULTS

- 4 genomic alterations
- 3 therapies associated with potential clinical benefit
- 0 therapies associated with lack of response
- 6 clinical trials

TUMOR TYPE: LUNG ADENOCARCINOMA

Genomic Alterations Identified†
ALK EML4-ALK fusion
TSC2 splice site 3285-1 G>A
CDKN2A/B loss
TP53 L194R

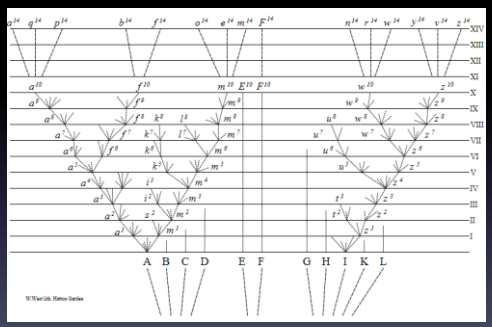
Additional Disease-relevant Genes with No Reportable Alterations Detected
EGFR
KRAS

†For a complete list of the genes assayed, please refer to the Appendix

Lecture Outline: Part 2

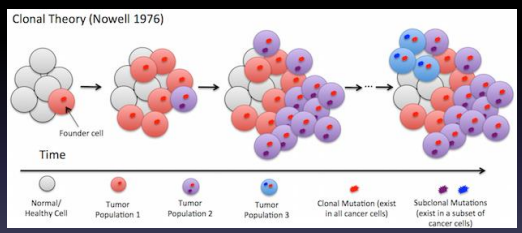
1. Cancer as a microevolutionary process
2. Pancreatic cancer as a model of cancer evolution
3. Limitations of targeted therapy
3. Immunotherapy for cancer

Charles Darwin: Origin of the species

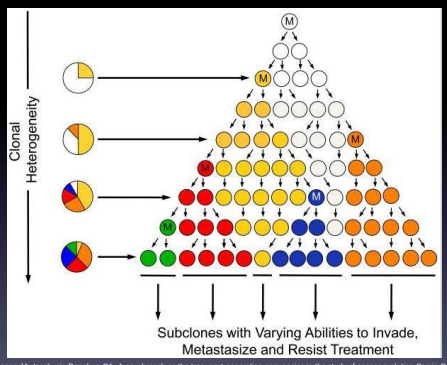


Darwin, Charles Robert. The Origin of Species. Vol. XI. The Harvard Classics. New York: P.F. Collier & Son, 1909–14; Bartleby.com, 2001. www.bartleby.com/11/.

Clonal diversity within tumors

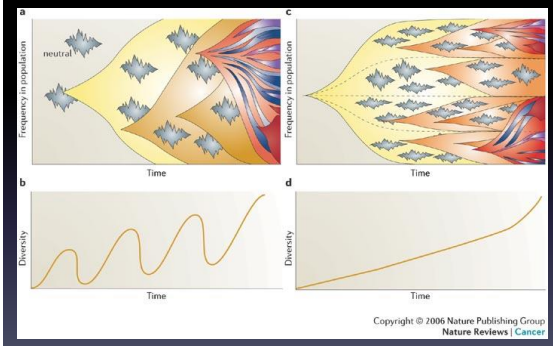


Intratumoral clonal evolution



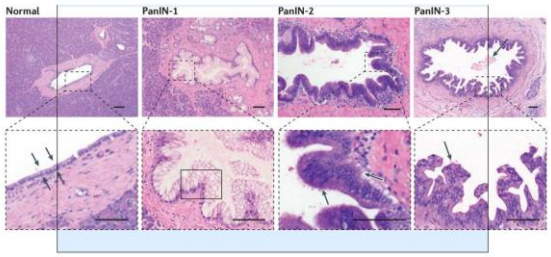
Brosnan JA, Iacobuzio-Donahue CA. A new branch on the tree: next-generation sequencing in the study of cancer evolution Semin Cell Dev Biol. 2012 Apr;23(2):237-42. doi: 10.1016/j.semcdb.2011.12.008. Epub 2012 Jan 8. Review. PMID: 22245832

Cancer as a microevolutionary process



Cancer as an evolutionary and ecological process. Cancer as an evolutionary and ecological process. Merlo LM, Pepper JW, Reid BJ, Maley CC. Nat Rev Cancer. 2006 Dec;6(12):924-35. Epub 2006 Nov 16.

Pancreatic cancer as a model of cancer evolution



Makohon-Moore A, Iacobuzio-Donahue CA. Pancreatic cancer biology and genetics from an evolutionary perspective. Nat Rev Cancer. 2016 Sep;16(9):553-65. doi: 10.1038/nrc.2016.66. Epub 2016 Jul 22. Review. PMID: 27444064

Next generation DNA sequencing applied to tumors in patients with advanced cancer

Johns Hopkins University Medical Center pancreatic cancer research group

- Rapid autopsy provides high quality DNA from tumors (both primary and metastatic)
- Next generation DNA sequencing technologies have revolutionized the study of cancer genomes
- Next generation DNA sequencing methods provide the technical resolution needed to decipher clonal evolution

Distant Metastasis Occurs Late during the Genetic Evolution of Pancreatic Cancer

Shinichi Yachida[1,†], **Siân Jones**[4,†], **Ivana Bozic**[5], **Tibor Antal**[5,6], **Rebecca Leary**[4], **Baojin Fu**[1], **Mihoko Kamiyama**[1], **Ralph H. Hruban**[1,2], **James R. Eshleman**[1], **Martin A. Nowak**[5], **Victor E. Velculescu**[4], **Kenneth W. Kinzler**[4], **Bert Vogelstein**[4], and **Christine A. Iacobuzio-Donahue**[1,2,3,*]

[1]Department of Pathology, The Sol Goldman Pancreatic Cancer Research Center, Johns Hopkins Medical Institutions, Baltimore Maryland 21231 USA

[2]Department of Oncology, The Sol Goldman Pancreatic Cancer Research Center, Johns Hopkins Medical Institutions, Baltimore Maryland 21231 USA

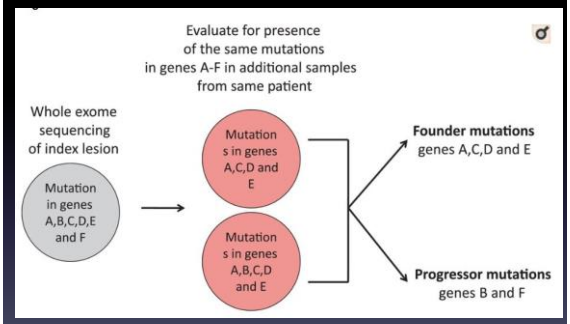
[3]Department of Surgery, The Sol Goldman Pancreatic Cancer Research Center, Johns Hopkins Medical Institutions, Baltimore Maryland 21231 USA

[4]The Ludwig Center for Cancer Genetics and Therapeutics and The Howard Hughes Medical Institute at The Johns Hopkins Kimmel Cancer Center, Baltimore, Maryland 21231 USA

[5]Program for Evolutionary Dynamics, Department of Mathematics, Department of Organismic and Evolutionary Biology, Harvard University, Cambridge, Massachusetts 02138 USA

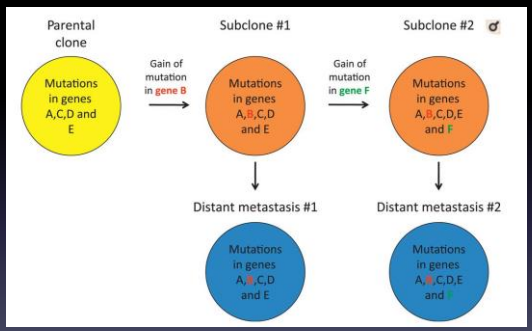
[6]School of Mathematics, University of Edinburgh, Edinburgh EH9-3JZ, UK

Comparative lesion sequencing



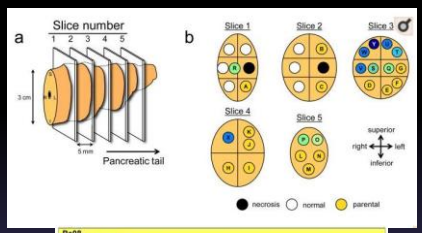
Iacobuzio-Donahue CA. Genetic evolution of pancreatic cancer: lessons learnt from the pancreatic cancer genome sequencing project. Gut. 2012 Jul;61(7):1085-94. doi: 10.1136/gut.2010.236026. Epub 2011 Jul 11.

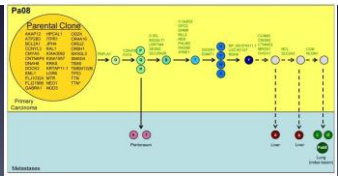
Clonal progression of pancreatic cancer



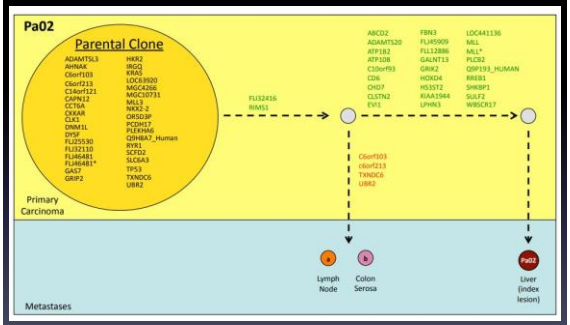
Iacobuzio-Donahue CA. Genetic evolution of pancreatic cancer: lessons learnt from the pancreatic cancer genome sequencing project. Gut. 2012 Jul;61(7):1085-94. doi: 10.1136/gut.2010.236026. Epub 2011 Jul 11.

Geographic mapping of metastatic clones within a primary pancreas tumor

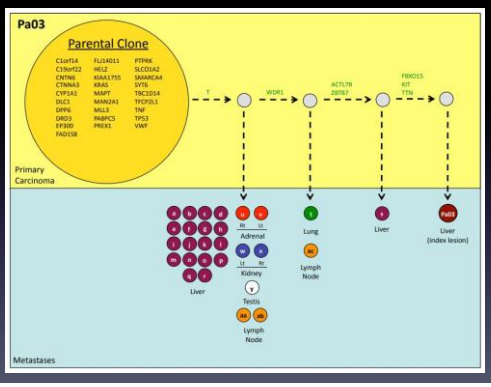




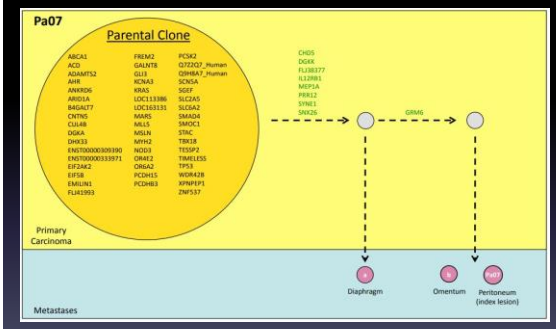
Patient # Pa02



Patient # Pa03



Patient # Pa07



Clonal progression of pancreatic cancer

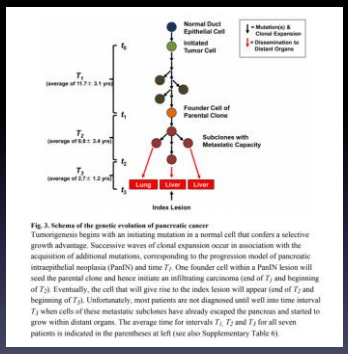


Fig. 3. Schema of the genetic evolution of pancreatic cancer
Tumorigenesis begins with an initiating mutation in a normal cell that confers a selective growth advantage. Successive waves of clonal expansion occur in association with the acquisition of additional mutations, corresponding to the progression model of pancreatic intraepithelial neoplasia (PanIN) and time T_1. One founder cell within a PanIN lesion will seed the parental clone and hence initiate an infiltrating carcinoma (end of T_1 and beginning of T_2). Eventually, the cell that will give rise to the index lesion will appear (end of T_2 and beginning of T_3). Unfortunately, most patients are not diagnosed until well into time interval T_3 when cells of these metastatic subclones have already escaped the pancreas and started to grow within distant organs. The average time for intervals T_1, T_2 and T_3 for all seven patients is indicated in the parentheses at left (see also Supplementary Table 6).

Yachida S1, Jones S, Bozic I, Antal T, Leary R, Fu B, Kamiyama M, Hruban RH, Eshleman JR, Nowak MA, Velculescu VE, Kinzler KW, Vogelstein B, Iacobuzio-Donahue CA. Distant metastasis occurs late during the genetic evolution of pancreatic cancer. Nature. 2010 Oct 28;467(7319):1114-7. doi: 10.1038/nature09515.

Limitations of targeted therapy

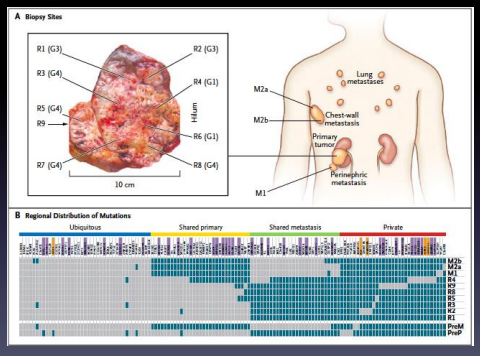
The NEW ENGLAND JOURNAL of MEDICINE

ESTABLISHED IN 1812 MARCH 8, 2012 VOL. 366 NO. 10

Intratumor Heterogeneity and Branched Evolution Revealed by Multiregion Sequencing

Marco Gerlinger, M.D., Andrew J. Rowan, B.Sc., Stuart Horswell, M.Math., James Larkin, M.D., Ph.D., David Endesfelder, Dip.Math., Eva Gronroos, Ph.D., Pierre Martinez, Ph.D., Nicholas Matthews, B.Sc., Aengus Stewart, M.Sc., Patrick Tarpey, Ph.D., Ignacio Varela, Ph.D., Benjamin Phillimore, B.Sc., Sharmin Begum, M.Sc., Neil Q. McDonald, Ph.D., Adam Butler, B.Sc., David Jones, M.Sc., Keiran Raine, M.Sc., Calli Latimer, B.Sc., Claudio R. Santos, Ph.D., Mahrokh Nohadani, H.N.C., Aron C. Eklund, Ph.D., Bradley Spencer-Dene, Ph.D., Graham Clark, B.Sc., Lisa Pickering, M.D., Ph.D., Gordon Stamp, M.D., Martin Gore, M.D., Ph.D., Zoltan Szallasi, M.D., Julian Downward, Ph.D., P. Andrew Futreal, Ph.D., and Charles Swanton, M.D., Ph.D.

One tumor, multiple subclones

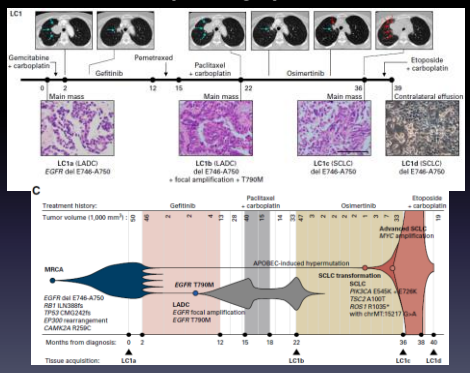


Gerlinger et al. *N Engl J Med.* 2012;366:883.

Intratumoral heterogeneity

St Luke's Mountain States Tumor Institute

Clonal (i.e. target) evolution



Lee et al. *J Clin Oncol.* 2017;35:3065.

Patient # 1: 50 year-old-woman with recurrent endometrial cancer

E, F, G. POSTERIOR WALL OF BLADDER, SIGMOID COLON, AND BILATERAL ADNEXAL REMNANT, *EN BLOC* RESECTION

BLADDER, PARTIAL CYSTECTOMY
- HIGH-GRADE, POORLY-DIFFERENTIATED CARCINOMA INVOLVING THE BLADDER ADVENTITIA AND MUSCULARIS PROPRIA
- TUMOR MEASURES 5.5 CM IN SIZE
- TUMOR FOCALLY PRESENT AT A PERIPHERAL MARGIN

COLON, SEGMENTAL RESECTION
- MULTIPLE NODULES OF HIGH-GRADE, POORLY-DIFFERENTIATED ADENOCARCINOMA INVOLVING THE COLONIC MESENTERY
- NO INVOLVEMENT OF THE COLONIC MUSCULARIS PROPRIA BY CARCINOMA
- PROXIMAL, DISTAL, AND MESENTERIC ROOT SURGICAL RESECTION MARGINS NEGATIVE FOR CARCINOMA
- 14 PERI-COLIC LYMPH NODES NEGATIVE FOR CARCINOMA (0/14)

BILATERAL ADNEXAL REMNANT, RESECTION
- MULTIPLE NODULES OF HIGH-GRADE, POORLY-DIFFERENTIATED CARCINOMA INVOLVING A REMNANT RIGHT OVARY AND BILATERAL REMNANT ADNEXAL TISSUE
- REMNANT OVARY AND ADNEXAL TUMORS MEASURE UP TO 5.5 CM IN SIZE
- MULTIPLE DISCRETE PERITONEAL TUMOR NODULES IDENTIFIED, MEASURING UP TO 3.5 CM IN SIZE

FOUNDATIONONE®CDx

NO REPORTABLE ALTERATIONS WITH COMPANION DIAGNOSTIC (CDx) CLAIMS
See professional services section for additional information

OTHER ALTERATIONS & BIOMARKERS IDENTIFIED

Results reported in this section are not prescriptive or conclusive for labeled use of any specific therapeutic product. See *professional services* section for additional information.

Microsatellite status MS-Stable §
Tumor Mutational Burden 6 Muts/Mb §
ARID1A Q780*
ATR L1017fs*9
CD274 (PD-L1) amplification §
JAK2 amplification §
PDCD1LG2 (PD-L2) amplification §
PIK3R1 F456_Q457del
PTEN Y88*
TP53 R249G

§ Refer to appendix for limitation statements related to detection of any copy number alterations, gene rearrangements, MSI or TMB result in this section.
Please refer to appendix for Explanation of Clinical Significance Classification and for variants of unknown significance (VUS).

Patient # 2: 70-year-old woman with a history of colon cancer 3 years ago, now with a liver mass

Final Diagnosis

RIGHT LIVER LESION, CT-GUIDED CORE NEEDLE BIOPSY:

METASTATIC ADENOCARCINOMA OF COLONIC ORIGIN. SEE COMMENT.
PENDING FOUNDATION ONE® CDx TESTING

FOUNDATIONONE®CDx

Companion Diagnostic (CDx) Associated Findings

GENOMIC FINDINGS DETECTED	FDA-APPROVED THERAPEUTIC OPTIONS
KRAS wildtype (codons 12 & 13)	Erbitux® (Cetuximab)
KRAS/NRAS wildtype (codons 12, 13, 59, 61, 117, & 146 in exons 2, 3, & 4)	Vectibix® (Panitumumab)

OTHER ALTERATIONS & BIOMARKERS IDENTIFIED

Results reported in this section are not prescriptive or conclusive for labeled use of any specific therapeutic product. See *professional services* section for additional information.

Microsatellite status MS-Stable §
Tumor Mutational Burden 9 Muts/Mb §
APC S457*
EPHB1 A922T
PIK3CA E542K
TP53 T125M

§ Refer to appendix for limitation statements related to detection of any copy number alterations, gene rearrangements, MSI or TMB result in this section.
Please refer to appendix for Explanation of Clinical Significance Classification and for variants of unknown significance (VUS).

Do targets behave the same in different tumors?

ORIGINAL ARTICLE

Vemurafenib in Multiple Nonmelanoma Cancers with *BRAF* V600 Mutations

Table 2. Preliminary Best Response According to Cohort.*

Variable	NSCLC (N=20)	Colorectal Cancer		Cholangiocarcinoma (N=8)	ECD or LCH (N=18)	Anaplastic Thyroid Cancer (N=7)
		Vemurafenib (N=10)	Vemurafenib + Cetuximab (N=27)			
Patients with ≥1 postbaseline assessment — no.	19	10	26	8	14	7
Complete response — no. (%)	0	0	0	0	1 (7)	1 (14)
Partial r						

In conclusion, we found that the *BRAF* V600 mutation is a targetable oncogene in some, but not all, cancer types. Histology-independent,

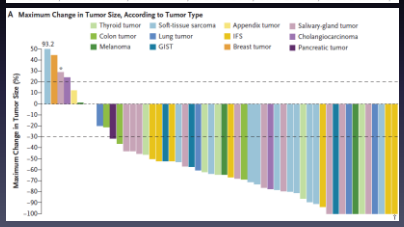
Hyman et al. *N Engl J Med.* 2015;373:726.

Do targets behave the same in different tumors?

ORIGINAL ARTICLE

Efficacy of Larotrectinib in *TRK* Fusion–Positive Cancers in Adults and Children

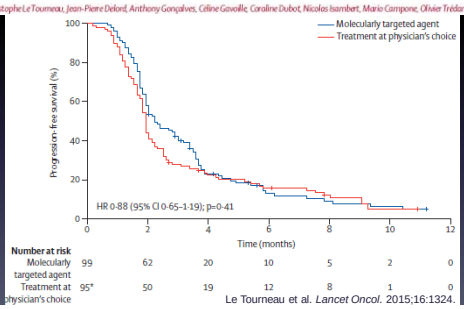
A. Drilon, T.W. Laetsch, S. Kummar, S.G. DuBois, U.N. Lassen, G.D. Demetri



Drilon et al. *N Engl J Med.* 2018;379:111.

Molecularly targeted therapy based on tumour molecular profiling versus conventional therapy for advanced cancer (SHIVA): a multicentre, open-label, proof-of-concept, randomised, controlled phase 2 trial

Christophe Le Tourneau, Jean-Pierre Delord, Anthony Gonçalves, Céline Gavoille, Caroline Dubot, Nicolas Isambert, Mario Campone, Olivier Trédan,

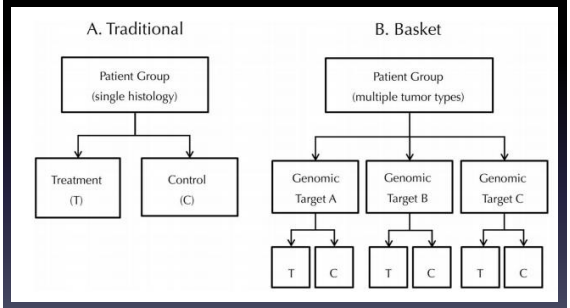


Le Tourneau et al. *Lancet Oncol.* 2015;16:1324.

NCI MATCH TRIAL 2016

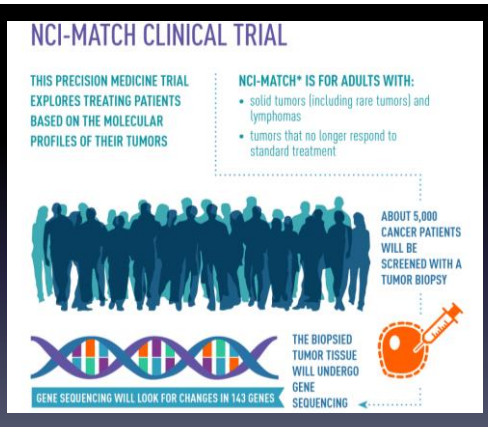
- **Molecular Analysis for Therapy Choice**
- **Trial run by NCI**
- **Basket trial**
- **24 treatment arms**
- **Enroll 5000 patients**
- **Open July 2015 – June 2022**
- **End points are ORR and PFS**

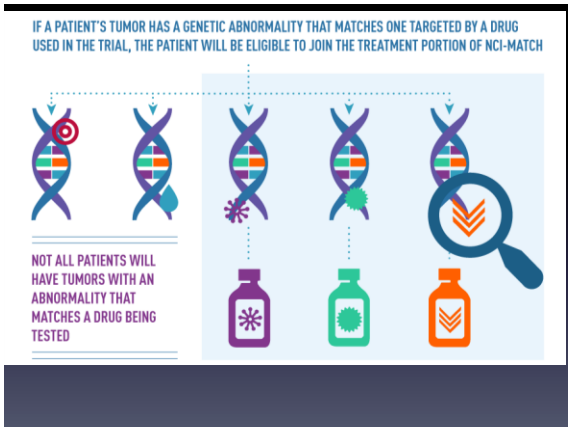
Clinical trial design: Traditional v. Basket



Wang DH, Park JY. Precision Medicine in Gastrointestinal Pathology. *Arch Pathol Lab Med.* 2016 May;140(5):449-60. doi: 10.5858/arpa.2015-0317-RA. PMID: 27128302

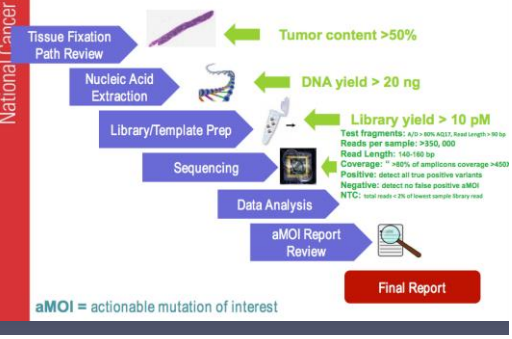
NCI-MATCH CLINICAL TRIAL





National Cancer Institute

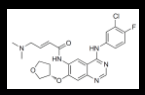
Workflow and Turnover Time of the Assay System



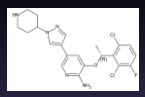
aMOI = actionable mutation of interest

NCI MATCH: targeted agents

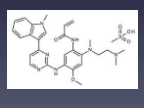
Afatanib



Crizotinib

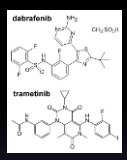


Osimertinib

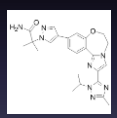


NCI MATCH: targeted agents

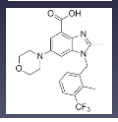
Dabrafenib + trametinib



Taselisib

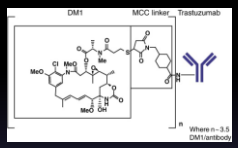


GSK2636771



NCI MATCH: Monoclonal antibody biologic therapies

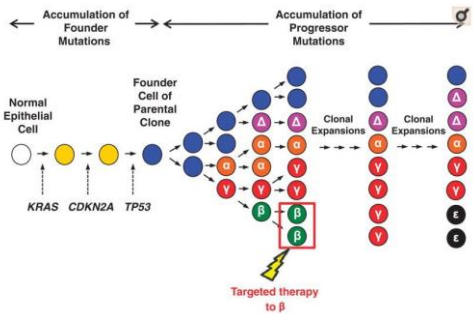
Ado-trastuzumab emtansine



Nivolumab

Humanized mouse monoclonal IgG4 anti-PD-1 antibody

Targeted treatment of the β clone in a pancreatic cancer

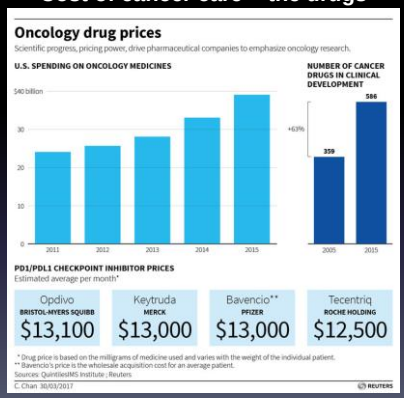


Implications of deep sequencing of tumors

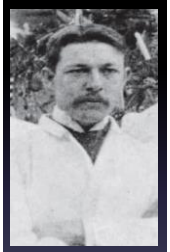
- Every cancer is a unique organism
- Clinically detectable cancers (>10^9 cells) are a complex mosaic of distinct clones
- The most common driver mutations identified in cancers are not targetable at the current time
- No matter how effective a targeted therapy, resistant cancer cells will always grow back to fill the void
- Targeted therapy can only slow down advanced cancers, you cannot hope to kill every single subclone in a genetically diverse tumor

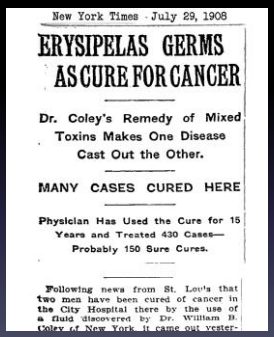
Navican precision cancer care©: Intermountain health in Utah

Cost of cancer care – the drugs



The first immunotherapy





New York Times - July 29, 1908

ERYSIPELAS GERMS AS CURE FOR CANCER

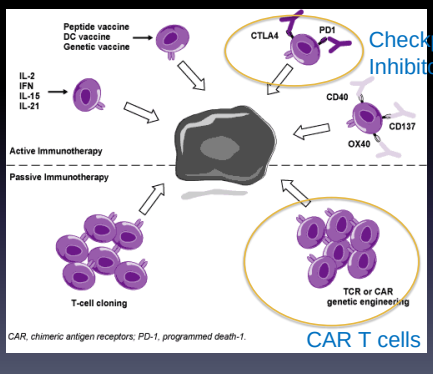
Dr. Coley's Remedy of Mixed Toxins Makes One Disease Cast Out the Other.

MANY CASES CURED HERE

Physician Has Used the Cure for 15 Years and Treated 430 Cases—Probably 150 Sure Cures.

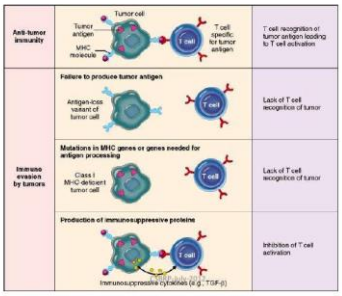
Following news from St. Louis that two men have been cured of cancer in the City Hospital there by the use of a fluid discovered by Dr. William B. Coley of New York, it came out yester-

Immunotherapy: overview

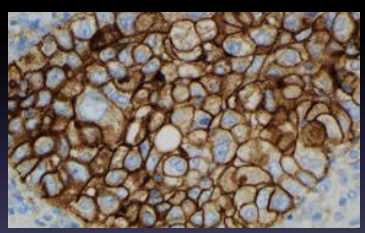


Immunotherapy in Oncology

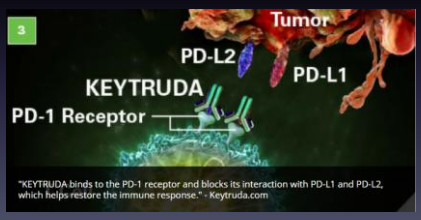
Mechanisms by which tumors evade the immune system



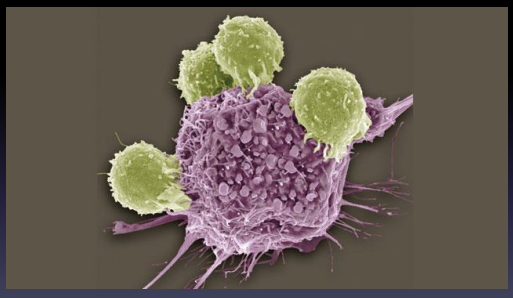
Immunohistochemistry: anti-PD-L1



KEYTRUDA®
(pembrolizumab) Injection 100 mg



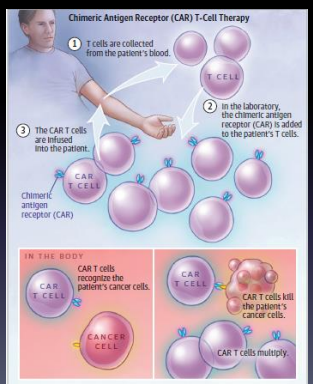
Chimeric antigen receptor (CAR) T-cells



Chimeric antigen receptor (CAR) T-cells

Chimeric antigen receptor (CAR) T-cells

Immunotherapy: CAR T cells



Pagel et al. *JAMA Oncol.* 2017;3:1595.

Grupp et al. *N Engl J Med.* 2013;368:1509.

Thank you

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