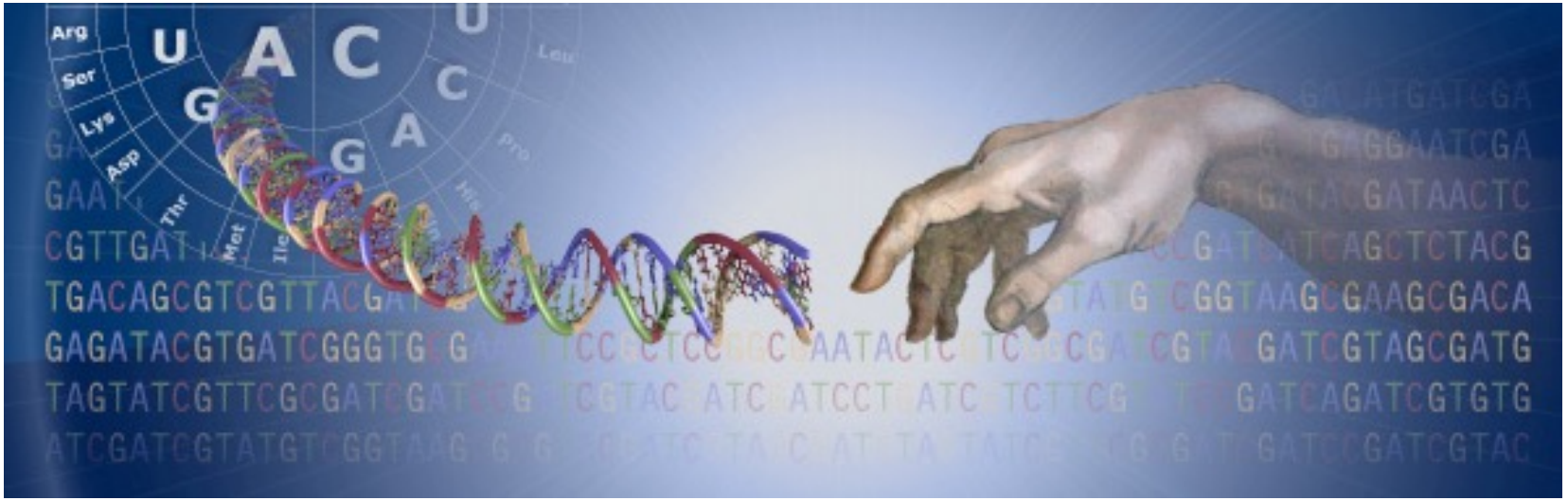
Arg
Ser
Lys
Asp
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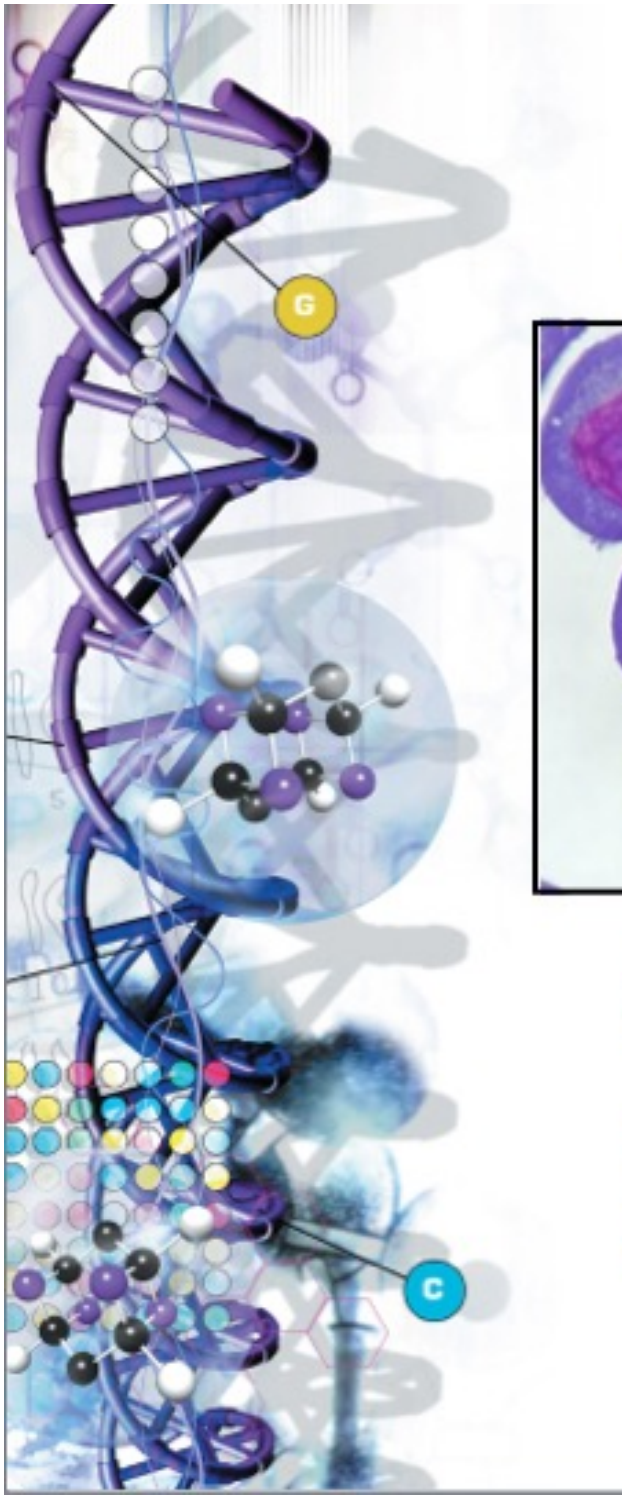
Pro


GENOME:
THE FUTURE IS NOW

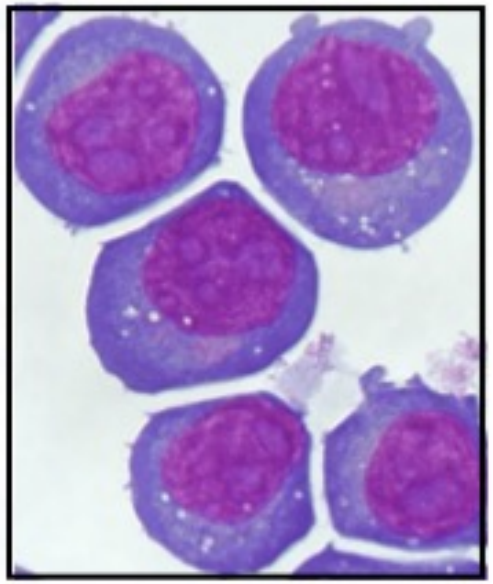
Exploration often gives a different perspective

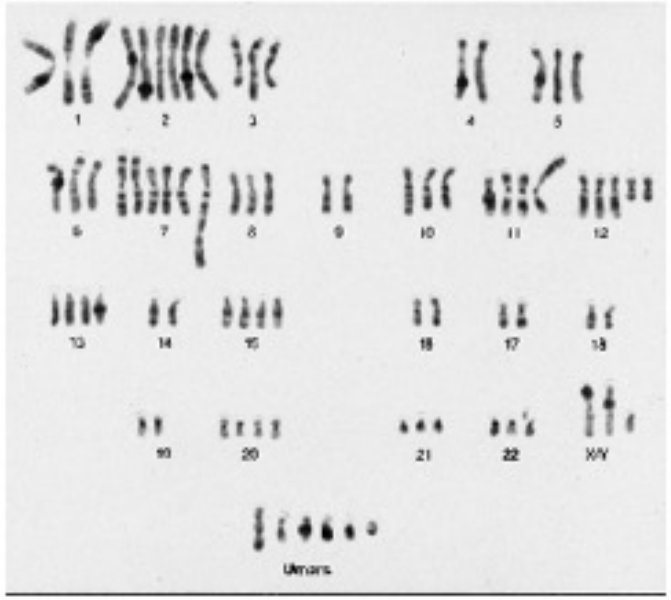
Earthrise from Apollo 11, 1969



Cancer

A Disease of the Genome





Challenge in Treating Cancer:

- Every tumor is different
- Every cancer patient is different

ICGC

Επιδημιολογία του καρκίνου

Figure 3. Leading Sites of New Cancer Cases and Deaths – 2018 Estimates

Estimated New Cases

Male			Female		
Prostate	164,690	19%	Breast	266,120	30%
Lung & bronchus	121,680	14%	Lung & bronchus	112,350	13%
Colon & rectum	75,610	9%	Colon & rectum	64,640	7%
Urinary bladder	62,380	7%	Uterine corpus	63,230	7%
Melanoma of the skin	55,150	6%	Thyroid	40,900	5%
Kidney & renal pelvis	42,680	5%	Melanoma of the skin	36,120	4%
Non-Hodgkin lymphoma	41,730	5%	Non-Hodgkin lymphoma	32,950	4%
Oral cavity & pharynx	37,160	4%	Pancreas	26,240	3%
Leukemia	35,030	4%	Leukemia	25,270	3%
Liver & intrahepatic bile duct	30,610	4%	Kidney & renal pelvis	22,660	3%
All sites	**856,370**	**100%**	**All sites**	**878,980**	**100%**

Estimated Deaths

Male			Female		
Lung & bronchus	83,550	26%	Lung & bronchus	70,500	25%
Prostate	29,430	9%	Breast	40,920	14%
Colon & rectum	27,390	8%	Colon & rectum	23,240	8%
Pancreas	23,020	7%	Pancreas	21,310	7%
Liver & intrahepatic bile duct	20,540	6%	Ovary	14,070	5%
Leukemia	14,270	4%	Uterine corpus	11,350	4%
Esophagus	12,850	4%	Leukemia	10,100	4%
Urinary bladder	12,520	4%	Liver & intrahepatic bile duct	9,660	3%
Non-Hodgkin lymphoma	11,510	4%	Non-Hodgkin lymphoma	8,400	3%
Kidney & renal pelvis	10,010	3%	Brain & other nervous system	7,340	3%
All sites	**323,630**	**100%**	**All sites**	**286,010**	**100%**

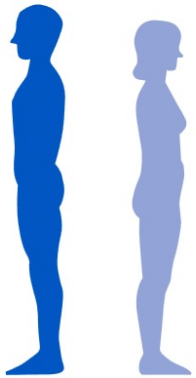
Estimates are rounded to the nearest 10, and cases exclude basal cell and squamous cell skin cancers and in situ carcinoma except urinary bladder. Ranking is based on modeled projections and may differ from the most recent observed data.

Επιδημιολογία του καρκίνου

Figure 3. Leading Sites of New Cancer Cases and Deaths – 2023 Estimates

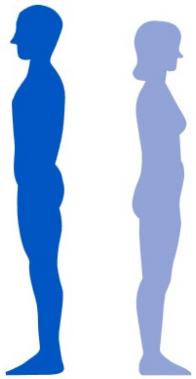
Estimated New Cases

Male			Female		
Prostate	288,300	29%	Breast	297,790	31%
Lung & bronchus	117,550	12%	Lung & bronchus	120,790	13%
Colon & rectum	81,860	8%	Colon & rectum	71,160	8%
Urinary bladder	62,420	6%	Uterine corpus	66,200	7%
Melanoma of the skin	58,120	6%	Melanoma of the skin	39,490	4%
Kidney & renal pelvis	52,360	5%	Non-Hodgkin lymphoma	35,670	4%
Non-Hodgkin lymphoma	44,880	4%	Thyroid	31,180	3%
Oral cavity & pharynx	39,290	4%	Pancreas	30,920	3%
Leukemia	35,670	4%	Kidney & renal pelvis	29,440	3%
Pancreas	33,130	3%	Leukemia	23,940	3%
All sites	**1,010,310**		**All sites**	**948,000**	



Estimated Deaths

Male			Female		
Lung & bronchus	67,160	21%	Lung & bronchus	59,910	21%
Prostate	34,700	11%	Breast	43,170	15%
Colon & rectum	28,470	9%	Colon & rectum	24,080	8%
Pancreas	26,620	8%	Pancreas	23,930	8%
Liver & intrahepatic bile duct	19,000	6%	Ovary	13,270	5%
Leukemia	13,900	4%	Uterine corpus	13,030	5%
Esophagus	12,920	4%	Liver & intrahepatic bile duct	10,380	4%
Urinary bladder	12,160	4%	Leukemia	9,810	3%
Non-Hodgkin lymphoma	11,780	4%	Non-Hodgkin lymphoma	8,400	3%
Brain & other nervous system	11,020	3%	Brain & other nervous system	7,970	3%
All sites	**322,080**		**All sites**	**287,740**	



Estimates are rounded to the nearest 10, and cases exclude basal cell and squamous cell skin cancers and in situ carcinoma except urinary bladder. Estimates do not include Puerto Rico or other US territories. Ranking is based on modeled projections and may differ from the most recent observed data.

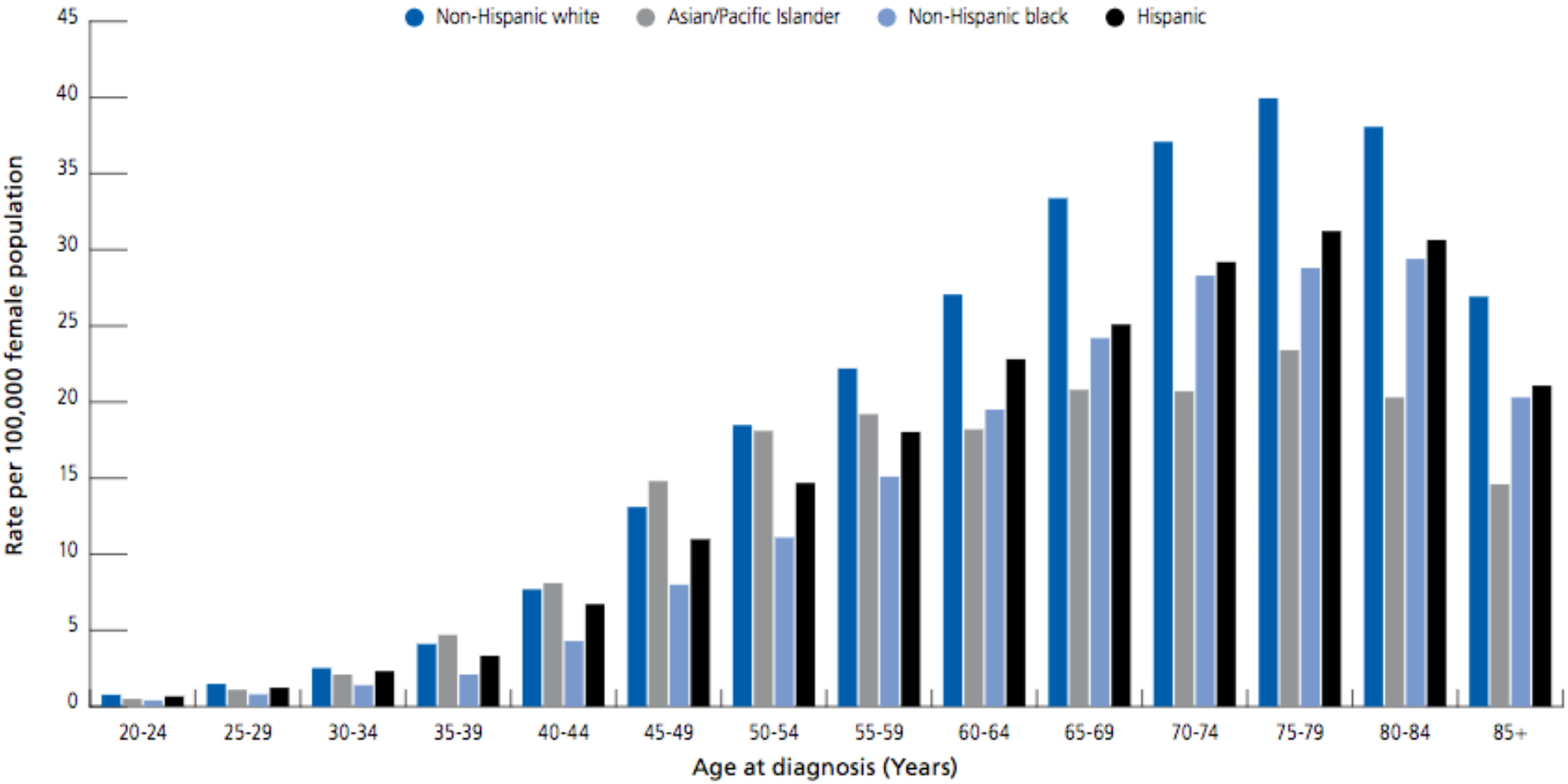
Table 1. Estimated Number* of New Cancer Cases and Deaths by Sex, US, 2023

	Estimated New Cases			Estimated Deaths		
	Both sexes	**Male**	**Female**	**Both sexes**	**Male**	**Female**
All sites	1,958,310	1,010,310	948,000	609,820	322,080	287,740
Oral cavity & pharynx	54,540	39,290	15,250	11,580	8,140	3,440
Tongue	18,040	13,180	4,860	2,940	1,950	990
Mouth	14,820	8,680	6,140	3,090	1,870	1,220
Pharynx	20,070	16,340	3,730	4,140	3,260	880
Other oral cavity	1,610	1,090	520	1,410	1,060	350
Digestive system	348,840	194,980	153,860	172,010	99,350	72,660
Esophagus	21,560	17,030	4,530	16,120	12,920	3,200
Stomach	26,500	15,930	10,570	11,130	6,690	4,440
Small intestine	12,070	6,580	5,490	2,070	1,170	900
Colon & rectum†	153,020	81,860	71,160	52,550	28,470	24,080
Colon	106,970	54,420	52,550			
Rectum	46,050	27,440	18,610			
Anus, anal canal, & anorectum	9,760	3,180	6,580	1,870	860	1,010
Liver & intrahepatic bile duct	41,210	27,980	13,230	29,380	19,000	10,380
Gallbladder & other biliary	12,220	5,750	6,470	4,510	1,900	2,610
Pancreas	64,050	33,130	30,920	50,550	26,620	23,930
Other digestive organs	8,450	3,540	4,910	3,830	1,720	2,110
Respiratory system	256,290	131,150	125,140	132,330	71,170	61,160
Larynx	12,380	9,900	2,480	3,820	3,070	750
Lung & bronchus	238,340	117,550	120,790	127,070	67,160	59,910
Other respiratory organs	5,570	3,700	1,870	1,440	940	500
Bones & joints	3,970	2,160	1,810	2,140	1,200	940
Soft tissue (including heart)	13,400	7,400	6,000	5,140	2,720	2,420
Skin (excluding basal & squamous)	104,930	62,810	42,120	12,470	8,480	3,990
Melanoma of the skin	97,610	58,120	39,490	7,990	5,420	2,570
Other nonepithelial skin	7,320	4,690	2,630	4,480	3,060	1,420
Other nonepithelial skin	7,320	4,690	2,630	4,480	3,060	1,420
Breast	300,590	2,800	297,790	43,700	530	43,170
Genital system	414,350	299,540	114,810	69,660	35,640	34,020
Uterine cervix	13,960		13,960	4,310		4,310
Uterine corpus	66,200		66,200	13,030		13,030
Ovary	19,710		19,710	13,270		13,270
Vulva	6,470		6,470	1,670		1,670
Vagina & other genital, female	8,470		8,470	1,740		1,740
Prostate	288,300	288,300		34,700	34,700	
Testis	9,190	9,190		470	470	
Penis & other genital, male	2,050	2,050		470	470	
Urinary system	168,560	117,590	50,970	32,590	22,680	9,910
Urinary bladder	82,290	62,420	19,870	16,710	12,160	4,550
Kidney & renal pelvis	81,800	52,360	29,440	14,890	9,920	4,970
Ureter & other urinary organs	4,470	2,810	1,660	990	600	390
Eye & orbit	3,490	1,900	1,590	430	240	190
Brain & other nervous system	24,810	14,280	10,530	18,990	11,020	7,970
Endocrine system	47,230	14,340	32,890	3,240	1,560	1,680
Thyroid	43,720	12,540	31,180	2,120	970	1,150
Other endocrine	3,510	1,800	1,710	1,120	590	530
Lymphoma	89,380	49,730	39,650	21,080	12,320	8,760
Hodgkin lymphoma	8,830	4,850	3,980	900	540	360
Non-Hodgkin lymphoma	80,550	44,880	35,670	20,180	11,780	8,400
Myeloma	35,730	19,860	15,870	12,590	7,000	5,590
Leukemia	59,610	35,670	23,940	23,710	13,900	9,810
Acute lymphocytic leukemia	6,540	3,660	2,880	1,390	700	690
Chronic lymphocytic leukemia	18,740	12,130	6,610	4,490	2,830	1,660
Acute myeloid leukemia	20,380	11,410	8,970	11,310	6,440	4,870
Chronic myeloid leukemia	8,930	5,190	3,740	1,310	780	530
Other leukemia‡	5,020	3,280	1,740	5,210	3,150	2,060
Other & unspecified primary sites‡	32,590	16,810	15,780	48,160	26,130	22,030

*Rounded to the nearest 10; cases exclude basal cell and squamous cell skin cancer and in situ carcinoma except urinary bladder. About 55,720 cases of female breast ductal carcinoma in situ and 89,070 cases of melanoma in situ will be diagnosed in 2023. †Cases and deaths for colon cancer include appendix. Deaths for colon and rectal cancers are combined because a large number of deaths from rectal cancer are misclassified as colon. ‡More deaths than cases may reflect lack of specificity in recording underlying cause of death on death certificates and/or an undercount in the case estimate.

Source: Estimated new cases are based on 2005-2019 incidence data reported by the North American Association of Central Cancer Registries (NAACCR). Estimated deaths are based on 2006-2020 US mortality data, National Center for Health Statistics, Centers for Disease Control and Prevention.

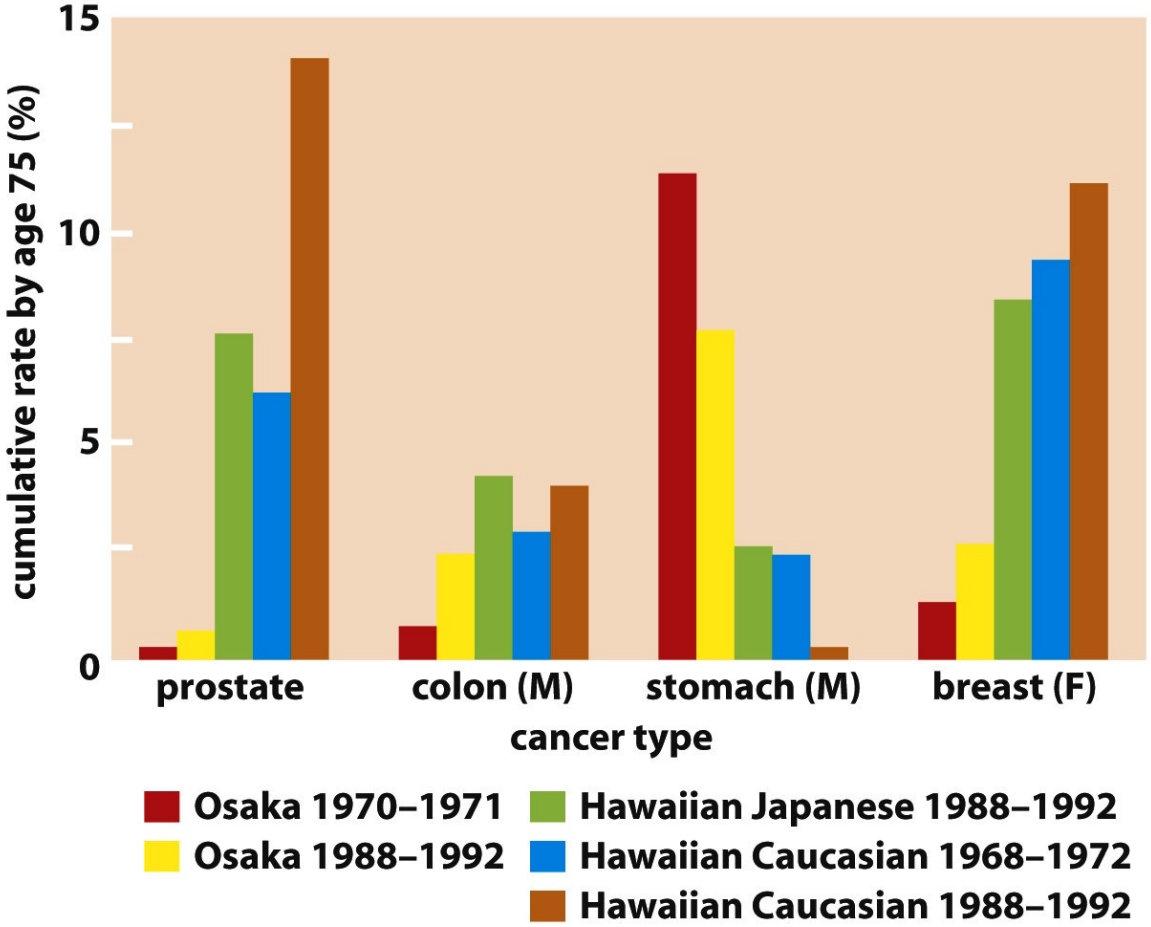
Figure S4. Epithelial Ovarian Cancer Incidence Rates* by Age and Race, US, 2010-2014



*Age adjusted to the 2000 US standard population. Persons of Hispanic origin may be of any race; Asians/Pacific Islanders include those of Hispanic and non-Hispanic origin. American Indians and Alaska Natives are not shown due to <25 cases reported for several age groups.

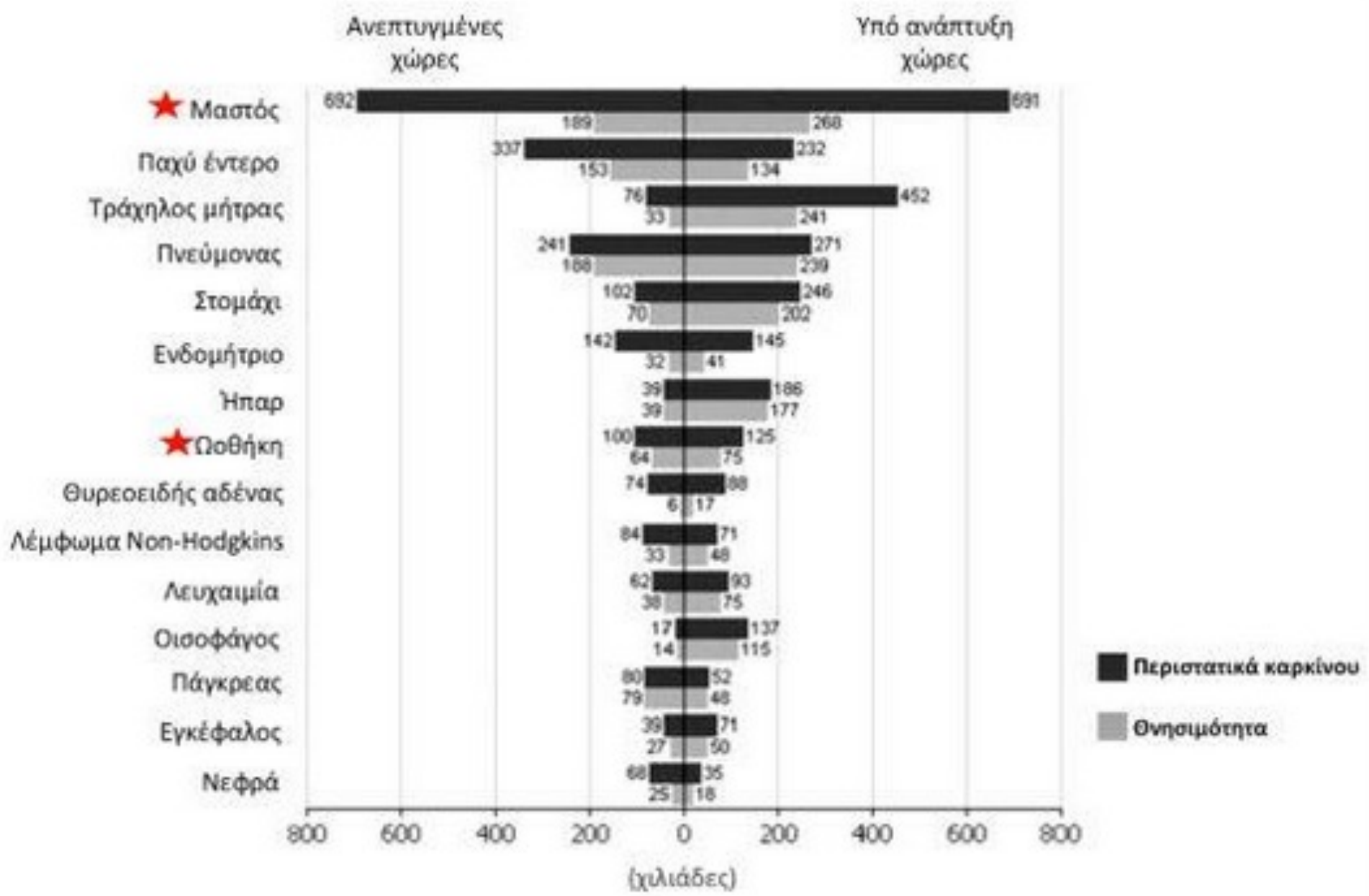
Source: NAACCR, 2017.

Country –to-country comparisons of cancer incidence



Geographic variation in cancer incidence and death rates

Countries showing highest and lowest incidence of specific types of cancer[a]			
Cancer site	Country of highest risk	Country of lowest risk	Relative risk H/L[b]
Skin (melanoma)	Australia (Queensland)	Japan	155
Lip	Canada (Newfoundland)	Japan	151
Nasopharynx	Hong Kong	United Kingdom	100
Prostate	U.S. (African American)	China	70
Liver	China (Shanghai)	Canada (Nova Scotia)	49
Penis	Brazil	Israel (Ashkenazic)	42
Cervix (uterus)	Brazil	Israel (non-Jews)	28
Stomach	Japan	Kuwait	22
Lung	U.S. (Louisiana, African American)	India (Madras)	19
Pancreas	U.S. (Los Angeles, Korean American)	India	11
Ovary	New Zealand (Polynesian)	Kuwait	8

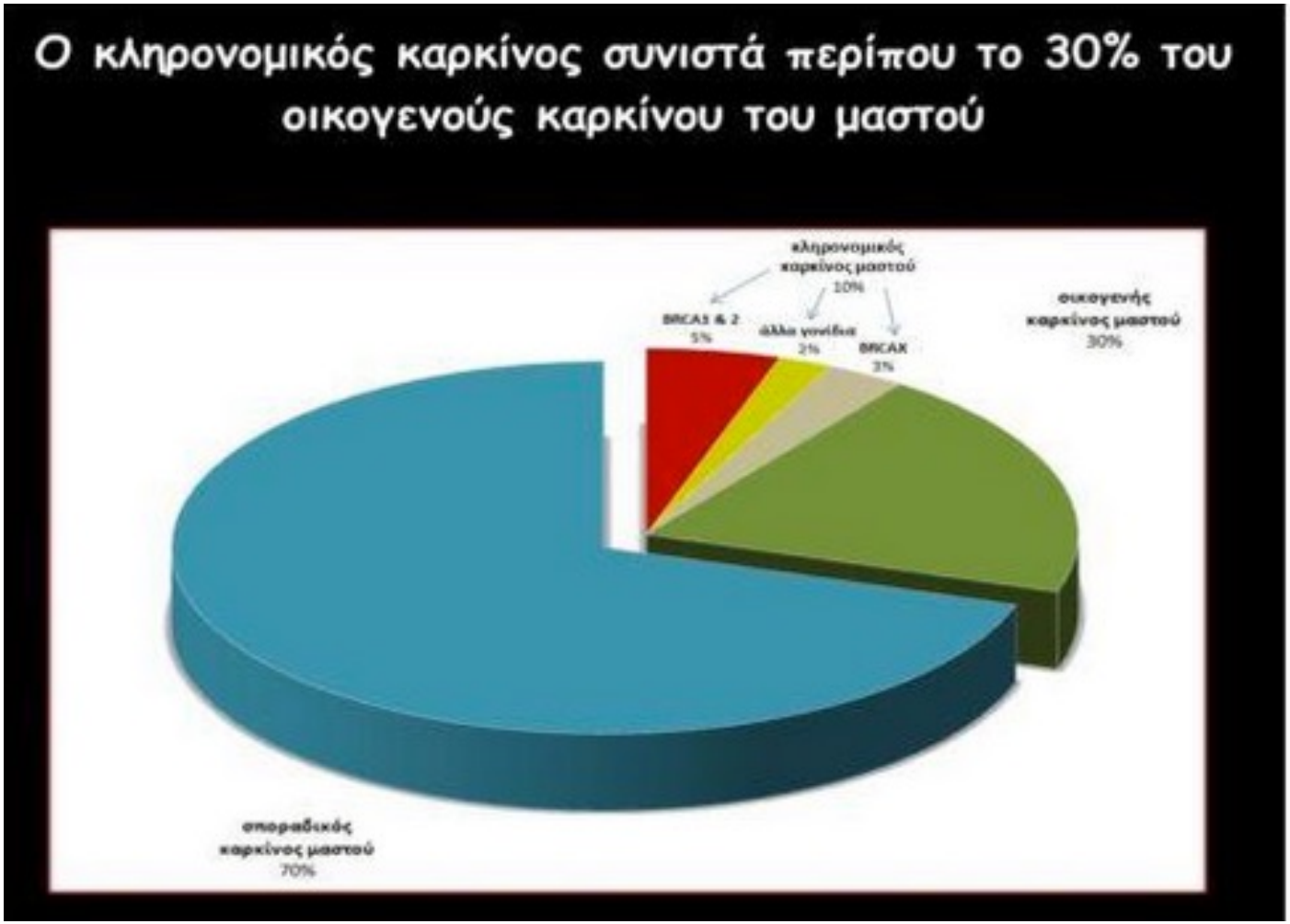

Ανεπτυγμένες χώρες
Υπό ανάπτυξη χώρες
Μαστός
Παχύ έντερο
Τράχηλος μήτρας
Πνεύμονας
Στομάχι
Ενδομήτριο
Ήπαρ
Ωοθήκη
Θυρεοειδής αδένας
Λέμφωμα Non-Hodgkins
Λευχαιμία
Οισοφάγος
Πάγκρεας
Εγκέφαλος
Νεφρά
692
189
691
268
337
153
232
134
76
33
452
241
241
168
271
239
102
70
246
202
142
32
145
41
39
39
186
177
100
64
125
75
74
6
88
17
84
33
71
48
62
38
93
75
17
14
137
115
80
79
52
48
39
27
71
50
68
25
35
18
800
600
400
200
0
200
400
600
800
(χιλιάδες)
Περιστατικά καρκίνου
Θνησιμότητα

Increase in Death Rates for Certain Cancers in Particular Geographical Areas

Cancer Type	Region	Possible Explanation
Breast	Northeast	*BRCA1* mutations, greater lifetime exposure to estrogens (early menstruation, late menopause, older age of first birth, exposure to pesticides)
Colon	Northeast	Dietary factors, medical screening
Lung	White men in south, white women in west, blacks in northern cities	Changes in regional trends in cigarette smoking
Lung	Men in southern coastal areas	Asbestos exposure while working in shipyards during World War II
Mouth, throat	Women in rural south	Smokeless tobacco
Esophagus	Washington, D.C., coastal South Carolina	Alcohol and tobacco, deficiencies of fruits and vegetables in diet

The risks of cancer often seem to be increased by assignable influences including lifestyle

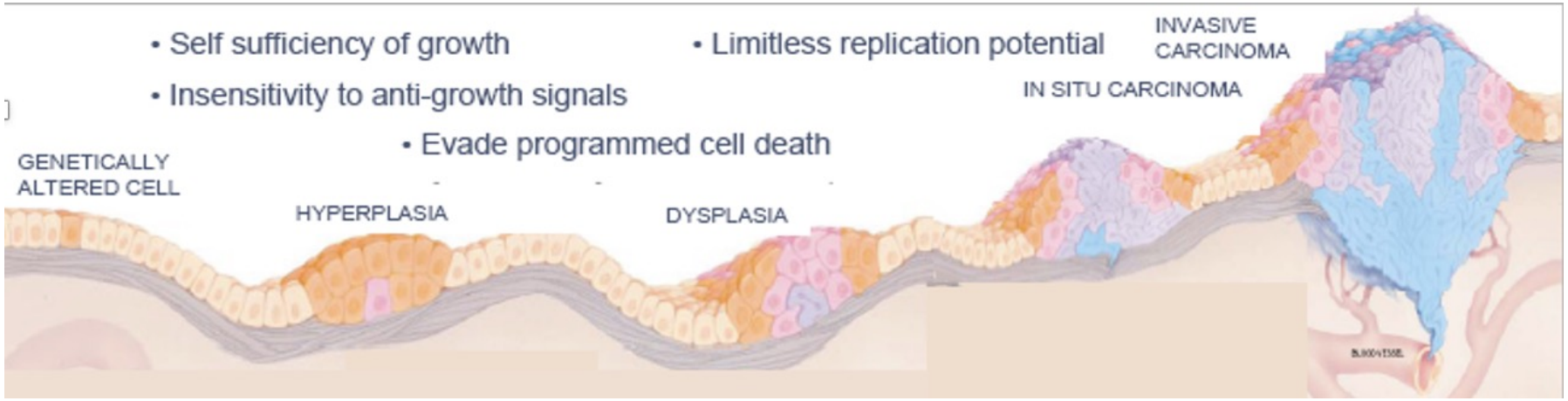
παραγοντες κινδύνου



Cancer in Families

Clues to the genetic basis of neoplasia

- **Certain types of tumors or groups of tumors are inherited within families**

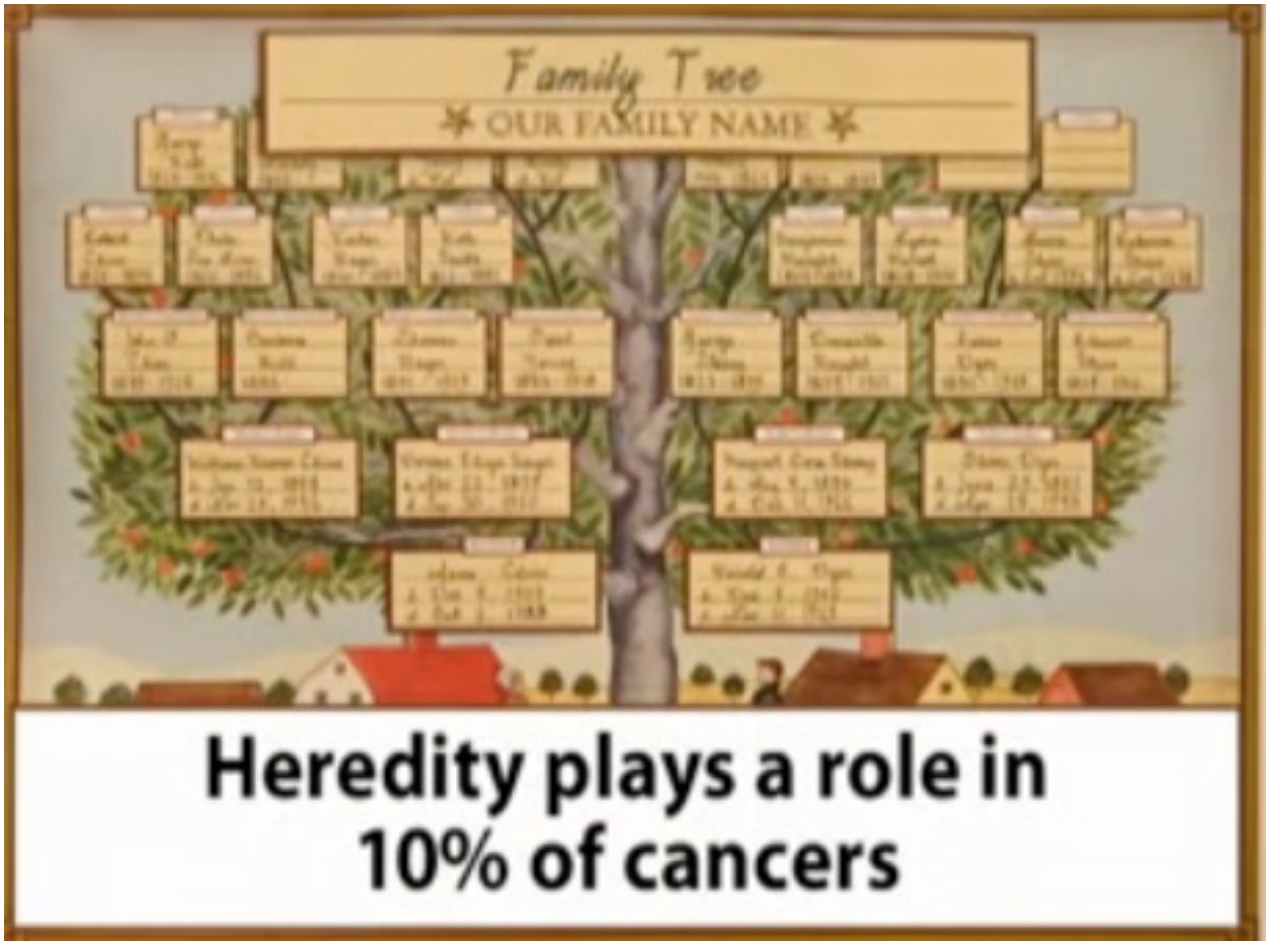


Cancer is a disease of the genome

What I mean by this is that all known cancers carry somatic DNA alterations that make it possible for the cells to grow without the normal limits.

Cancer risk can be familial, due to inherited mutations that are present in every cell.

Κληρονομικότητα


Family Tree
OUR FAMILY NAME
Heredity plays a role in
10% of cancers

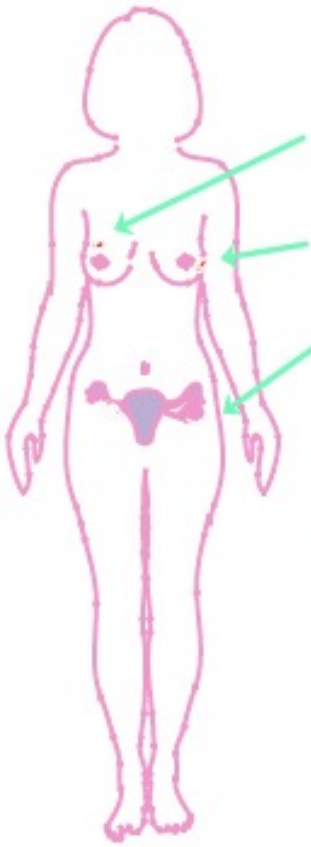

Some types of cancer can be hereditary:
Prostate Cancer
Breast Cancer

Hereditary breast cancer

Early onset, familial breast cancer

- Observation: women with **early onset breast** cancer sometimes have multiple affected family members
- After 17 years, identified BRCA1 in 1993 as a strong genetic risk factor for breast and ovarian cancer

Risk of cancer in carriers of BRCA1 and BRCA2 mutations



Breast cancer: 40%-85% (often early age at dx)

Opposite breast cancer: 40%-60%

Ovarian cancer: 15%-40%

In BRCA2 males, risk of breast cancer is elevated, and the risk of early prostate cancer may also be elevated.

Begg CB. *J Natl Cancer Inst.* 2002; 94:1221-1226.

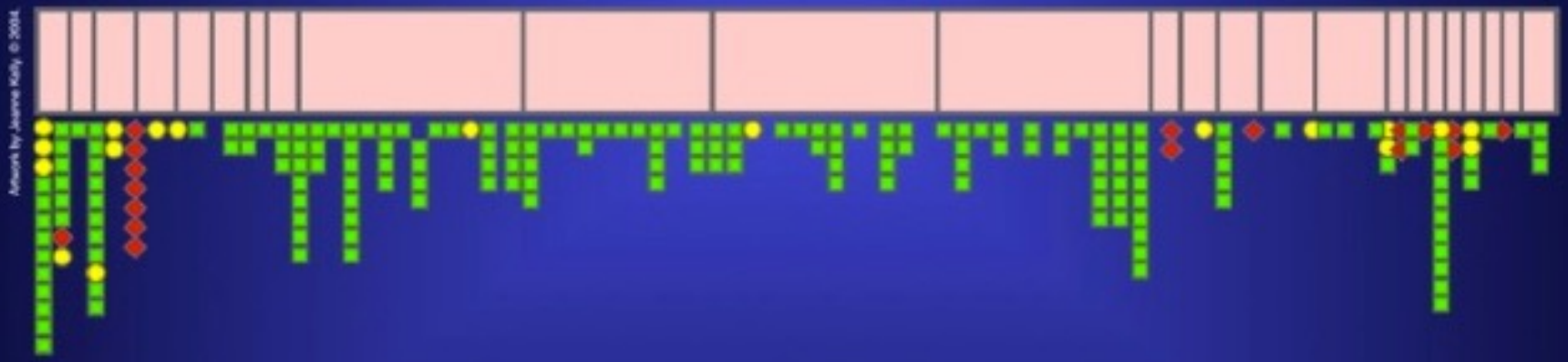
Breast Cancer Linkage Consortium. *J Natl Cancer Inst.* 1999;91:1310-1316.

Ford D, DF Easton, Stratton M, et al. *Am J Hum Genet.* 1998;62:676-689.

Slide courtesy Judy Garber, ASCO

Mutations in Cancer Susceptibility Genes: *BRCA1*

- On chromosome 17
- Protein has role in genomic stability
- Autosomal dominant transmission
- ~500 different mutations reported



- Nonsense/Frameshift
- Missense
- Splice-site

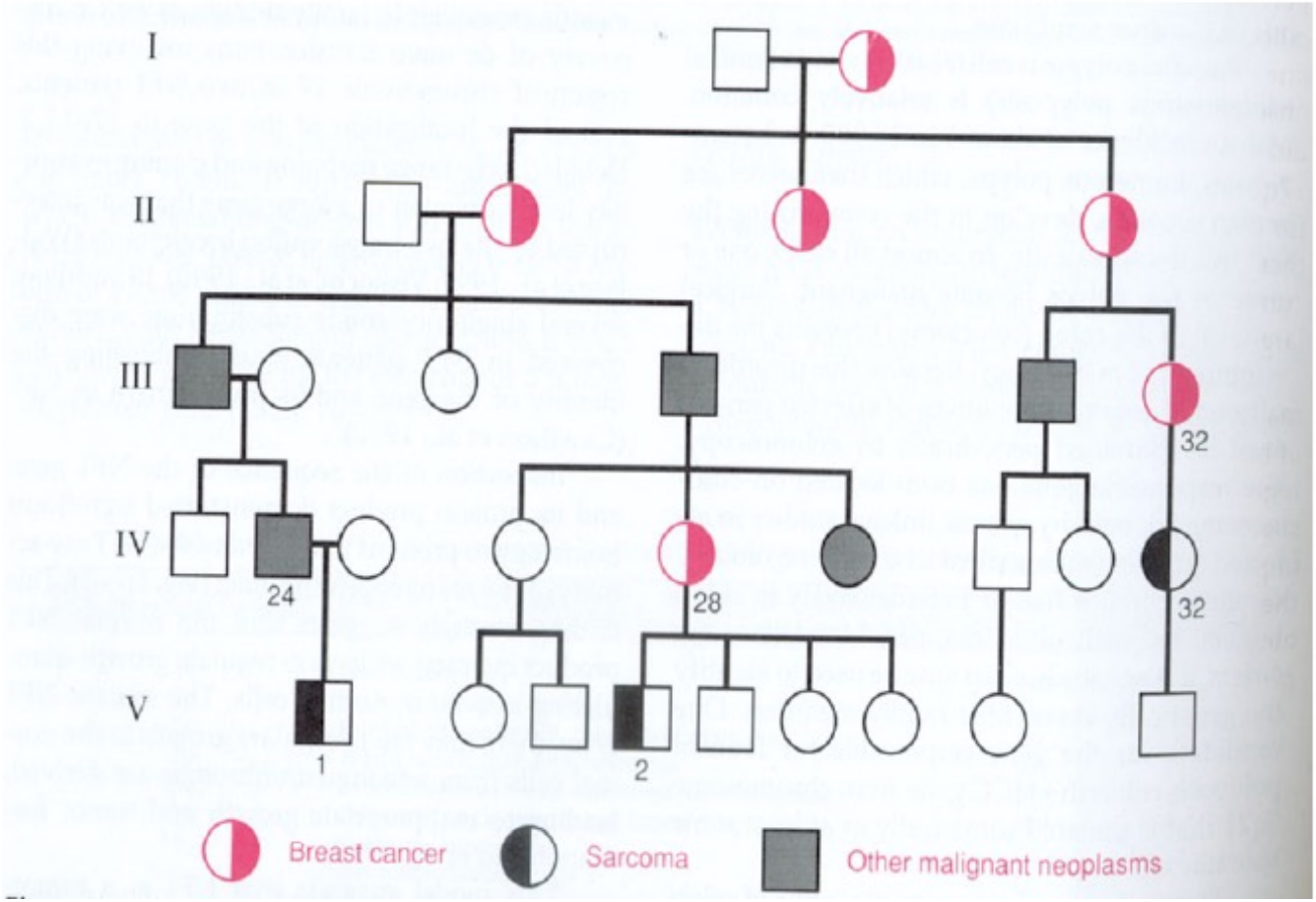
NATIONAL CANCER INSTITUTE

Data Sharing

BRCA Exchange aggregates data on thousands of BRCA variants to inform understanding of cancer risk

Li-Fraumeni Syndrome

Mutations identified in p53 in 75% of cases



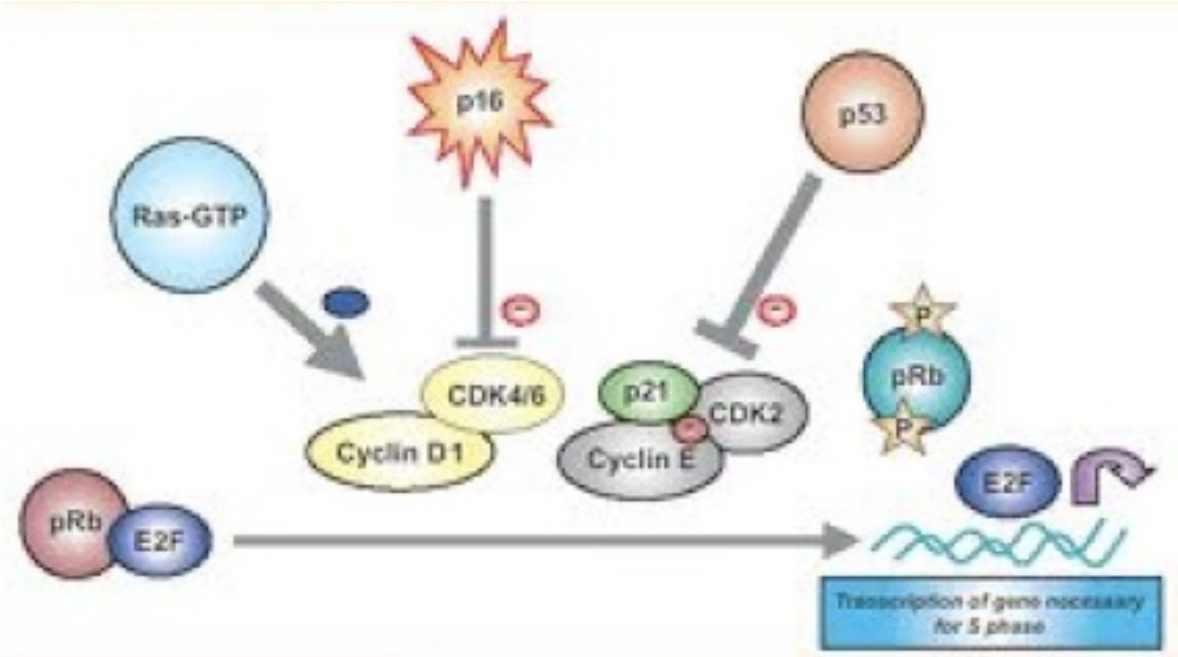
Li-Fraumeni syndrome is very rare

But p53 mutations are observed in a large fraction of sporadic tumors

- Lung 56%
- Colon 50%
- Esophagus 44%
- Pancreas 44%
- Gastric 41%
- Sarcoma 31%
- Prostate 30%
- Breast 30%
- Thyroid 13%
- Melanoma 9%

p53 Tumor Suppressor Gene: Guardian of the Genome

- Germline mutations found in the Li-Fraumini cancer syndrome
- Sporadic mutations found in over 50% of human



Hereditary non-polyposis colorectal cancer (HNPCC)

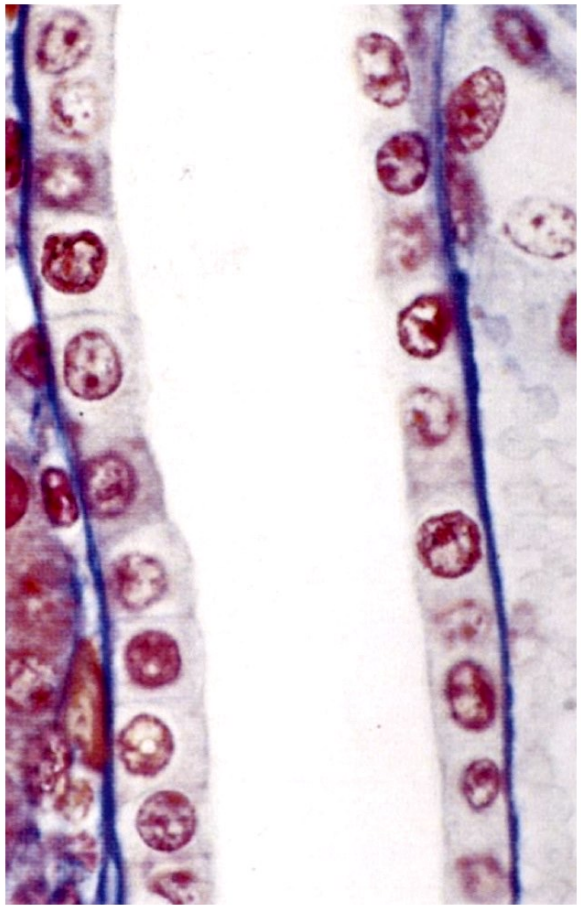
- Colorectal carcinoma in at least 3 relatives in 2 or more generations.
- Age of onset for at least one patient less than 50
- Most cases: mutations in DNA repair genes MSH1 and MLH1.
  - Homologues of DNA repair genes first identified in E. coli (MutS and MutL, respectively)

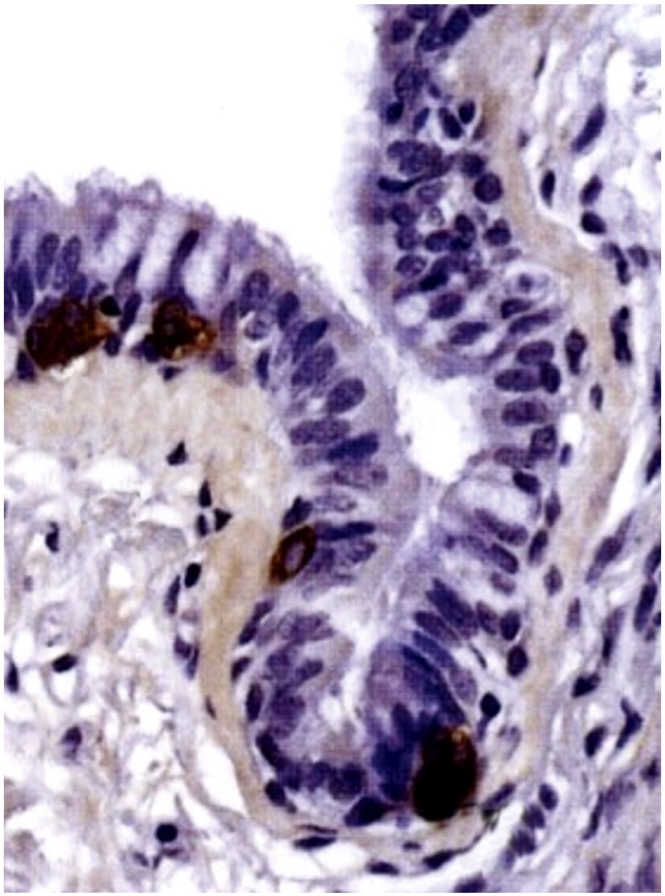
Degree of aggressive growth

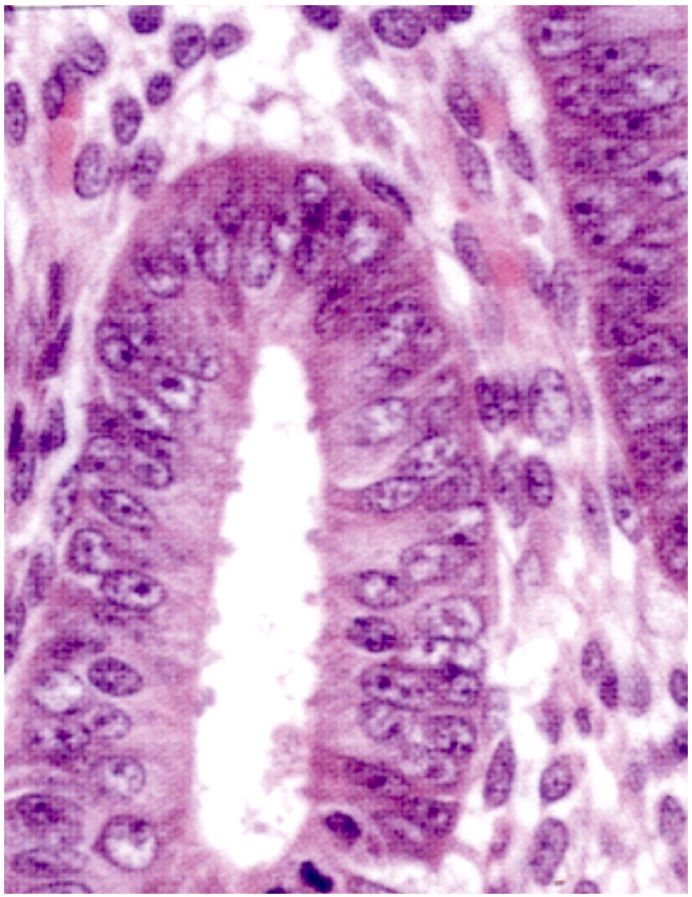
*Benign

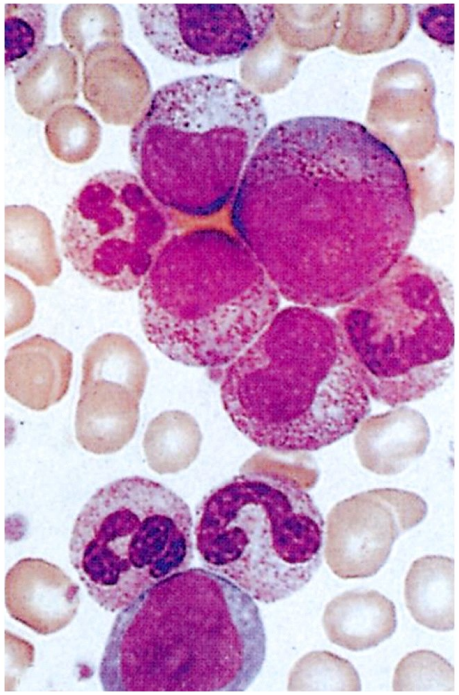
*malignant

Architecture of epithelial tissues



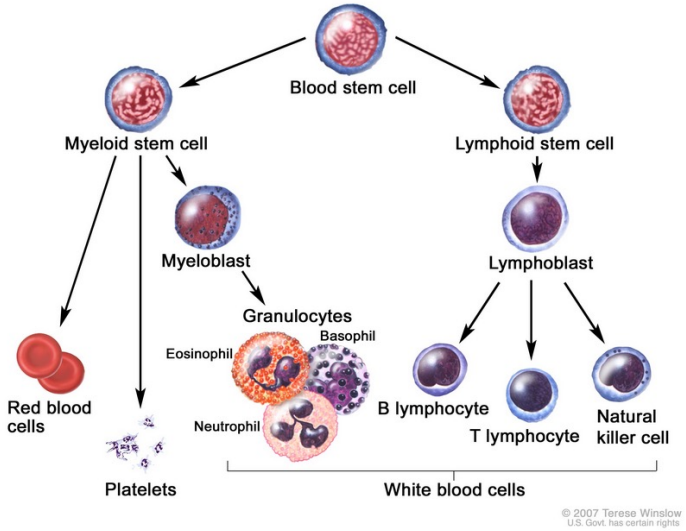


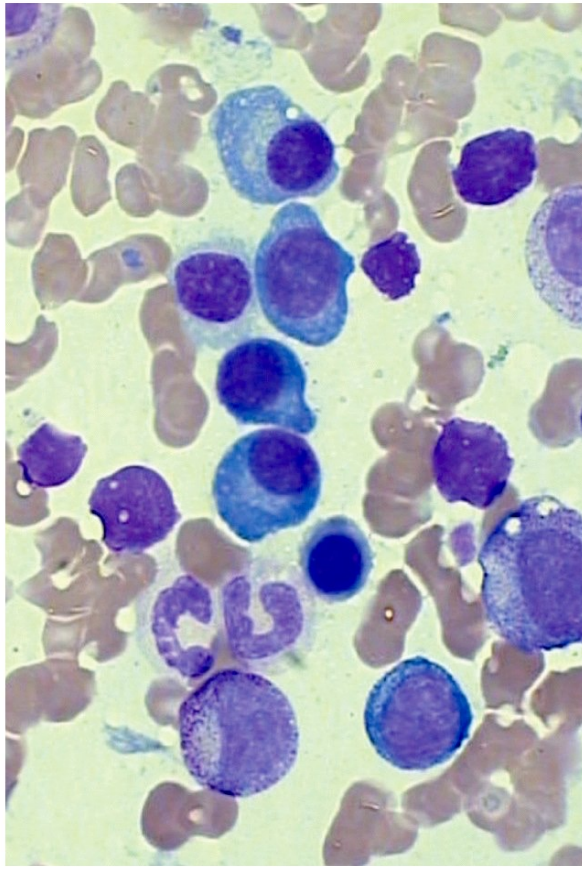




Hematopoietic malignancies

acute lymphocytic leukemia
acute myelogenous leukemia
chronic myelogenous leukemia
chronic lymphocytic leukemia
multiple myeloma
non-Hodgkin's lymphoma[a]
Hodgkin's disease





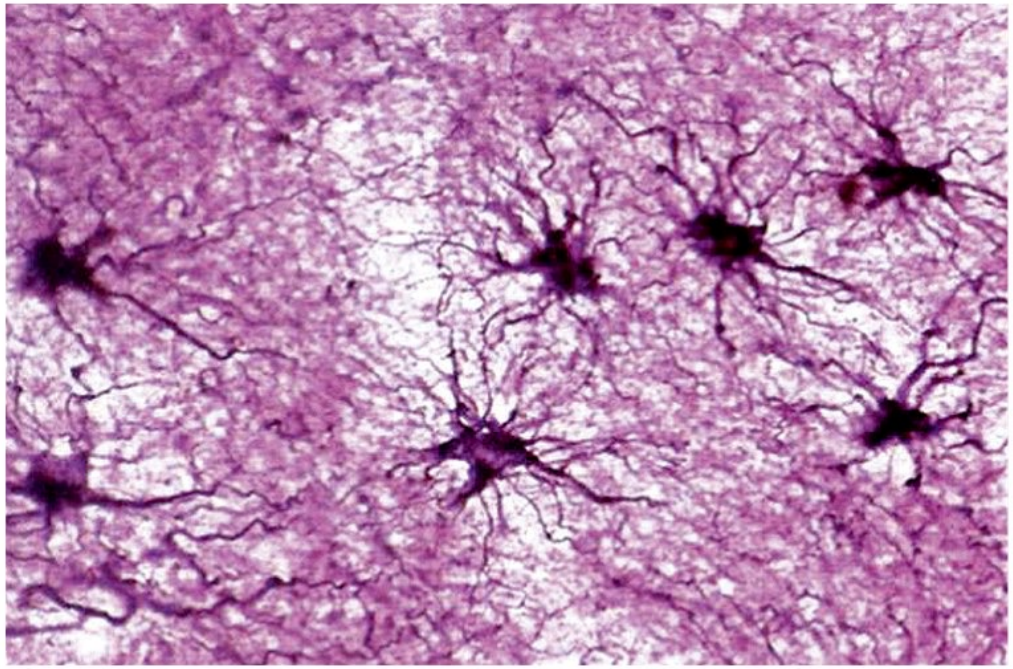
Multiple myeloma is a malignancy of the B-cell lineage which are responsible for producing and secreting antibody molecules,hence, their relative large cytoplasms. Seen here are plasma cells of MM at various stages of differentiation (purple nuclei)

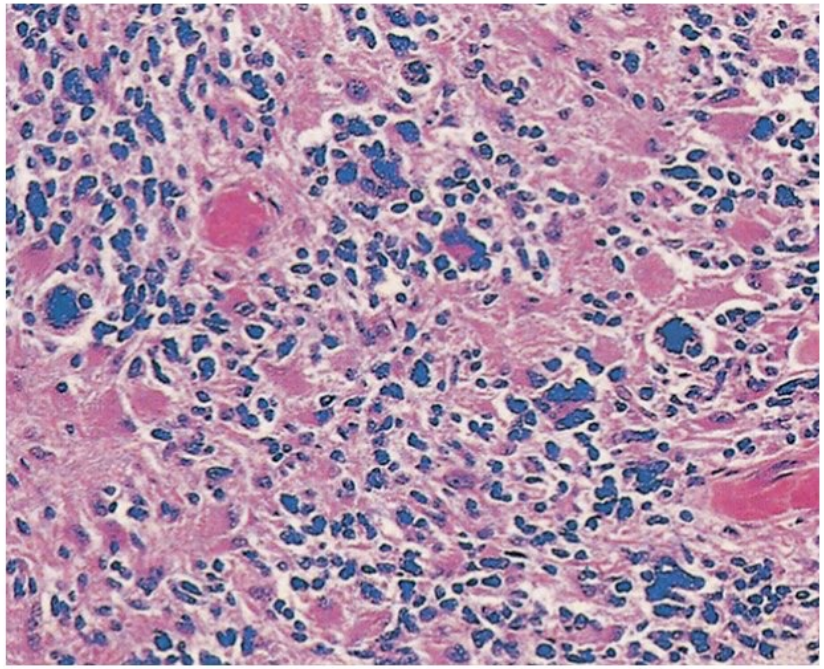
Neuroectodermal malignancies

glioblastoma multiforme
astrocytoma
meningioma
neurinoma
retinoblastoma
neuroblastoma
ependymoma
oligodendroglioma
medulloblastoma

Astrocytes nonneuronal,supporting cells of the brain (dark purple, left panel)-are the presumed precursors of astrocytomas and glioblastomas (right panel).

Glioblastoma multiforme takes its name from the multiple distinct neuroectodermal cell types that constitute the tumor. The tumor cells are seen to have nuclei of various sizes (purple).





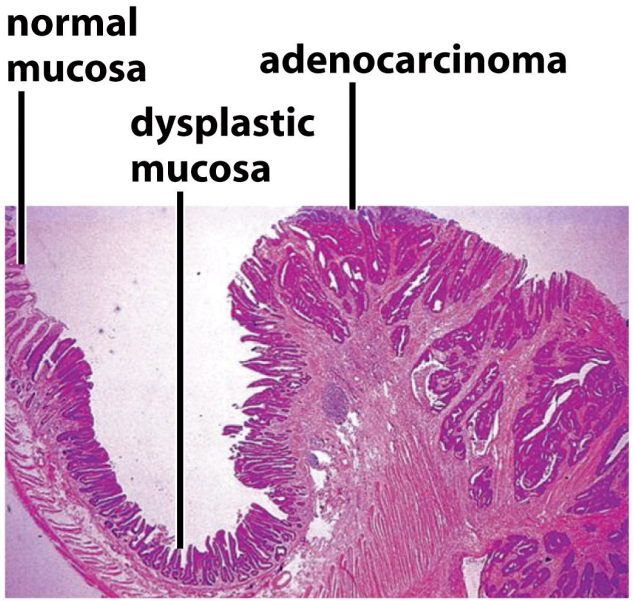
The Nature of Cancer

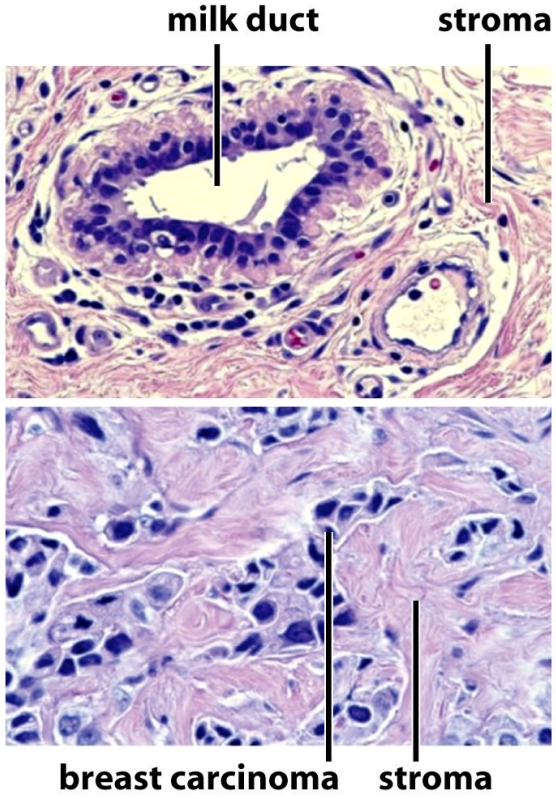
The tumors arise from normal tissues.

Normal versus neoplastic tissue

The great majority of mutations affecting huma
are acquired.

Acquired vs inherited





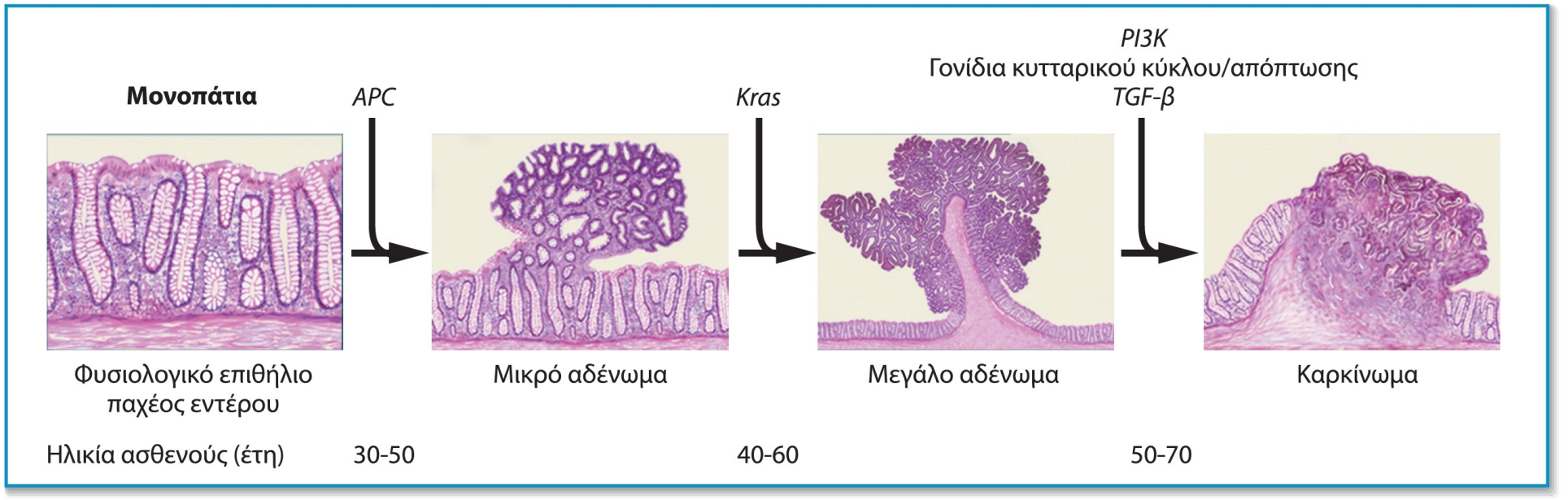
Metastasis of cancer cells to distant sites

Metastases(white) in the liver often arise in patients with advanced colon carcinomas. The portal vein which drains blood from the colon into the liver provides a route for metastasizing colon cancer cells to migrate directly into the liver

Multi step Tumorigenesis

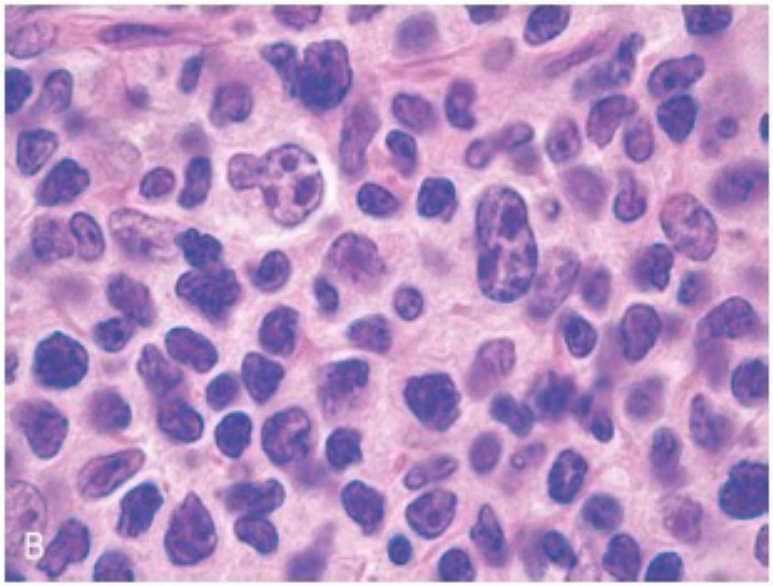
Most human cancers develop
over many decades of time

Τα στάδια ανάπτυξης του καρκίνου του παχέος εντέρου

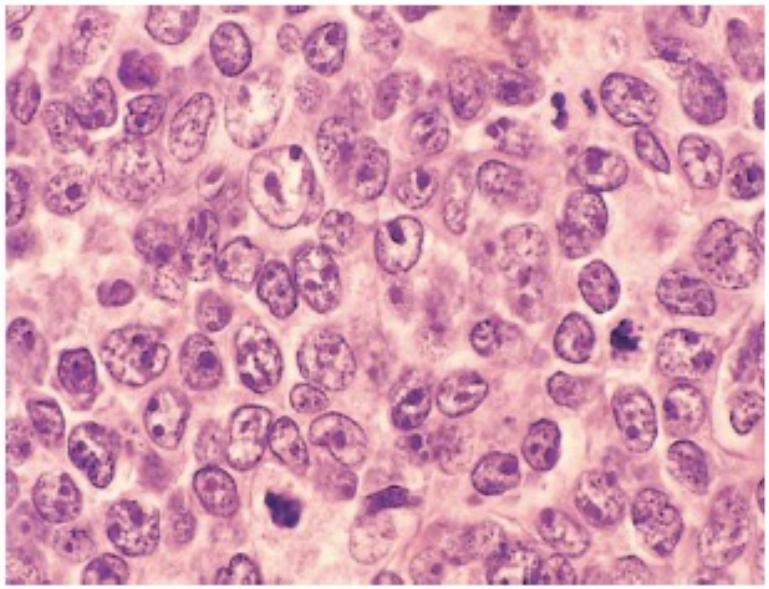


Στο πάνω μέρος της εικόνας αναφέρονται ορισμένα από τα γονίδια στα οποία εντοπίζονται μεταλλαγές-οδηγοί που οδηγούν βαθμιαία στην ανάπτυξη ορθοκολικού καρκίνου. Με το πέρασμα του χρόνου αθροίζονται όλο και περισσότερες μεταλλαγές-οδηγοί. Η πορεία προς τον σχηματισμό ενός κακοήθους όγκου μπορεί να διαρκέσει 40 ή περισσότερα χρόνια.

As a rule, *all cancers become more aggressive over time*



+ mutations

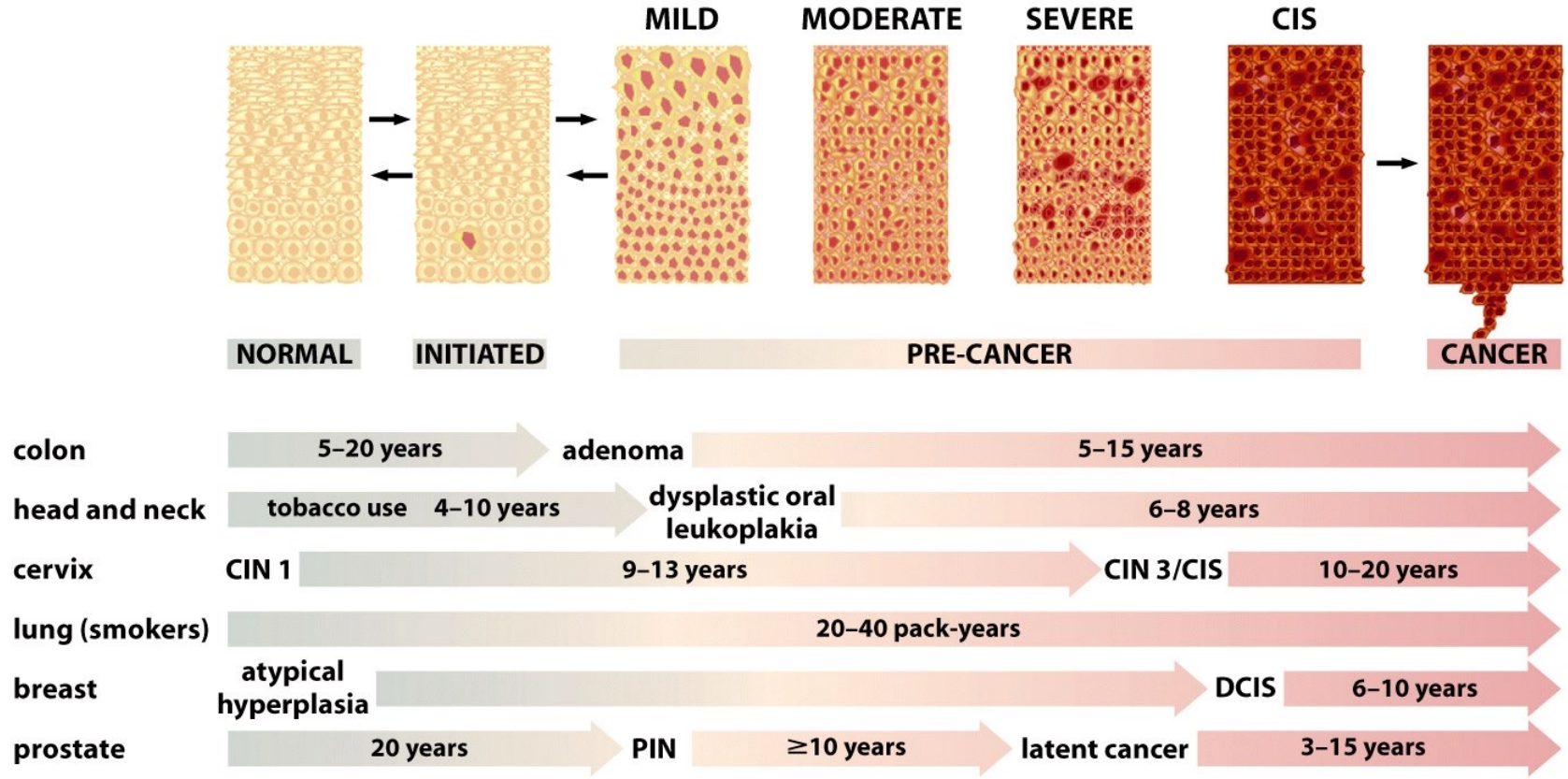


Follicular lymphoma (B cell)
-median survival: 7-9 year

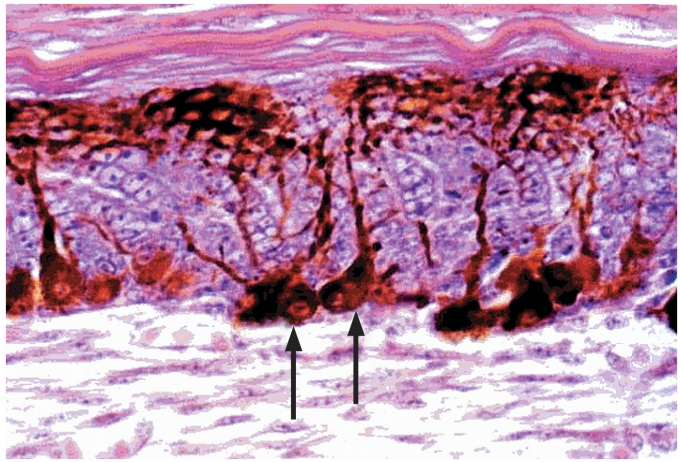
Diffuse large B cell lymphoma
-median survival: ~1 year

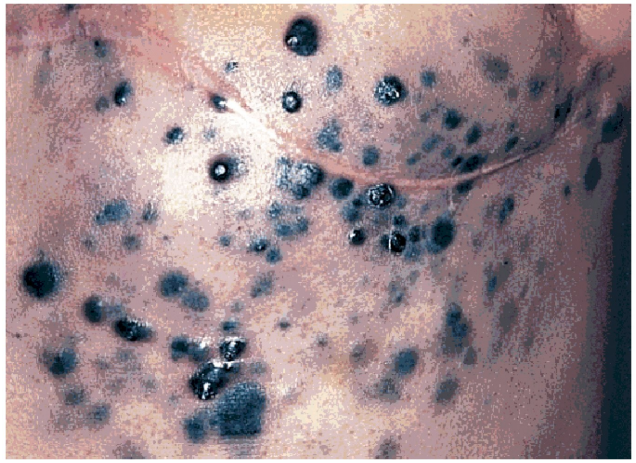
Tumor progression is denoted by spread to more sites, more rapid growth, resistance to therapy

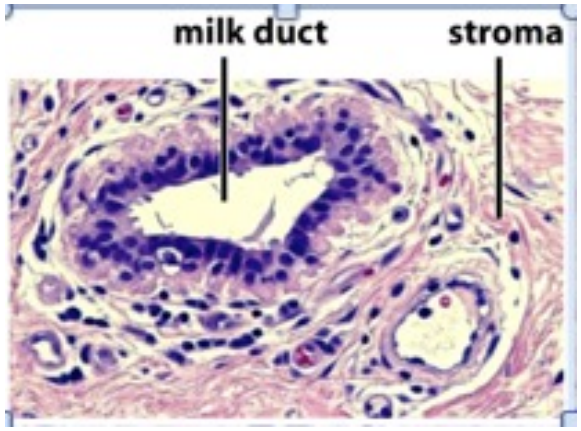
Multi-step tumorigenesis in a variety of organ sites

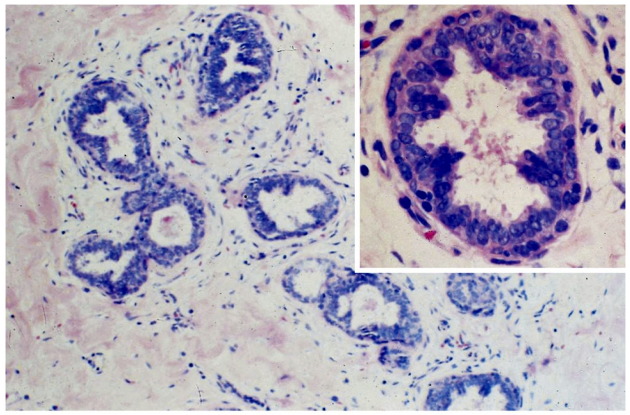


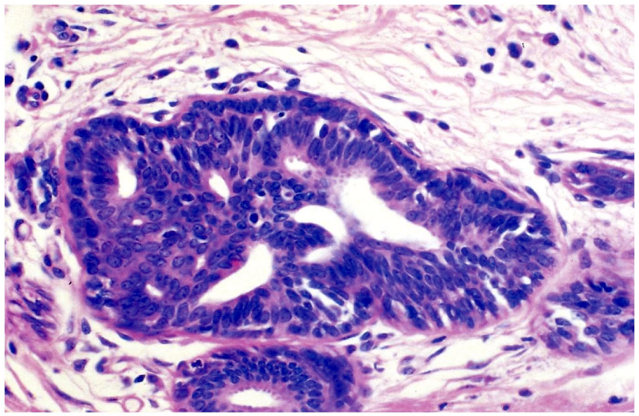
Melanocytes and melanomas







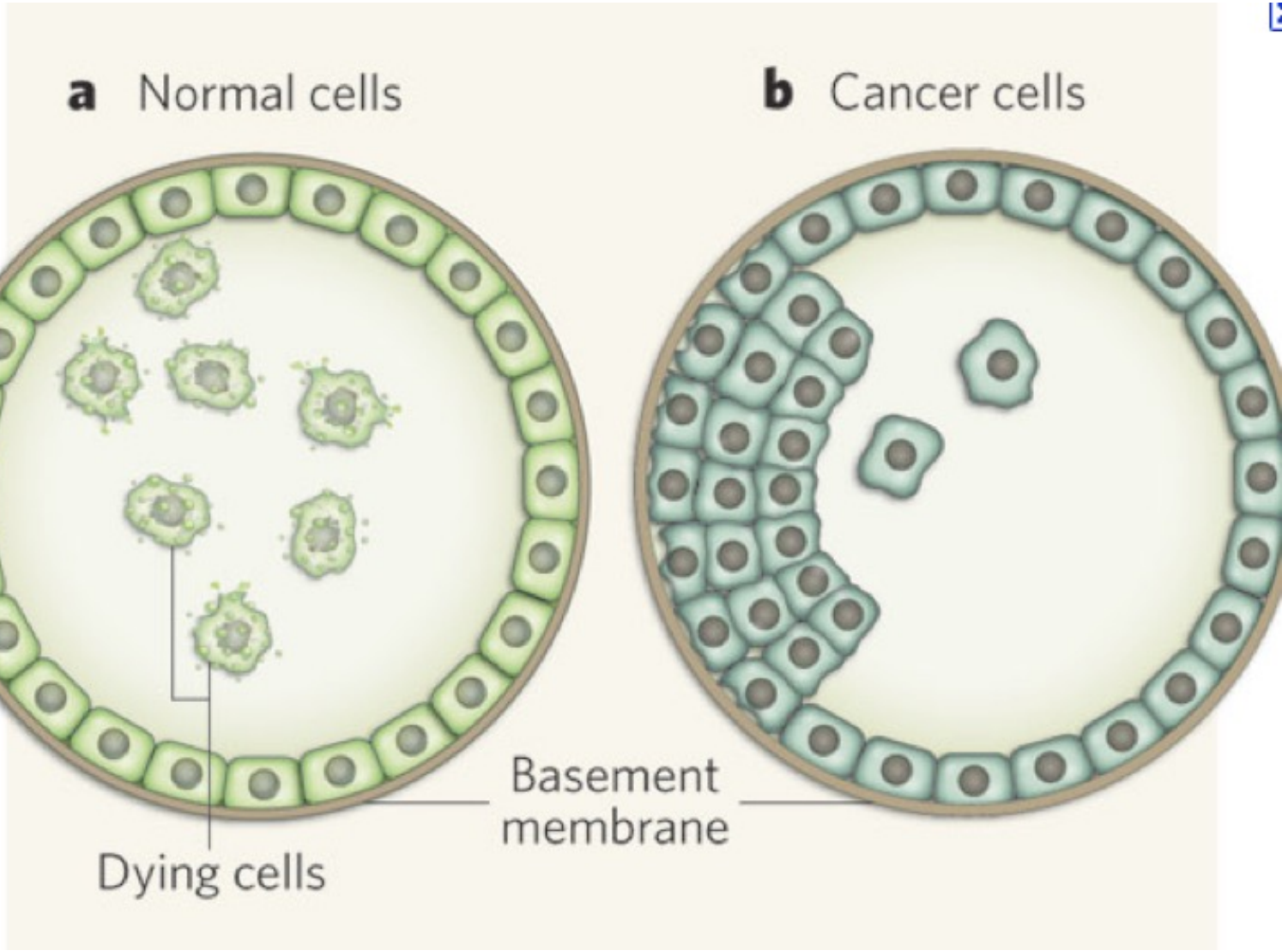


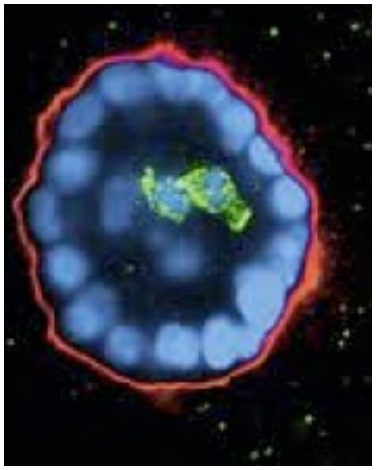


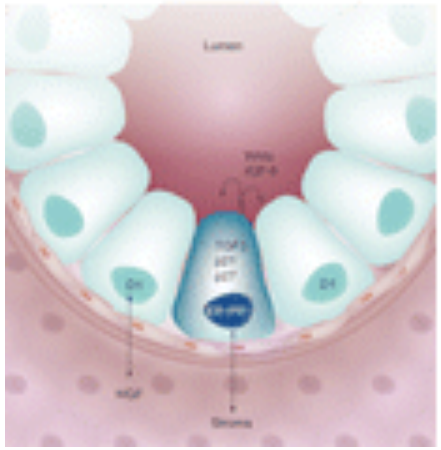
Normal versus hyperplastic epithelim

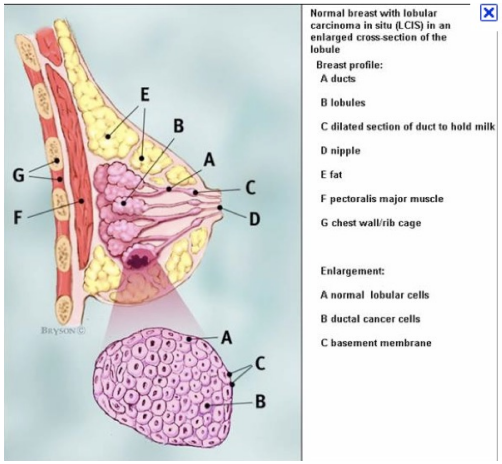
The morphology of the normal ductal epithelum of the mammary gland can be compared with different degrees of hyperplasia

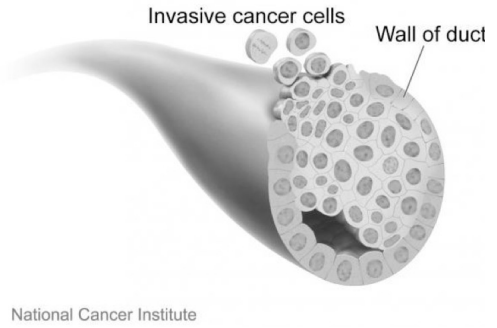
Tissue *architecture*: the ultimate regulator of *breast epithelial* function



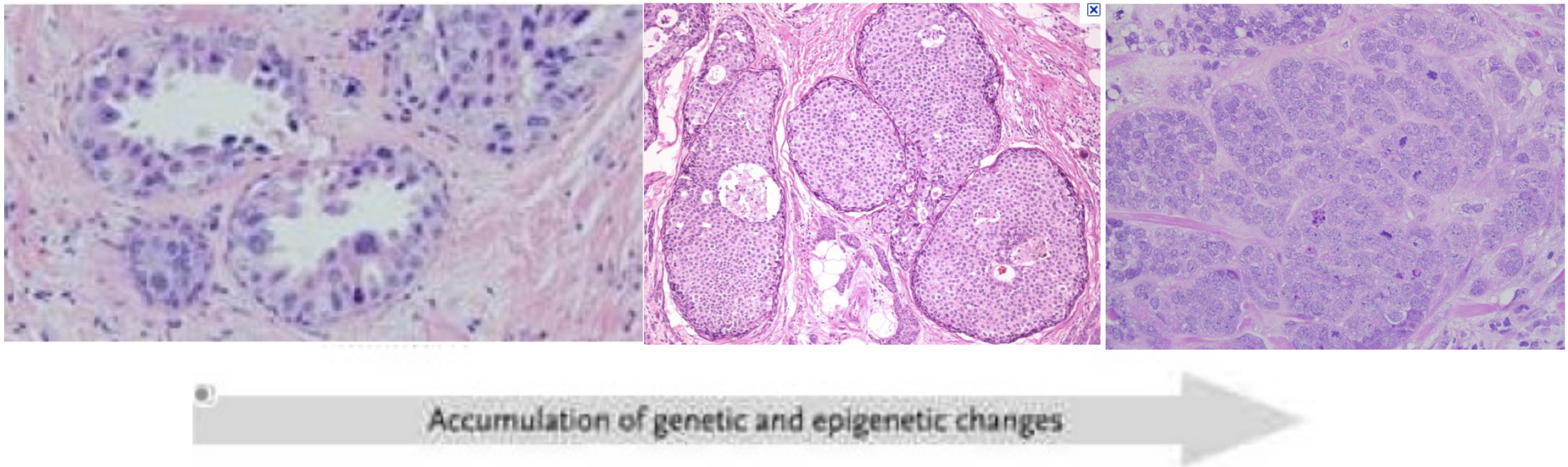








Breast cancer progression

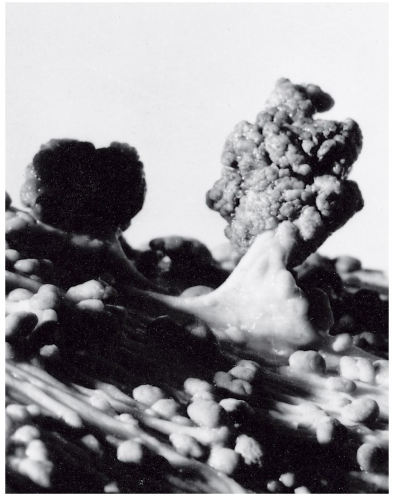


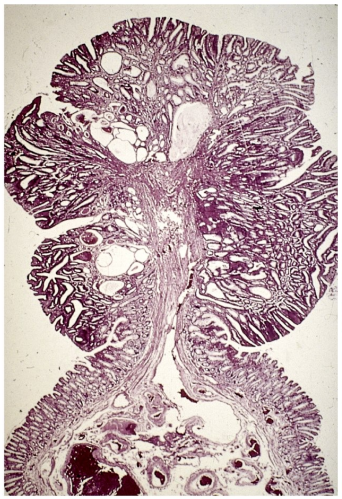
Hyperplasia : too many cells (=look like normal)

Benign tumors-non- aggressive, non-destructive, no potential to spread

Malignant tumor(cancer) aggressive , destructive, potential to spread

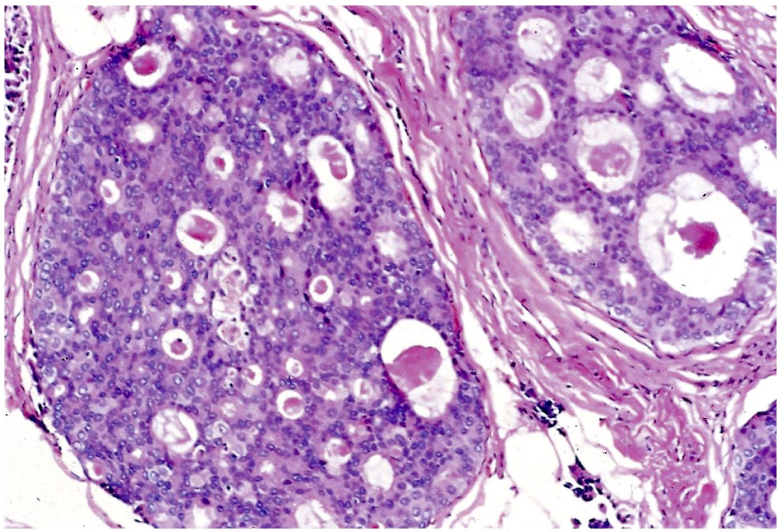
Pro-invasive adenomas and carcinomas

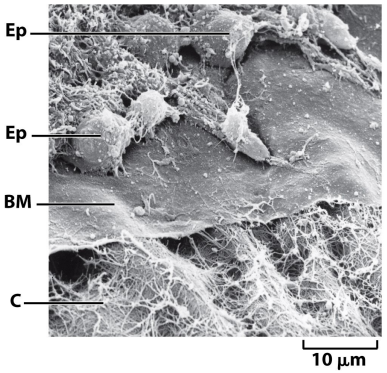




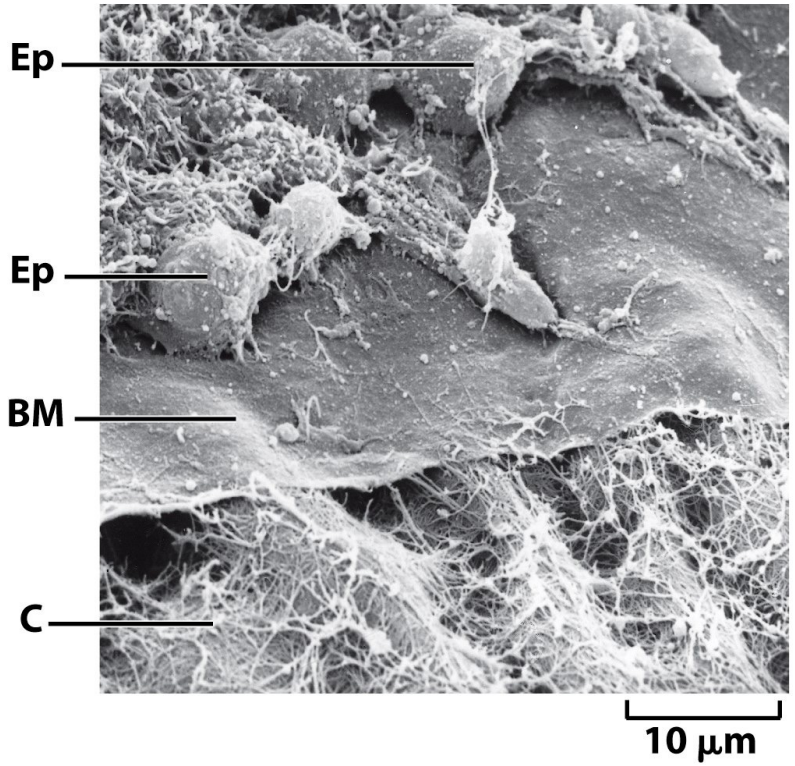
Polyps are here in a photograph (left)

and a micrograph section (right)



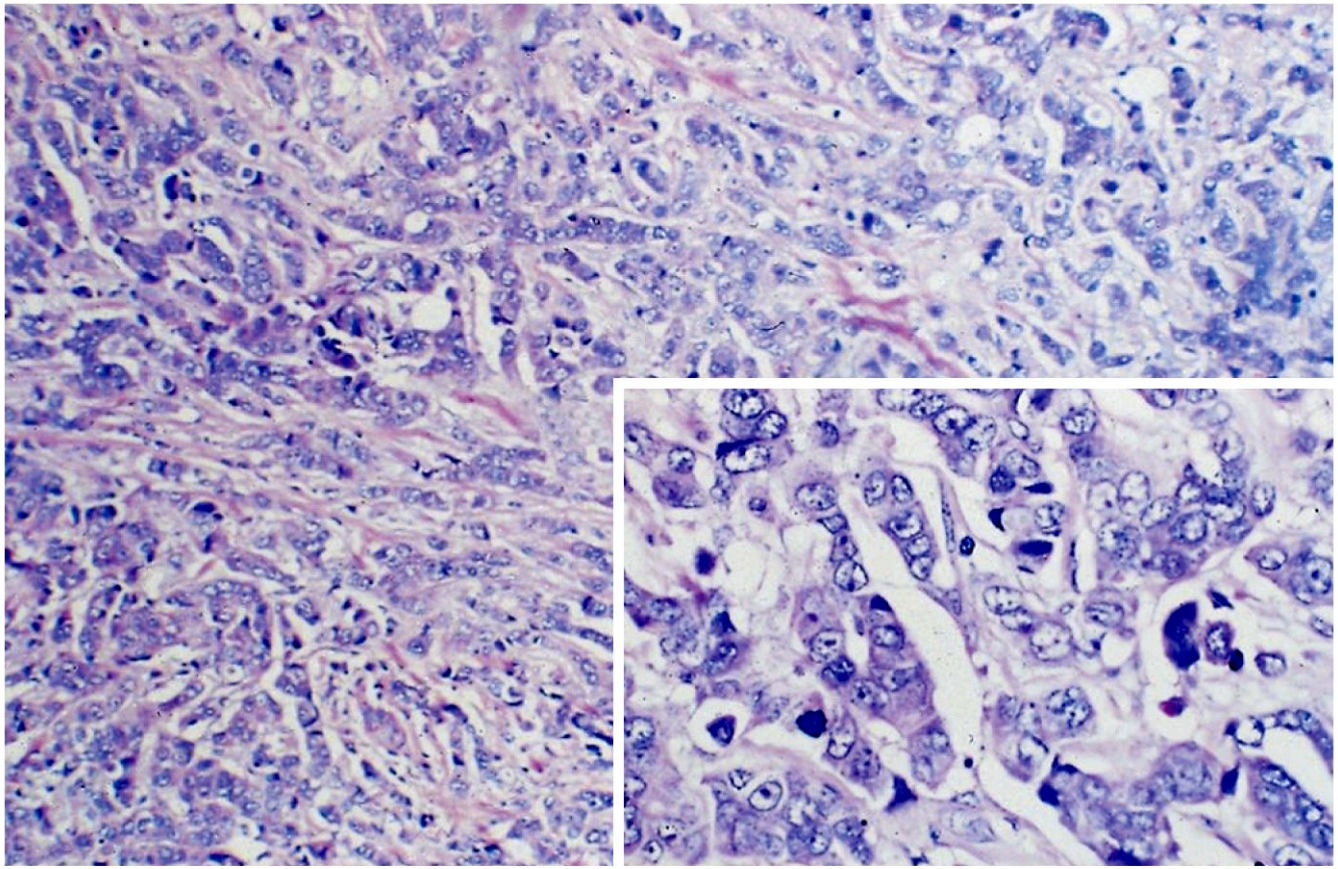


TUMORS ARISE FROM MANY SPECIALIZED CELL TYPES THROUGHOUT THE BODY

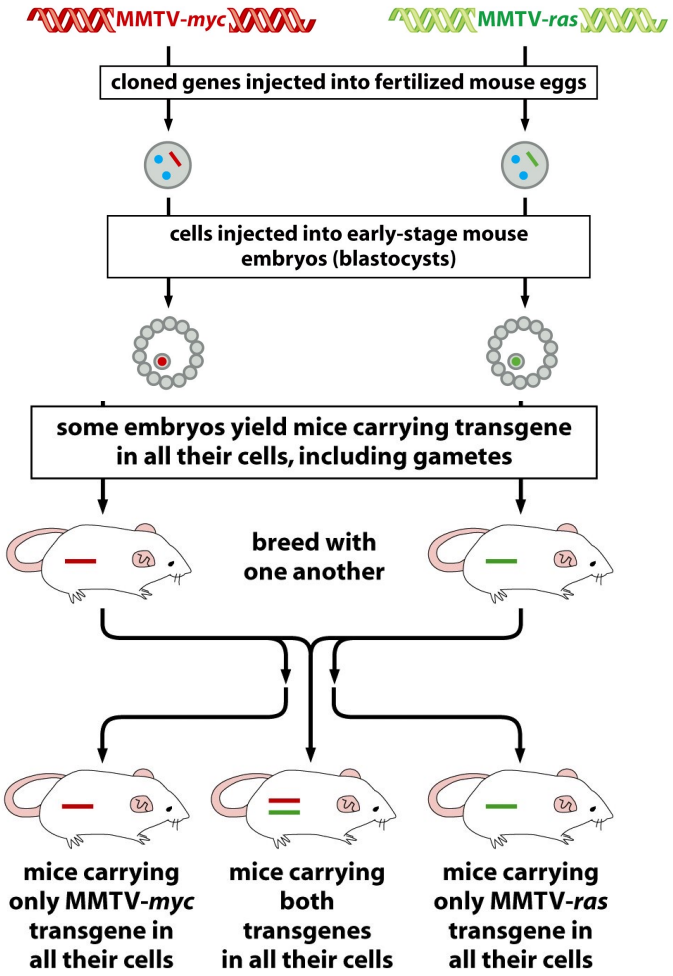


Invasive carcinomas

Invasive ductal carcinomas of the breast, islands of of epithelial cancer cells are intermingled with stromal cells.



Transformation usually requires collaboration between two or more mutant genes



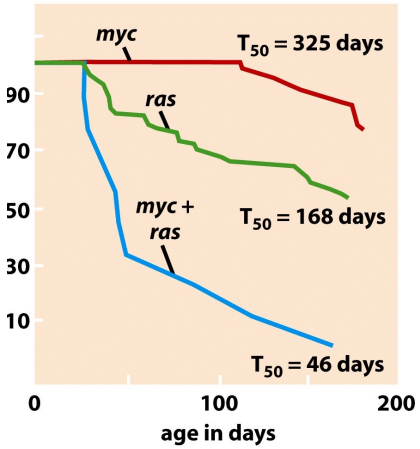


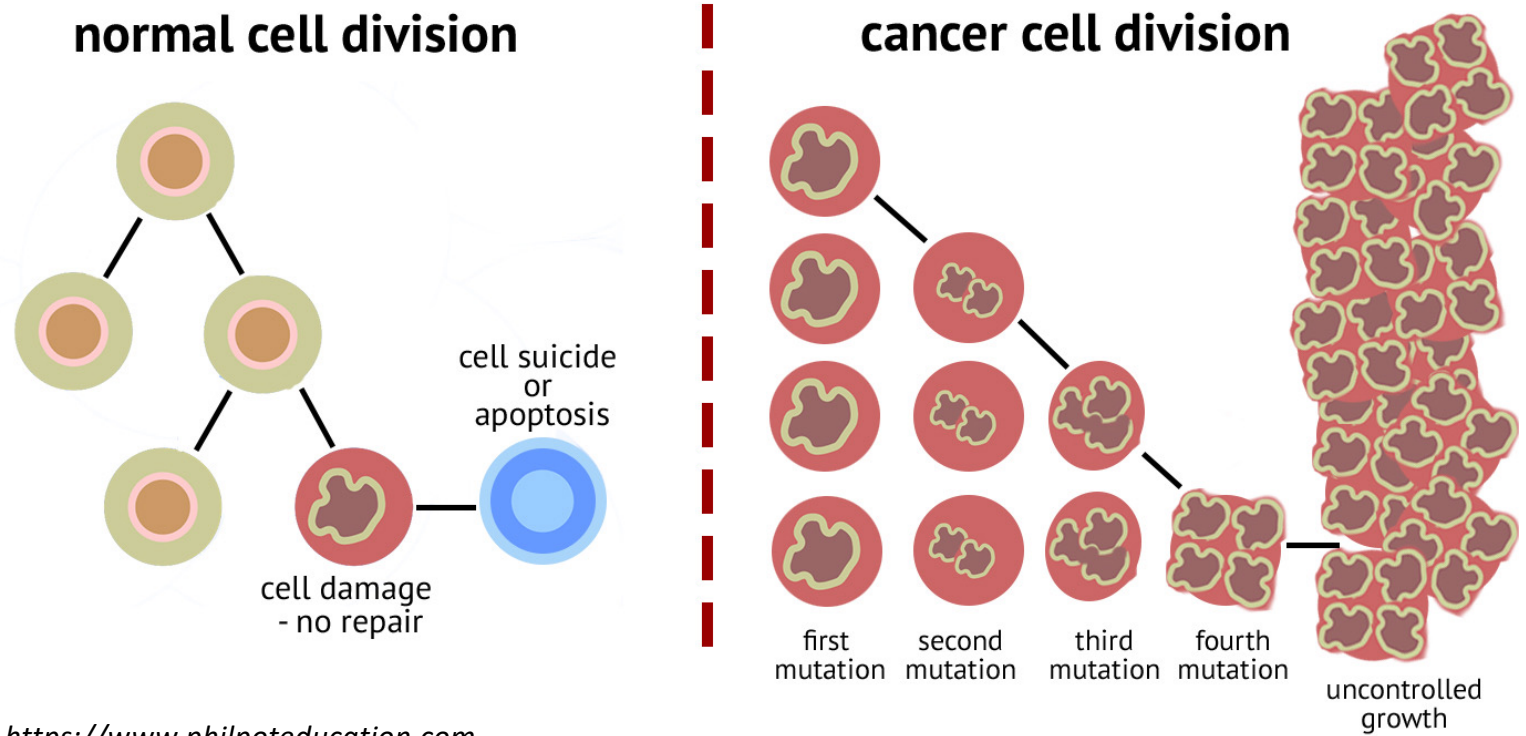
Table 11.2 Physiologic mechanisms of oncogene collaboration[a]

Oncogene pair	Cell type	Mechanisms of action
ras + SV40 *large T*	rat Schwann cells	*ras*: proliferation + proliferation arrest *large T*: prevents proliferation arrest and reduces mitogen requirement
ras + *E1A*	mouse embryo fibroblasts	*ras*: proliferation and senescence *E1A*: prevents senescence
erbB + *erbA*	chicken erythroblasts	*erbB*: induces GF-independent proliferation *erbA*: blocks differentiation
TGF-α + *myc*	mouse mammary epithelial cells	*TGF-α*: induces proliferation and blocks apoptosis *myc*: induces proliferation and apoptosis
v-*sea* + v-*ski*	avian erythroblasts	v-*sea*: induces proliferation v-*ski*: blocks differentiation
bcl-2 + *myc*	rat fibroblasts	*bcl-2*: blocks apoptosis *myc*: induces proliferation and apoptosis
ras + *myc*	rat fibroblasts	*ras*: induces anchorage independence *myc*: induces immortalization
raf + *myc*	chicken macrophages	*raf*: induces growth factor secretion *myc*: stimulates proliferation
src + *myc*	rat adrenocortical cells	*src*: induces anchorage and serum independence *myc*: prolongs proliferation

[a]In each pair, the first oncogene encodes a cytoplasmic oncoprotein while the second oncogene encodes a nuclear oncoprotein.

The origin of tumor cells

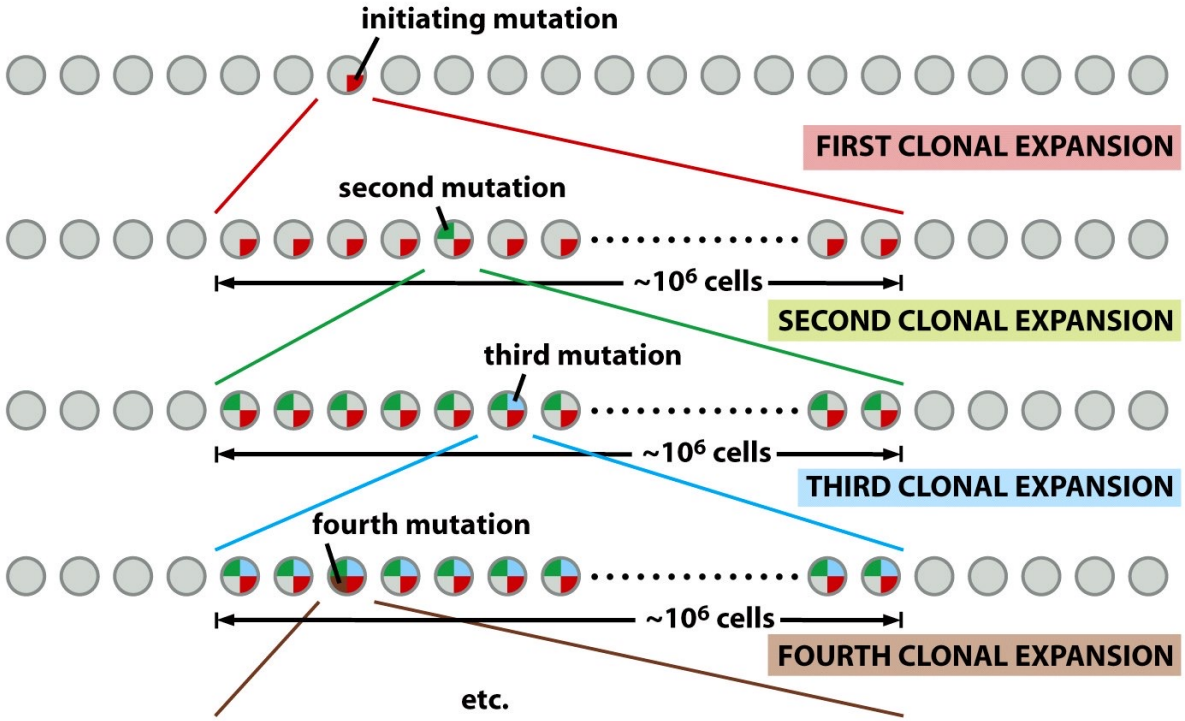
Ο καρκίνος ως αποτέλεσμα κλωνικής εξέλιξης (clonal evolution)



https://www.philpoteducation.com

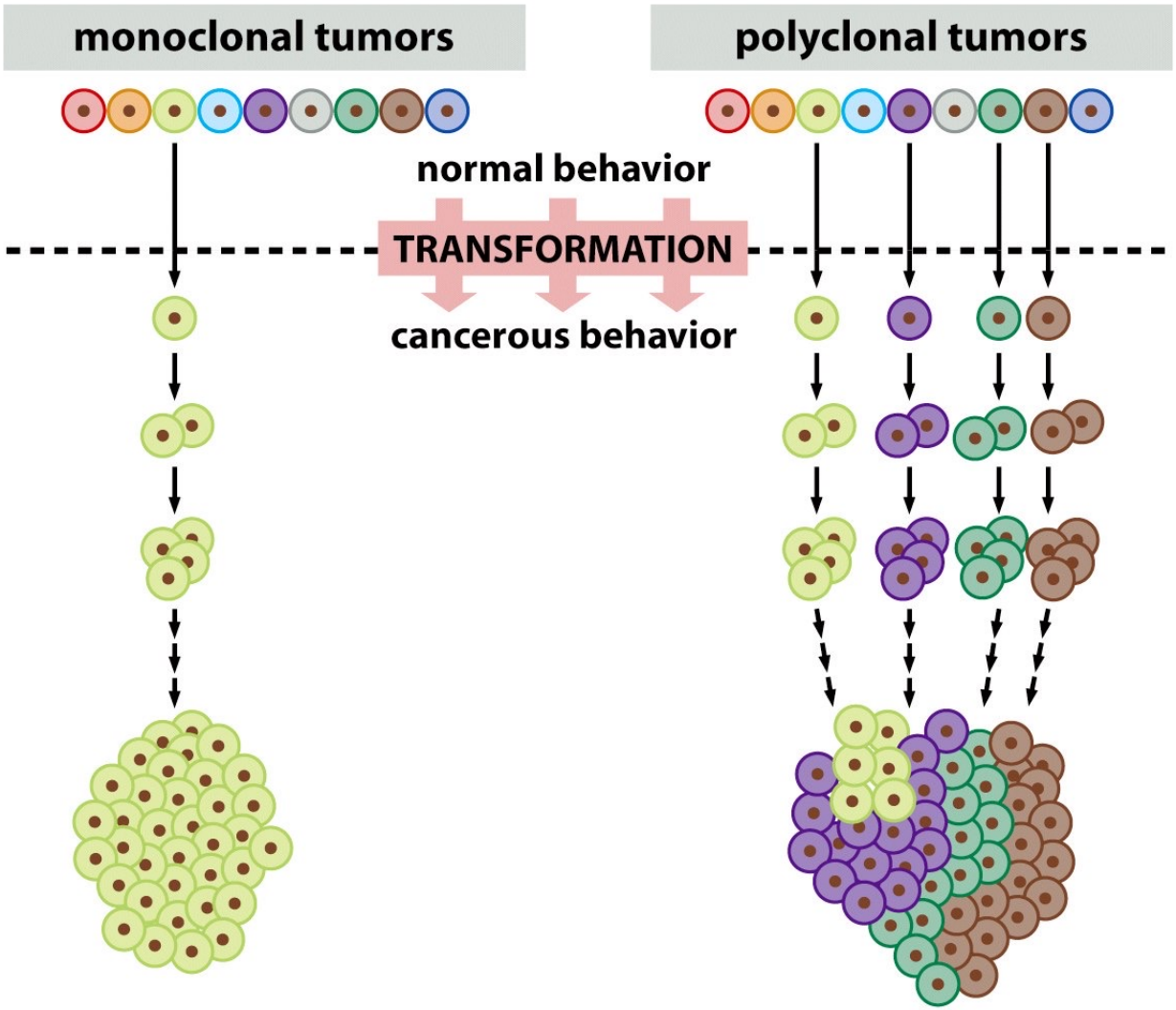
Μια **αρχική μετάλλαξη** περνάει από ένα κύτταρο στους απογόνους του και σε όλες τις επόμενες γενιές.
Τελικά, **ικανός αριθμός συσσωρευμένων μεταλλάξεων** σε ένα κύτταρο-απόγονο οδηγεί στην **καρκινογένεση**

Cancer development seems to follow the rules of Darwinian evolution

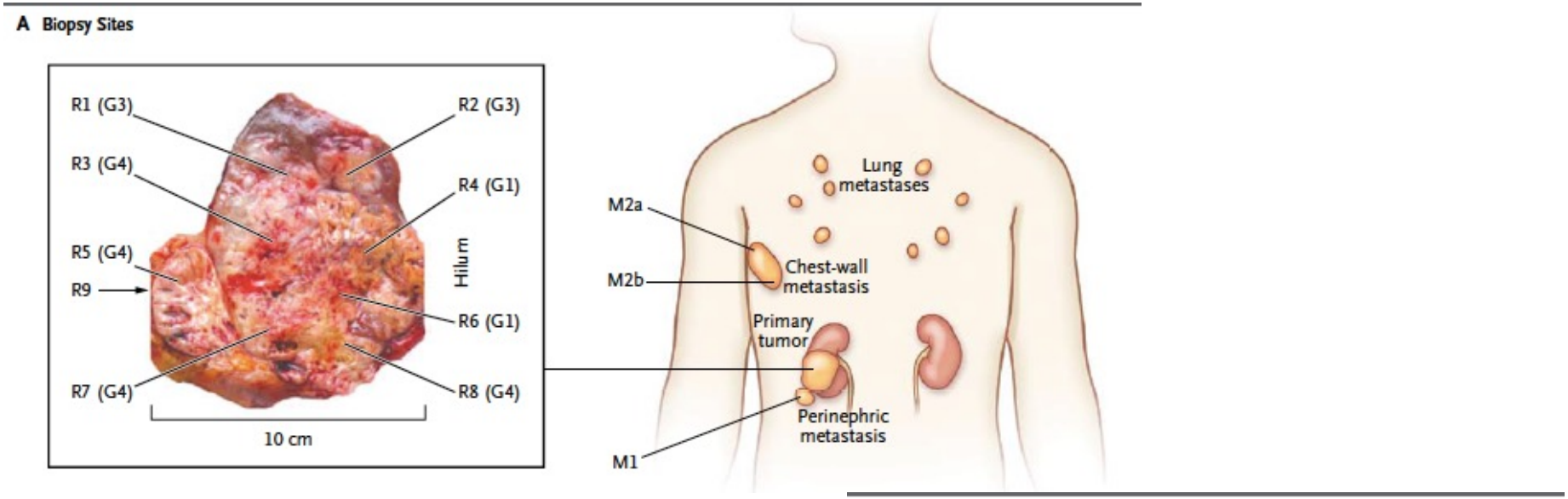


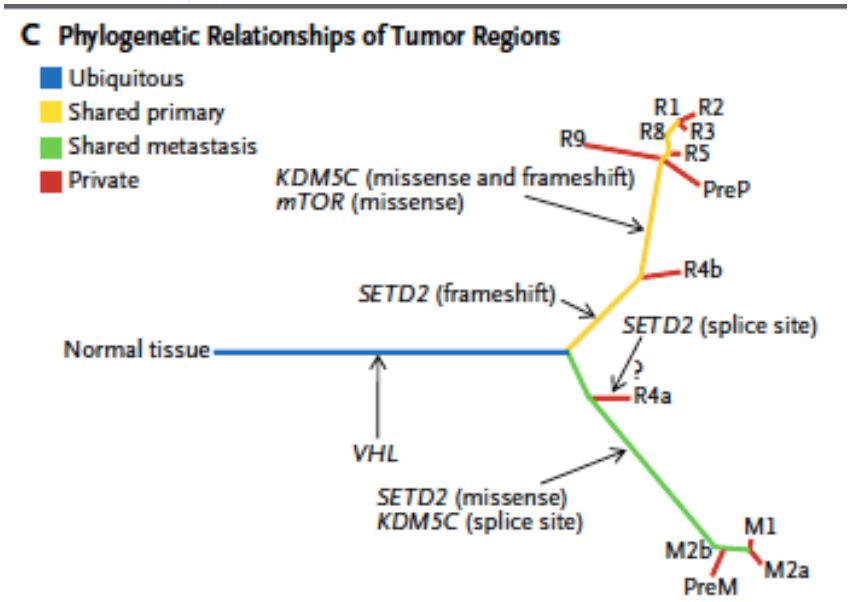
Darwinian evolution and clonal successions

Monoclonality versus polyclonality of tumors

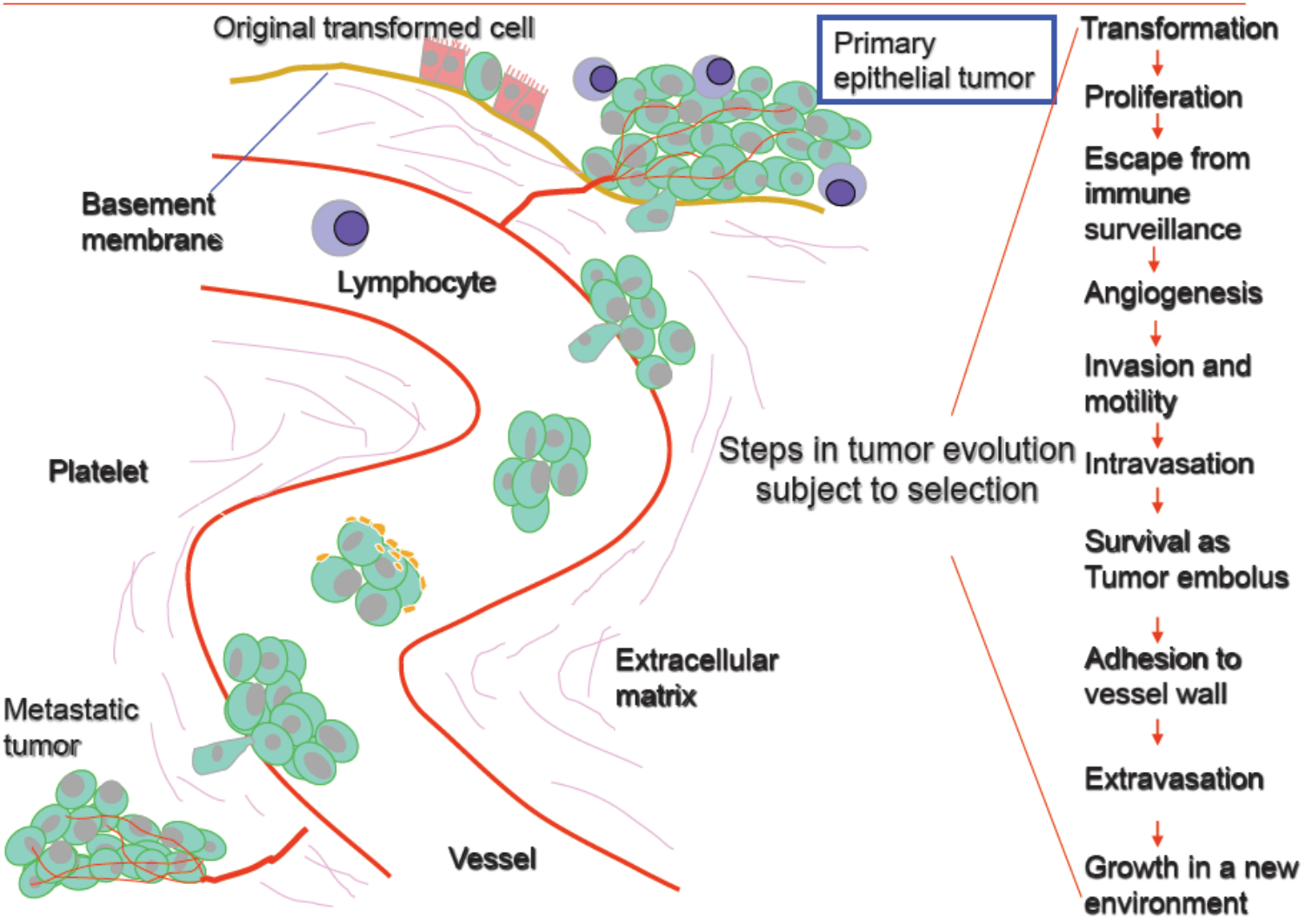


Cancer Genomics informs on clonal evolution

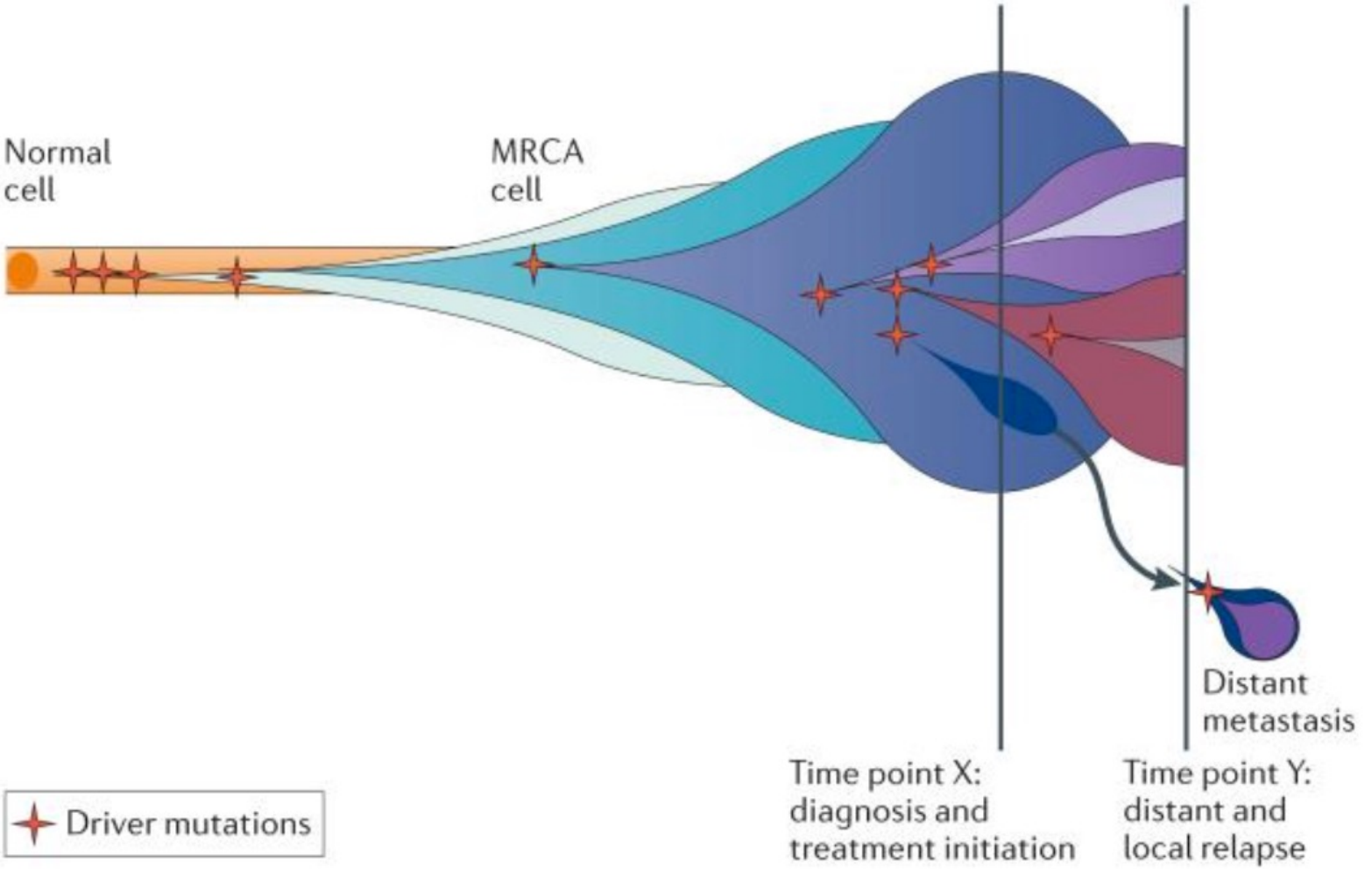




Tumor Progression and Natural Selection



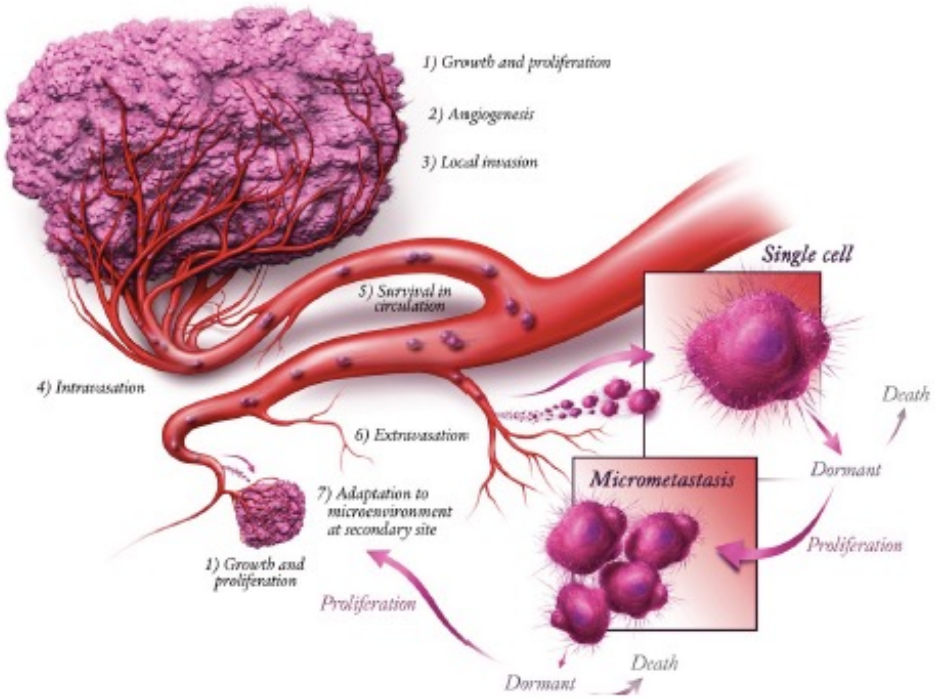
Cancer is an evolutionary process



Yates and Campbell et al, Nat Rev Genet 2012

Single cell expression profiling challenges

Cancer Metastasis and Circulating Tumor Cells

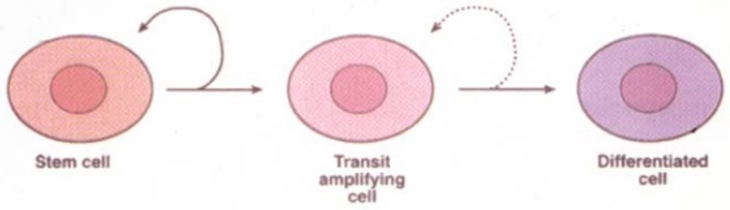


- Metastasis is a result of rare cells migrating from the primary tumor through the lymphatic or hematogenous route
- Metastasis depends on cross talk between selected cancer cells (the seed) and specific organ microenvironment (the soil)
- Correlates but does not depend on the presence of CTC (CTC heterogeneity)

Cancer Stem Cells (CSCs)

Stem Cells
&
Cancer Stem Cells

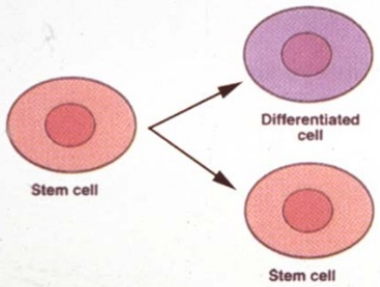
Stem Cells



Stem Cells: Cells capable (1) of self renewal and (2) of giving rise to at least one differentiated cell type

Stem cells are generally a slow dividing cell type. Often they directly give rise to an intermediate rapidly dividing cell type

Self renewal of Stem Cells



a) Invarient asymmetric cell division yeilds one stem daughter and one which undergoes differentiation

b) Stem cell gives rise to daughters that have finite probabilities of being either stem cells or committed progenitors

Roles of Stem Cells

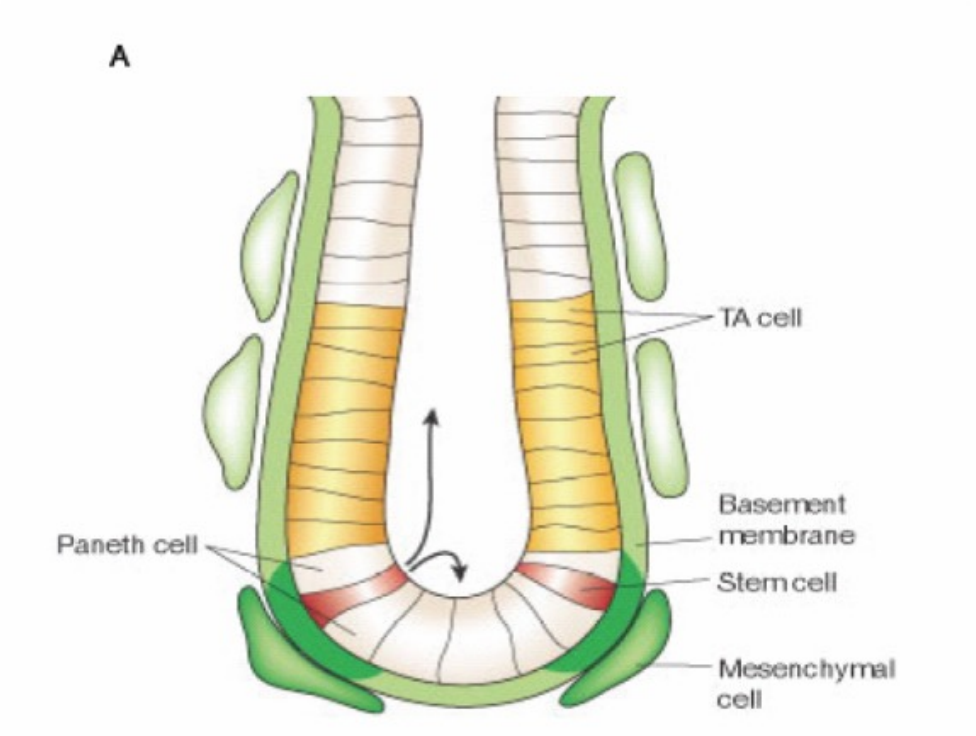
Embryogeneis: generation of fetal tissues

Renewing adult tissues: heamatopoietic, epidermis, hair, small intestine

Regeneration: liver, skin

Adult stem cells exist in specific "niches" (specific cellular microenvironment in specific location in a tissue)

Intestinal stem cells



From Leedham et al., 2004

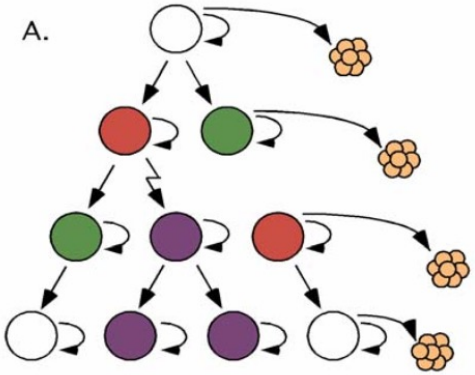
Tissue-specific stem cells

Tissue	Stem cell	Differentiated progeny
Blood	HSC	All lineages of blood cells
Brain	NSC	Neurons, glia
Intestine	ISC	Intestinal epithelium
Skin	Bulge cell	Hair, sebaceous gland, epidermis
Muscle	Satellite cell	Myoblasts, myofibers
Germline	Germ cell	Oocyte, sperm
Liver	Oval cell	Hepatocyte, bile duct
Heart	Cardiac progenitor	Cardiomyocytes, smooth muscle,
Blood vessels	EPC	Endothelium
Lung	BASC	Alveoli, pneumocytes
Kidney	?	Renal tubule
Pancreas	?	Exocrine/endocrine cells
Fat	?	adipocytes

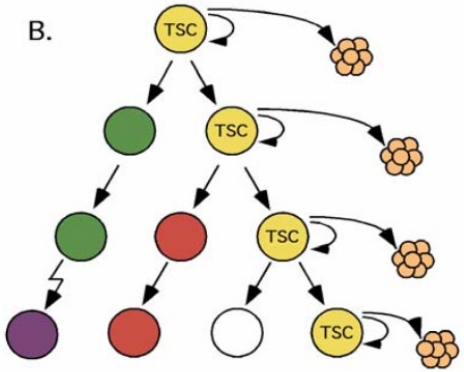
Cancer Stem Cells (CSCs)

Cancer stem cells:

Cancers may be viewed as a type of disregulated organogenesis, in which self-renewing cancer stem cells (CSC) give rise to abnormal differentiated progeny.



Tumor cells are heterogeneous, but most cells can proliferate extensively and form new tumors



Tumor cells are heterogeneous and only the tumor stem cell subset (TSC; yellow) has the ability to proliferate extensively and form new tumors.

What are cancer stem cells (CSCs)?

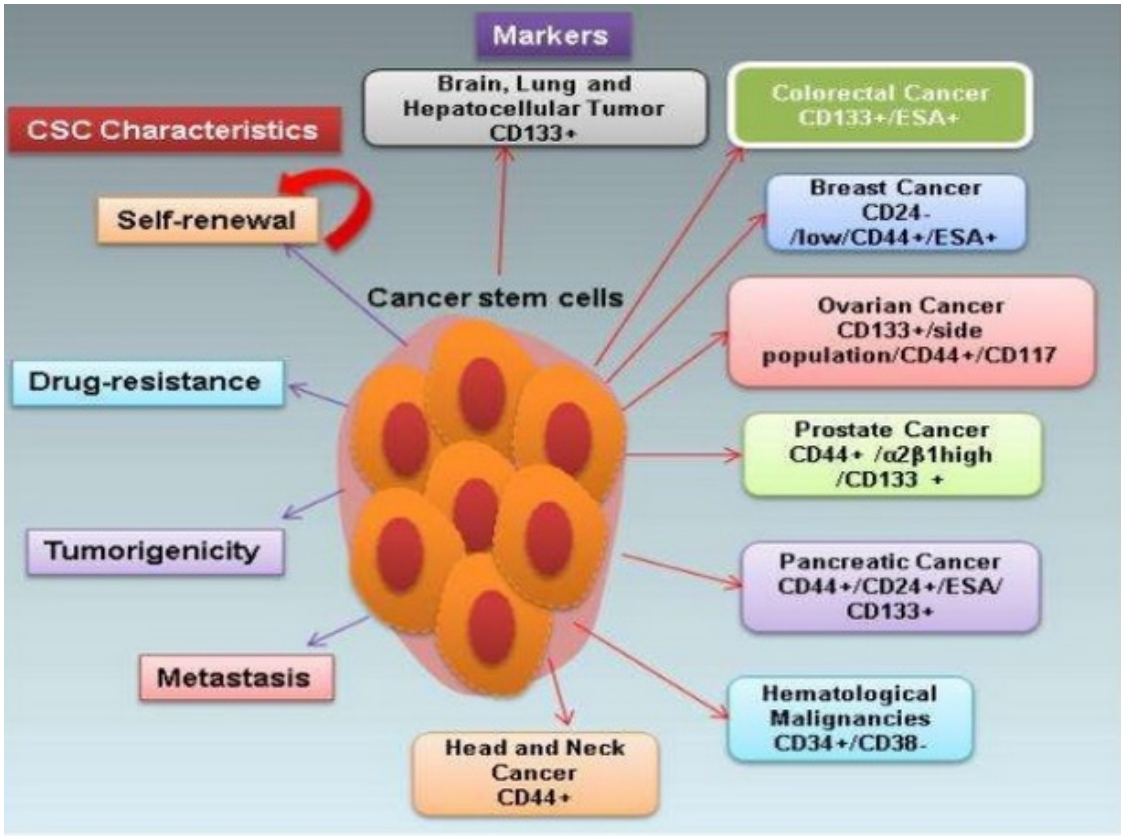
- immortal tumor-initiating cells that can self-renew and have pluripotent capacity
- can generate tumor cells with different phenotypes, which results in the growth of the primary tumor and emergence of new tumors.
- Found in multiple malignancies, including leukemia and various solid cancers (breast, lung cancer, colon cancer, prostate cancer, ovarian cancer, brain cancer, and melanoma).

CSC biomarkers

the most common method used to identify CSCs is fluorescence-activated cell sorting (FACS) based on cell surface markers or intracellular molecules.

Cell surface phenotypes of CSCs.

Tumor type	Phenotype of CSCs markers
Leukemia	$CD34^{+}CD38^{-}$HLA-DR-$CD71^{-}CD90^{-}CD117^{-}CD123^{+}$
Breast cancer	$ESA^{+}CD44^{+}CD24^{-/low}Lineage^{-}$, $ALDH\text{-}1^{high}$
Liver cancer	$CD133^{+}$, $CD49f^{+}$, $CD90^{+}$
Brain cancer	$CD133^{+}$, $BCRP1^{+}$, $A2B5^{+}$, $SSEA\text{-}1^{+}$
Lung cancer	$CD133^{+}$, $ABCG2^{high}$
Colon cancer	$CD133^{+}$, $CD44^{+}$, $CD166^{+}$, $EpCAM^{+}$, $CD24^{+}$
Multiple myeloma	$CD138^{-}$
Prostate cancer	$CD44^{+}$, $\alpha2\beta1^{high}$, $CD133^{+}$
Pancreatic	$CD133^{+}$, $CD44^{+}$, $EpCAM^{+}$, $CD24^{+}$
Melanoma	$CD20^{+}$
Head and neck cancer	$CD44^{+}$


Markers
Brain, Lung and Hepatocellular Tumor CD133+
Colorectal Cancer CD133+/ESA+
CSC Characteristics
Self-renewal
Breast Cancer CD24-/low/CD44+/ESA+
Cancer stem cells
Ovarian Cancer CD133+/side population/CD44+/CD117
Drug-resistance
Prostate Cancer CD44+ /α2β1high /CD133 +
Tumorigenicity
Pancreatic Cancer CD44+/CD24+/ESA/ CD133+
Metastasis
Hematological Malignancies CD34+/CD38-
Head and Neck Cancer CD44+

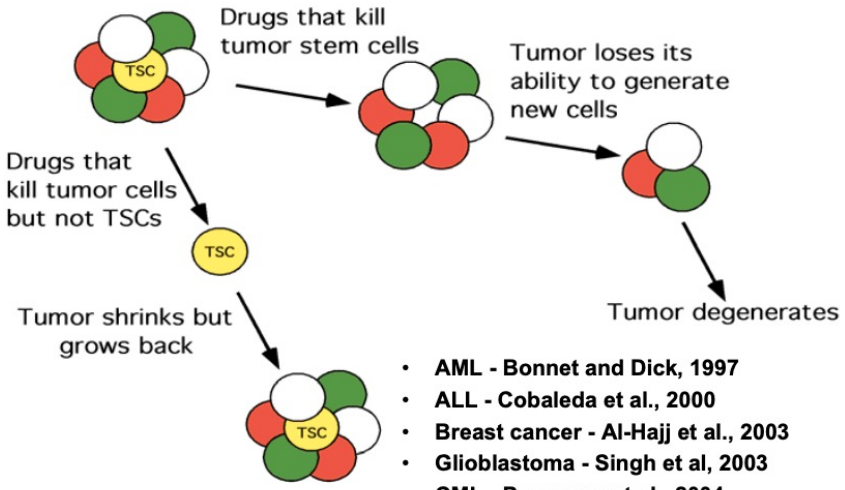
“Cancer Stem Cells”

Old view: All cells of a malignant tumor are dividing endlessly. New mutations are selected that give some cells a proliferative advantage in this enlarging target pool

Emerging view: In at least some cases….Most dividing cells in a tumor are not immortalized. They are equivalent to the rapidly dividing transient amplifying cells in normal tissue. A much smaller population of slowly dividing cells within the tumor act as stem cells

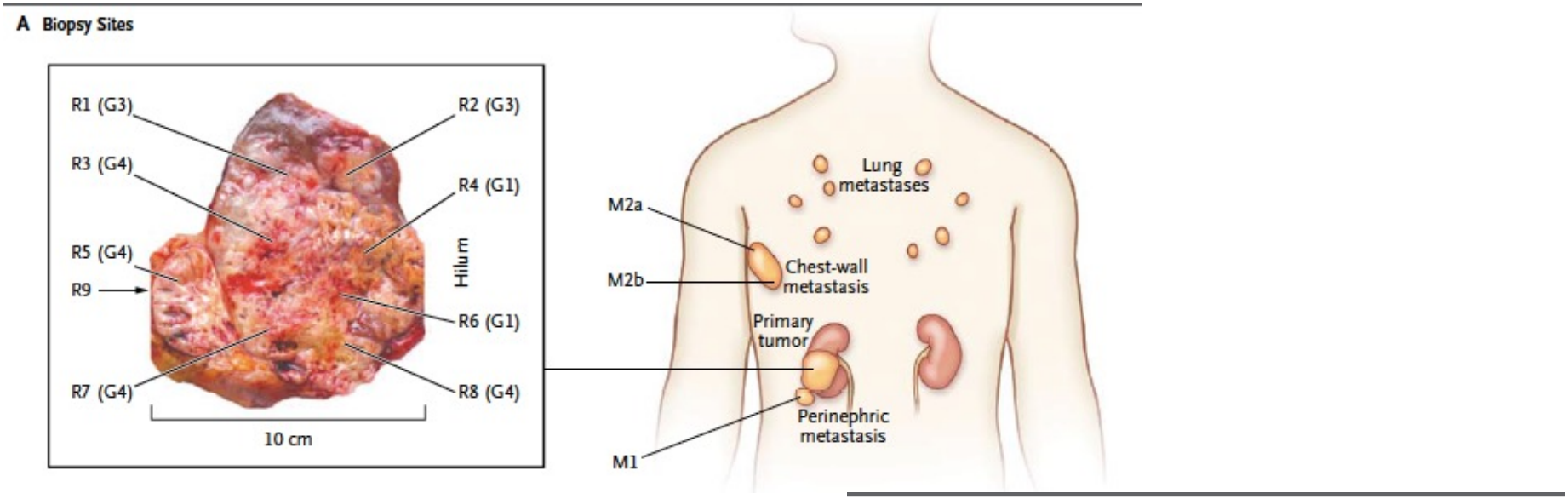
Implication: Traditional chemo and radiation therapy targets the rapidly dividing bulk of the tumor but may entirely miss the slow growing stem cells at its root.

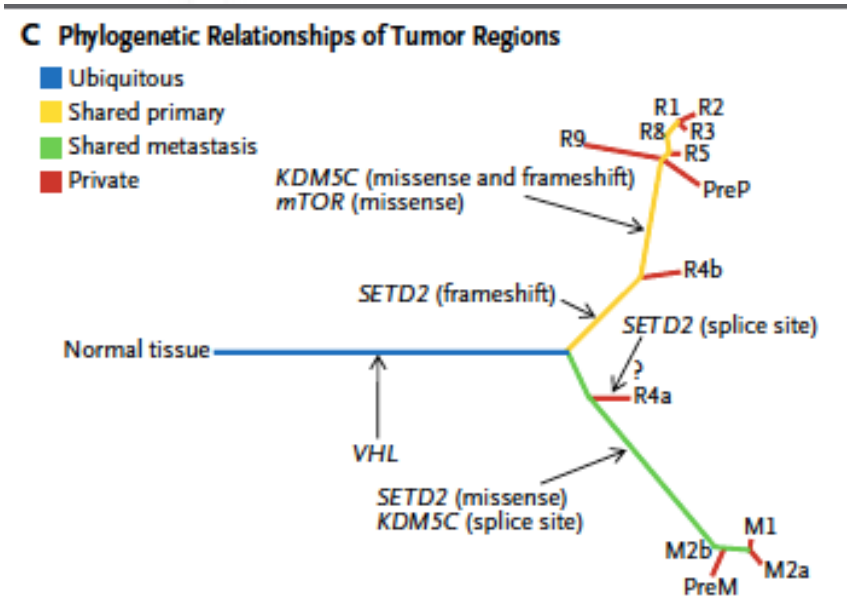
Implications of CSC for chemotherapy



- **AML - Bonnet and Dick, 1997**
- **ALL - Cobaleda et al., 2000**
- **Breast cancer - Al-Hajj et al., 2003**
- **Glioblastoma - Singh et al, 2003**
- **CML - Passegue et al., 2004**

Cancer Genomics informs on clonal evolution





The Genome Gets Personal!

The cells of human body

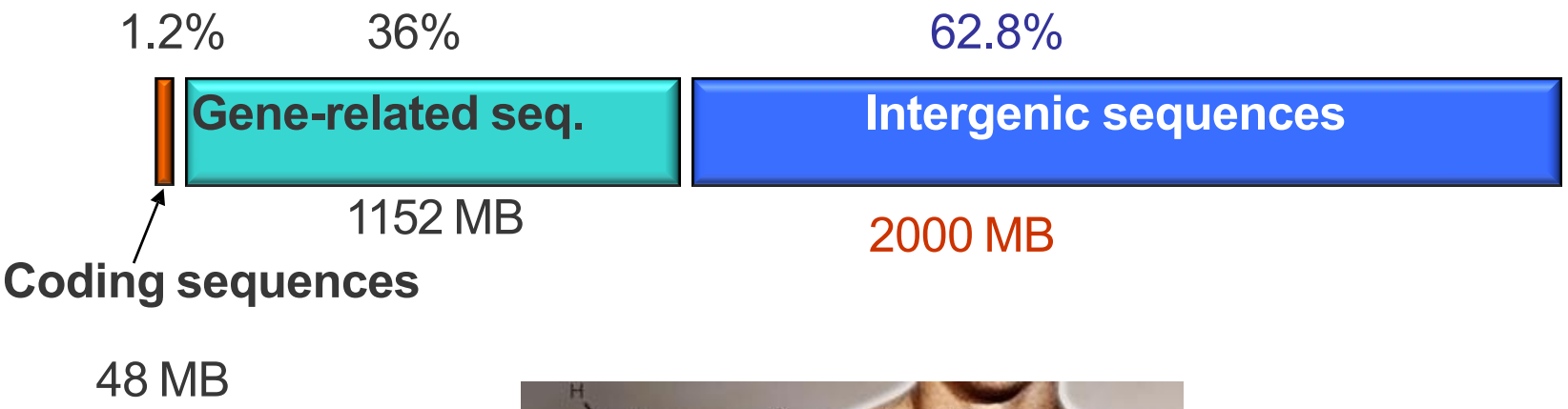
Cells:

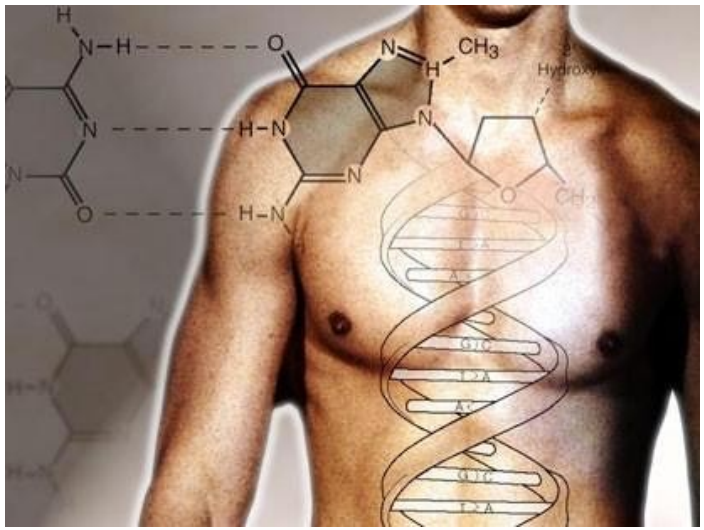
Bacterial: **100 trillion** **(10^{14})**
Human: **10 trillion** **(10^{13})**

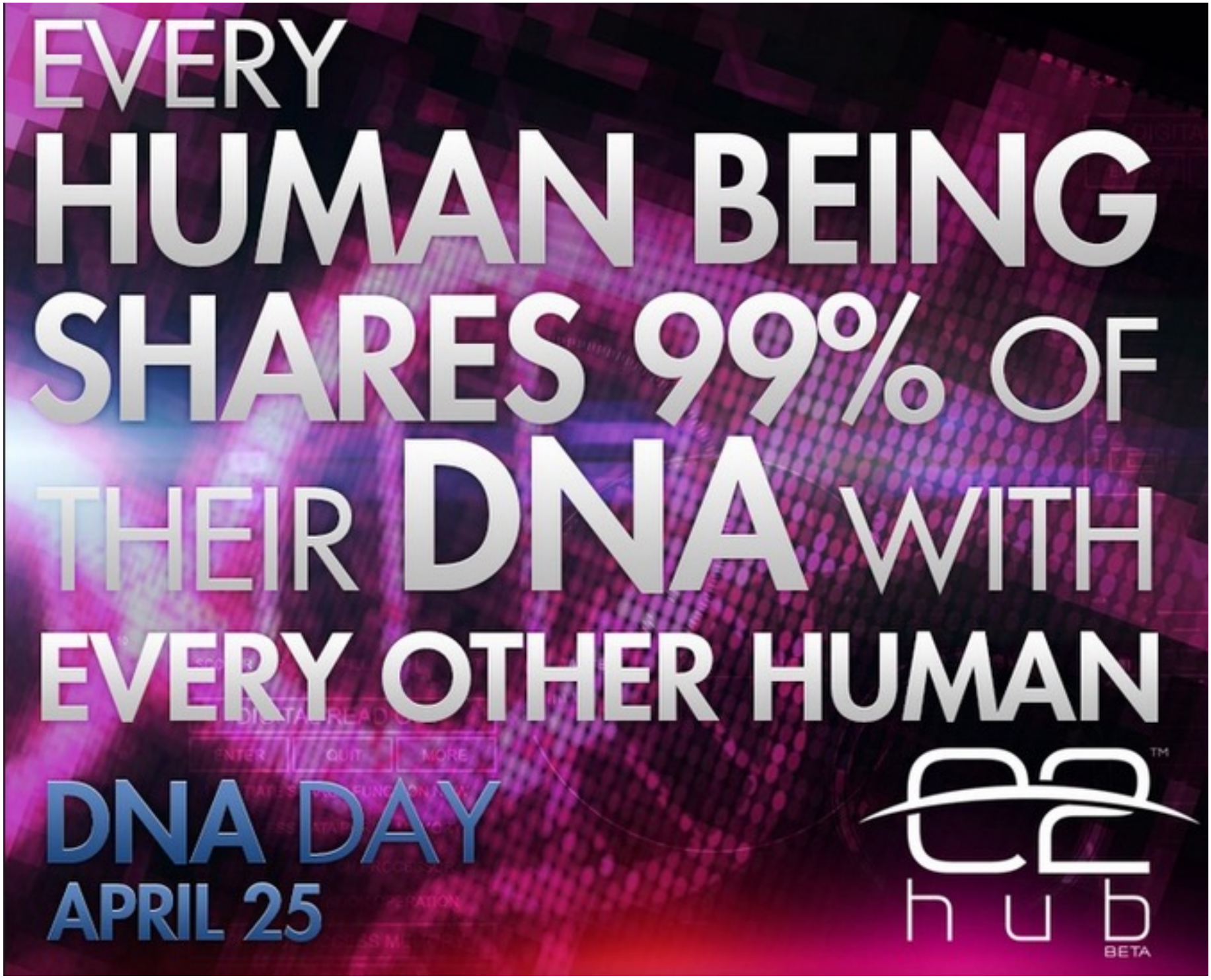
Genes:

100x more virus & bacterium than human

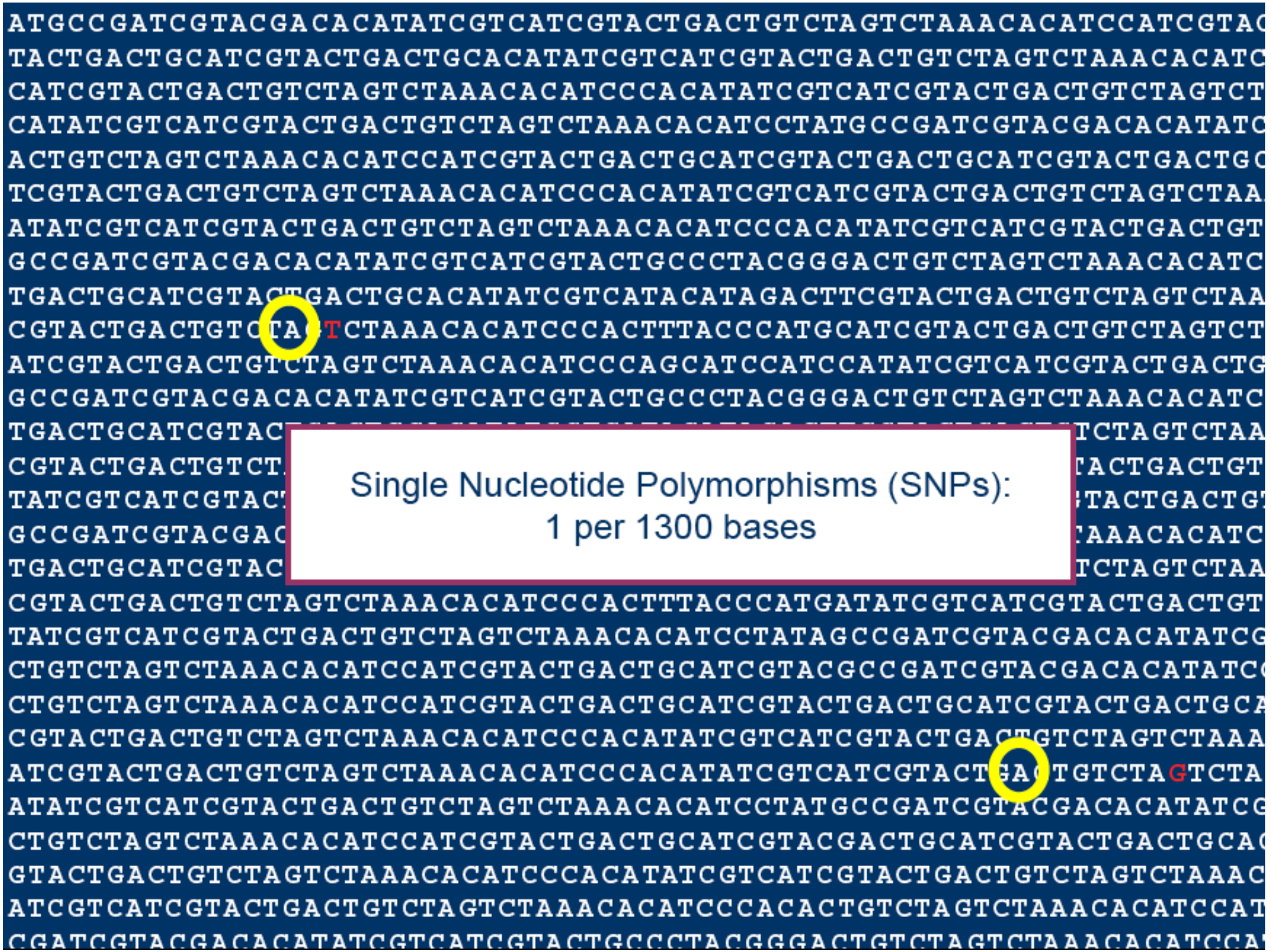
Human genome



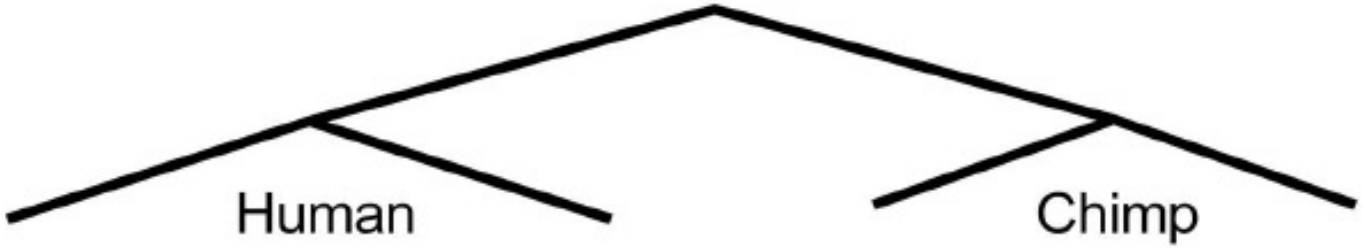



EVERY
HUMAN BEING
SHARES 99% OF
THEIR DNA WITH
EVERY OTHER HUMAN
DNA DAY
APRIL 25
hub
BETA

Human Genetic Variation


Single Nucleotide Polymorphisms (SNPs):
1 per 1300 bases

How much do genomes vary?



← one in a 1,000 bp →

← one in a 300 bases →

← one in a 100 bases →

Common Variant Important in Risk of Common Disease

ApoE4	Alzheimer's disease
Factor V Leiden	Venous thrombosis
HFE	Hemachromatosis
PPARγ	Type 2 Diabetes
MTHFR 667T	Cardiovascular disease
CCR5	HIV resistance
HLA-DQα	Type 1 Diabetes

Molecular Basis for biomarkers:
Human genetic variation susceptibility to diseases

Two individuals share 99.9% of DNA sequence

The remaining 0.1%reportedly has an enormous range of genetic variations and is responsible for a predisposition to asthma, diabetes, cancer, heart disease, schizophrenia, and many other diseases

Richard Cotton

Human Variom Project

Collection of variable sequences from different individuals

- primary focus on medical application

The FUTURE: personalized medicine

THE HUMAN VARIOME PROJECT

The human variome:

disease predisposition

Published Genome-Wide Associations through 07/2012
Published GWA at p≤5X10^{-8} for 18 trait categories

NHGRI GWA Catalog
www.genome.gov/GWAStudies
www.ebi.ac.uk/fgpt/gwas/

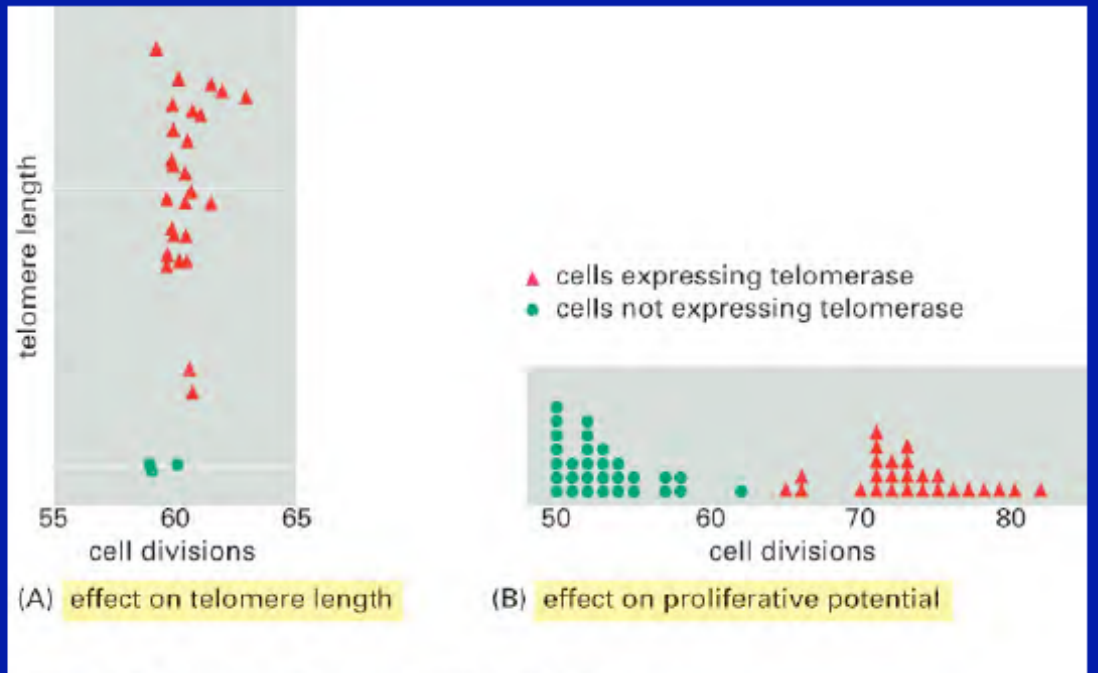
- Digestive system disorder
- Cardiovascular disorder
- Metabolic disorder
- Immune system disorder
- Neurological disorder
- Liver enzyme measurement
- Lipid or lipoprotein measurement
- Inflammatory marker measurement
- Hematological measurement
- Body measurement
- Cardiovascular measurement
- Other measurement
- Chemical compound
- Biological process
- Cancer
- Other disease
- Other trait
- Trait mapping in progress

National Human Genome Research Institute

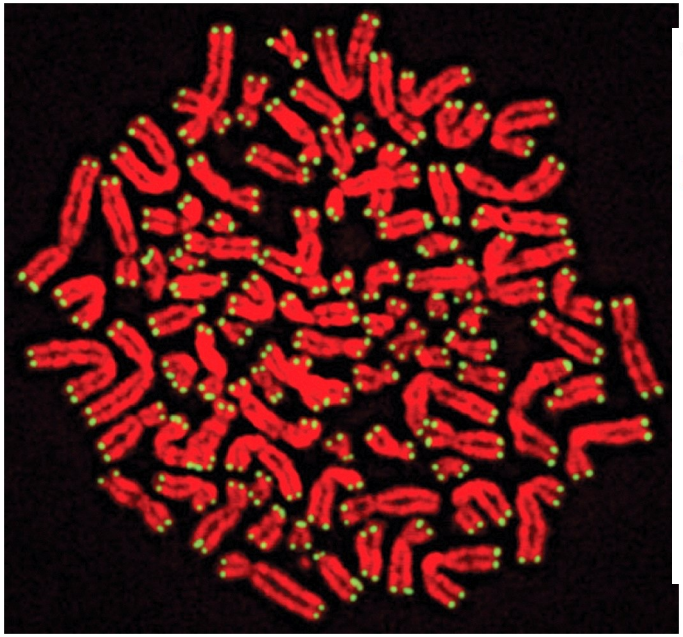
EMBL-EBI

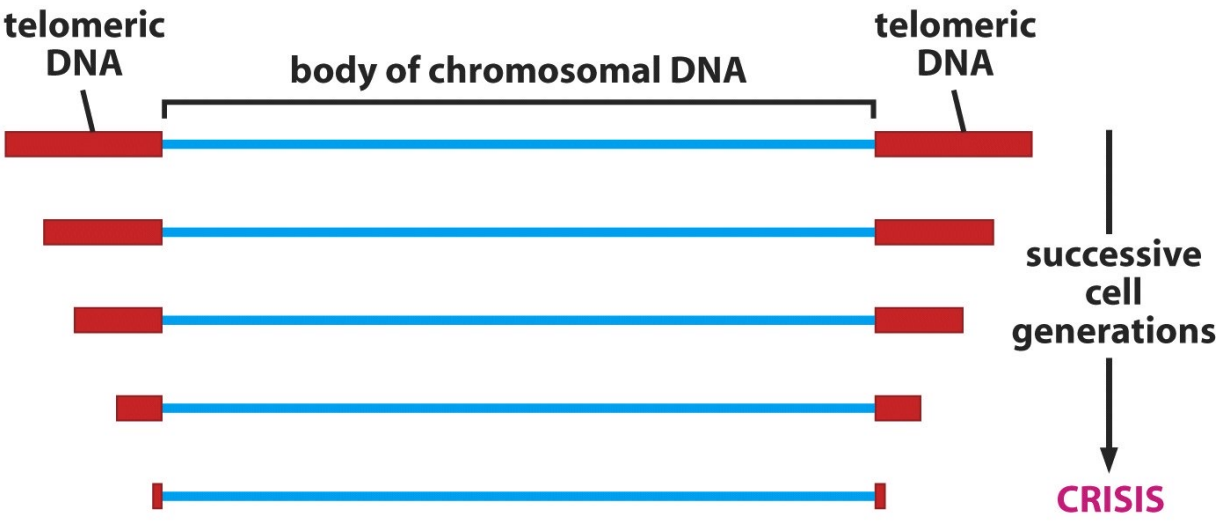
Τα ζωϊκά κύτταρα δεν διαιρούνται απεριόριστα

- **Μία εξήγηση για την κυτταρική γήρανση είναι τα τελομερίδια. Όταν τα τελομερίδια μικρύνουν κάτω από ένα ορισμένο επίπεδο, τότε το κύτταρο δεν μπορεί να διαιρεθεί πια. Όταν αποκαθίσταται η δράση της τελομεράσης, τότε το κύτταρο μπορεί να γίνει αθάνατο.**



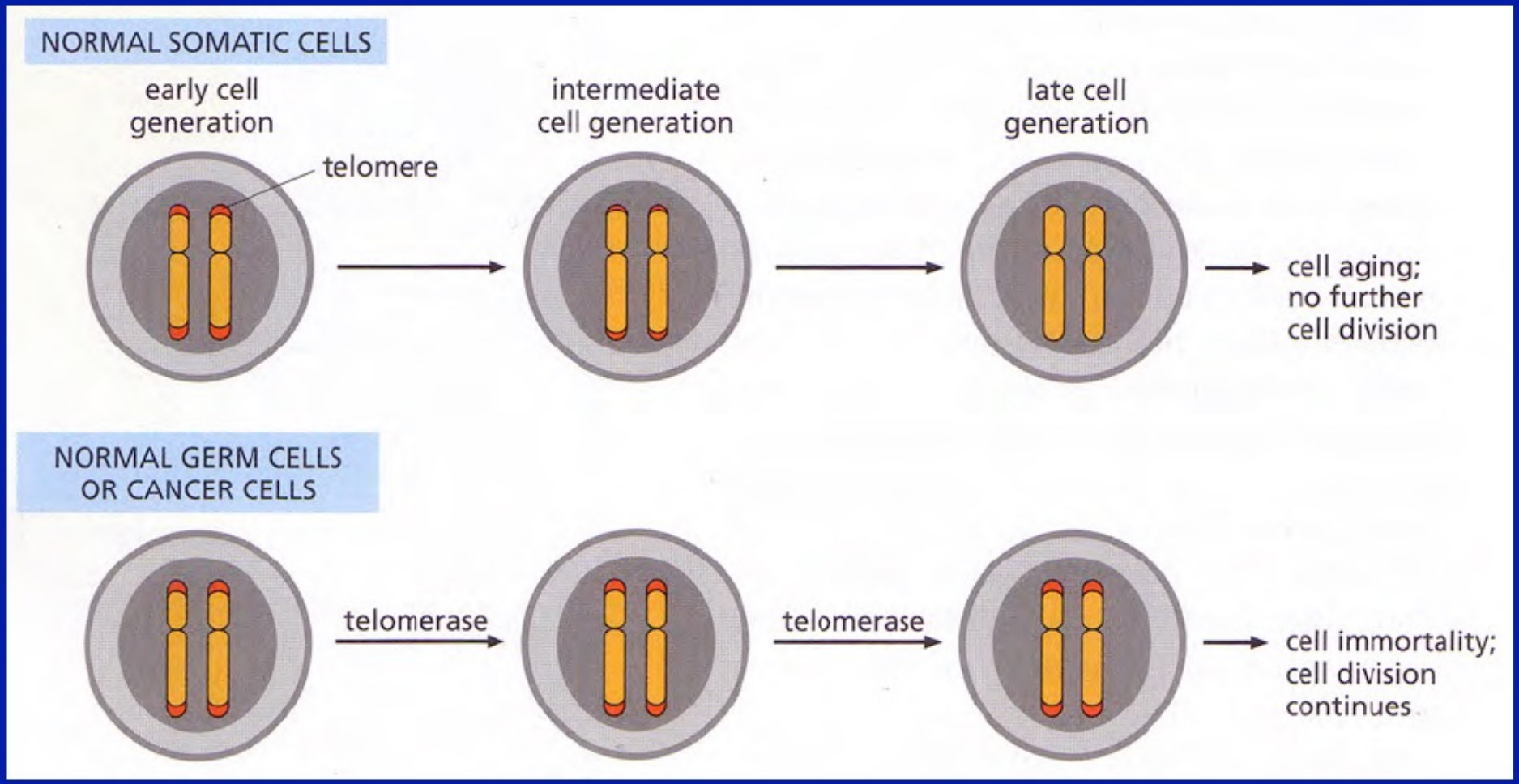
Cancer cells need to become immortal in order to form tumors



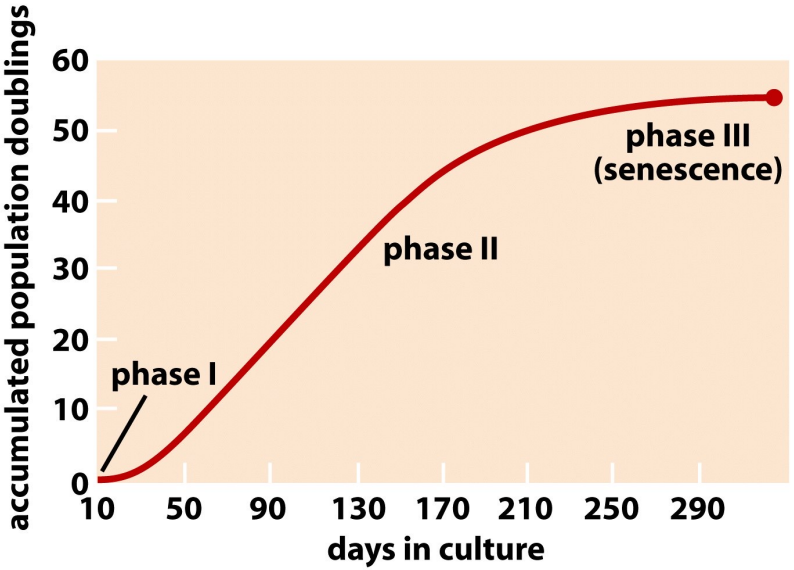


Shortening of telomeric DNA in concert with cell proliferation

Τα ζωϊκά κύτταρα δεν διαιρούνται απεριόριστα



Eternal Life : Cell Immortalization and Tumorigenesis

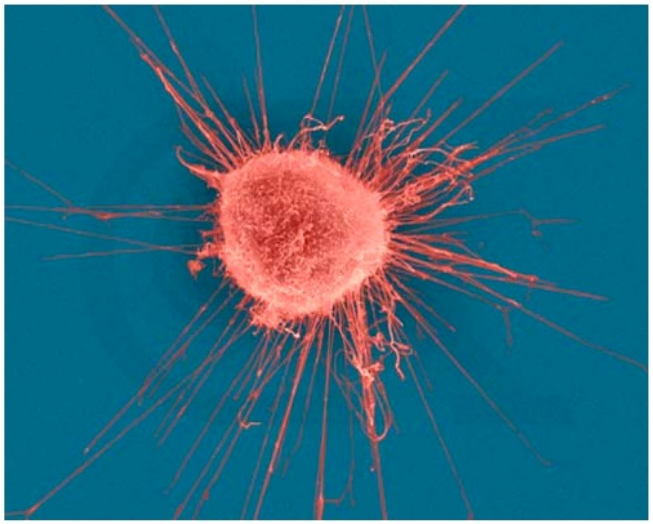


The proliferative capacity of cells passage extensively in culture

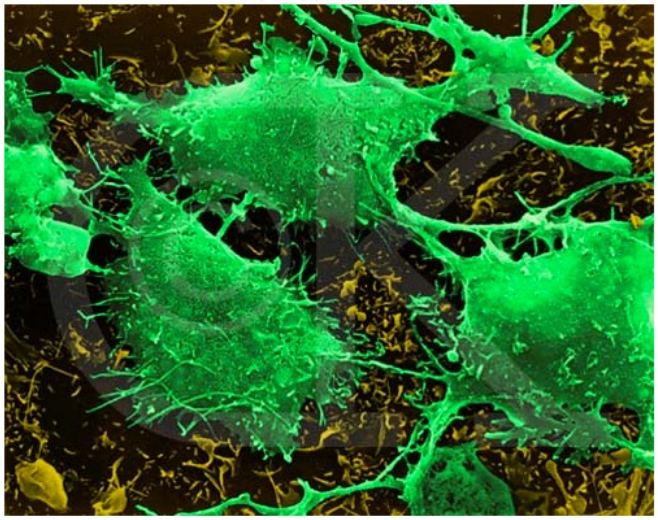
Cancer cells need to become immortal in order to form tumors

Marc Rosenthal

Breast Cancer Cell



Brain Cancer Cell

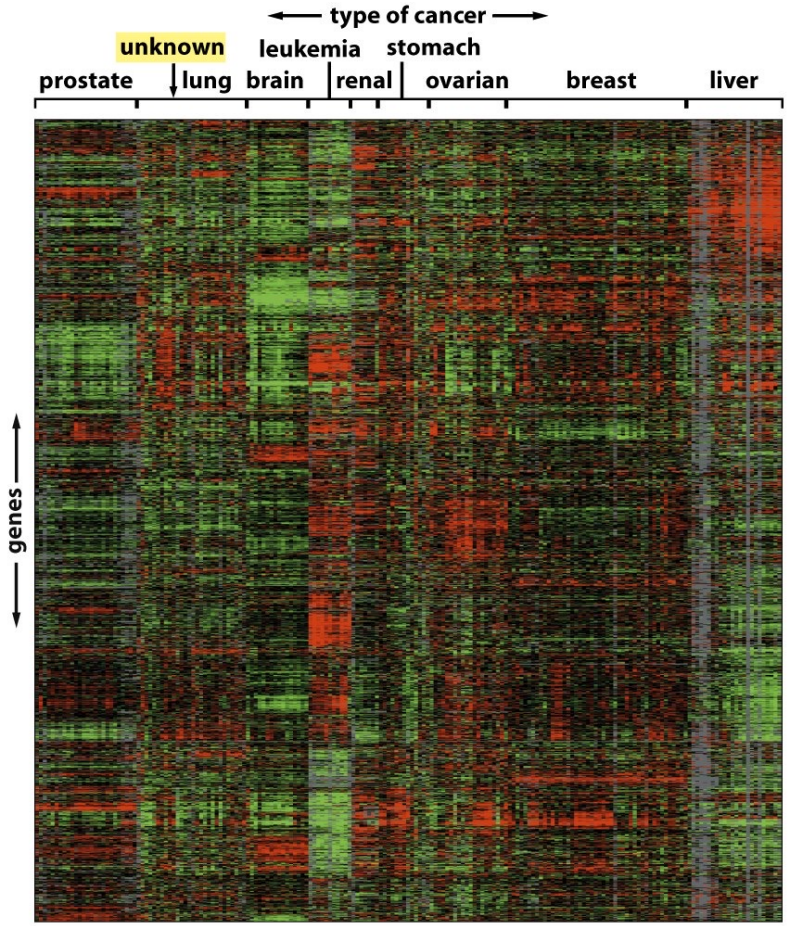


Cancer Cell Biology

Normal cell

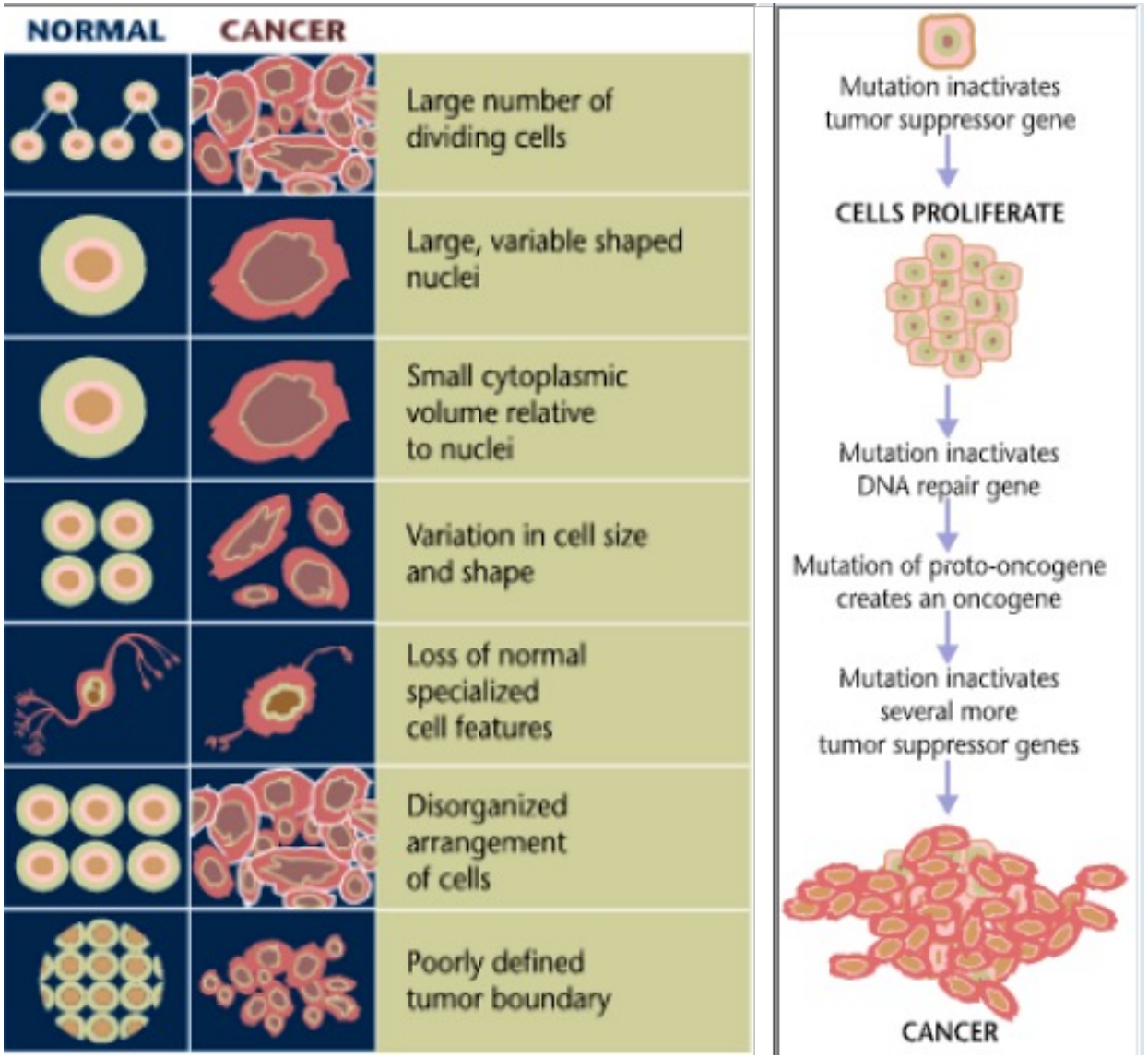
Example of one type of abnormal or cancerous cell

Gene expression patterns also control phenotype



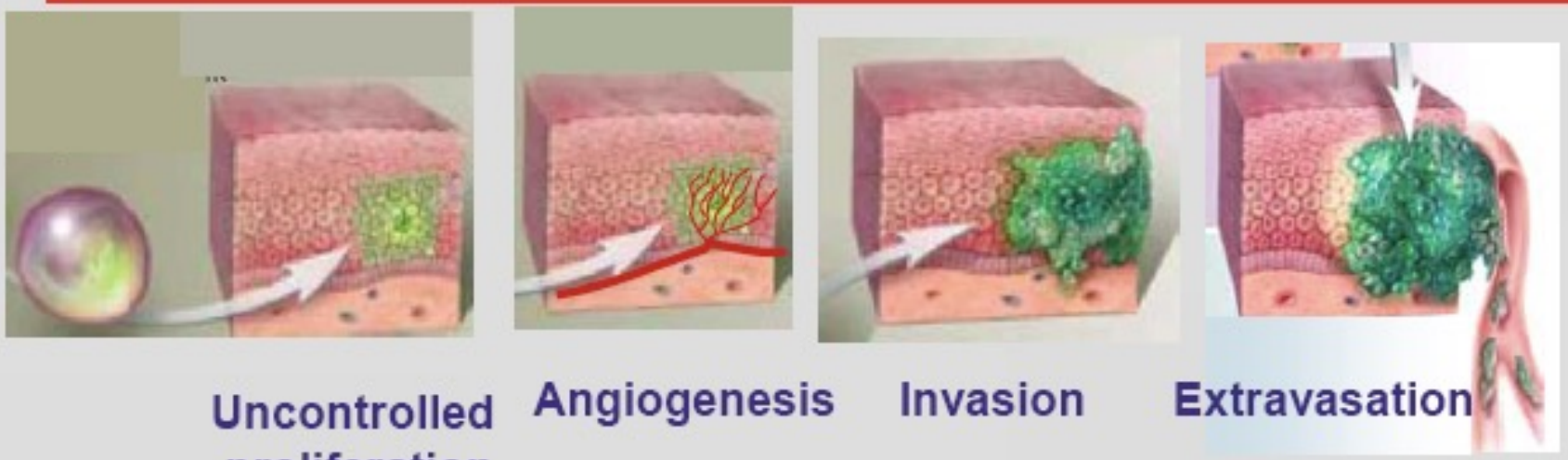
Global surveys of gene expression arrays

Μορφολογικές διαφορές μεταξύ φυσιολογικού και καρκινικού κυττάρου



https://universe-review.ca/I10-90-OncoDiff.png

The Development of Cancer Involves both 'Loss of Function' and 'Gain of Function' Alterations



Uncontrolled proliferation | Angiogenesis | Invasion | Extravasation

Metastasis

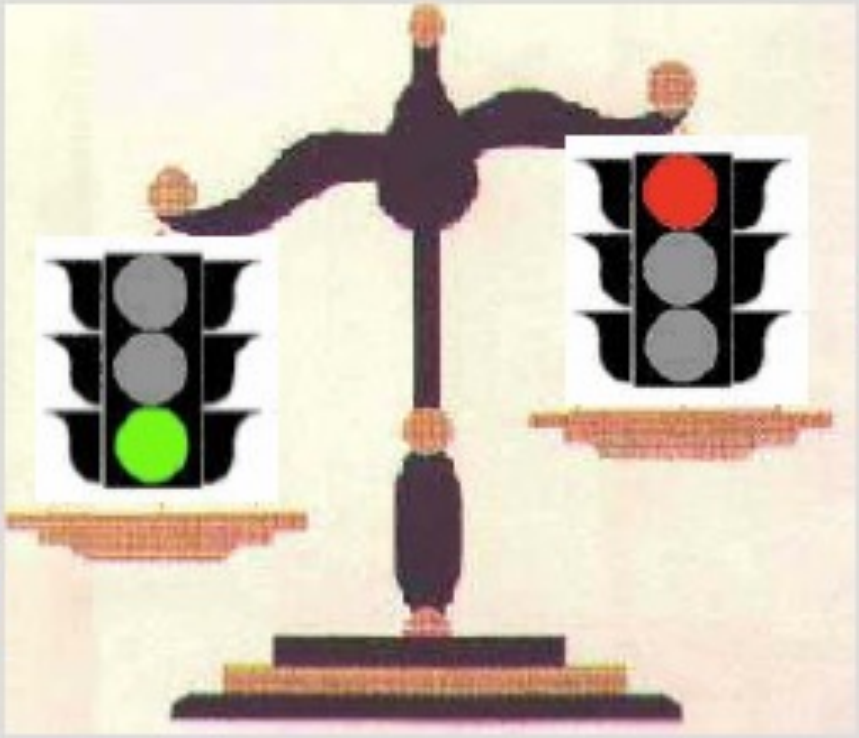
In general -- two types of alterations:

- 'Gain of function' alterations that "drive" the specific step

- 'Loss of function' alterations that inactivate checkpoints that normally prevent aberrant events

Delicate Balance of "off" and "on" Controls

Some proteins tell promote cells to proliferate, survive, or move.



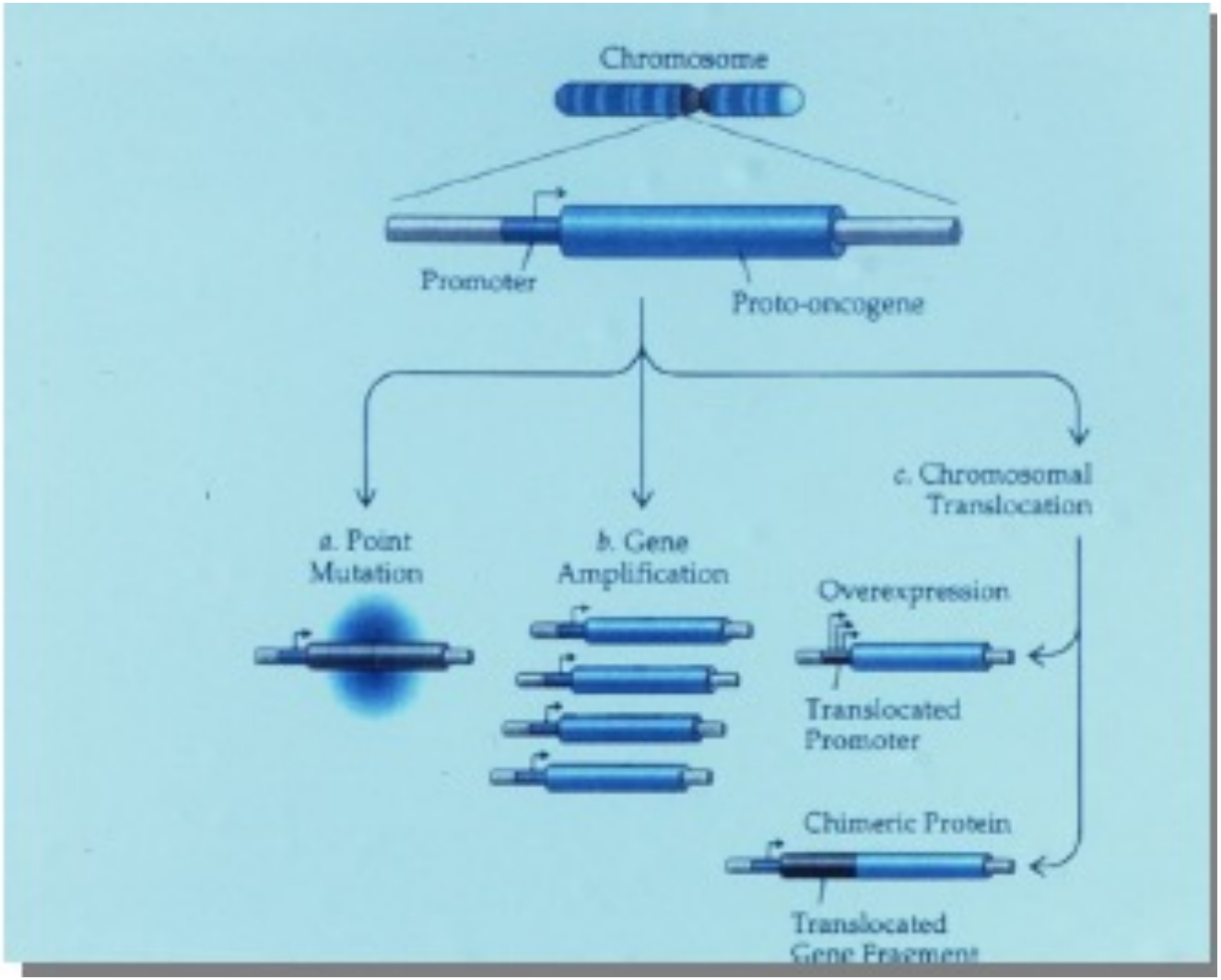
Other proteins halt division or tell cells to die.

These are over-produced or mutated causing them to always ON in cancer.

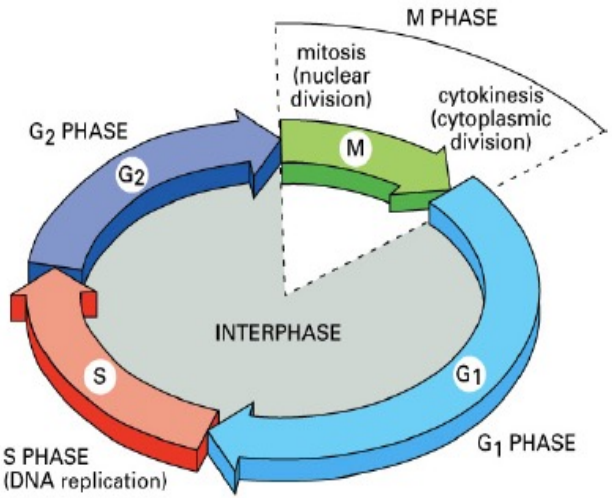
These are lost or underproduced in cancer.

Cellular Oncogenes

Activation of Proto-oncogenes

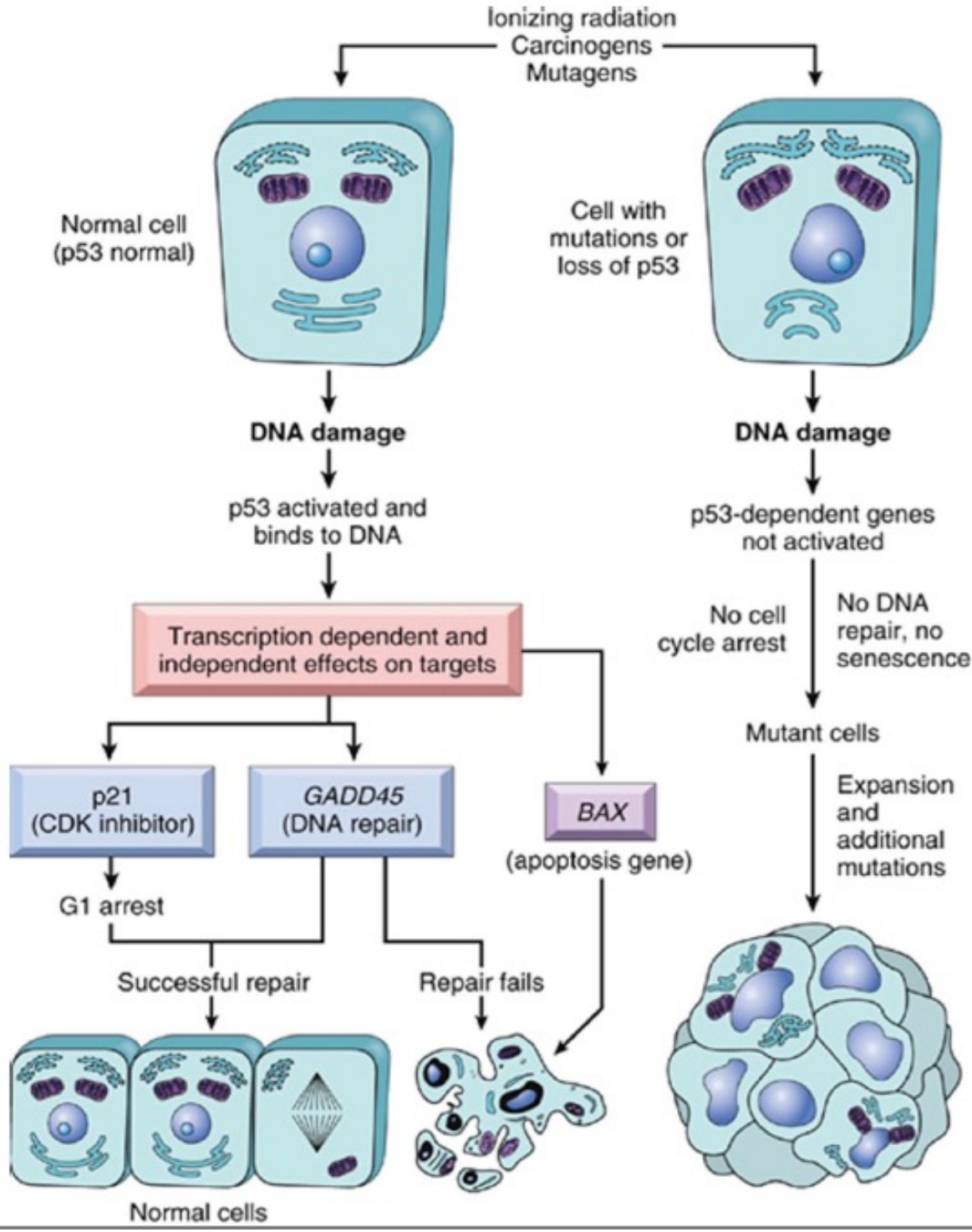


Tumor Suppressor Genes



- **Tumor suppressor genes encode proteins that inhibit the proliferation of cells and/or prevent the accumulation of mutations that can lead to cancer**

- **Both alleles must be inactivated to relieve the block in tumor development imposed by these genes; therefore, mutations in tumor suppressor genes are recessive**



p53 Tumor Suppressor Gene: Guardian of the Genome

Loss of p53 Function

↓

Accelerated Mutation Rate (genomic instability)

↓

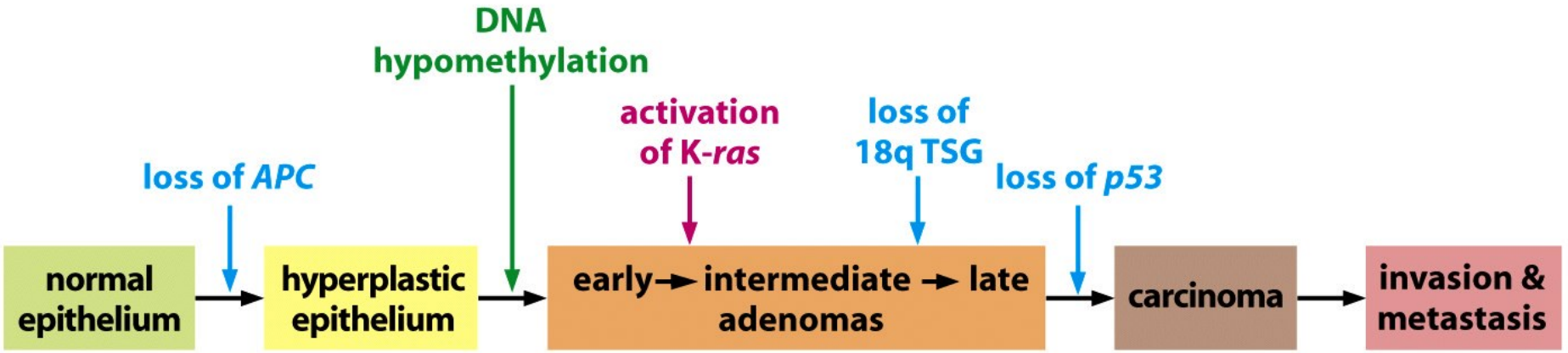
Genetic Heterogeneity

↓

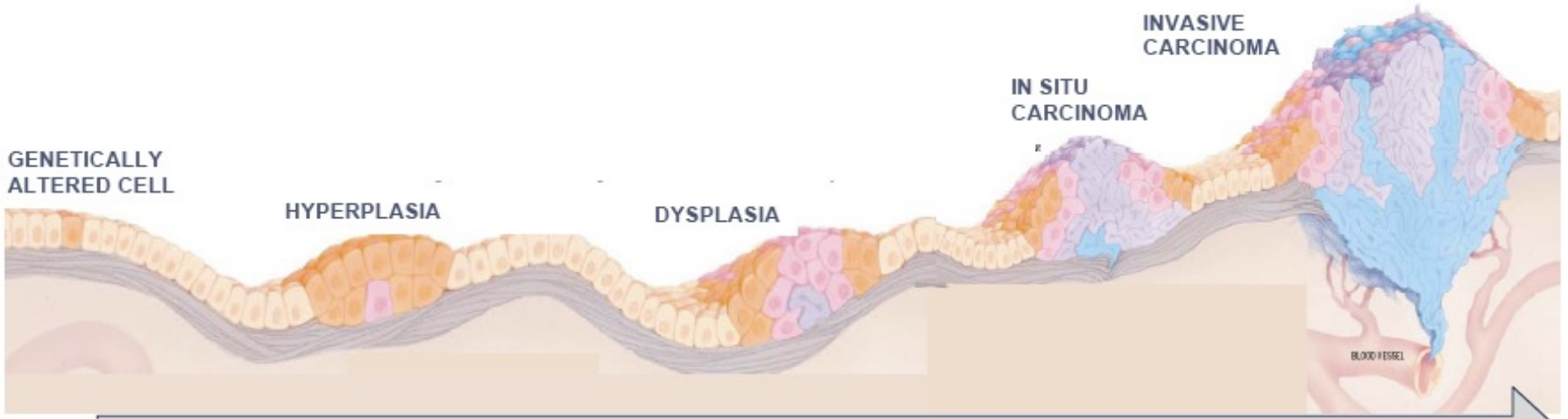
Accelerated Tumor Evolution

Gene Name	*Pathways/Function*	*Gain or Loss of Function?*
BRAF	**RAS/RAF/ERK/MEK**	**Gain**
NF1	**RAS**	**Loss**
Kras	**RAS/RAF/ERK/MEK AKT/PI3K**	**Gain**
Nras	**RAS/RAF/ERK/MEK AKT/PI3K**	**Gain**
Hras	**RAS/RAF/ERK/MEKAKT/PI3K**	**Gain**
AKT1	**AKT/PI3K**	**Gain**
AKT2	**AKT/PI3K**	**Gain**
AKT3	**AKT/PI3K**	**Gain**
PIK3CA	**AKT/PI3K/RAS/RAF/ERK/MEK**	**Gain**
PTEN	**AKT/PI3K/RAS/RAF/ERK/MEK**	**Loss**
P53	**DNA Repair**	**Loss**
FBXW7	**DNA Repair**	**Loss**
ATM	**DNA Repair**	**Loss**
PARP1	**DNA repair**	**Loss**
PARP2	**DNA repair**	**Loss**
ERCC1	**DNA repair**	**Loss**
MLH1	**DNA repair**	**Loss**
MSH2	**DNA repair**	**Loss**
NBN	**DNA Repair**	**Loss**
ATR	**DNA repair**	**Loss**
MGMT	**DNA repair**	**Loss**

Tumor suppressor genes and colon progression



Cancer Progression



- Self sufficiency of growth
- Insensitivity to anti-growth signals
- Evade programmed cell death/apoptosis
- Limitless replication potential
- Develop and sustain angiogenesis
- Tissue invasion and metastasis

In tumors cells, there is a loss of checkpoint control proteins that prevent inappropriate cell cycle progression

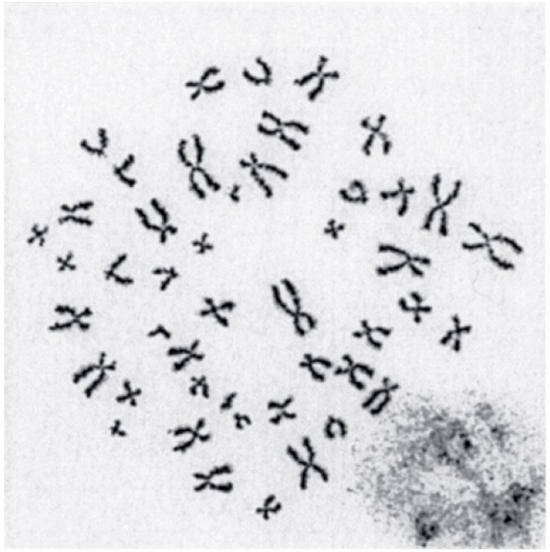
Checkpoints:

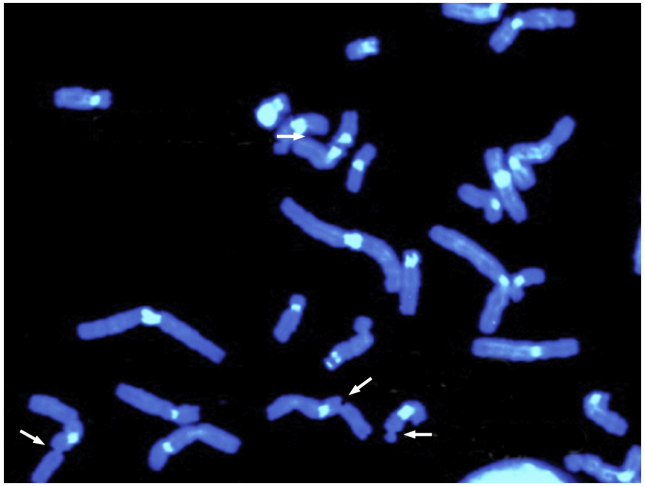
- Monitor proper progression of cell cycle processes
- Induce cell-cycle delay
- Help activate repair pathways
- Maintenance of cell-cycle arrest until repair complete
- Re-initiate cell-cycle progression

Examples: ATM,ATR, Chk1,Chk2,BRCA1, Mad, Mre11,Nbs1, p53, p16, Rb --

Loss of all these is known to cause familiar cancer or clinical syndromes that are cancer prone and mice lacking these genes show increased incidence of tumors.

Consequences of loss checkpoint controls





Programmed cell death

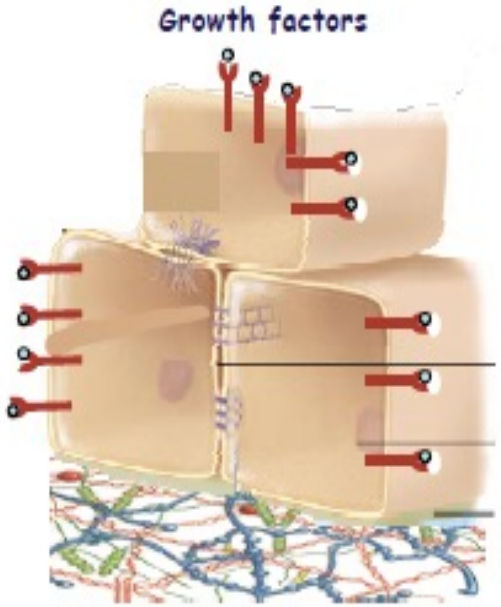
Programmed cell death:

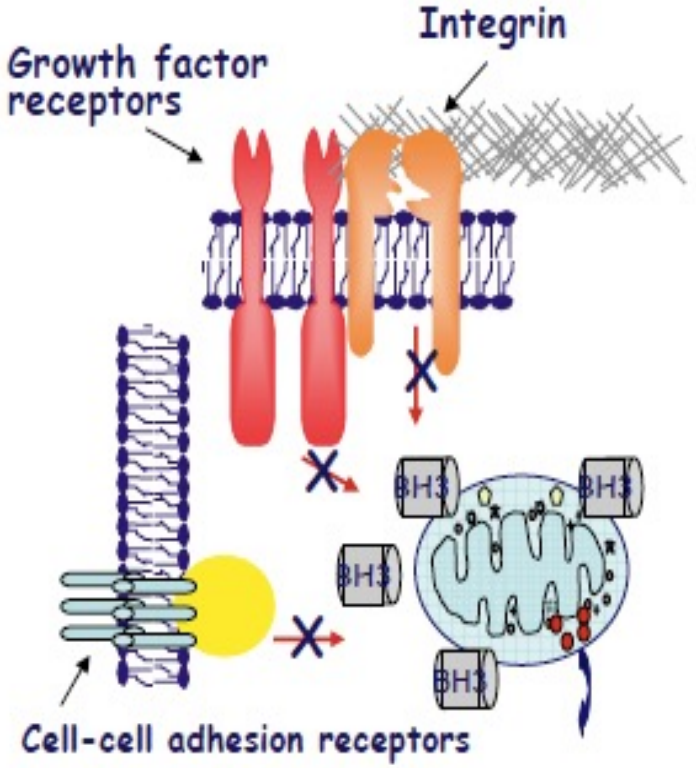
- ensures that tissues maintain an appropriate cell number
- ensures that aberrant cells are destroyed to avoid pathologic consequences
- to maintain homeostasis, ~10 billion cells are made each day to balance cell death

Therefore, programmed cell death can be considered as significant as cell proliferation.

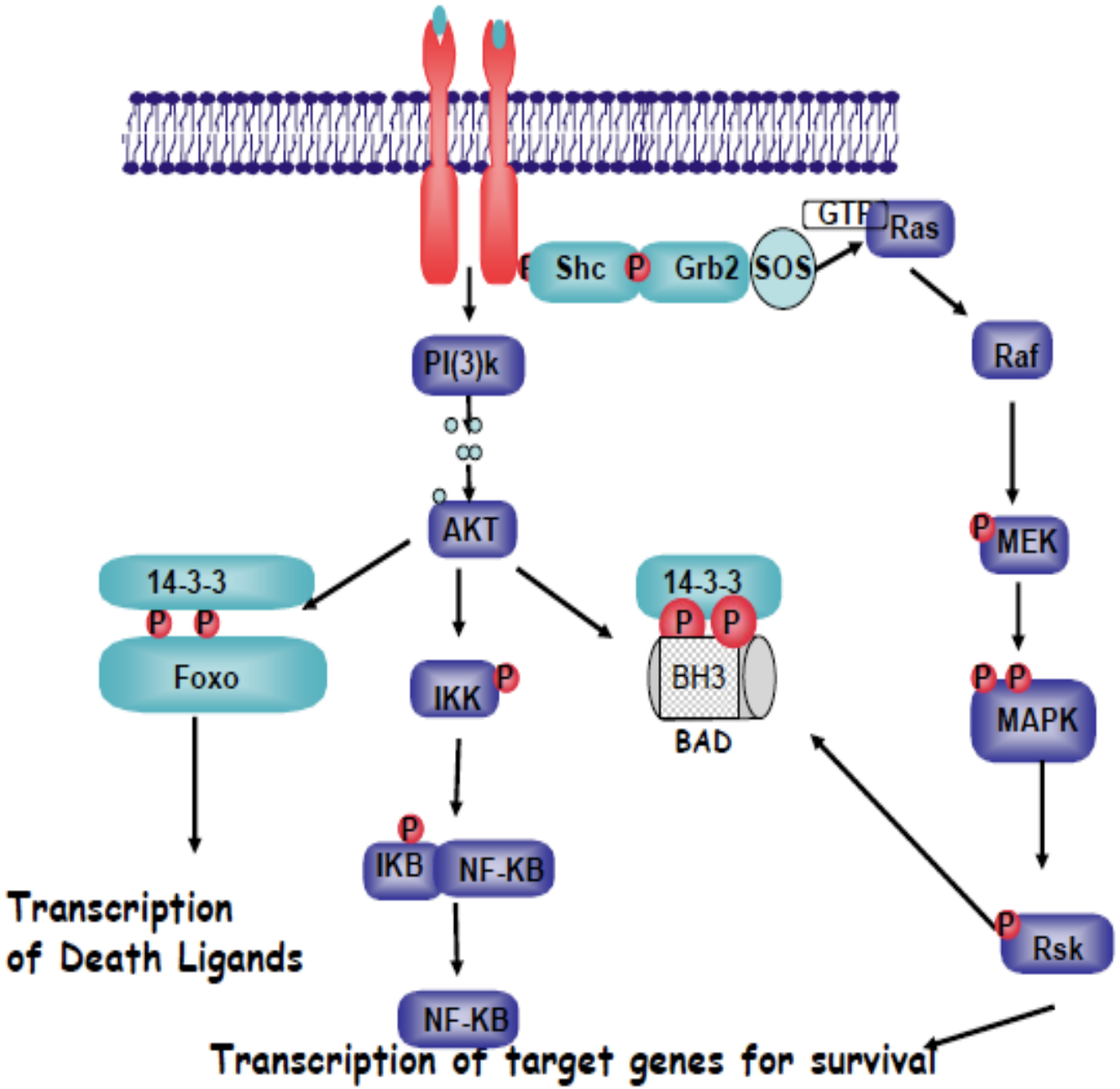
Pathways That Protect Cells from Apoptosis

- Most evidence suggests that cell death is a default pathway
- Cells need to actively keep these pathways off in order to prevent apoptosis
- Many cellular pathways control apoptosis

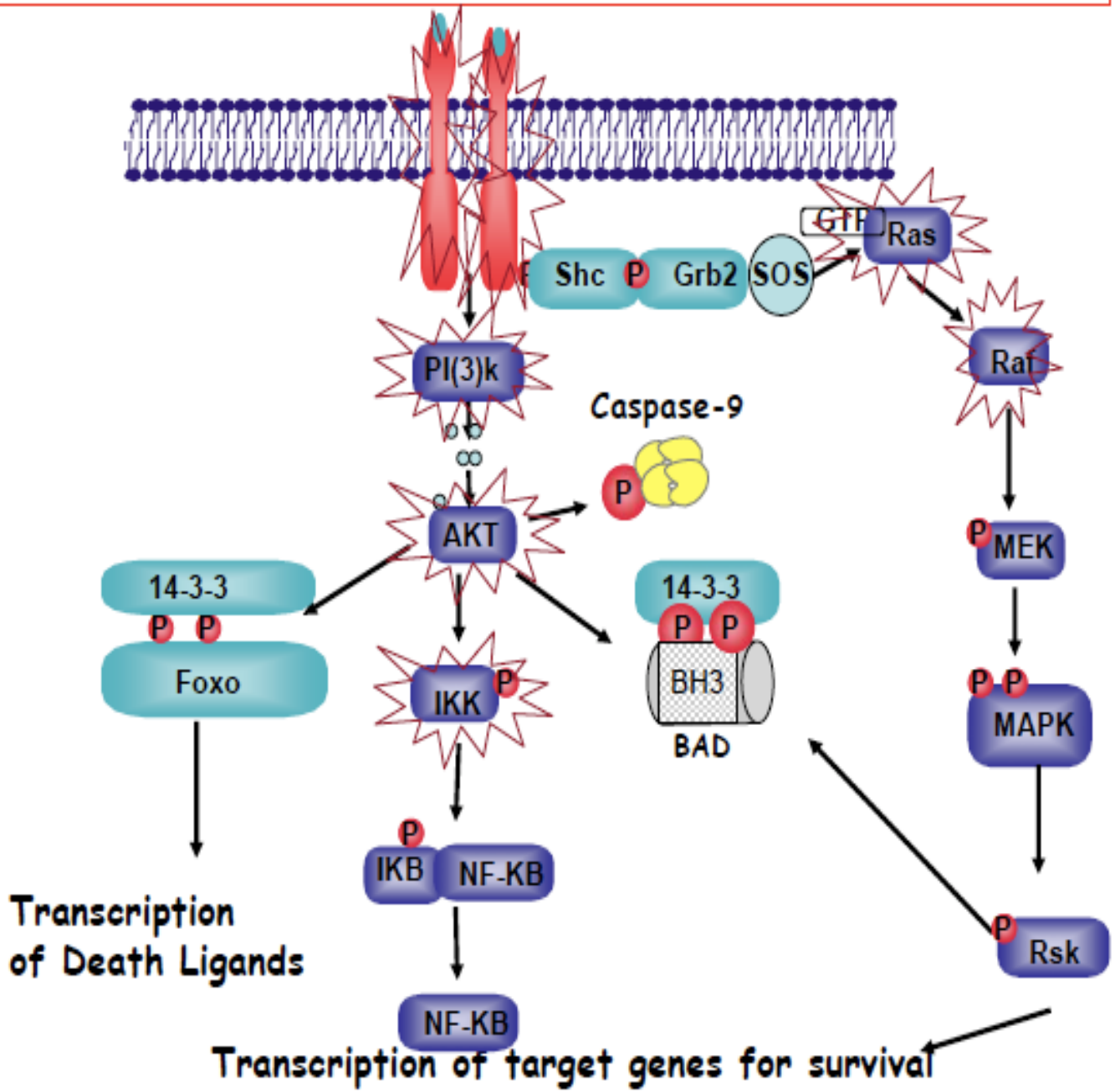




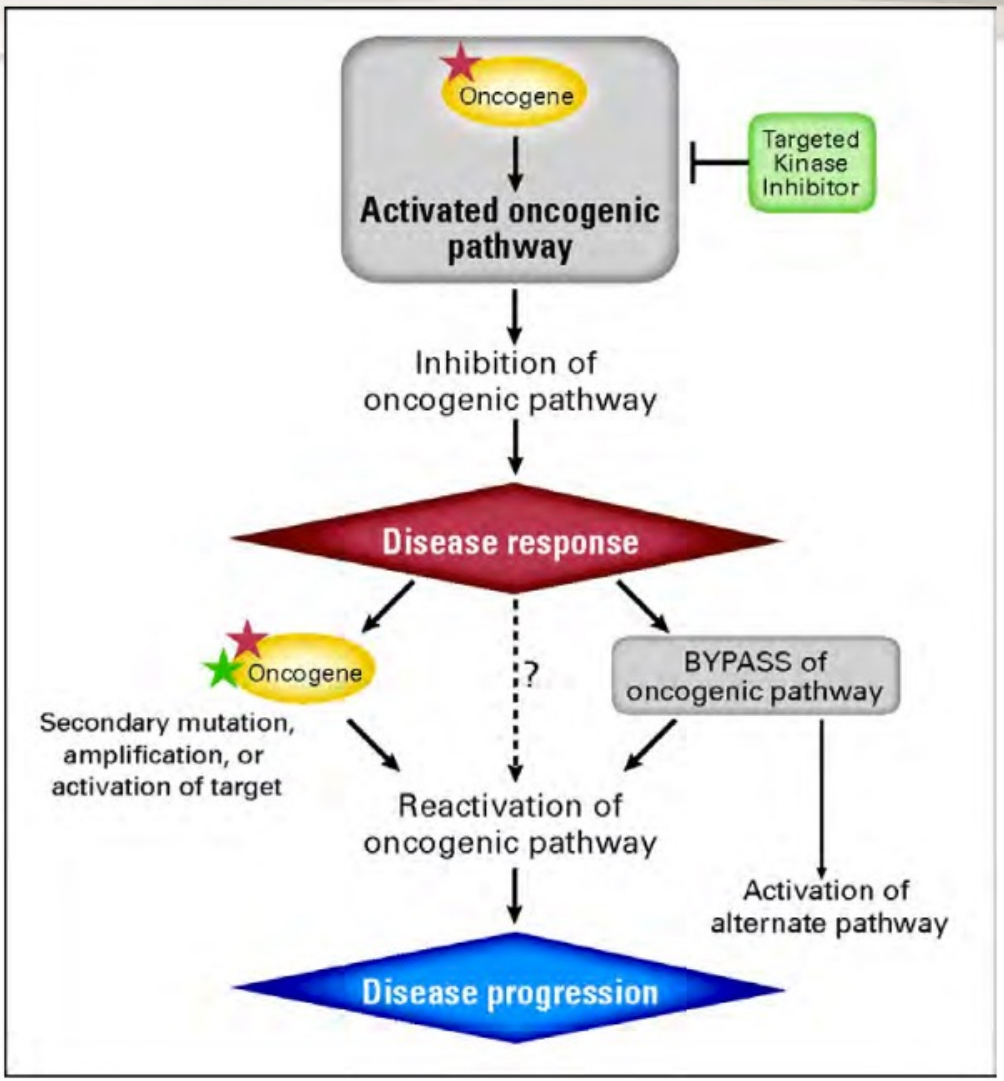
Pathways That Regulate Survival



Pathways That Regulate Survival

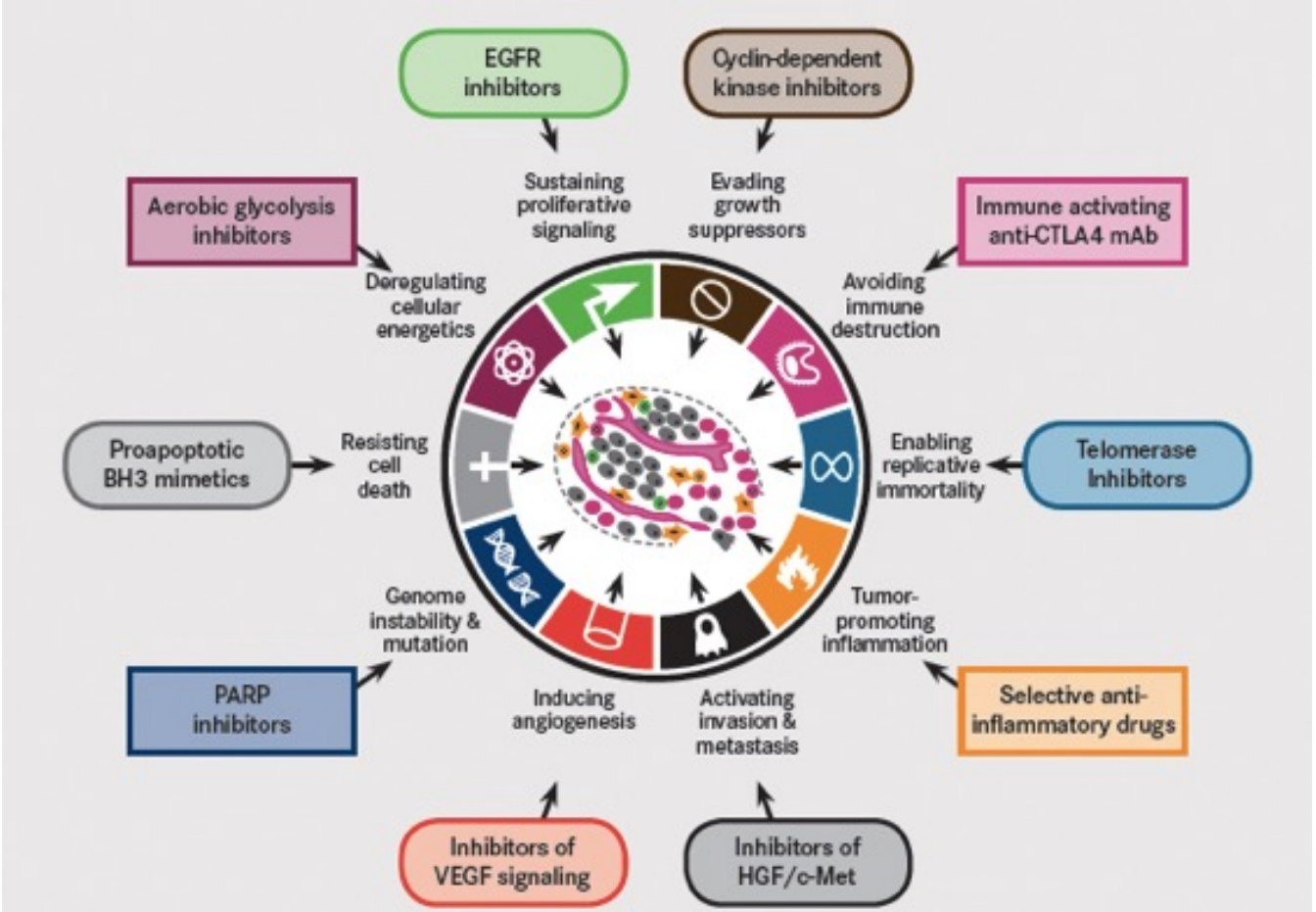


Kinase oncogene dependence and principles of drug resistance.



Wagle N et al. JCO 2011;29:3085-3096

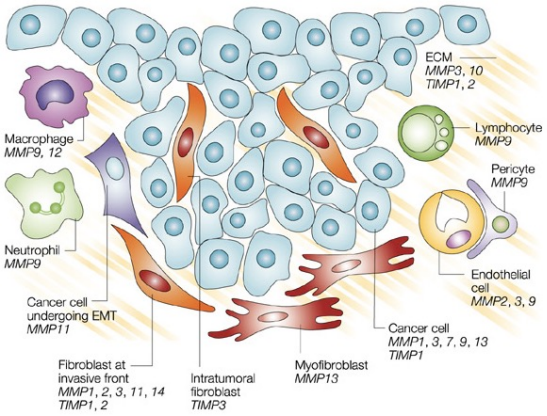
Hallmarks of Cancer : Therapeutic Opportunities

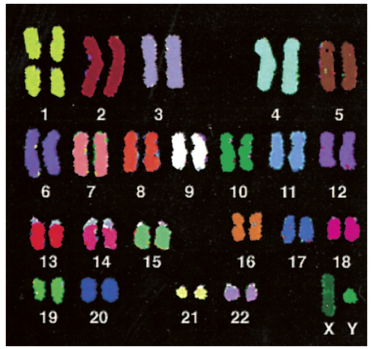


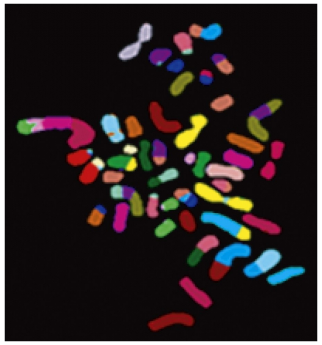
This figure illustrates some of the many approaches employed in developing therapeutics targeted to the known and emerging hallmarks of cancer.

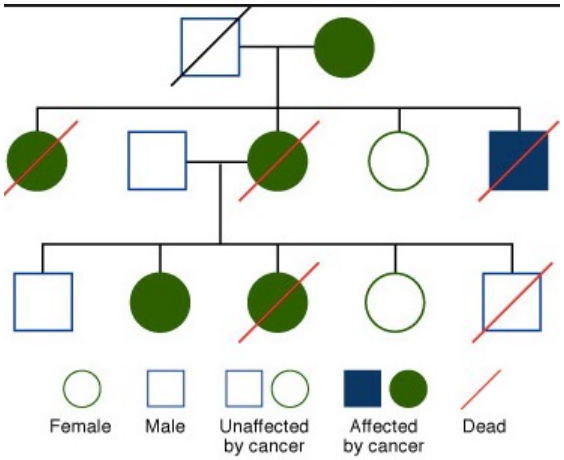
EGFR indicates epidermal growth factor receptor; CTLA4, cytotoxic T lymphocyte-associated antigen 4; mAb, monoclonal antibody; HGF, hepatocyte growth factor; VEGF, vascular endothelial growth factor; PARP, poly-(ADP ribose) polymerase.

Source: Hanahan D, Weinberg RA. Hallmarks of cancer: the next generation. *Cell*. 2011;144:646-674. Reprinted with permission.

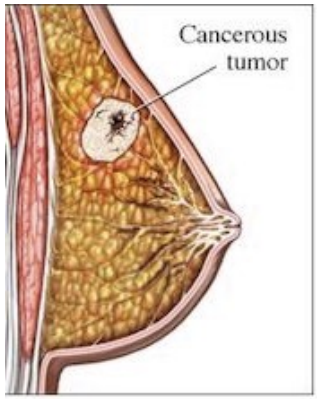


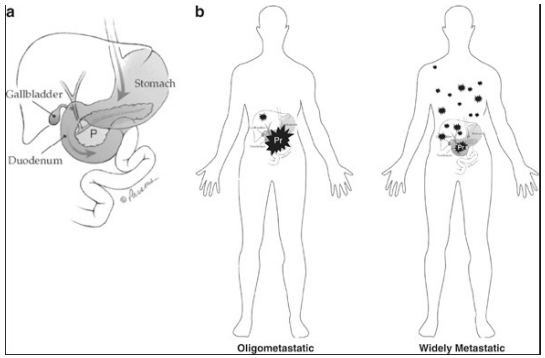




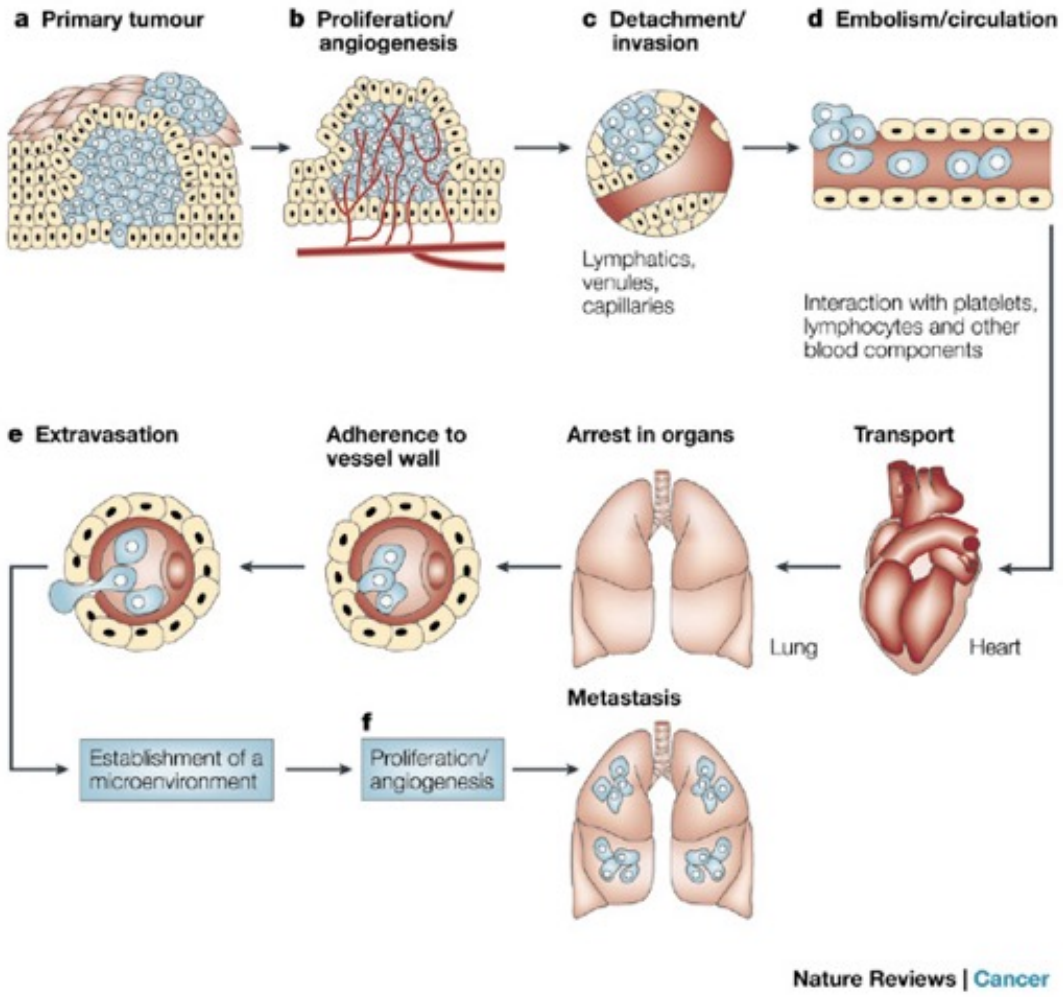


Cancer is Complex disease



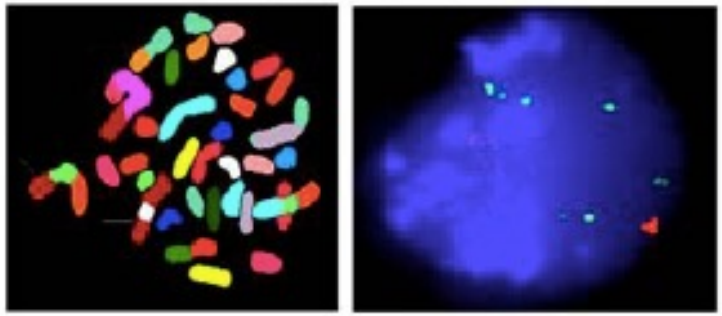


Early invasion in primary tumors->recurrence and metastasis despite surgical therapy

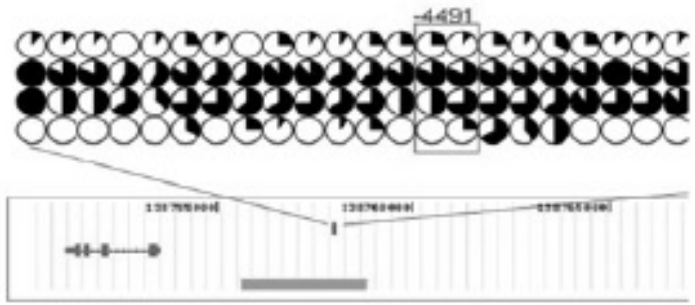


Cancer genes are dysregulated by multiple mechanisms

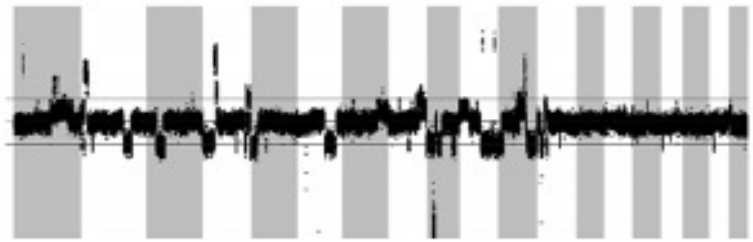
Aneuploidy; Re-arrangement; Translocation



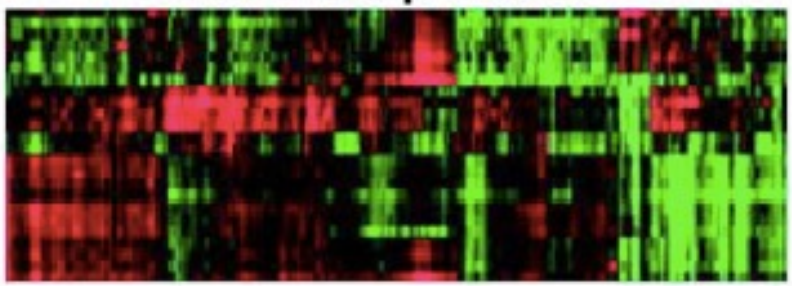
Methylation or histone modification



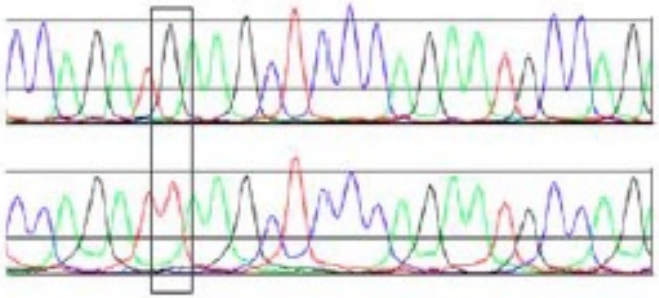
Copy number aberrations



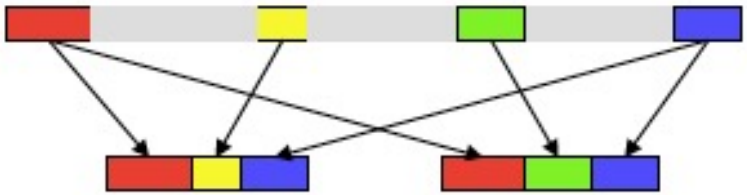
Altered expression



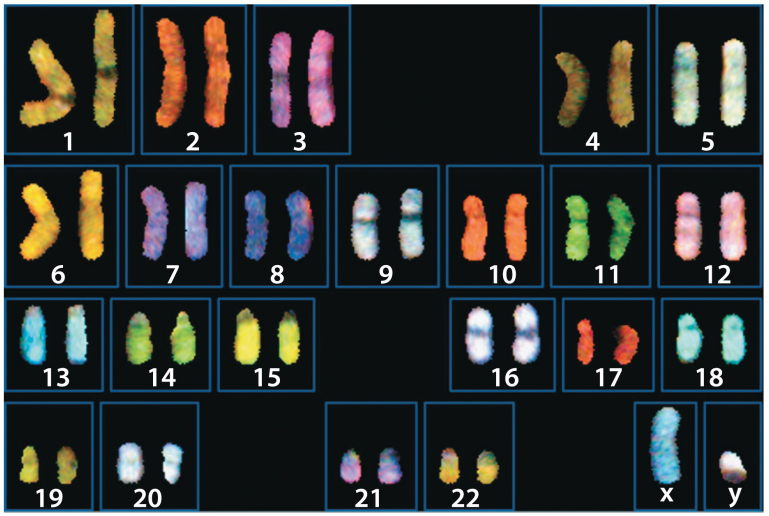
Somatic mutations



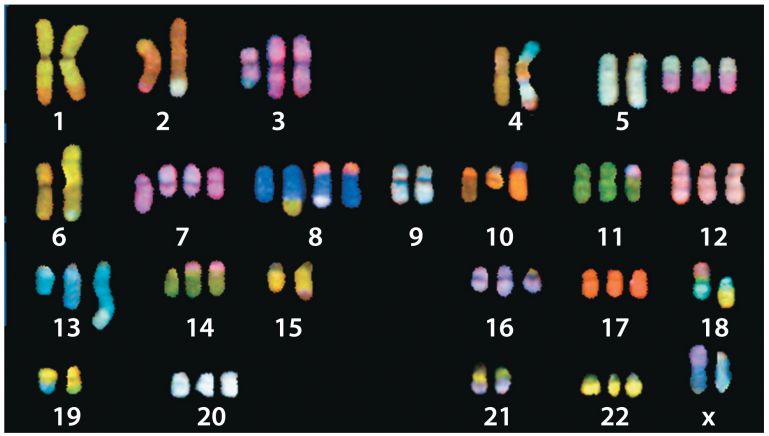
Gene Splicing Alterations



(α)



(β)

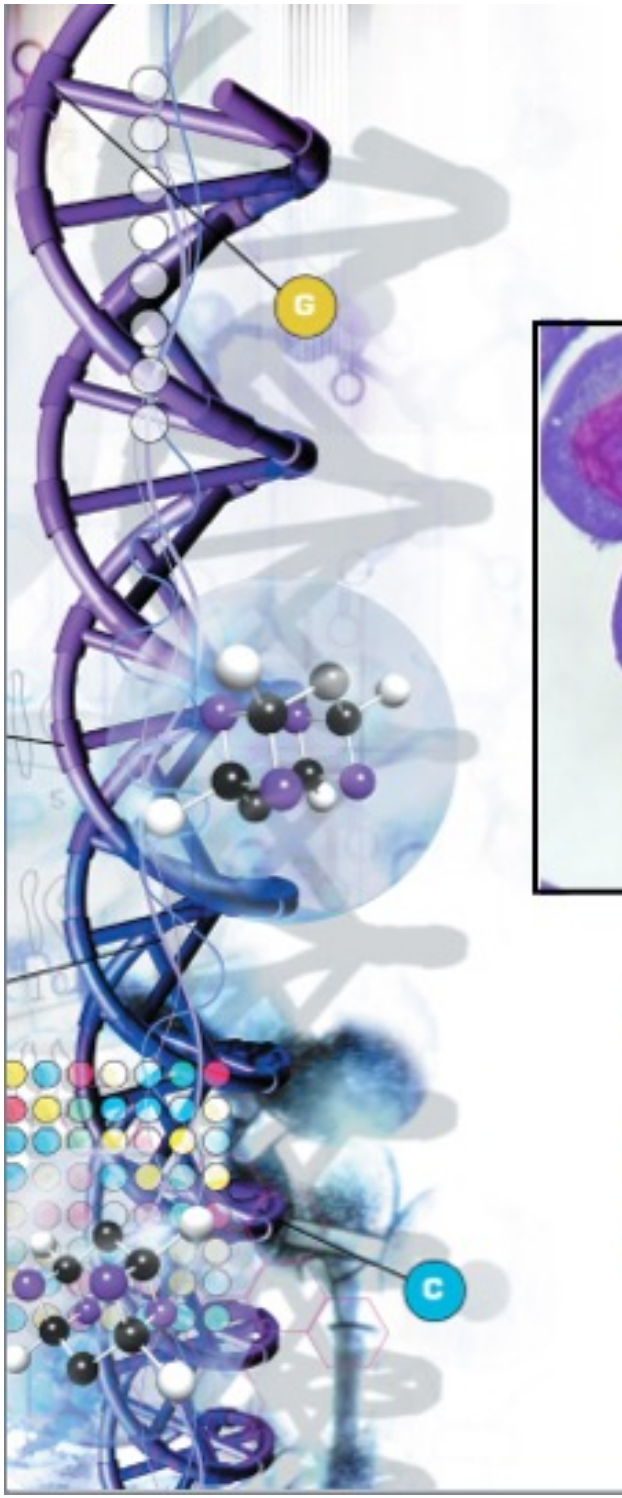


(α) Φασματικός καρυότυπος φυσιολογικού κυττάρου. (β) Καρυότυπος καρκινικού κυττάρου στον οποίο φαίνονται διάφορες μεταθέσεις και ελλείμματα καθώς και ανευπλοειδίες. Οι χρωμοσωμικές ανωμαλίες αποτελούν τυπικό χαρακτηριστικό των καρκινικών κυττάρων.

Abnormal Chromosome Number

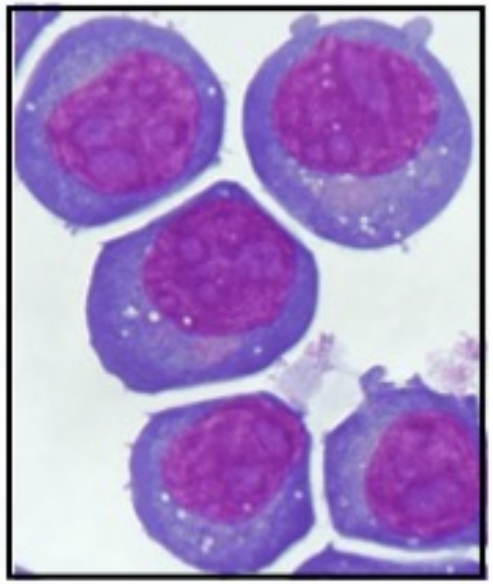
Chromosome Abnormalities

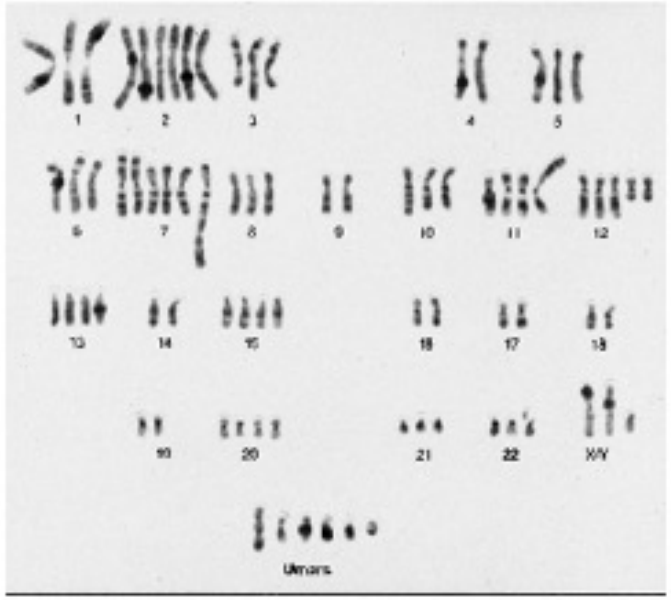
Type of Abnormality	Definition
Polyploidy	Extra chromosome sets
Aneuploidy	An extra or missing chromosome
Monosomy	One chromosome absent
Trisomy	One chromosome extra
Deletion	Part of a chromosome missing
Duplication	Part of a chromosome present twice
Inversion	Segment of chromosome reversed
Translocation	Two chromosomes join long arms or exchange parts



Cancer

A Disease of the Genome





Challenge in Treating Cancer:

- Every tumor is different
- Every cancer patient is different

ICGC

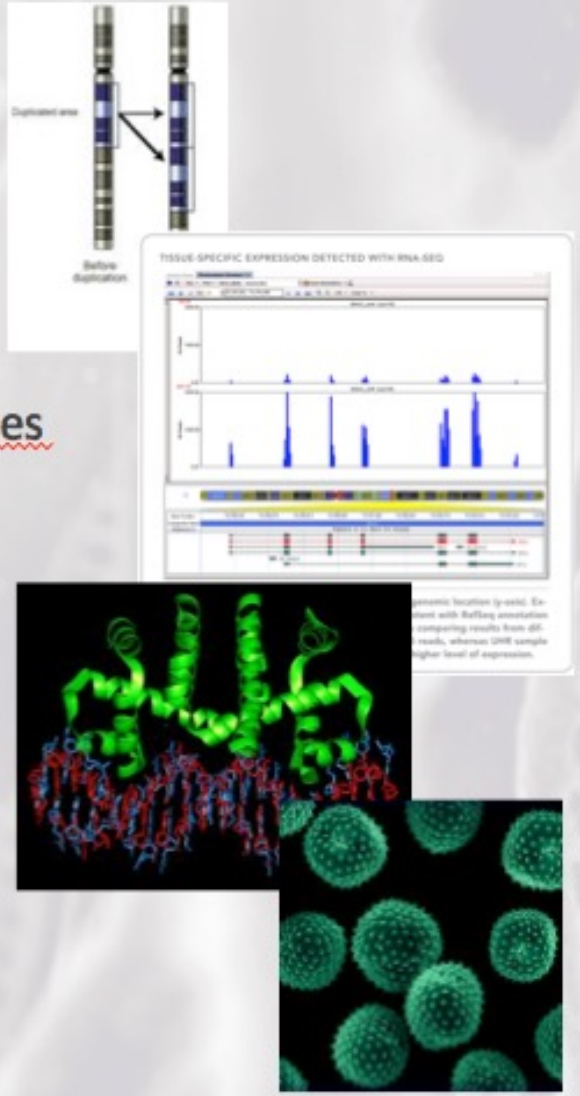
So what kinds of information are available to me through sequencing?

- **Qualitative** information
  - Mutations or changes from a standard reference.

    SNPs, insertions, deletions, duplications, inversions.
    - E.g. Cancer, heritable disorders
  - Pairwise differences. What is it about their genetic makeup makes sample A and sample B different?
    - E.g. Disease resistance, genetic rick factors, morphological differences
  - Validation. Did my breeding, genetic modification, or construct come out as planned?
    - E.g. Genetic engineering, agriculture, synthetic biology, cloning
  - Time series. How is genetic information changing over time?
    - E.g. Evolutionary studies, pathogen monitoring

So what kinds of information are available to me through sequencing?

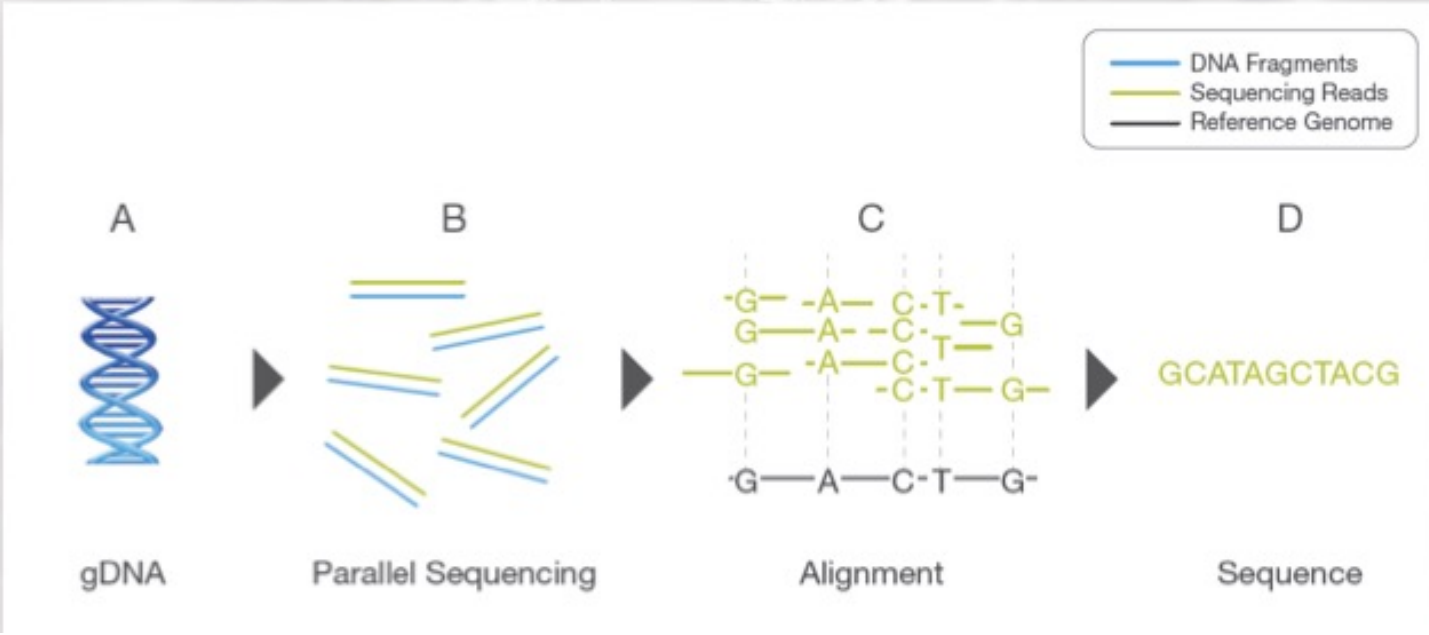
- **Quantitative** information
  - Copy number variation
    - E.g. Reproductive health, genetic engineering
  - Gene expression
    - E.g. Host/pathogen interactions, novel traits, drug response, developmental biology, metatranscriptomes
  - Gene regulation
    - Small RNA "degradome" sequencing, antisense expression
  - Protein/DNA interactions
    - E.g. DNA binding sites, chromatin architecture, regulatory pathways in cell biology
  - Epigenetics
    - E.g. DNA methylation
  - Metagenomics
    - Microbial community profiling, environmental changes



SafeBlood

Next Generation Sequencing

- Basic Principle: Sanger Sequencing
  - ✓ Base recognition of DNA fragments while the fragment is re-synthesized
- Massive Parallel Sequencing
  - ✓ Millions DNA fragments



Our Mission

To improve human health by unlocking the power of the genome

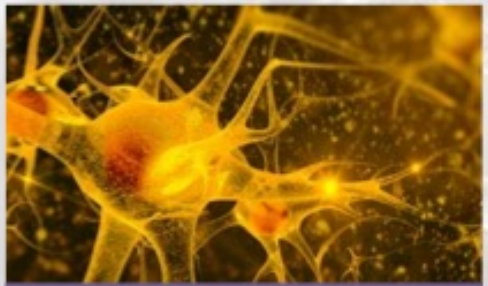
illumina®

Reproductive Health

Oncology

Population Sequencing

Research



Complex Disease

Consumer

Infectious Disease

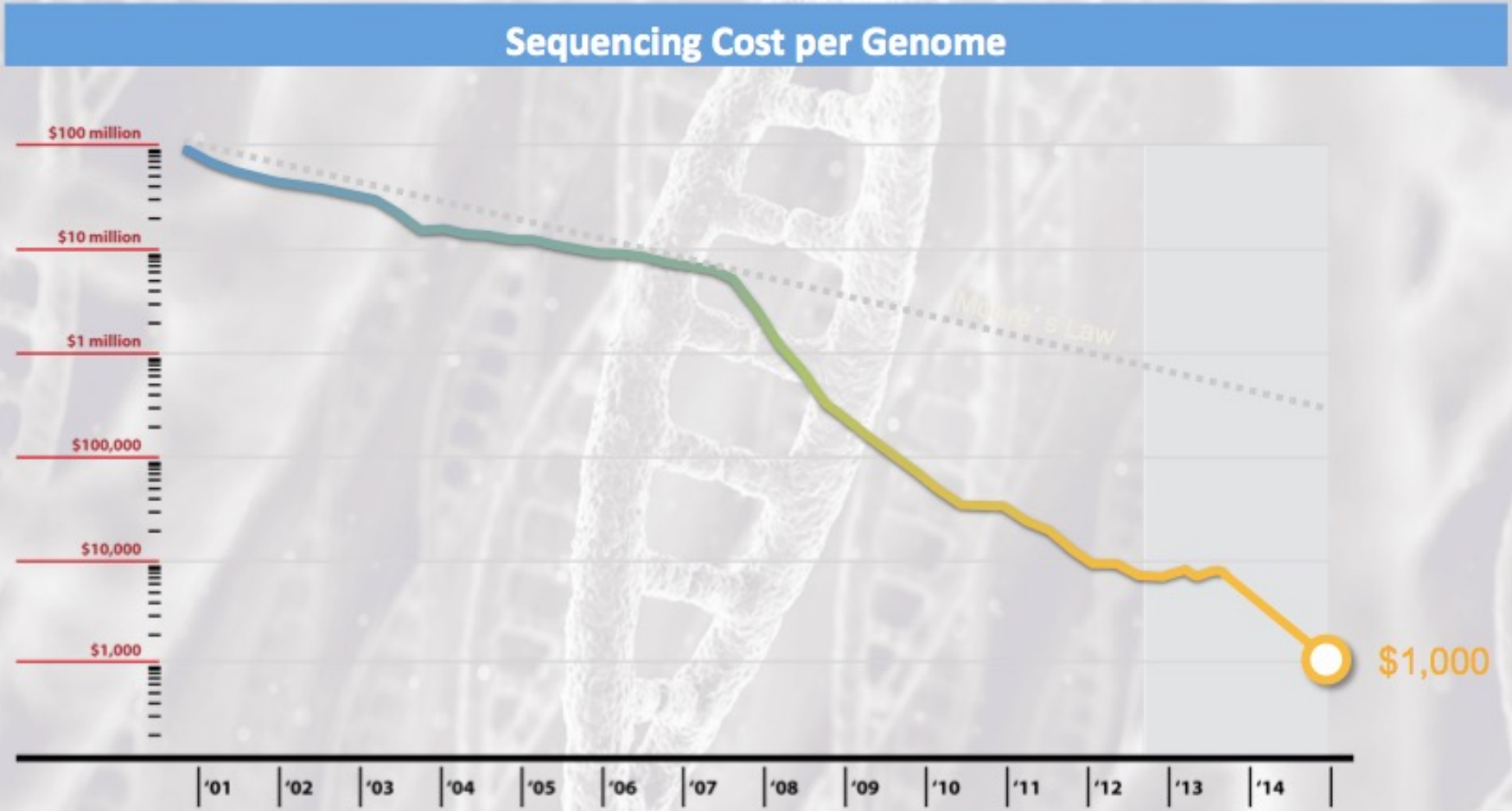
Forensics

Agriculture

Genetic Health

BioPharm

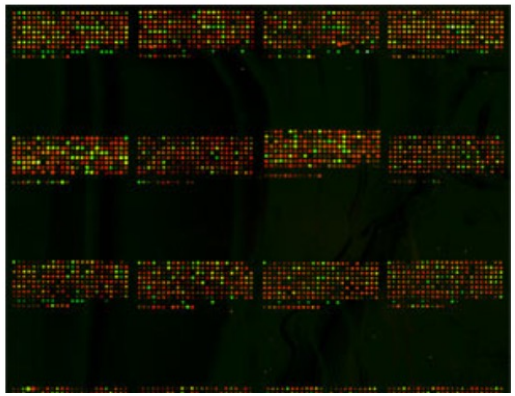
Recent Trends in Sequencing



1. National Human Genome Research Institute. DNA Sequencing Costs http://www.genome.gov/sequencingcosts/ accessed 10/22/2015. Updated October 2, 2015.

Cancer Genomics

Arrays



Parallel Sequencing

```
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```
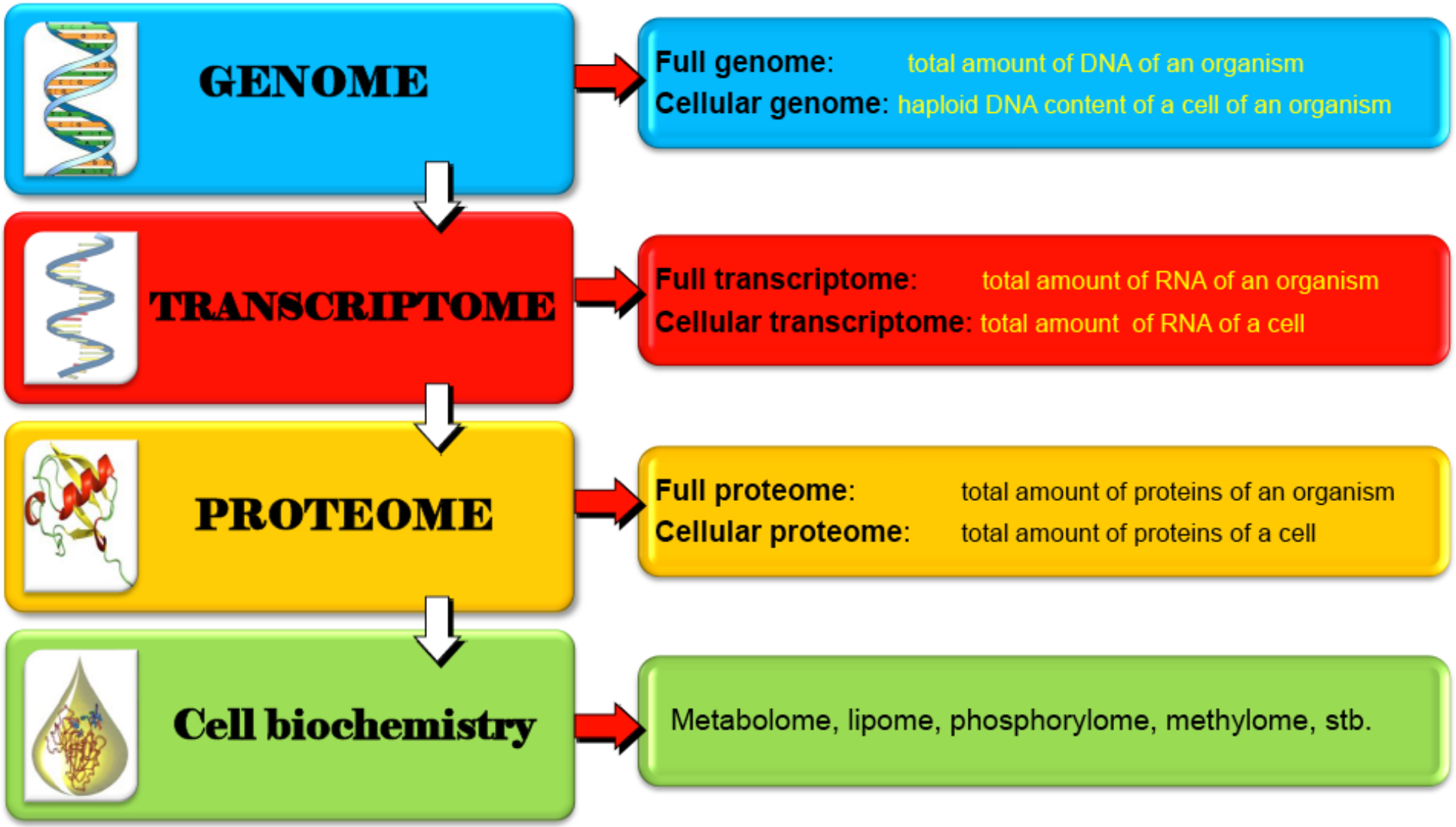
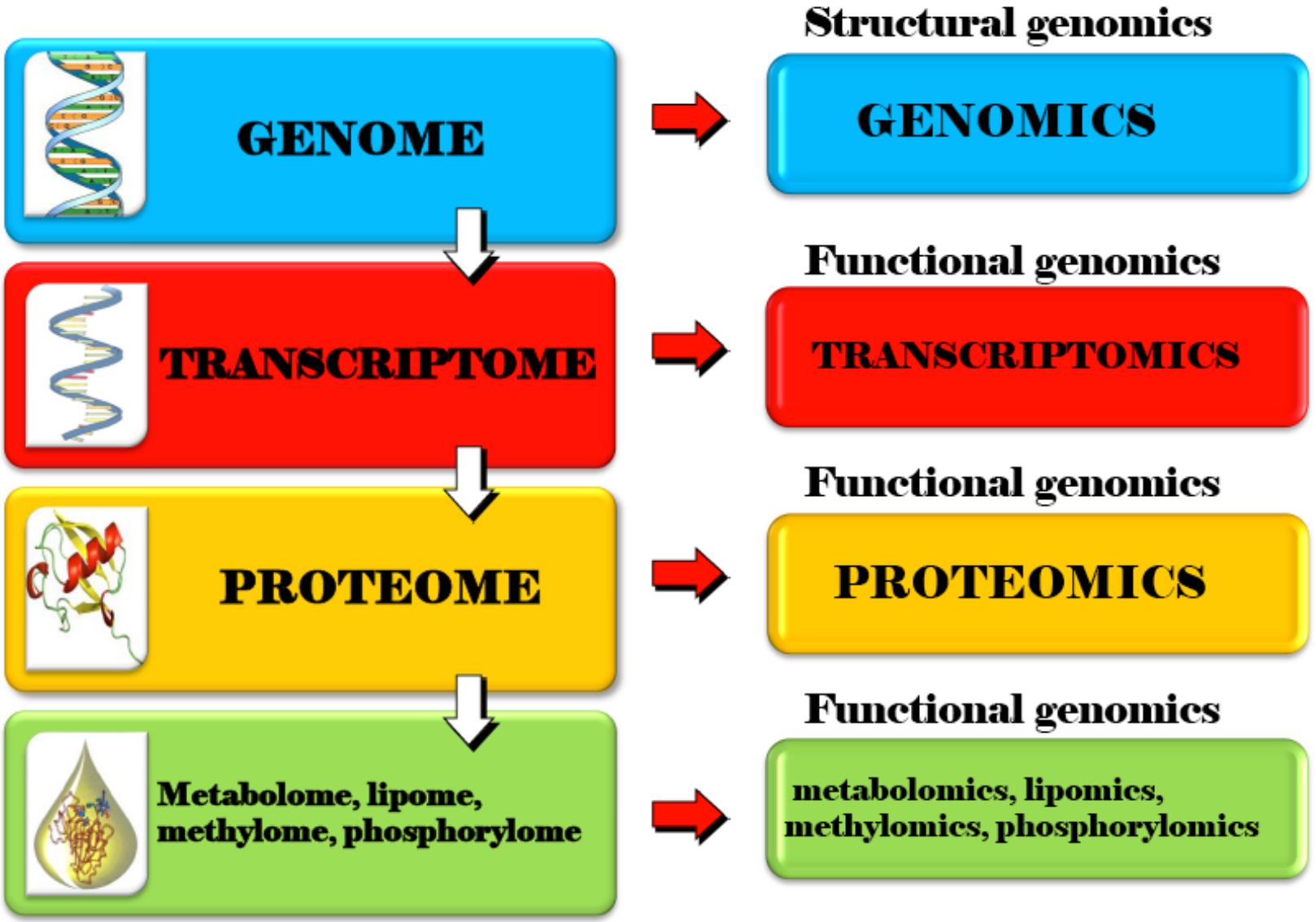

From genome to cell biochemistry 5.



OMICs – version 1,2



Terms/Vocabulary

- **Single Nucleotide Polymorphisms (SNP)**
  - Variation in single base in DNA in germline, most common variants in genome (over 50 million identified)
  - SNP arrays interrogate the entire genome-uses DNA from germ-line (blood)
- **Used in Genome Wide Association Studies (GWAS)**
  - Typically uses SNP arrays to compare populations (with disease or not)
  - Determines risk or susceptibility to some state

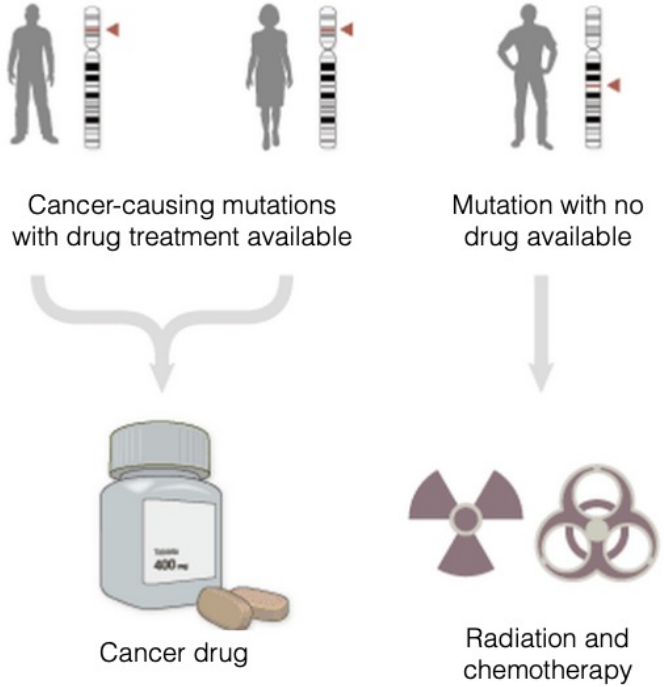
Terms (con't)

- **RNA expression profiles-determines global messenger RNA expression in a sample-using hybridization of mRNA to a Chip**
- **Methylation arrarys-determines global methylation of the genome-an epigenetic change typically inserts a methyl group at CpG islands in DNA and alters transcription-using hybridization of DNA to a Chip**
- **Massively parallel sequencing-allows for rapid sequencing of entire exome (WES) on whole genome (WGS) or cDNA (RNA-seq)**

Translating the Cancer Genome

Treatment of Cancer

Targeted Cancer Therapy

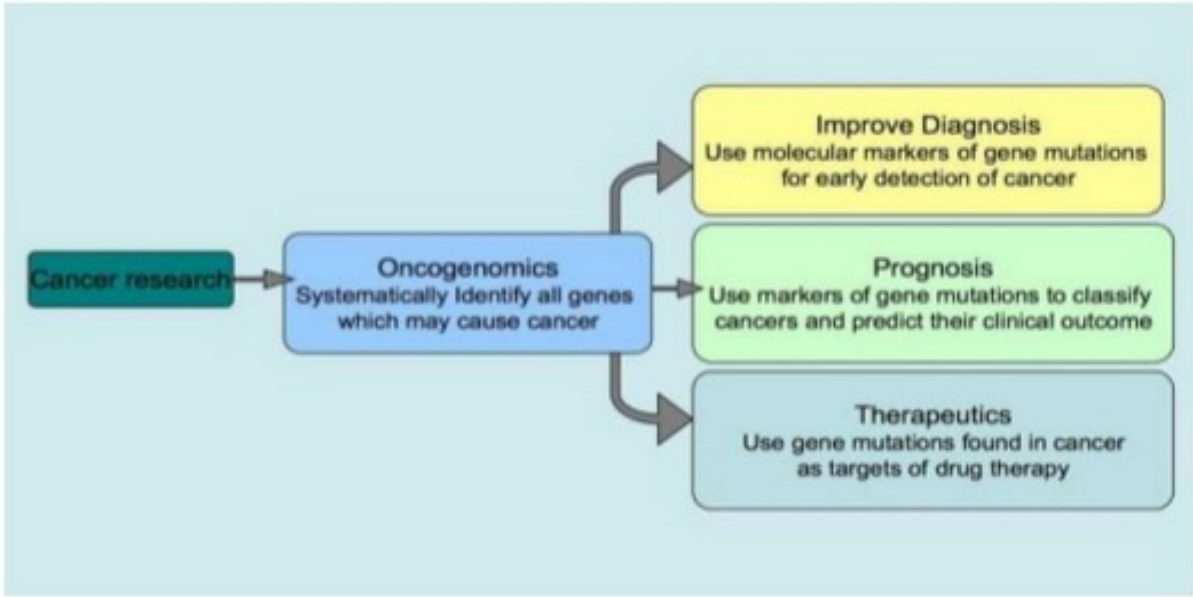


Schema adapted from NY times

Evolution of Treatments for Breast Cancer

Strategy	Examples
Remove or destroy cancerous tissue	Surgery, radiation, chemotherapy
Use phenotype to select drug	Estrogen-receptor-positive women take Tamoxifen
Use genotype to select drug	*Her-2/neu*-positive cancers take Herceptin (monoclonal antibody)
Genomic level	Gene expression profile on DNA microarray used to guide drug choice

Goal of Oncogenomics

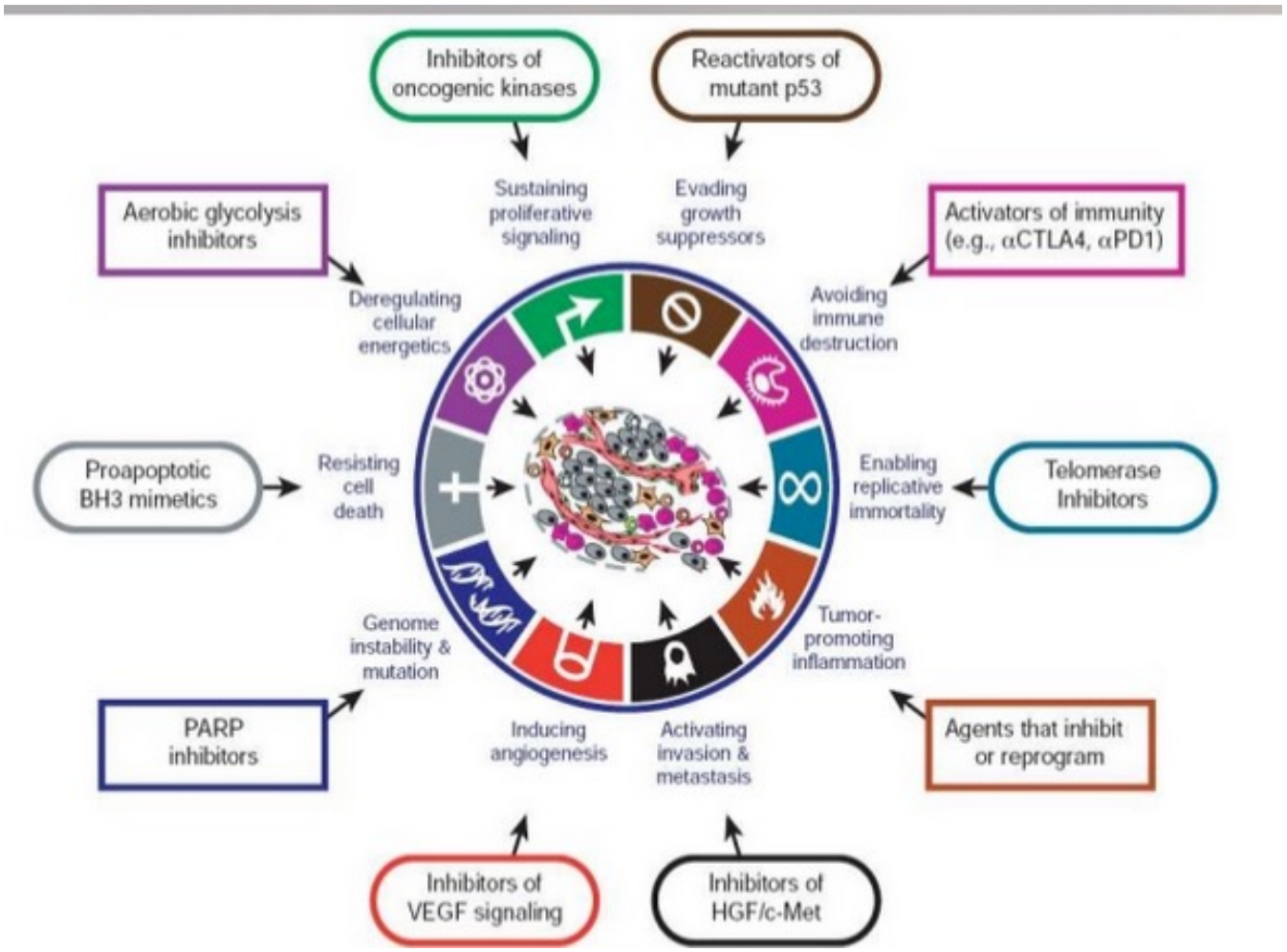


Cancer Genomics: What for?

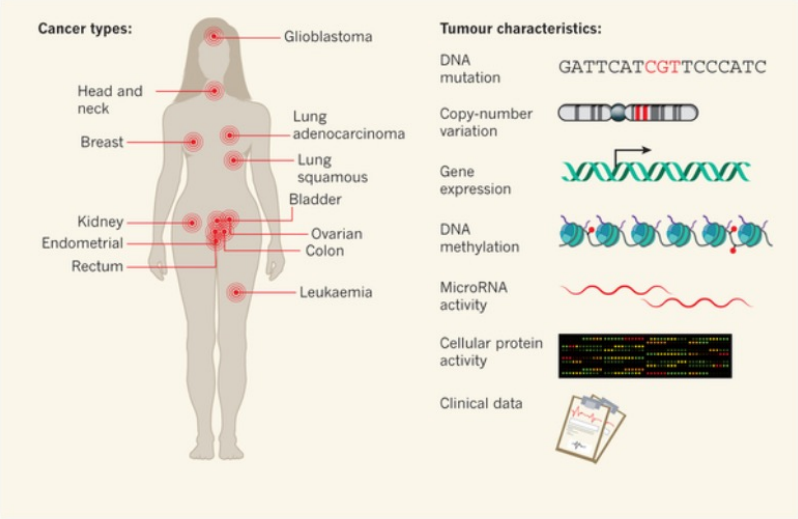
- Finding new cancer genes (cancer drivers)
- Finding new therapeutic targets
- Identify molecular signatures to stratify tumors
- Move towards personalized cancer treatment

The Rational Treatment of Cancer

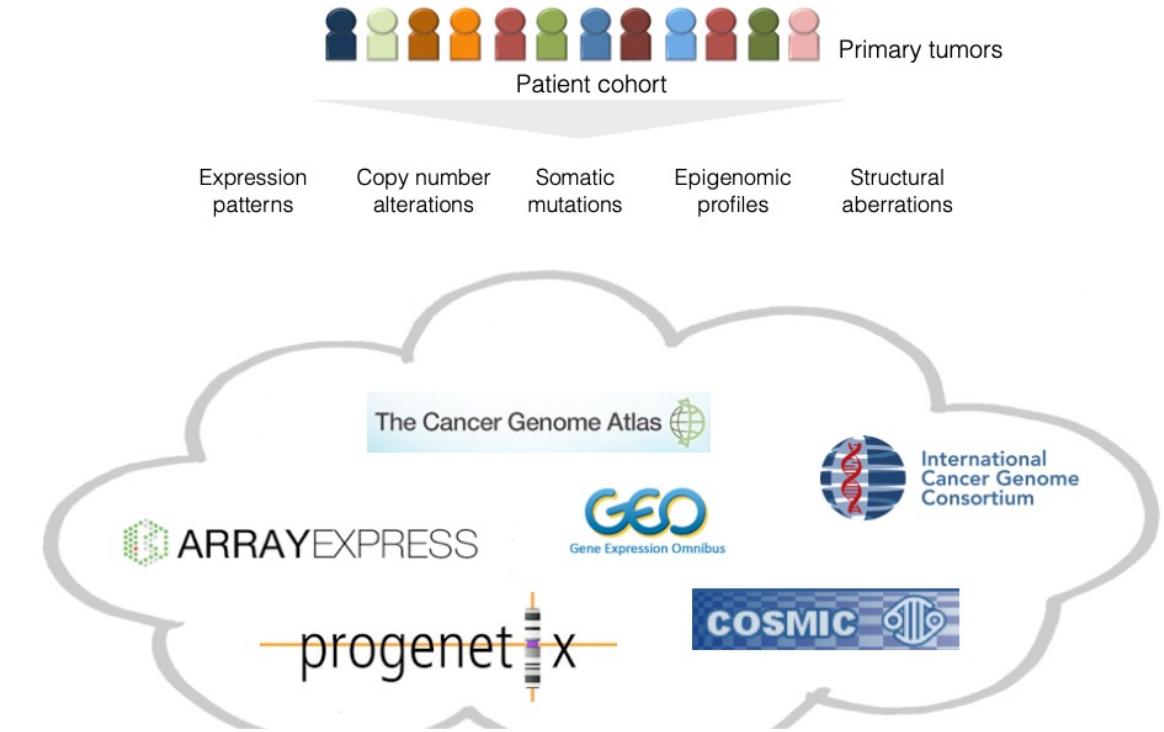

Moving towards personalized
cancer medicine


Inhibitors of oncogenic kinases
Reactivators of mutant p53
Sustaining proliferative signaling
Evading growth suppressors
Aerobic glycolysis inhibitors
Activators of immunity (e.g., αCTLA4, αPD1)
Deregulating cellular energetics
Avoiding immune destruction
Proapoptotic BH3 mimetics
Resisting cell death
Enabling replicative immortality
Telomerase Inhibitors
Genome instability & mutation
Tumor-promoting inflammation
PARP inhibitors
Inducing angiogenesis
Activating invasion & metastasis
Agents that inhibit or reprogram
Inhibitors of VEGF signaling
Inhibitors of HGF/c-Met

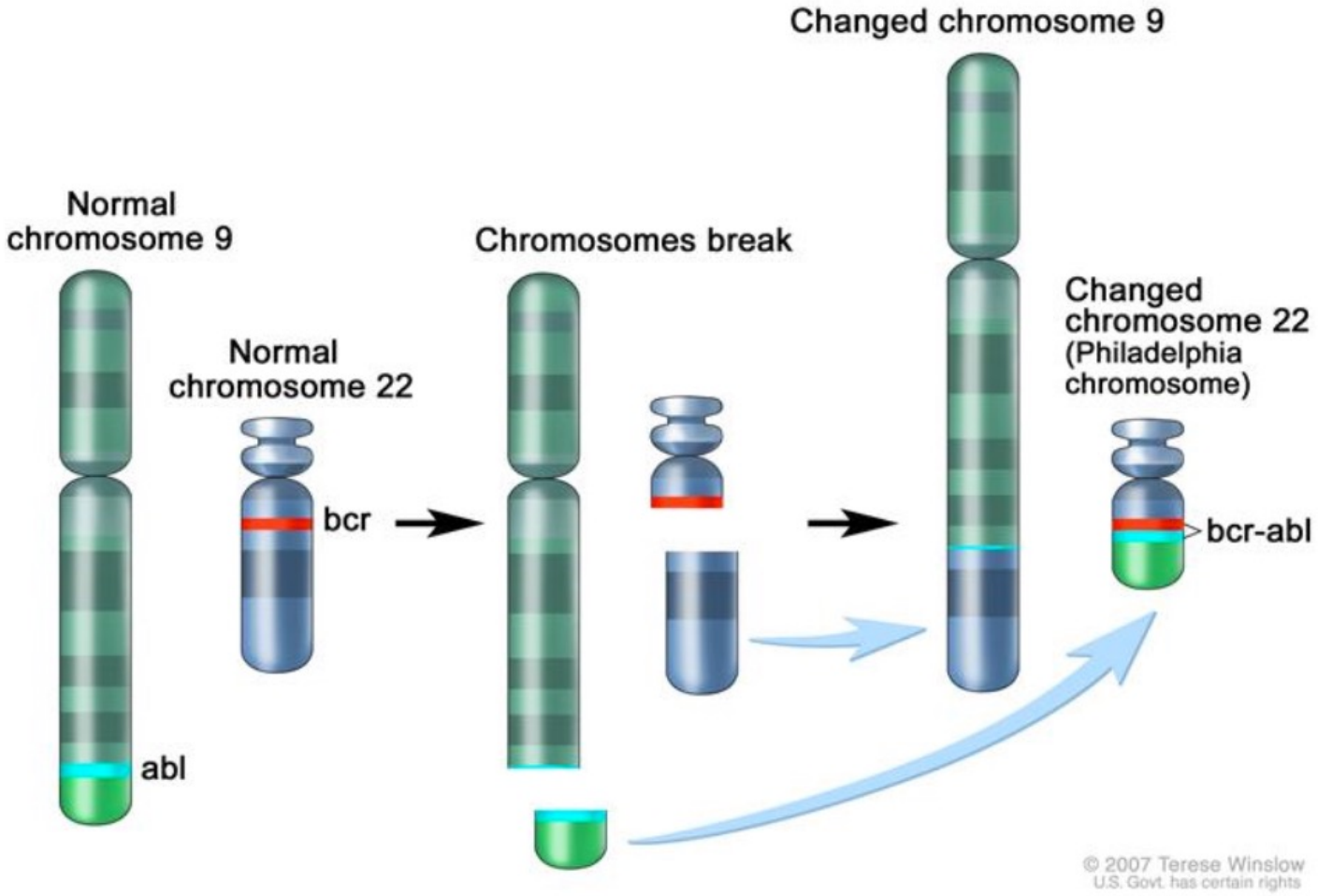
Cancer Genomics Projects



Cancer Genomics Projects

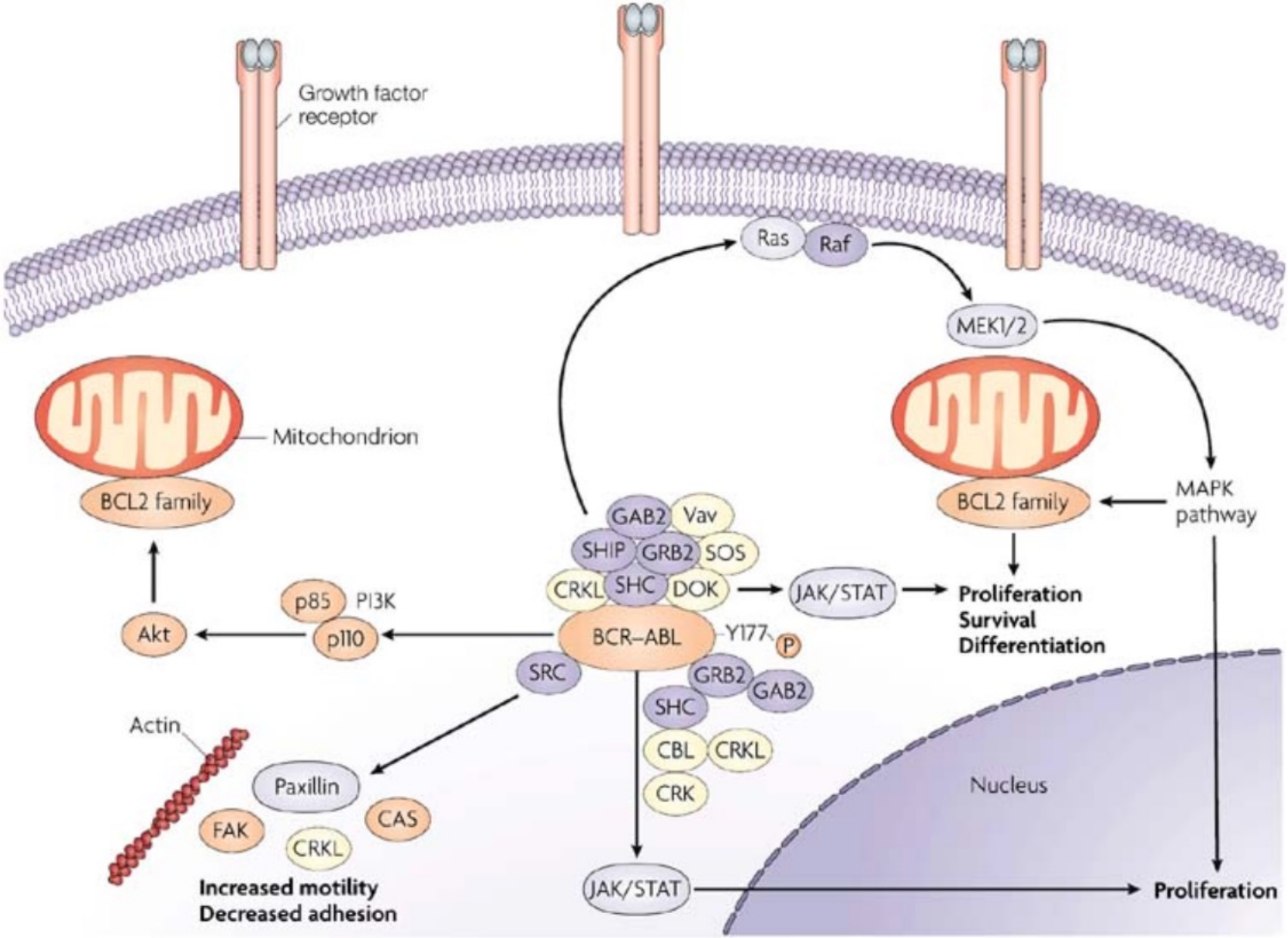


BCR-ABL fusion cause Chronic Myelogenous Leukemia (CML)



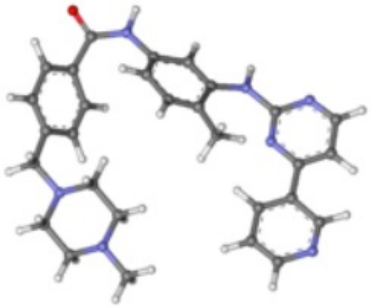
1985

BCR-ABL: constitutive active ABL kinase activity

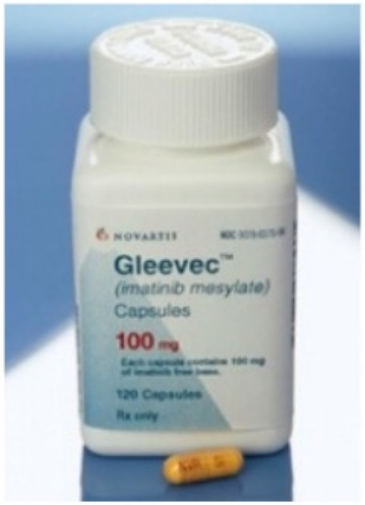


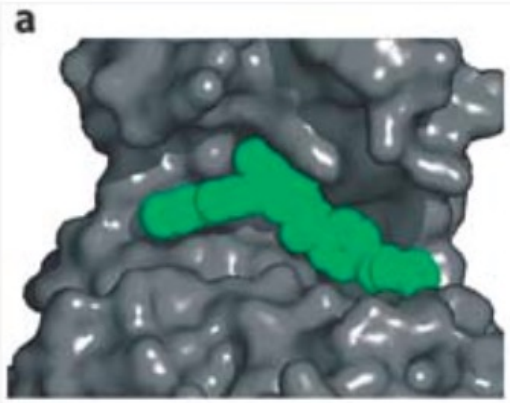
Weisberg et al., Nature Reviews Cancer 2007

Imatinib inhibits tyrosine-kinase activity of ABL

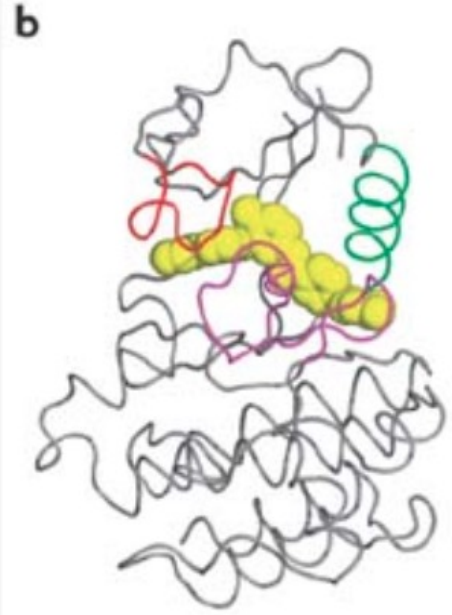


Imatinib



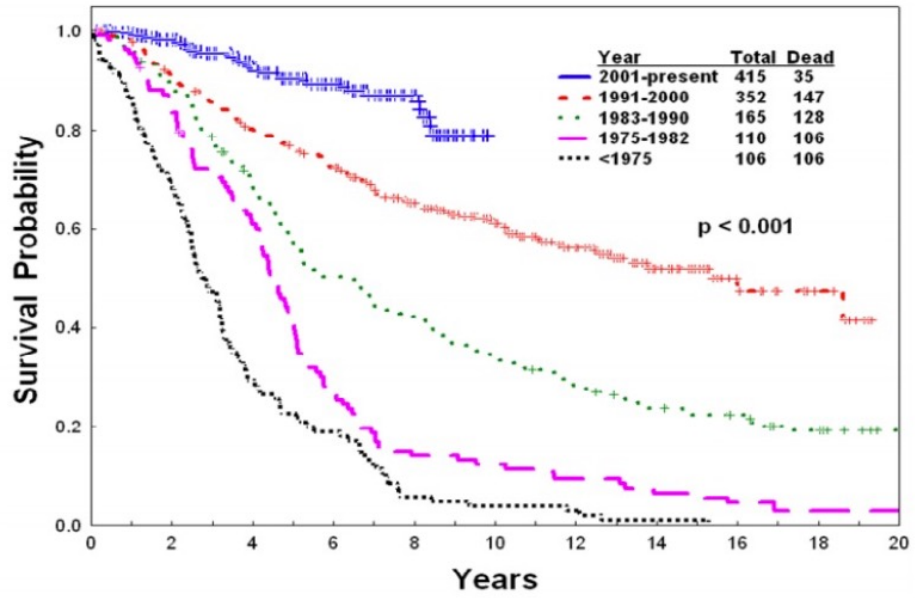


Imatinib

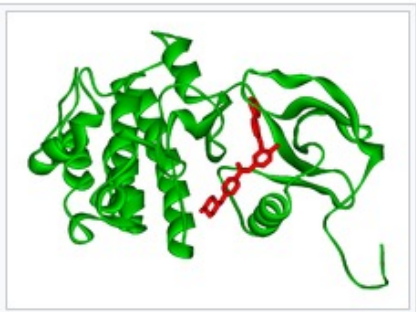


Imatinib

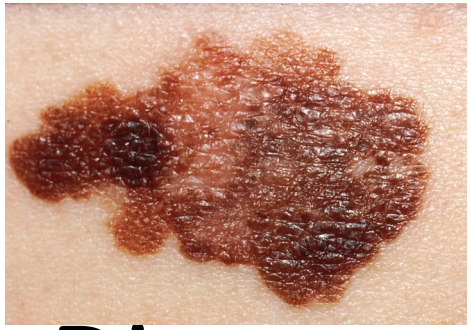
Dramatically improved long term survival rates (95.2%) since the introduction of Gleevec in 2001

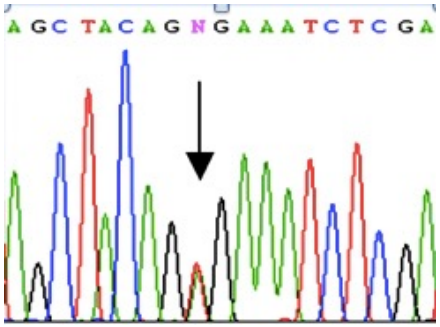


Kantarjian et al., Blood 2012



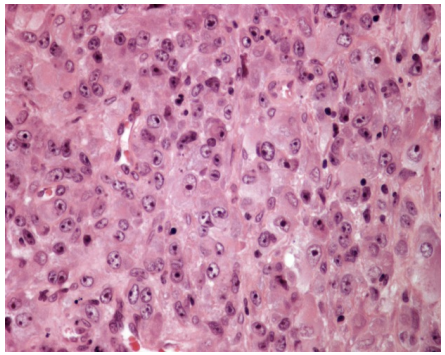
bcr-abl kinase (green), which causes CML, inhibited by imatinib (red; small molecule).

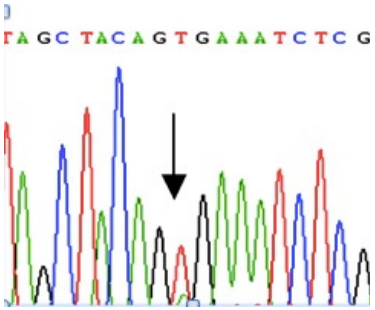


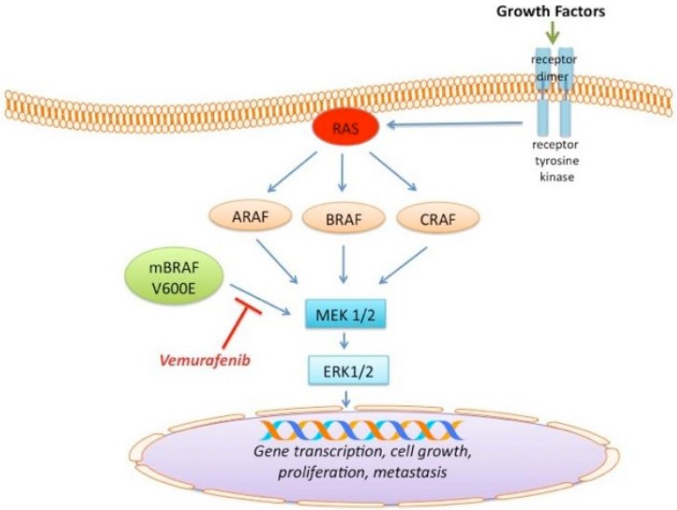


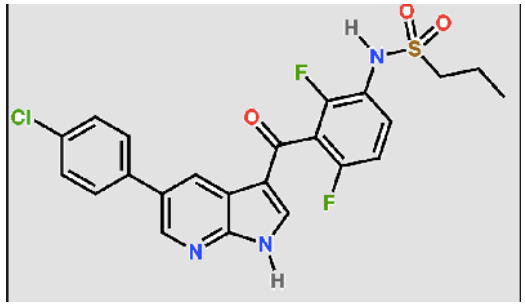
Disease

Pathogenesis



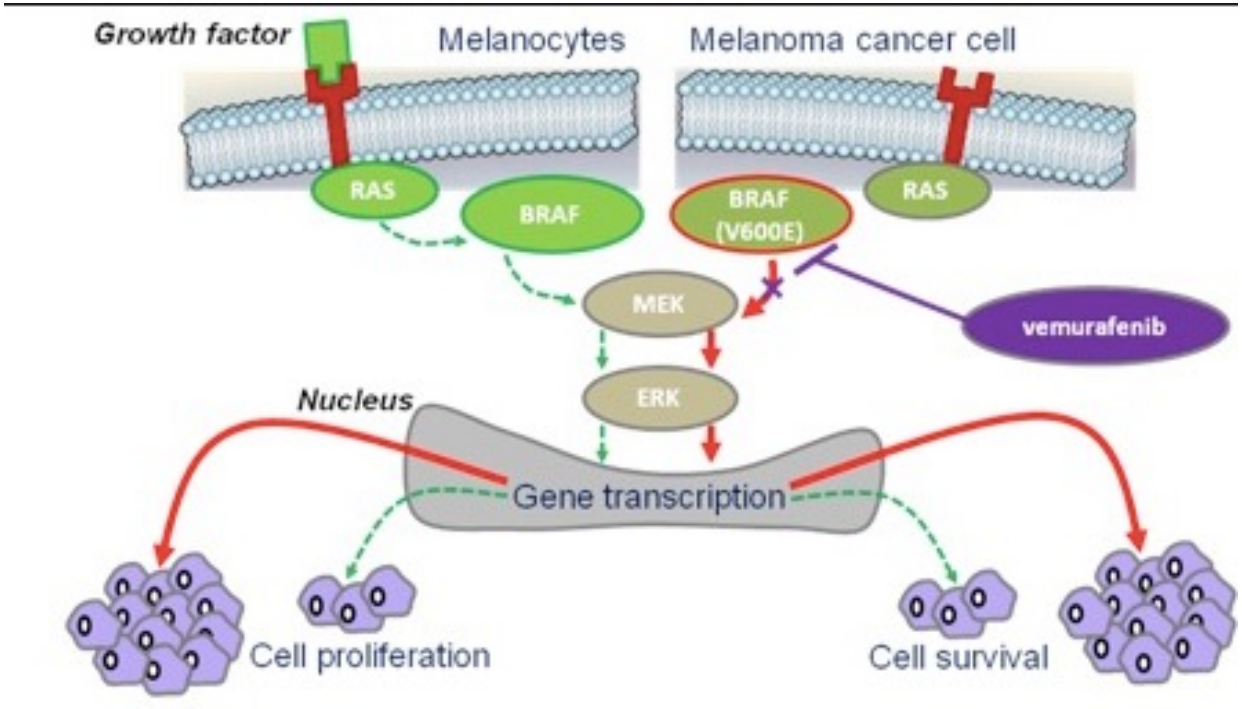




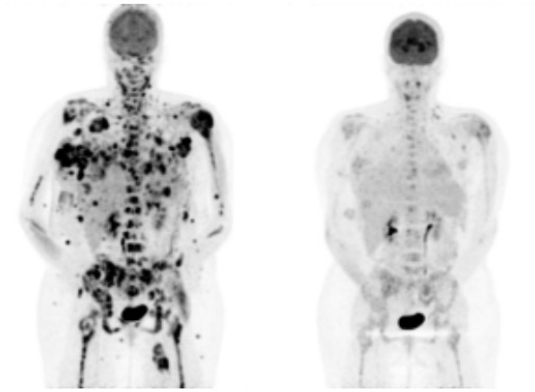


Vemurafenib

Treatment



2 setmanes
Vemurafenib

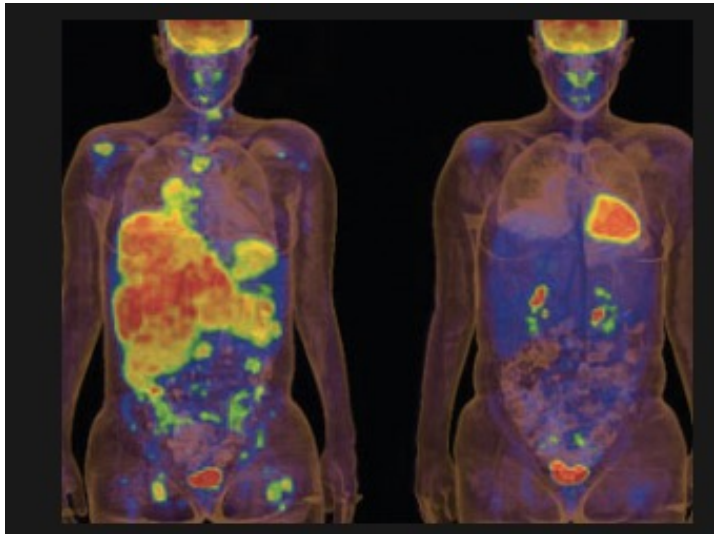


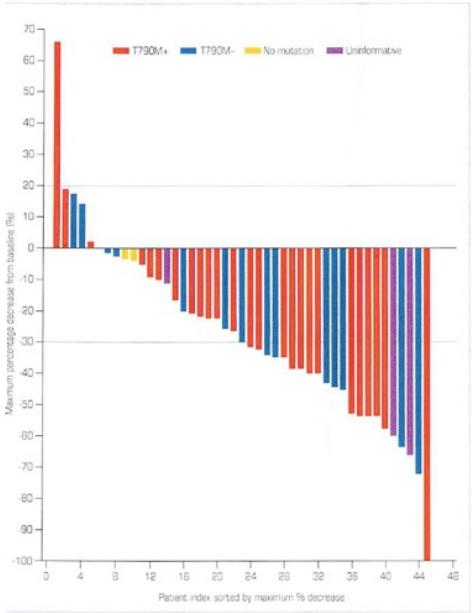
Vemurafenib

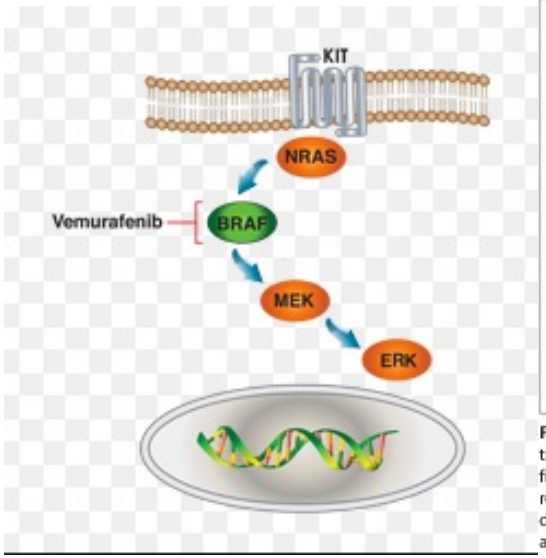
Personalized medicine / Precision medicine

Promise of Personalized Medicine

Vemurafenib







Crizotinib

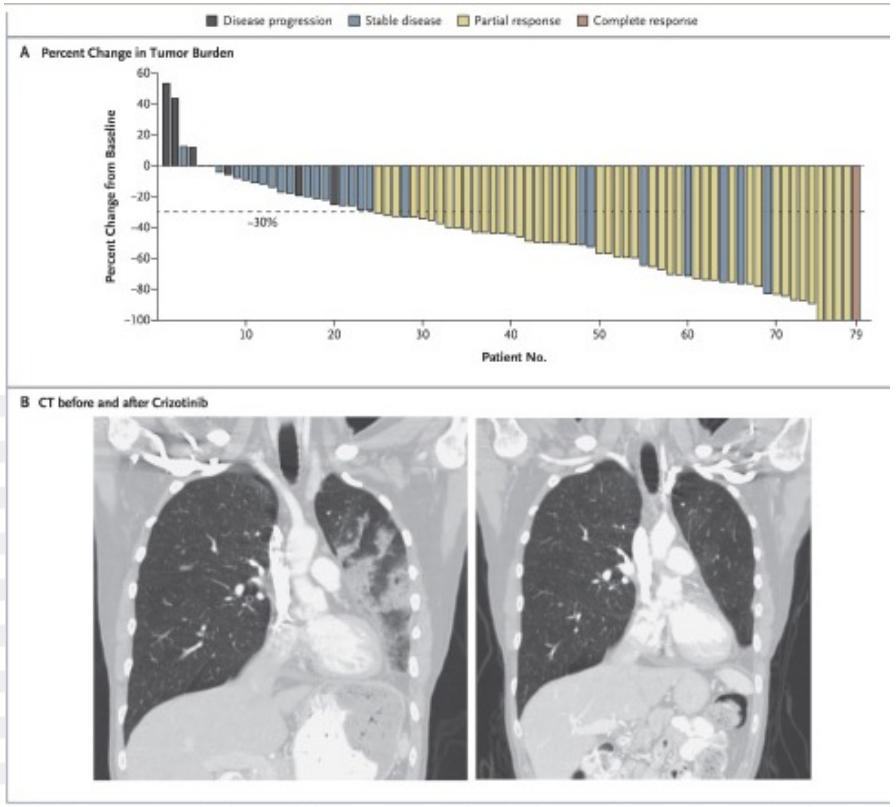


Fig. 1: Response to ALK Inhibition—**(A)** Best response of patients with *ALK*-positive tumors who were treated with crizo tinib, as compared with pretreatment baseline. Numbers along the x axis indicate arbitrarily assigned subject number from 1 to 79. The bars indicate the percent change in tumor burden from baseline. **(B)** The results of CT with corona reconstruction in a representative patient at baseline (left) and after two cycles of therapy (right). This patient had ur dergone previous left lower lobectomy. ©Massachusetts Medical Society. Reprinted with permission from Kwak EL, e al: N Engl J Med 363:1693-1703. 2010.

Example of Vemurafenib

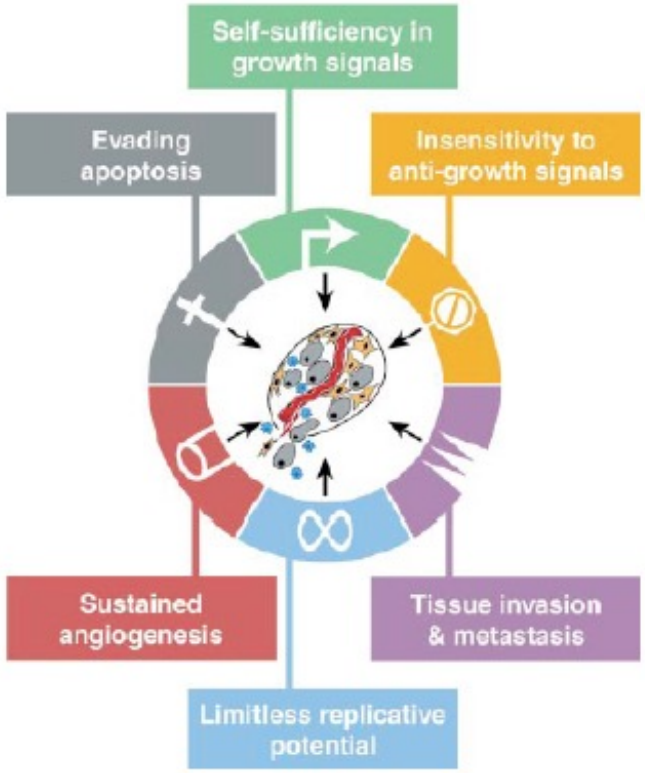
- 50-60% of melanoma patients have driver mutations in BRAF (V600E)
- At doses of vemurafenib that inhibit 90% of B-RAF activity, most patients respond rapidly with tumor shrinkage
- Median duration of response is less than 12 months due to resistance
- What are the mechanisms of resistance?

Example-BRAF (V600E) mutations in colon cancer

- ***Unresponsiveness of colon cancer to BRAF (V600E) inhibtion through feedback activation of EGFR*** ***Prahallad A, et al. Nature Jan 26 2012***
- **Mechanism-appears to be inhibition of BRAF leads to inhibition of MEK and ERK, leading to reduced phosphatase activity of CDC25C, leading to reduced dephosphorylation of EGFR, leading to increased activation and EGFR signaling**

Defining Biologically Relevant Molecular Modules

Hallmarks of breast cancer



Proliferation genes (GGI)

Immune Response

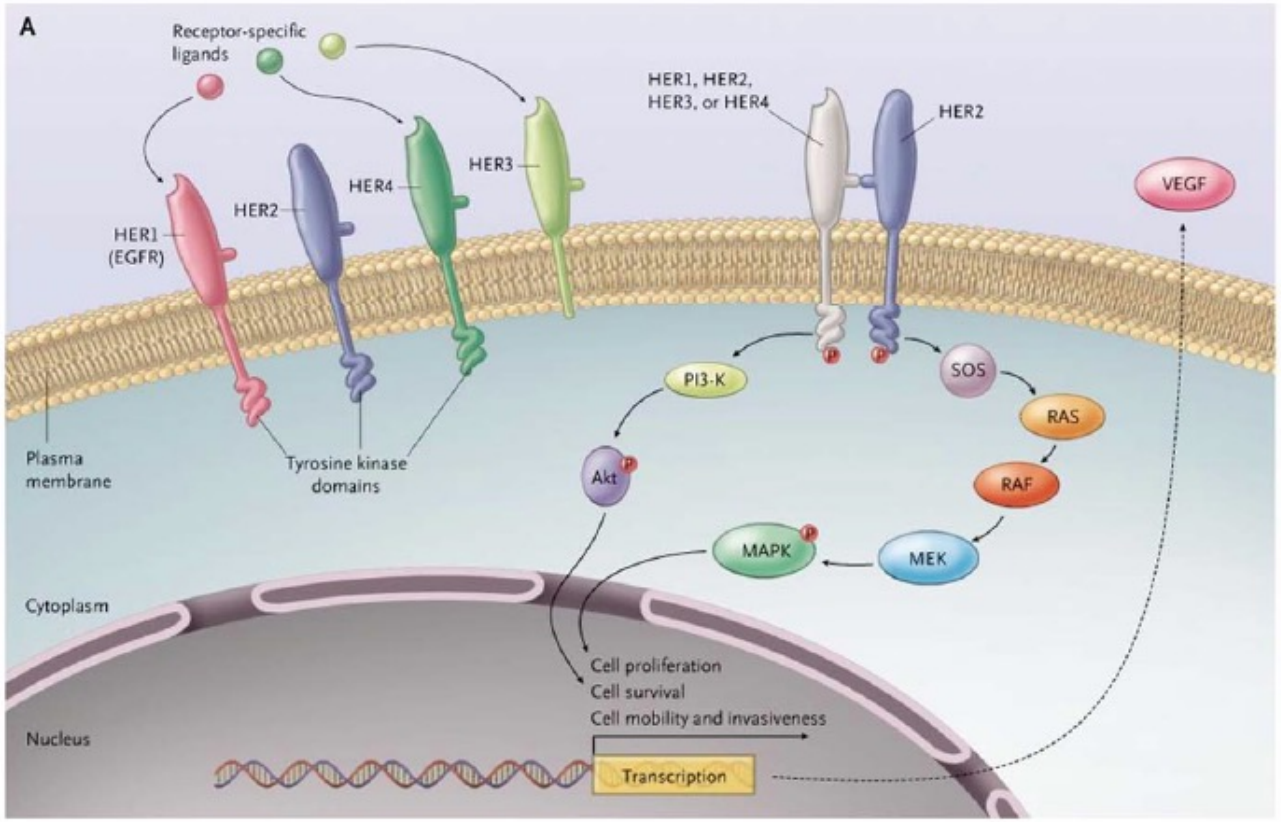
HER-2 signaling

ER signaling

Invasion

Hanahan D, Weinberg R, Cell 2000

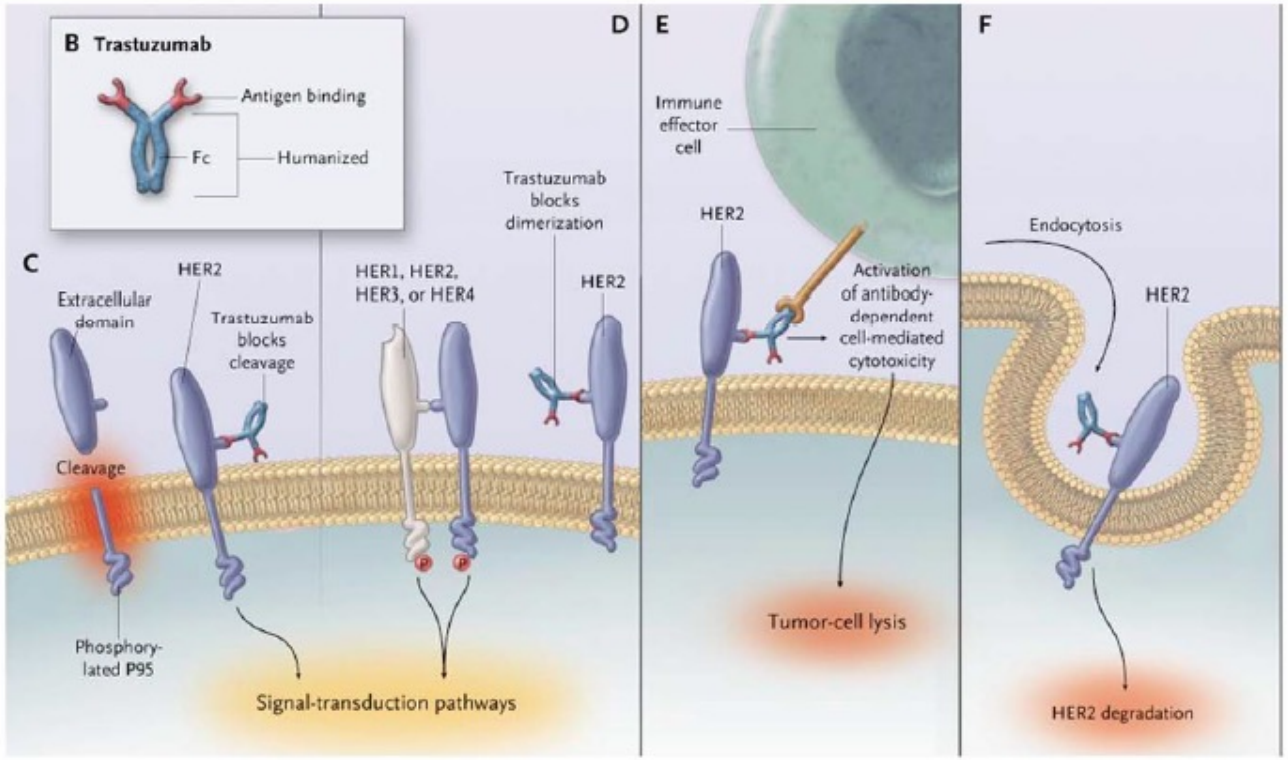
Her2 in breast cancer



Hudis, N Engl J Med 357:39, 2007

One example is measurement of Her2 in breast cancer. Her2 is a cell surface receptor that is involved in cell proliferation and survival. Certain tumors express high levels of Her2, while others do not.

Trastuzumab as a threrapy for Her2+ breast cancer



Hudis, N Engl J Med 357:39, 2007

An antibody that binds to Her2 is used to target therapy to breast cancer cells that express Her2.

HER-2 Protein and Herceptin

- Herceptin (trastuzumab):
  - Metastatic breast cancer
  - Targets tumor cells that over-express the human epidermal growth factor receptor 2 (HER2) protein
  - Best response attained in women who over-express the HER2 protein
  - HER-2 over-expression in breast cancer cells should be done before patients receive the drug


Made-to-Order
Medicine

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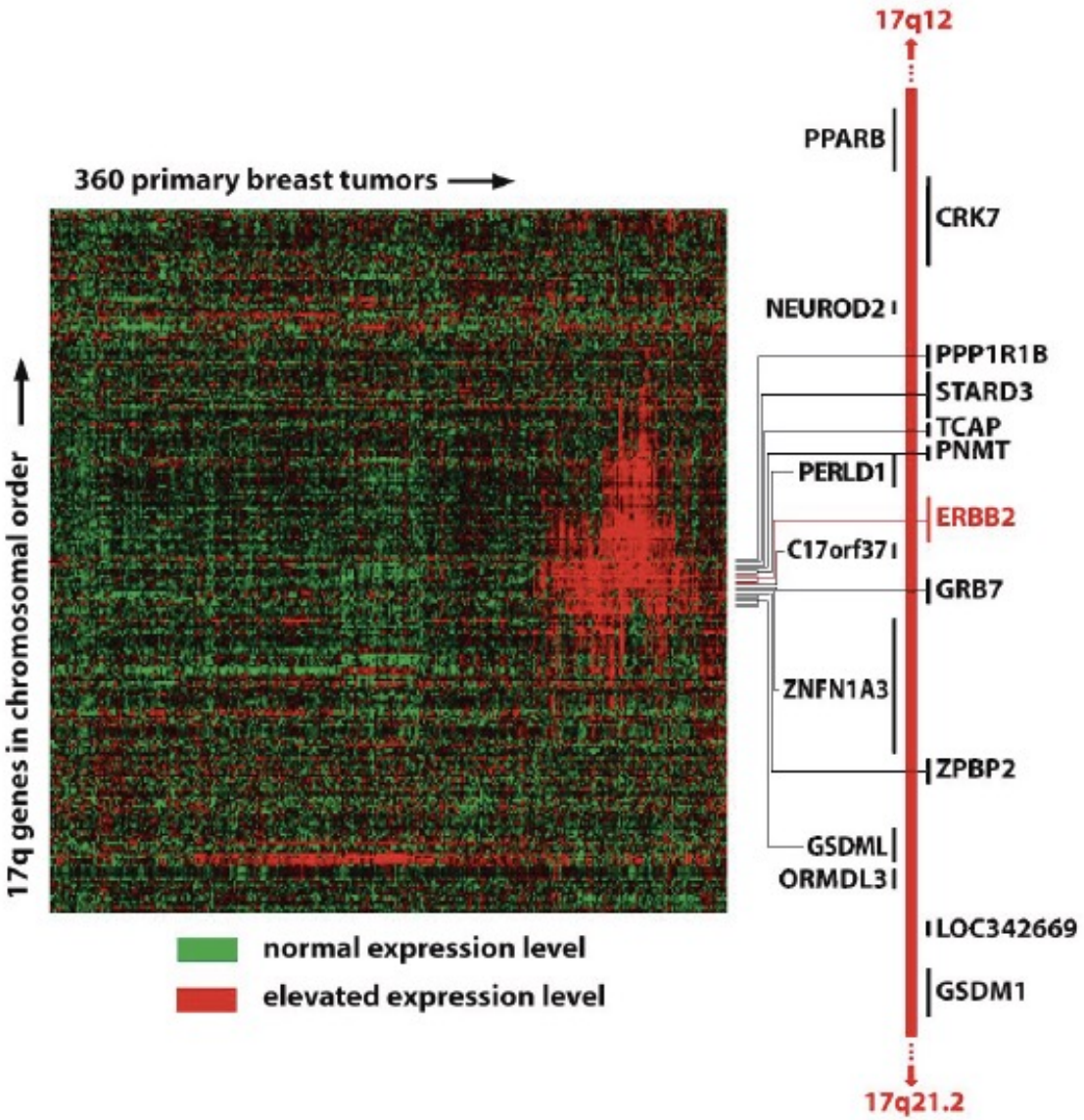
Trastuzumab
HERCEPTIN®


17q12
PPARB
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PPP1R1B
STARD3
TCAP
PNMT
PERLD1
ERBB2
C17orf37
GRB7
ZNFN1A3
ZPBP2
GSDML
ORMDL3
LOC342669
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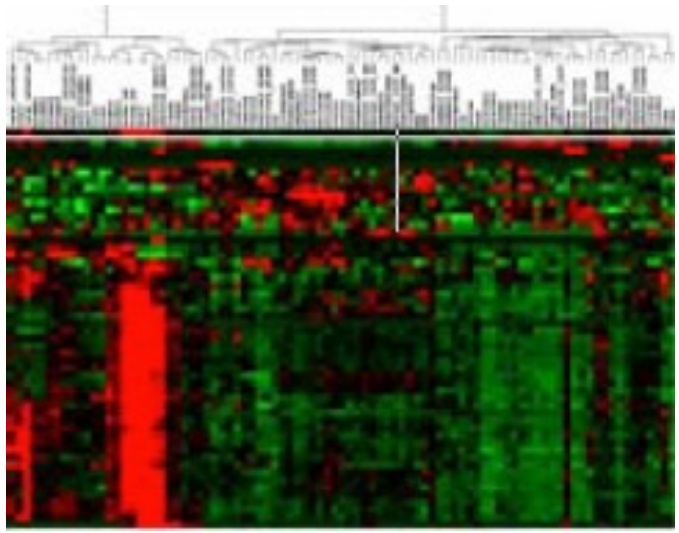
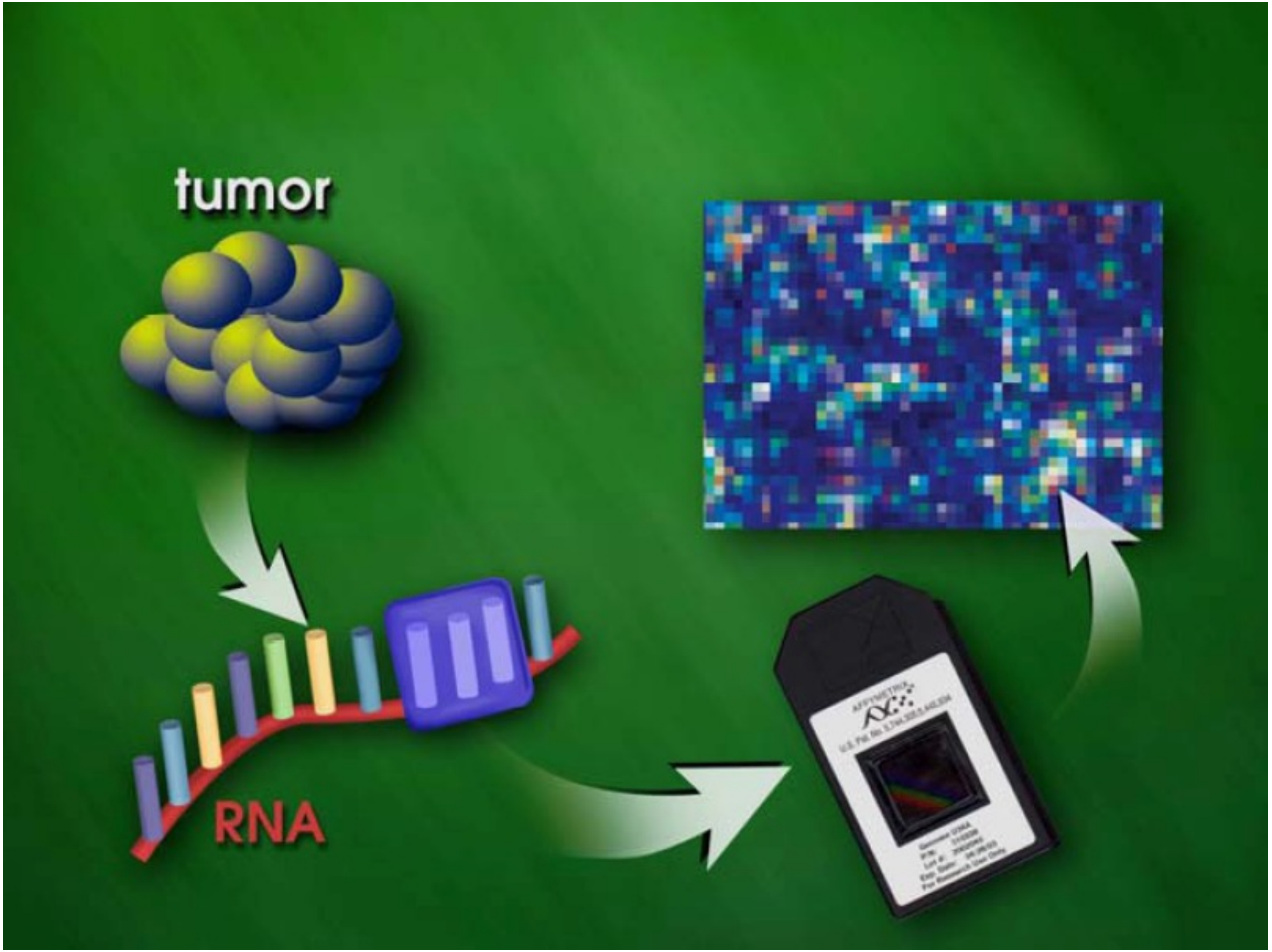
17q21.2
360 primary breast tumors
17q genes in chromosomal order
normal expression level
elevated expression level

Cellular Signatures




tumor
RNA

Leukemias

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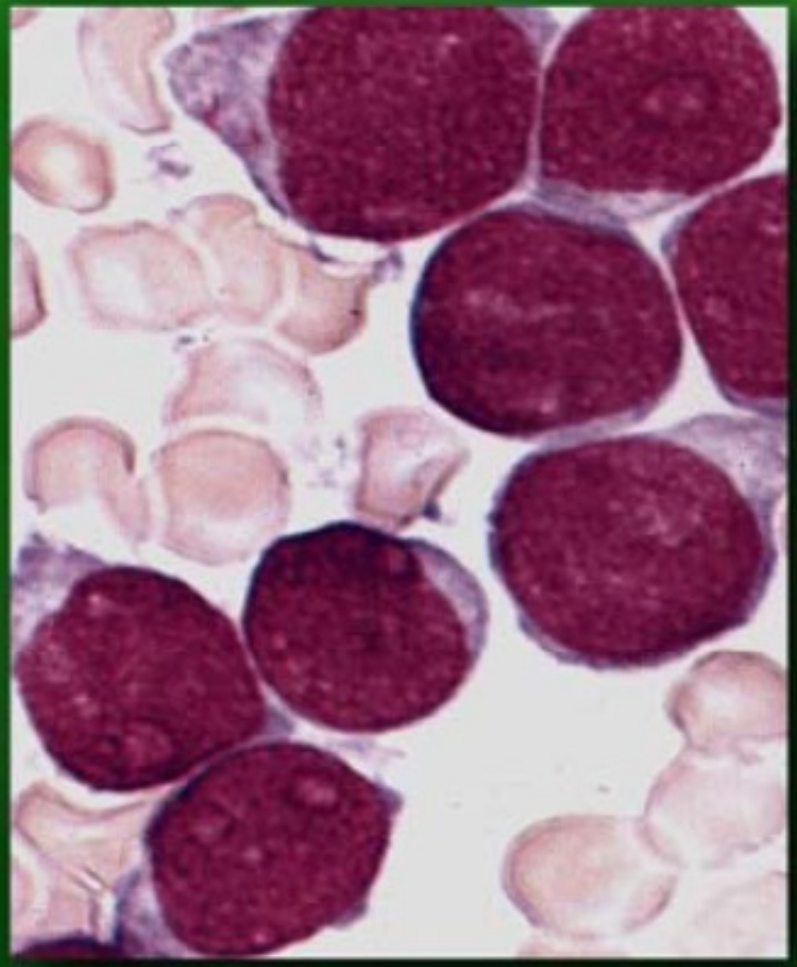

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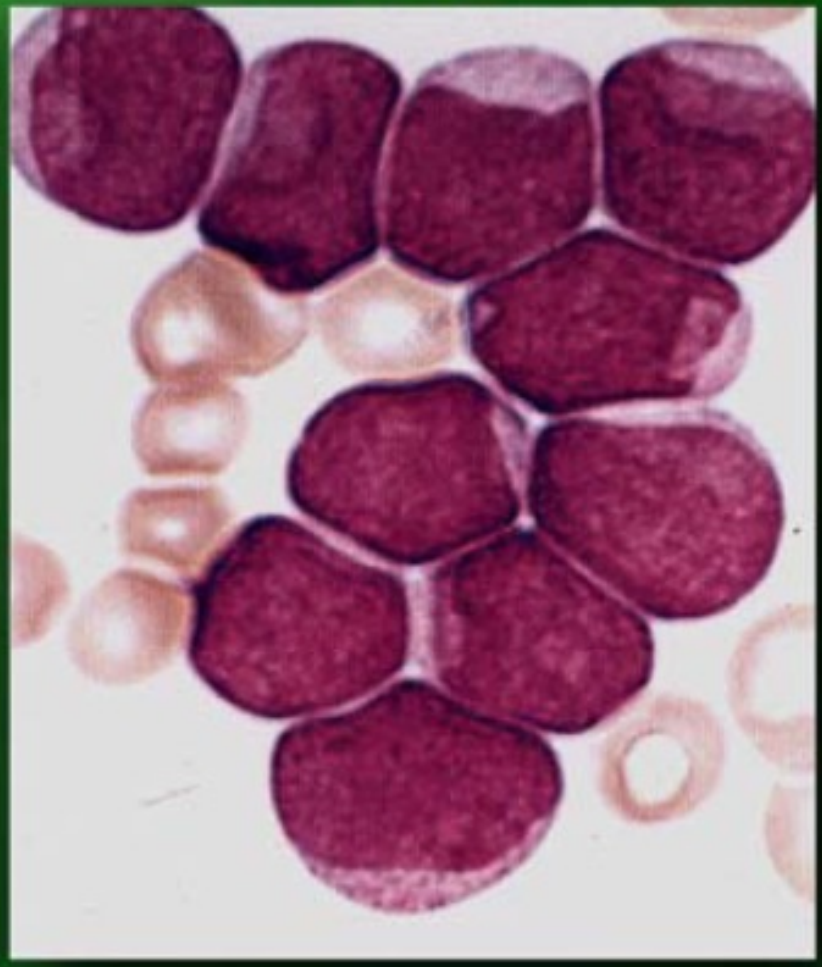

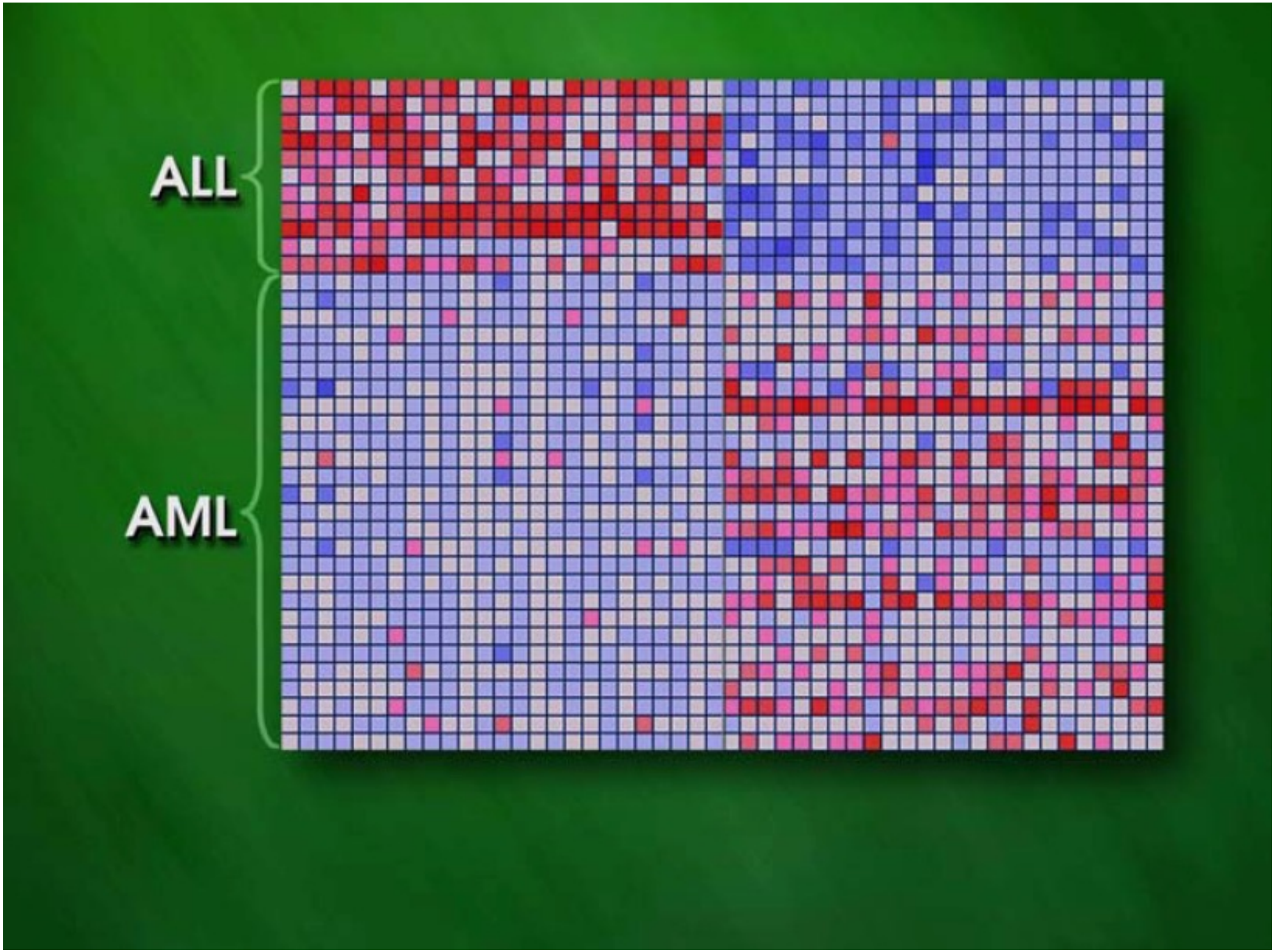
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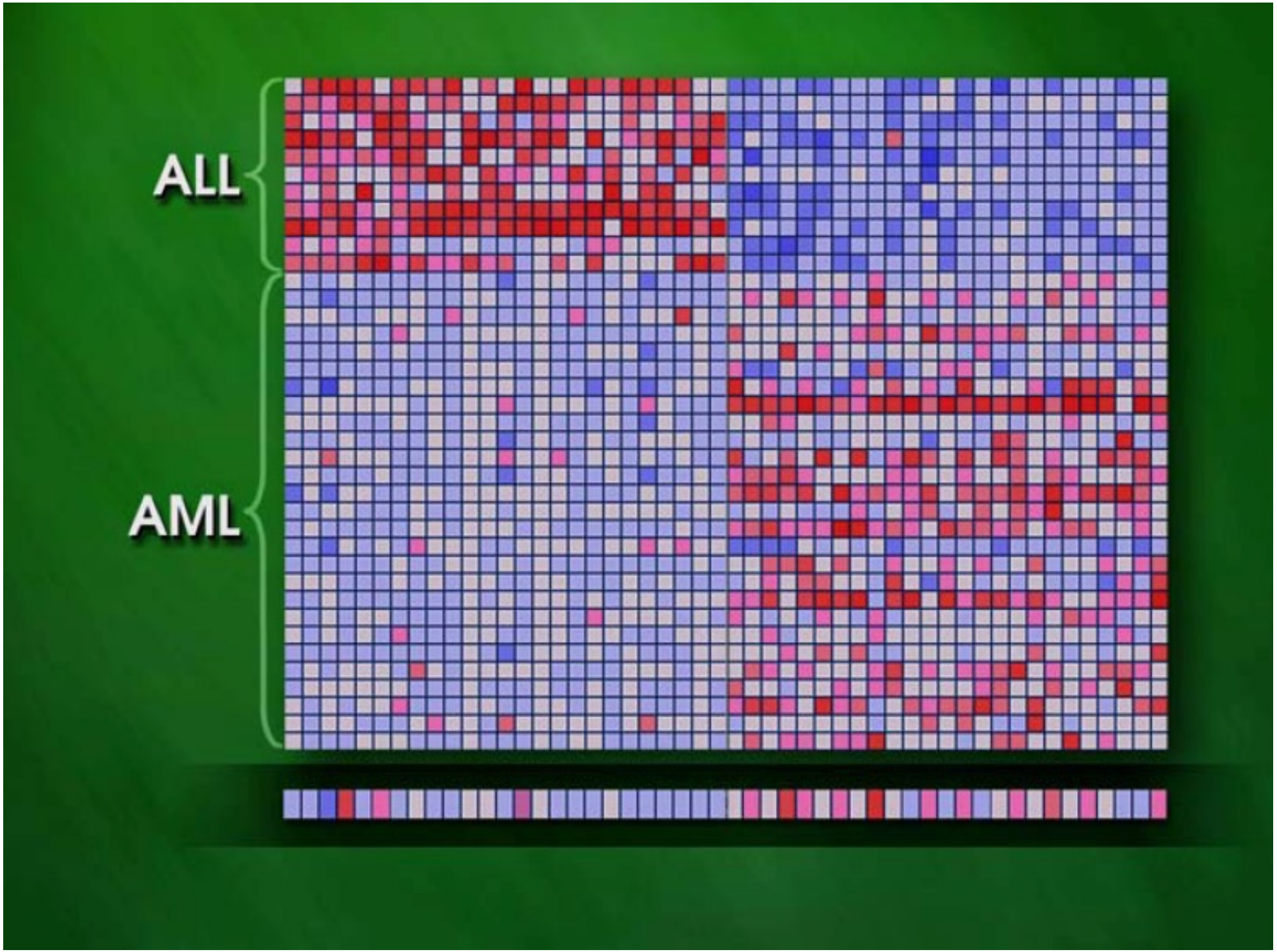
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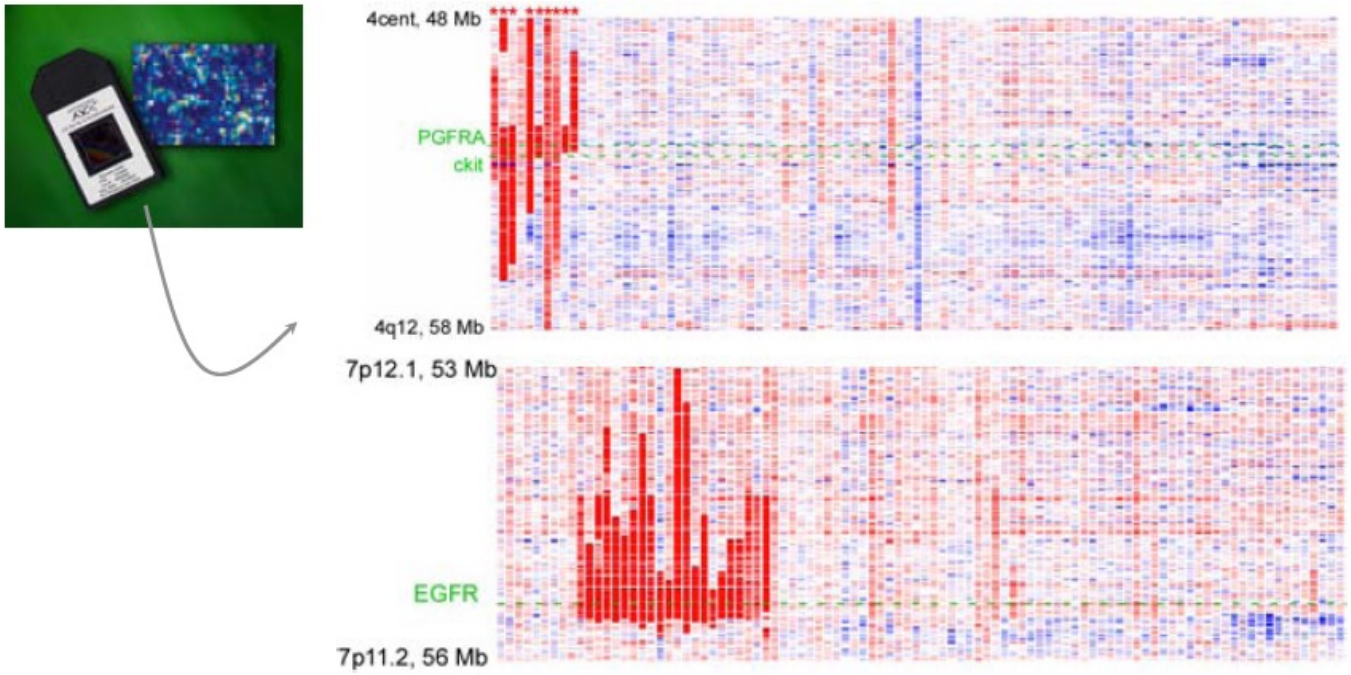
Cancer Genomes are different-context is important for developing novel molecularly targeted therapies

Genome deletion and amplification

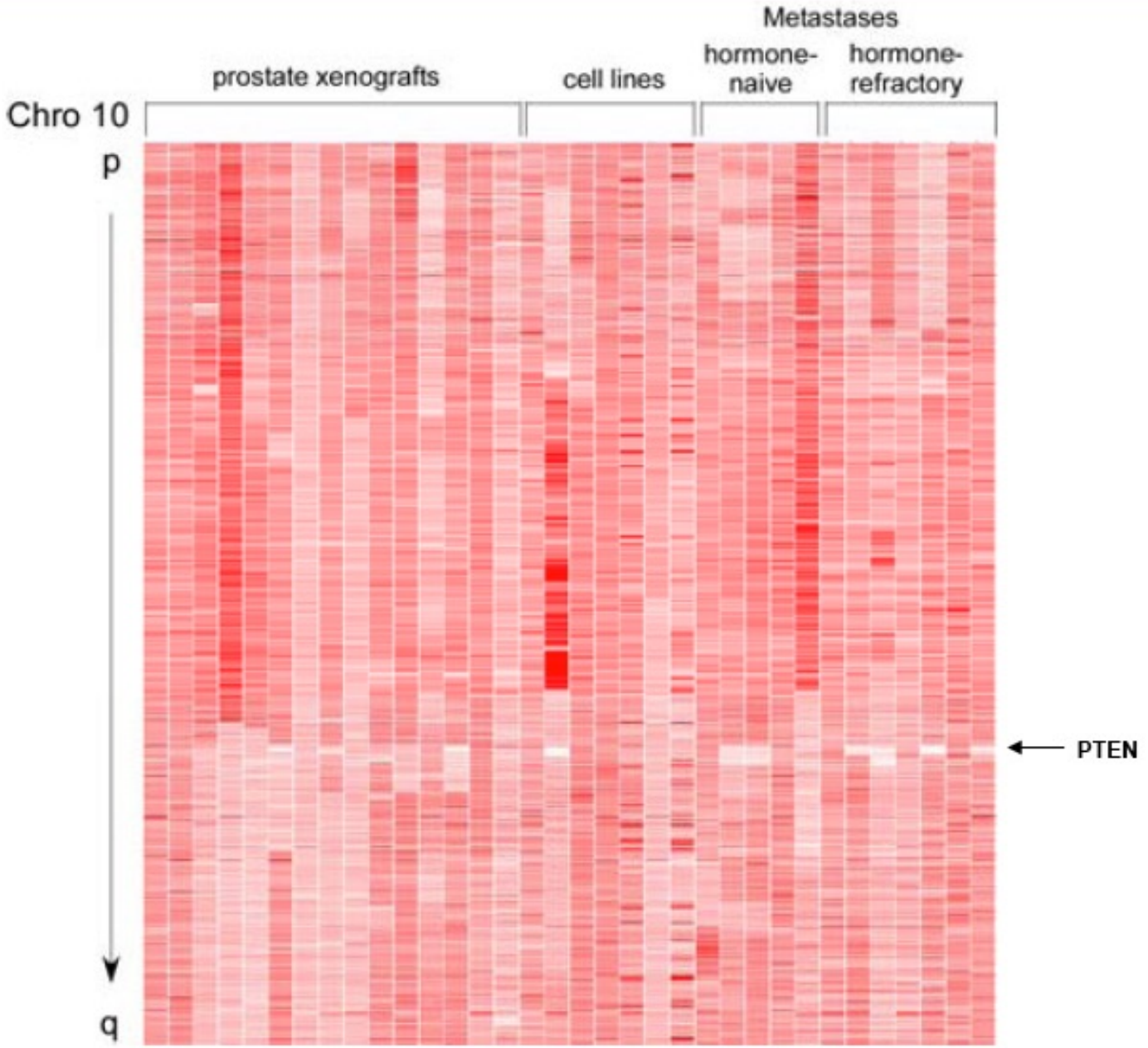
Matt Meyerson, Bill Sellers

Gliomas



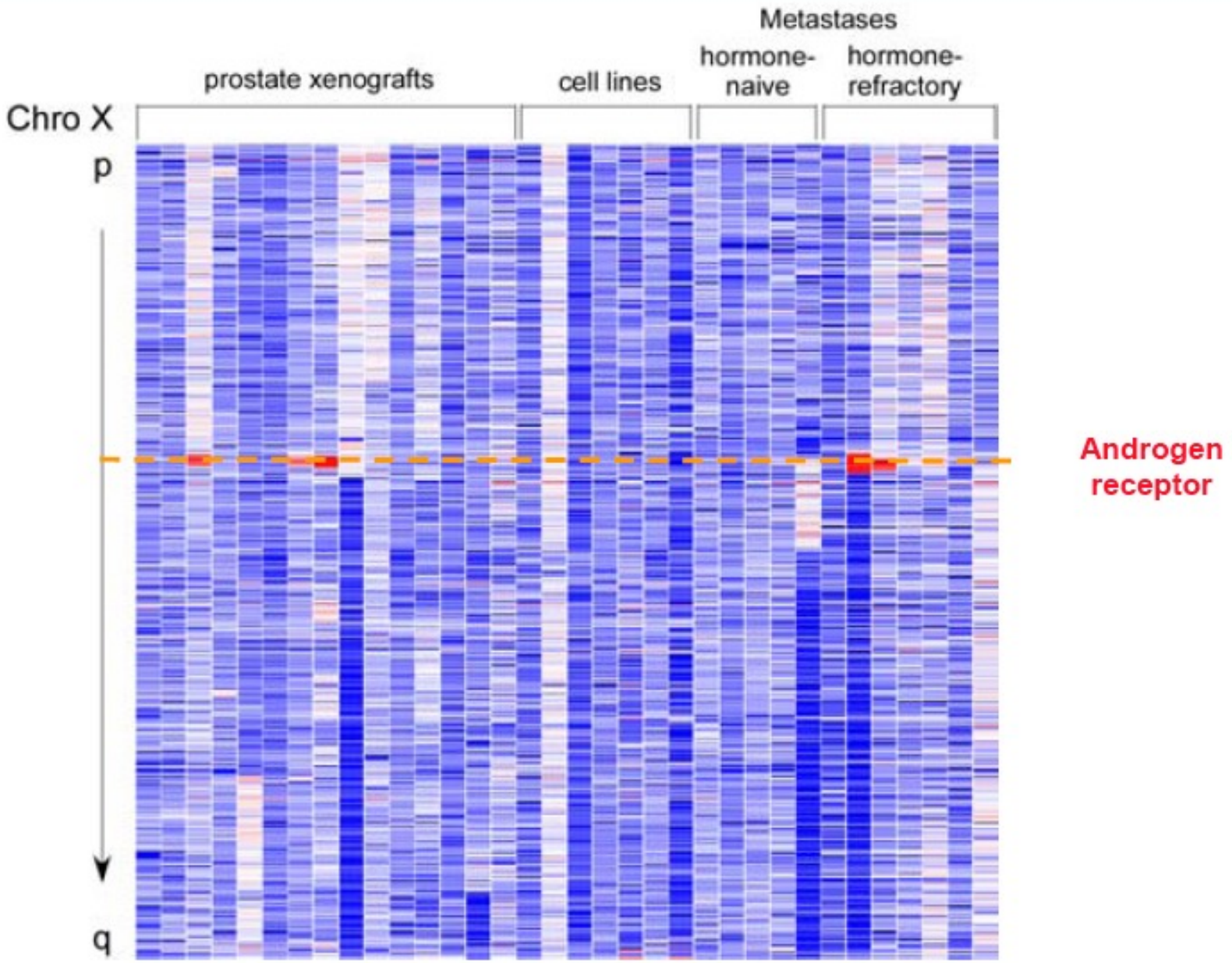
Deletions in Prostate Cancers

(Sellers, Meyer



Amplification in Prostate Cancer

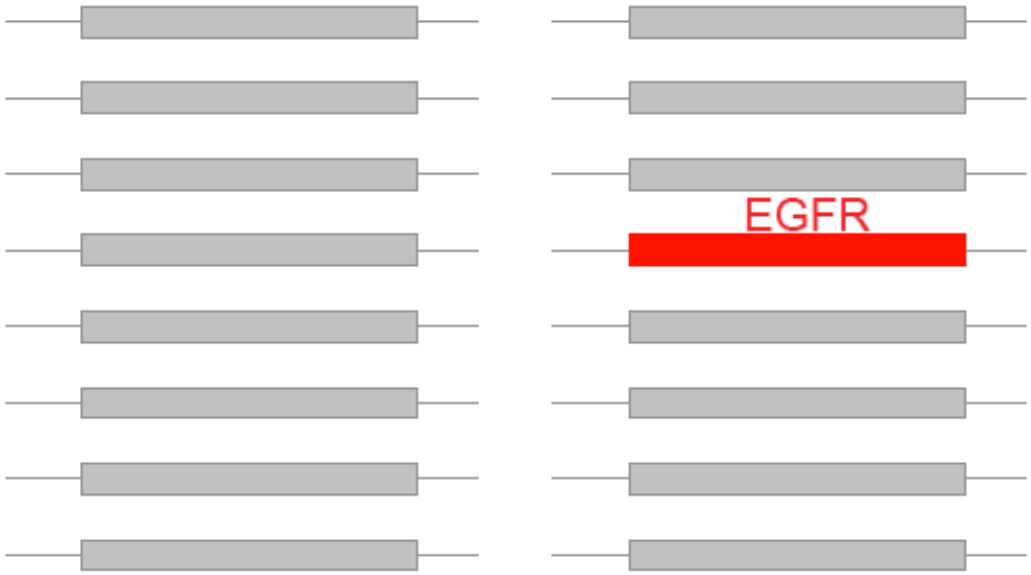
(Sellers, Meyerson)

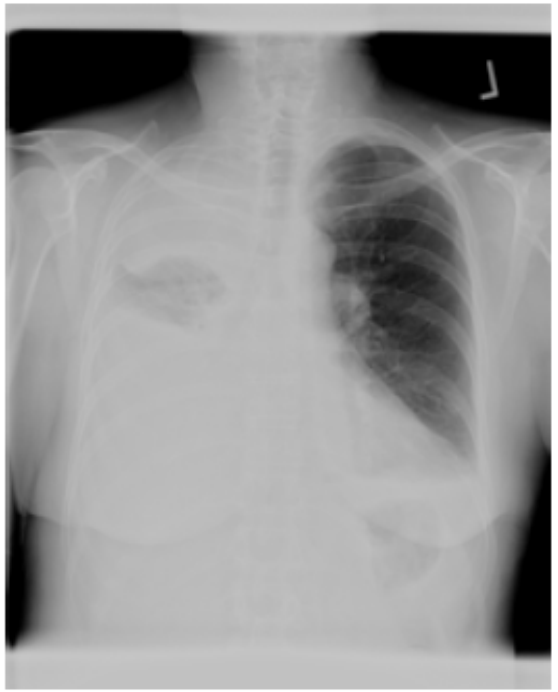


Systematic Search for Mutations in Lung Cancer

Matt Meyerson, Bill Sellers, DFC

Resequencing ~50 Kinase Genes in Tumors



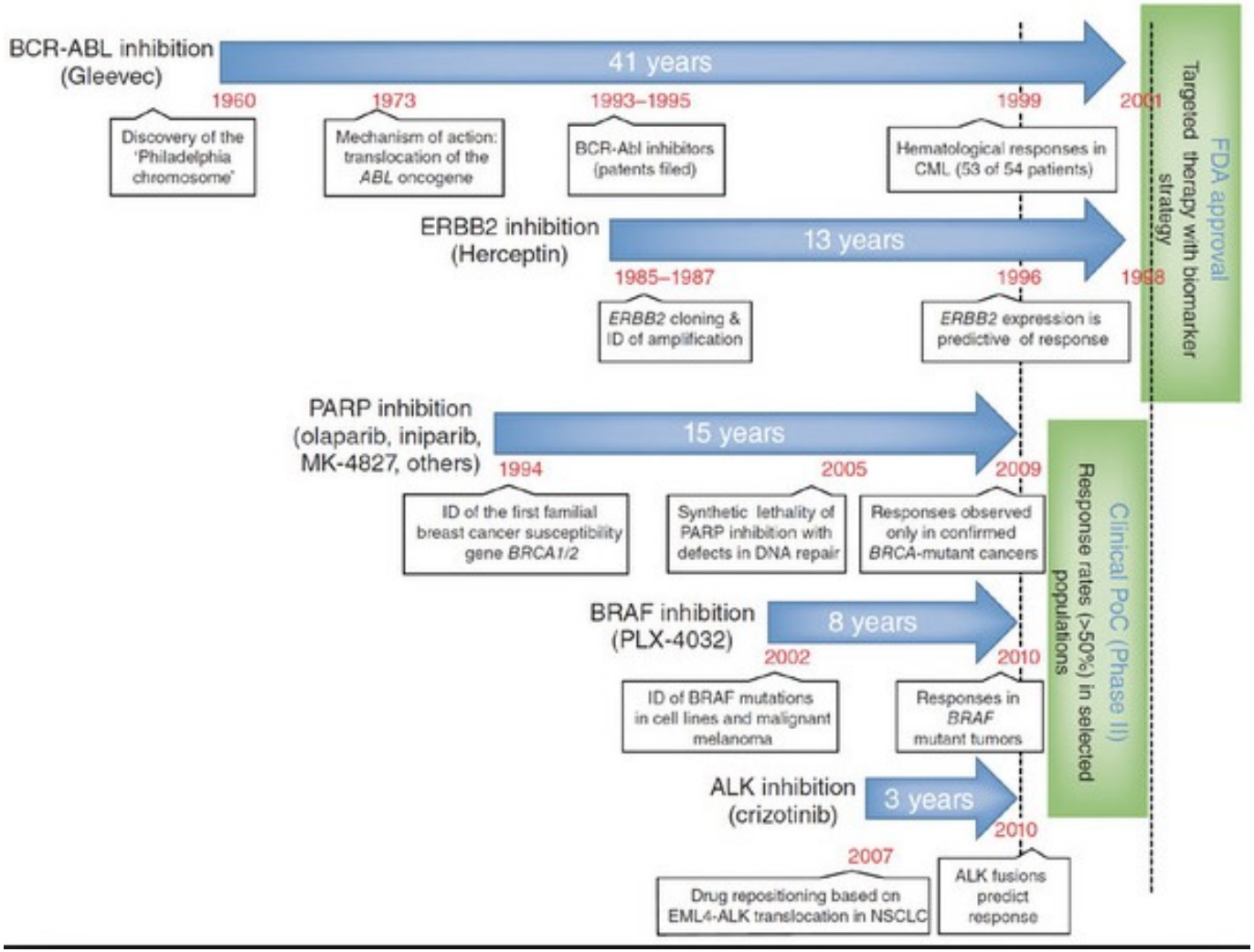


Epidermal Growth Factor Receptor (EGFR) Mutations in Tumors:

- Japanese
- Non-smokers
- Women
- Adenocarcinomas

Matches Reponse Profile of New Drug, Iressa

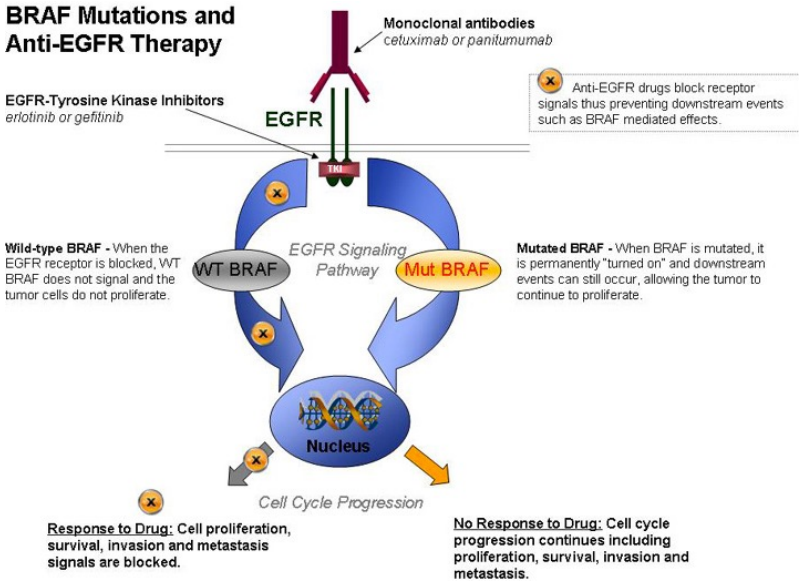
Cancer Genetics are accelerating the time from "target discovery" to clinical Proof of Concept



Lack of efficacy accounts for about 50% of failure in clinical trials, 29%Strategic , Safety (toxic)19%, Efficacy 51%

Need to better understand the context (cellular and genetic) in which a target is rate limiting

It is not only about the drug need to understand How to use it

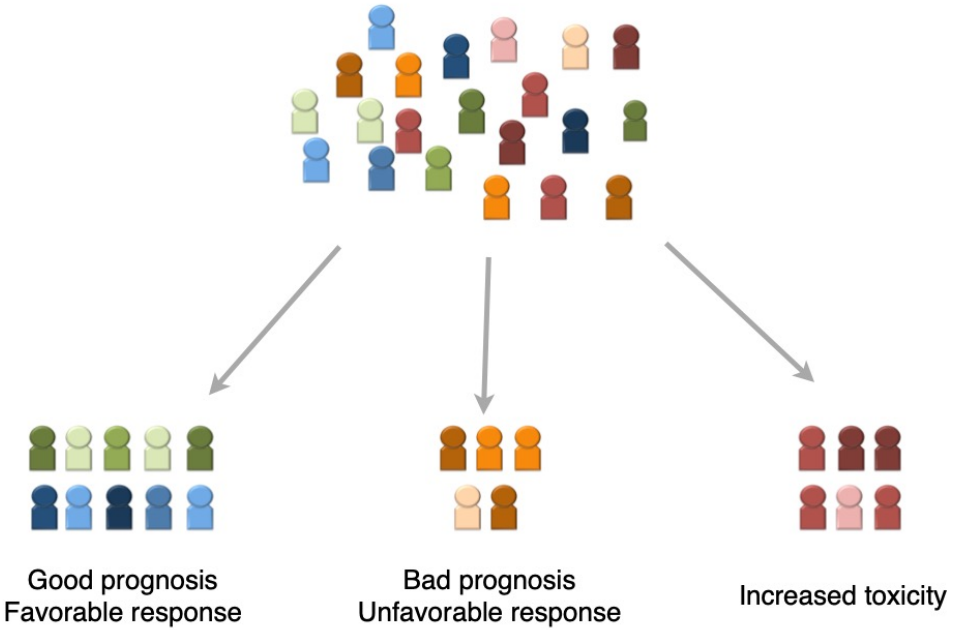


Right Target (Patient Omics, Validation,function) , Right Drug(molecules,Assays, Biology& mechanismof action), **Right Patient** (biomarkers,Rx comBination, predictive evidence-based decision) → clinical Success

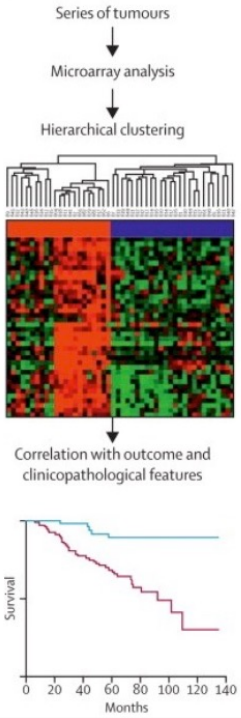
Cancer Genomics: What for?

- Finding new cancer genes (cancer drivers)
- Finding new therapeutic targets
- Identify molecular signatures to stratify tumors
- Move towards personalized cancer treatment

Stratify tumors based on molecular patterns

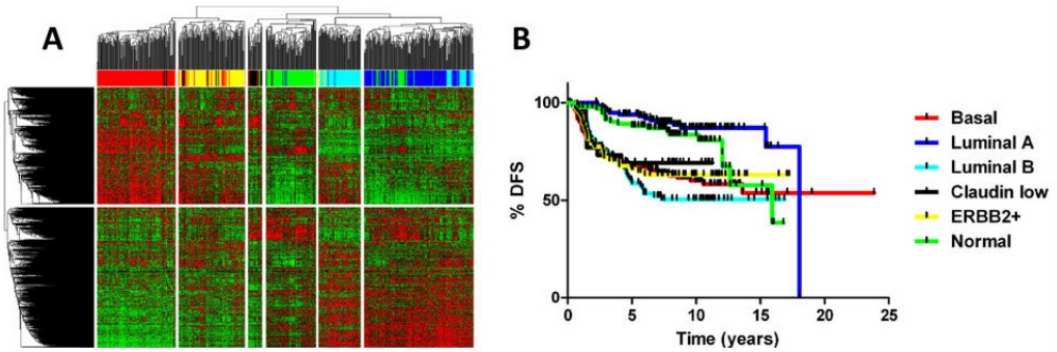


Stratify tumors based on molecular patterns



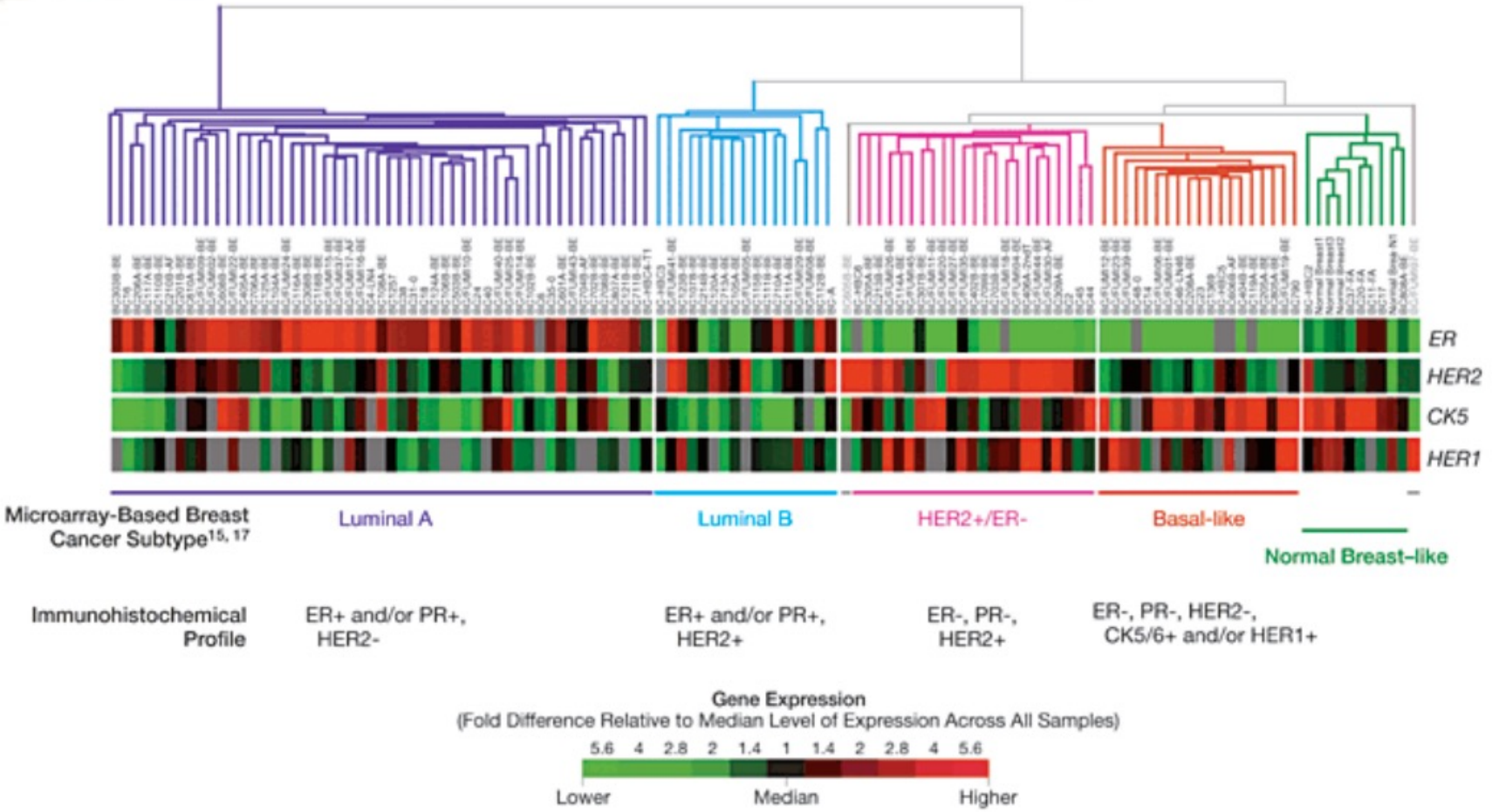
Stratify tumors based on molecular patterns

One example: Breast Cancer Intrinsic Subtypes



Expression profile Identification of Breast Tumor Intrinsic Subtypes

CENTER FOR CANCER RESEARCH



Carey, L. A. et al. JAMA 2006;295:2492-2502.

JAMA

SOMATIC MUTATION: MOLECULAR PORTRAITS OF BREAST CANCER AND PRECISION MEDICINE

The different
molecular subtypes are:
1. ER-positive group is divided into
a. luminal A: PgR high, HER2 negative
b. luminal B: PgR low, HER2 negative
2. HER2 type: HER2 positive (particularly aggressive)
3. Basal like: often referred to as triple negative breast cancer (TNBC): ER negative, PgR negative, HER2 negative.

Table 1 Gene alterations detected in the patient with HER2 resistance and relevant therapies to the alterations

Pathway	Gene, variant	Relevant therapies	Status of development
ERBB2	*ERBB2*, amplified	Trastuzumab Ado-trastuzumab	Approved therapy
CDK	*CDKN2A*, deleted	Palbociclib	Unapproved therapy
CDK	*CDKN2B*, deleted	Palbociclib	Unapproved therapy
P53	*TP53*, T125P	Investigational	Under development
mTOR	*STK11*, deleted	Everolimus	Unapproved therapy

The gene alterations were determined by NGS-based gene panel test (CANCERPLEX, KEW Inc., MA)

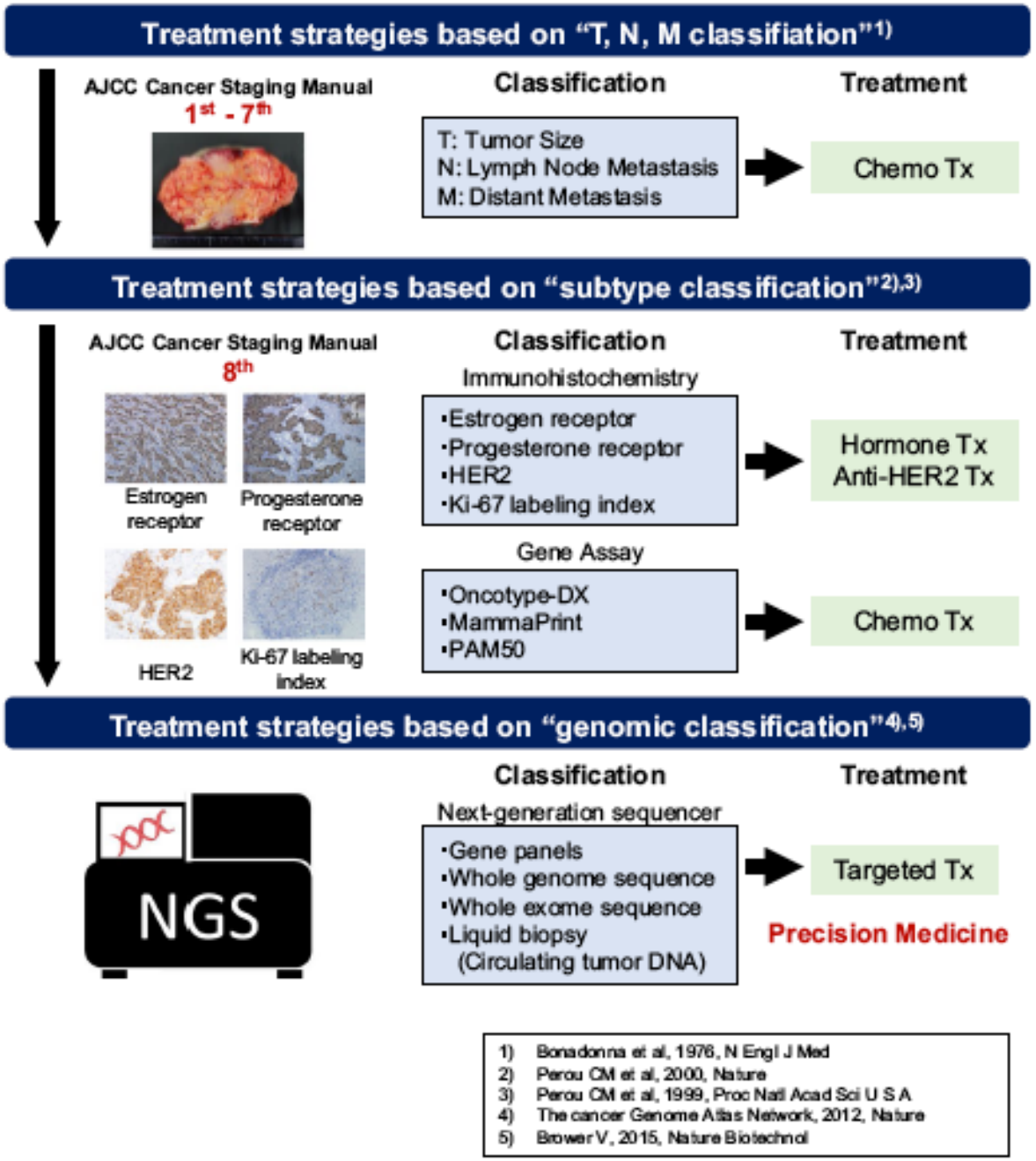


Fig. 1 Paradigm shift of breast cancer treatment. Initially, indications for chemotherapy for breast cancer were determined exclusively by tumor size, lymph node metastasis and distant metastasis. Currently, breast cancer is categorized by the expression of estrogen receptor, progesterone receptor, human epidermal growth factor receptor 2 (HER2) protein, and Ki-67 labeling index, given the availability and efficacy of specifically tailored therapies to each. More recently, gene assays predict the benefit of chemotherapy. Lately, genomic test utilizing next-generation sequencer enables one to select patients who are expected to respond better to each drug. *Ad* adjuvant; *AJCC* American Joint Committee on Cancer; *Tx* treatment

1) Bonadonna et al, 1976, N Engl J Med
2) Perou CM et al, 2000, Nature
3) Perou CM et al, 1999, Proc Natl Acad Sci U S A
4) The cancer Genome Atlas Network, 2012, Nature
5) Brower V, 2015, Nature Biotechnol

Cancer Genomics: What for?

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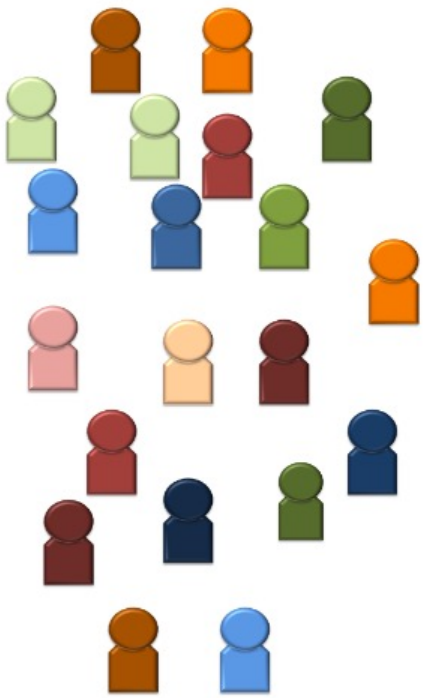
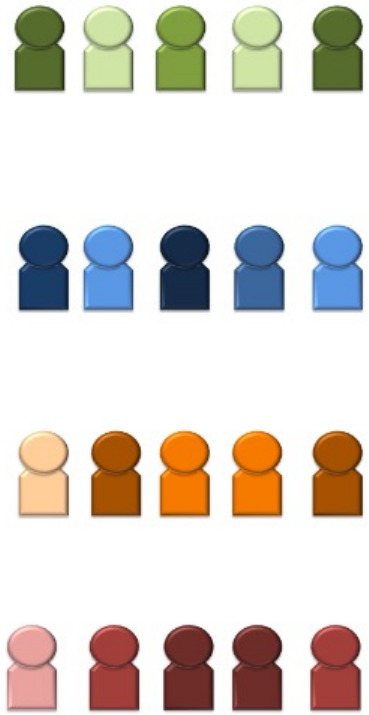
- Identify molecular signatures to stratify tumors
- Move towards personalized cancer treatment

Move towards personalized cancer treatment



YESTERDAY



TODAY

Find the right treatment
for the right patient
at the right time

TOMORROW

The Genome Gets Personal

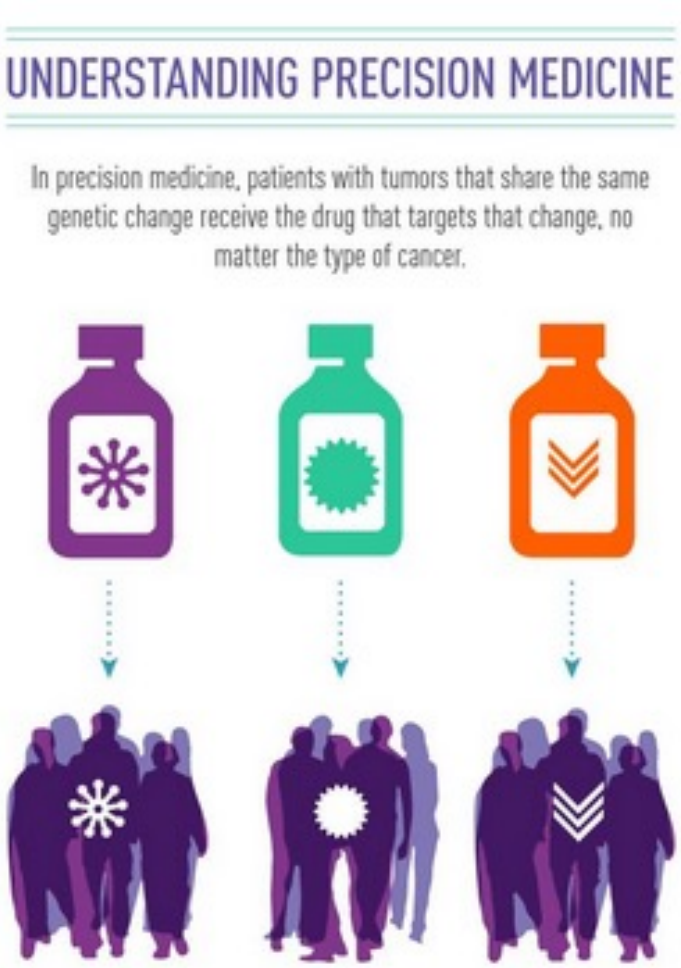
Genomics Help us Look at the Patients Individual Tumor Biology

Genomics

- Genomics is the study of how genes interact and are expressed as a whole
- Genomics and gene expression profiling tools focus on the *cancer itself* and can help determine
  - How aggressive is the cancer (prognosis)
  - What is the likely benefit from treatment (prediction)

Clinical Applications of Cancer Genomics

- **Prevention**: prediction of disease risk based on inherited or early somatic changes before neoplastic transformation
- **Diagnostic**: early disease diagnosis
- **Therapeutic**: identify cancer subtypes likely to respond; treatment selection-sensitivity or resistance to an agent
- **Prognostic**: Identify subsets with good or poor prognosis



Using the genetic changes in a patient's tumor to determine their treatment is known as precision medicine.

Credit: National Cancer Institute

Growing importance of genomic biomarkers for personalized medicine

A genomic biomarker is a measurable DNA and/or RNA characteristic that is an indicator of normal biologic processes, pathogenic processes, and/or response to therapeutic or other interventions.

Predisposition	• Risk of disease • e.g. genetic risk factors
Screening	• Presence of disease or precursors of disease. • e.g. detection markers of disease risk (PSA)
Diagnosis	• Classifying disease, Sub-typing disease • e.g. Benign vs. malignant.
Prognosis	• Stage of disease / likely outcome • e.g. Localized vs. metastatic
Therapeutic Choice	• Which drug is best for this tumor (CDx) • E.g. Her2Neu up-regulation → Herceptin
Monitoring	• Progression, remission, response to treatment

Definitions taken from 'Guidance for Industry – E15 Definitions for Genomic Biomarkers, Pharmacogenomics, Pharmacogenetics, Genomics Data and Sample coding categories,' US, FDA

More Diagnostic Microarrays (coming soon…)

Breast cancer

- **Almac Prognosis** using frozen or FFPE tissue and custom DSA expression array
- **bioMérieux Detection** using expression array
- **Ipsogen Genomic grade** and HER2 using tissue and expression array
- **Roche** Tamoxifen **metabolism** using blood and custom gene array AmpliChip® CYP450
- **Veridex Prognostic** using tissue and expression array

Colorectal cancer

- **Almac** Stage II **chemotherapy decision** using frozen or FFPE tissue & custom DSA expression array
- **bioMérieux Detection** using expression array

Prostate cancer

- **Almac Prognosis** using frozen or FFPE tissue and custom DSA expression array
- **Epigenomics Metastasis prediction** using prostate tissue and a custom PITX2 gene methylation array

Lung cancer

- **Almac** Tissue for **adjuvant chemotherapy** DSA expression array

Ovarian Cancer

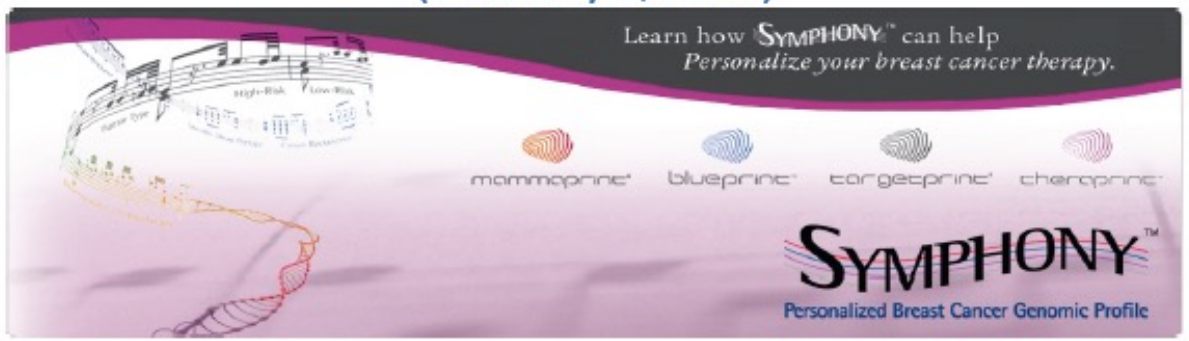
- **Almac** Tissue for **adjuvant chemotherapy** using frozen or FFPE tissue and custom DSA expression

Blood cancer

- **Skyline Diagnosis** of Acute Myeloid Leukemia (AML) on blood samples using custom expression array

FDA approves MammaPrint for cancer diagnosis

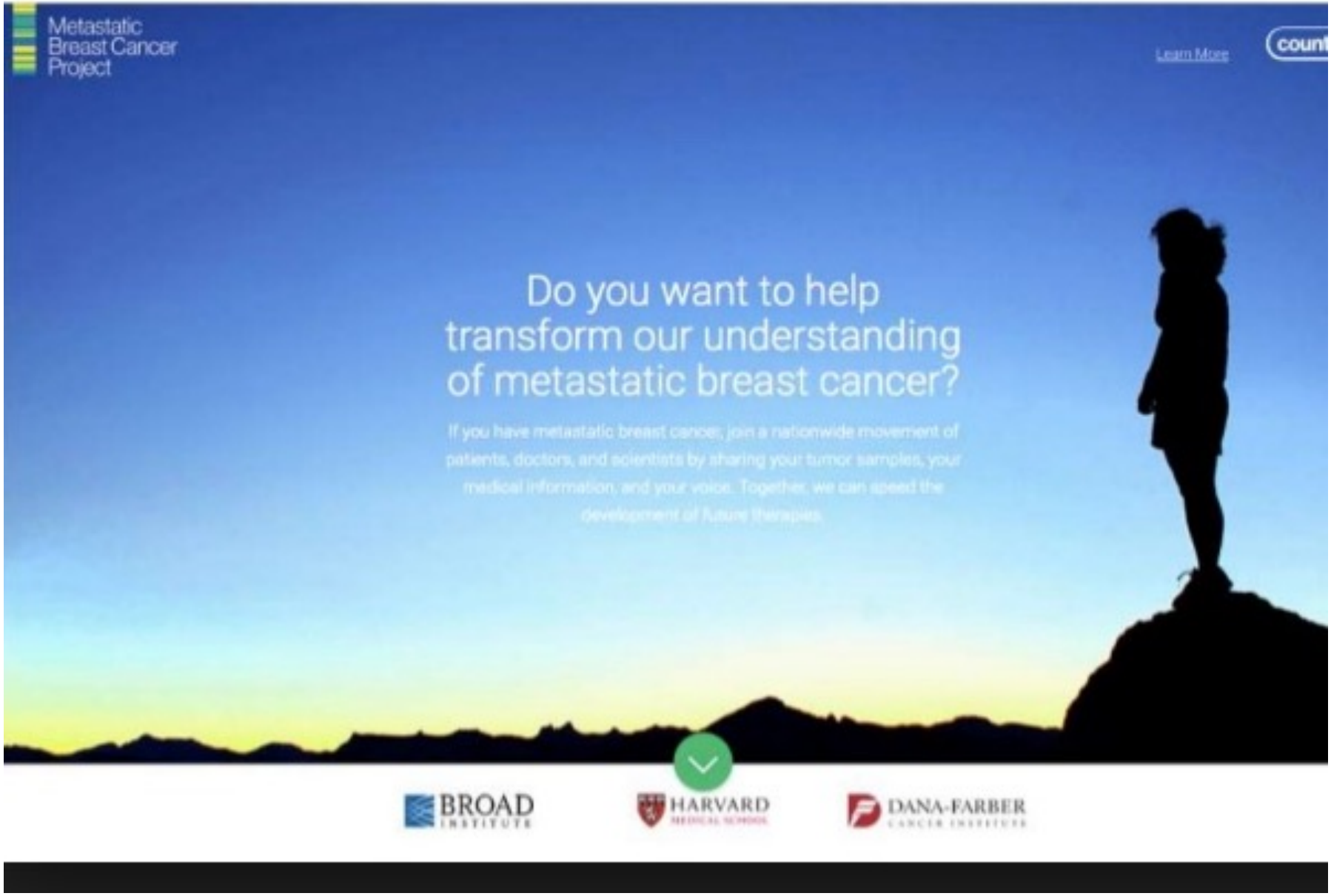
(February 7, 2007)



- **MammaPrint** interrogates all of the critical molecular pathways involved in the breast cancer metastatic cascade. It analyzes 70 critical genes that comprise a definitive gene expression signature and stratifies patients into two distinct groups — low risk or high risk of distant recurrence.
- **TargetPrint** is a microarray-based gene expression test which offers a quantitative assessment of the patient's level of estrogen receptor (ER), progesterone receptor (PR) and HER2/neu overexpression within her breast cancer.
- **BluePrint** is an 80-gene expression signature which classifies breast cancer into Basal-type, Luminal-type and ERBB2-type cancer
- **TheraPrint** is a microarray-based gene expression panel of 56 genes that have been identified as potential targets for prognosis and therapeutic response to a variety of therapies.

The Metastatic Breast Cancer Project

MBCproject.org



Άλλα γενετικά τεστ

Κάποια γενετικά τεστ που διατίθενται για τον καρκίνο του μαστού:

- Oncotype Dx
- MammaPrint
- BluePrint
- Prosigna
- bioTheranostics Breast Cancer Index
- Endopredict
- FoundationOne

Το MammaPrint και το BluePrint είναι της ίδιας εταιρείας, της Agendia.

Άλλα γενετικά τεστ: Oncotype Dx

Oncotype Dx: Γενετικό τεστ που με τη χρήση 21 γονιδίων (16 γονίδια σχετικά με τον καρκίνο, 5 reference) μπορεί να έχει συμβουλευτικό χαρακτήρα σε ό,τι αφορά τη θεραπεία και να προβλέψει την επανεμφάνιση της νόσου.

Αυτή η δοκιμή εξετάζει μια ομάδα γονιδίων για να προσδιορίσει την πιθανότητα υποτροπής του καρκίνου του μαστού και τα πιθανά οφέλη της χημειοθεραπείας σε πρώιμο στάδιο, θετικό σε ορμονικούς υποδοχείς καρκίνο του μαστού.

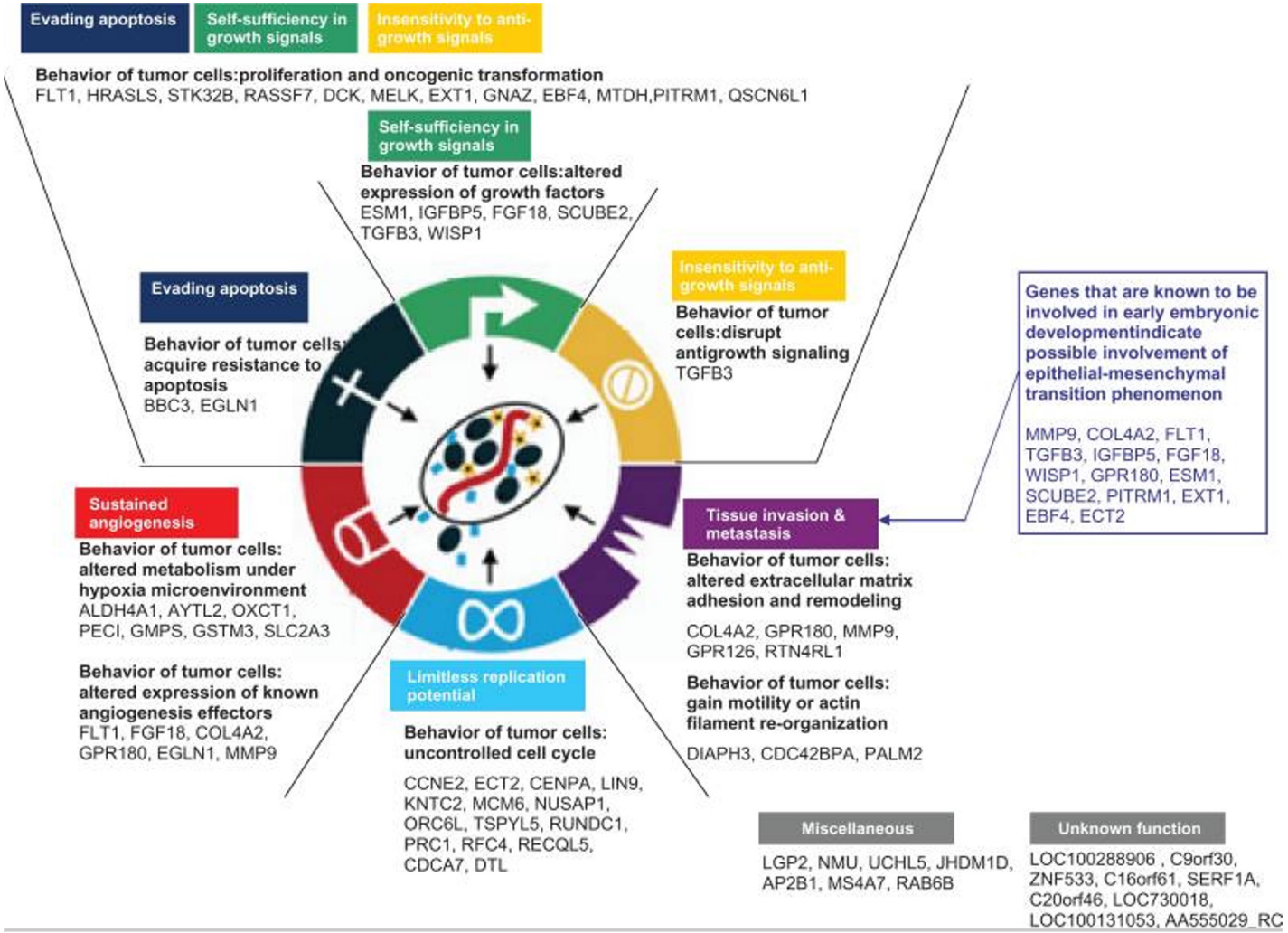
CANCER RELATED GENES (16)	REFERENCE GENES (5)
Proliferation genes: *Ki67; STK15; Survivin; CCNB1 (Cyclin B1); MYBL2*	*ACTB (b-actin)*
Invasion genes: *MMP11 (Stromolysin 3); CTSL2 (Cathepsin L2)*	*GAPDH*
HER2 genes: *GRB2; HER2*	*RPLPO*
Estrogen genes: *ER; PGR; BCL2; SCUBE2*	*GUS*
Other cancer related genes: *GSTM1; CD68; BAG1*	*TFRC*

Άλλα γενετικά τεστ: Oncotype Dx

- Ασθενείς κατάλληλοι για το συγκεκριμένο τεστ: early stage (μέχρι stage IIIa), ER+ HER2-, node+ ή node- (άρα αν έχει πάει ή όχι σε λεμφαδένες).
- Τα αποτελέσματα εμφανίζονται ως σκορ από το 1 ως το 100. Όσο υψηλότερο είναι το σκορ, τόσο πιο υψηλή είναι η πιθανότητα επανεμφάνισης, αλλά και το όφελος της χημειοθεραπείας στον ασθενή (αντίθετα, ένα χαμηλό σκορ δείχνει μεγαλύτερη ανταπόκριση σε ορμονοθεραπεία).
    - Σε γυναίκες άνω των 50: σκορ μέχρι 25 θεωρείται χαμηλό και πάνω από 25 υψηλό.
    - Σε γυναίκες κάτω των 50: μέχρι 15 θεωρείται χαμηλό, μέχρι 20 χαμηλό προς μέτριο, μέχρι 25 μέτριο και πάνω από 25 υψηλό.
- Επιπλέον, έχει και ποσοτικές μετρήσεις σε σκορ (μέσω RT-PCR) για ER, PGR, HER2.
- Υπάρχει και τεστ ειδικό για DCIS, που προβλέπει επανεμφάνιση (είτε σαν DCIS, είτε σαν επιθετικό καρκίνωμα), καθώς και ανταπόκριση σε ακτινοθεραπεία. Ένα σκορ μικρότερο από 39 θεωρείται χαμηλό, μέχρι 54 μέτριο και πάνω από 54 υψηλό.

Άλλα γενετικά τεστ: MammaPrint

70 γονίδια σχετικά με την επανεμφάνιση του καρκίνου.



Άλλα γενετικά τεστ: MammaPrint

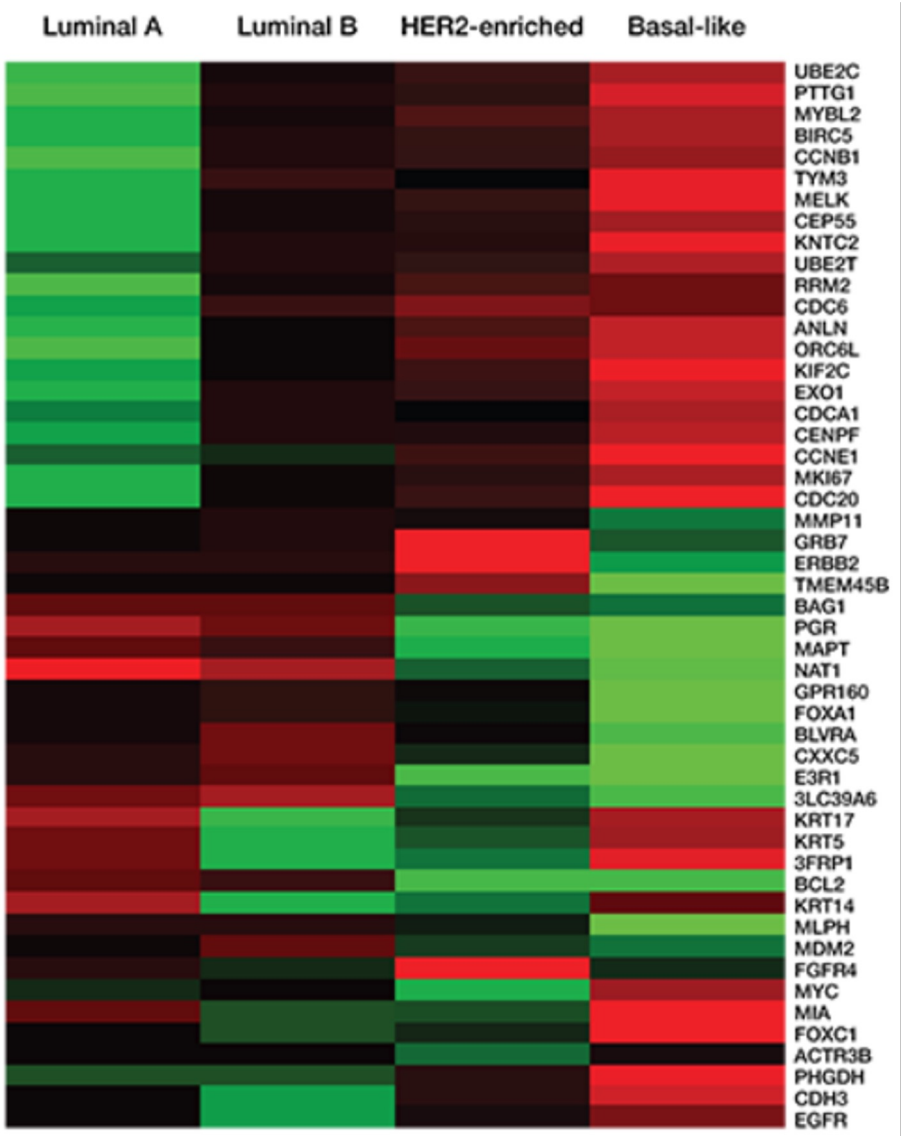
MammaPrint: Αυτό το τεστ αξιολογεί τη δραστηριότητα ενός συνόλου γονιδίων
και παρέχει ένα προφίλ γονιδιωματικού κινδύνου που προβλέπει τον κίνδυνο επανεμφάνισης
του καρκίνου του μαστού εντός 10 ετών σε γυναίκες με καρκίνο του μαστού πρώιμου σταδίου.

- Χρησιμοποιείται σε άτομα με καρκίνο stage I, II ή χειρουργήσιμο III, που είτε δεν έχει πάει σε λεμφαδένες είτε έχει 1-3 και ο όγκος είναι μέχρι 5 εκατοστά.
- Η ηλικία του ασθενούς δεν περιορίζει το τεστ.
- Από την ίδια την Agendia προτείνεται το τεστ να προηγηθεί της επέμβασης και γενικότερα να αποτελέσει βάση της στρατηγικής της θεραπείας (επέμβαση και μετέπειτα θεραπεία).
- Το ενδιαφέρον με αυτό το τεστ είναι ότι δεν περιλαμβάνει ούτε το ER, ούτε το HER2. Όμως, 12 από αυτά τα γονίδια που έχει δείχνουν έμμεσα την έκφραση του ER, οπότε υπάρχει και πληροφορία για αυτό.
- Η Agendia παρέχει το BluePrint ως τεστ για την κατηγοριοποίηση του τύπου του καρκίνου. Αυτό ελέγχει 80 γονίδια και οι ασθενείς που μπορούν να το κάνουν πρέπει να πληρούν τα ίδια κριτήρια με το MammaPrint. Από τα 80 γονίδια, τα 58 βοηθούν στην κατηγοριοποίηση ως luminal, τα 28 ως triple negative και τα 4 ως HER2-like.

Άλλα γενετικά τεστ: Prosigna

- Εξέταση 50 γονιδίων, γνωστών ως PAM50 gene signature.
- Μαζί με άλλες κλινικές ενδείξεις, προβλέπεται η πιθανότητα επανεμφάνισης του όγκου μέσα στα επόμενα 10 χρόνια σε ασθενείς που είναι hormone receptor positive και σε αρχικό στάδιο.
- Το τεστ μπορεί να χρησιμοποιηθεί σε γυναίκες μετά την εμμηνόπαυση, θετικές σε υποδοχείς ορμονών, είτε χωρίς λεμφαδένες (στάδια I & II), είτε με λεμφαδένες σε στάδια II & IIIA.
- Τα αποτελέσματα χωρίζονται σε 4 ενδογενείς υποτύπους: luminal A, luminal B, HER2-enriched, basal-like.

Άλλα γενετικά τεστ: Prosigna



- Ο τρόπος ερμηνείας των αποτελεσμάτων προέρχεται από έναν αλγόριθμο που βασίζεται στο PAM50 gene signature, τον ενδογενή υπότυπο, το μέγεθος του όγκου, την παρουσία ή όχι λεμφαδένων (nodal status) και το σκορ πολλαπλασιασμού (που υπολογίζεται μέσω γονιδίων υπεύθυνων για τον πολλαπλασιασμό των κυττάρων).
- Έτσι λοιπόν προκύπτει ένας αριθμός από το 1 ως το 100 που δείχνει την πιθανότητα επανεμφάνισης του όγκου μέσα στα επόμενα 10 χρόνια.

Άλλα γενετικά τεστ: FoundationOne

- FoundationOne: Εγκεκριμένο από τον FDA τεστ που μπορεί να χρησιμοποιηθεί σε ποικιλία συμπαγών όγκων.
- Χρήσιμο για την εύρεση της κατάλληλης θεραπείας και την κατανόηση αποτελεσμάτων σχετικών με την ανθεκτικότητα σε συγκεκριμένες θεραπείες.
- Για τον καρκίνο του μαστού υπάρχουν οι εξής biomarkers:

BIOMARKERS	FDA-APPROVED THERAPY‡
ERBB2 (HER2) amplification	**Herceptin**® (trastuzumab), **Kadcyla**® (ado-trastuzumab-emtansine), or **Perjeta**® (pertuzumab)
PIK3CA C420R, E542K, E545A, E545D [1635G>T only], E545G, E545K, Q546E, Q546R, H1047L, H1047R, and H1047Y alterations	**Piqray**® (alpelisib)

Άλλα γενετικά τεστ: FoundationOne

FoundationOne CDx: Αυτό το ολοκληρωμένο τεστ γονιδιωματικού προφίλ αναλύει πολλαπλά γονίδια, συμπεριλαμβανομένων εκείνων που είναι γνωστό ότι σχετίζονται με τον καρκίνο του μαστού, για να εντοπίσει στοχευμένες επιλογές θεραπείας και πιθανή καταλληλότητα κλινικών δοκιμών.

Άλλα γενετικά τεστ: FoundationOne

FOUNDATIONONE® CDx

PATIENT

TUMOR TYPE
Breast carcinoma (NOS)

REPORT DATE

ORDERED TEST #

PATIENT
DISEASE Breast carcinoma (NOS)
NAME
DATE OF BIRTH
SEX
MEDICAL RECORD #

PHYSICIAN
ORDERING PHYSICIAN
MEDICAL FACILITY
ADDITIONAL RECIPIENT
MEDICAL FACILITY ID
PATHOLOGIST

SPECIMEN
SPECIMEN SITE
SPECIMEN ID
SPECIMEN TYPE
DATE OF COLLECTION
SPECIMEN RECEIVED

Companion Diagnostic (CDx) Associated Findings

GENOMIC FINDINGS DETECTED	FDA-APPROVED THERAPEUTIC OPTIONS
PIK3CA E542K	Piqray® (Alpelisib)

For Microsatellite Instability (MSI) results, confirmatory testing using a validated orthogonal method should be performed.

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 5 Muts/Mb §
CDK4 amplification §
ESR1 Y537S
FGFR2 amplification §
PTEN T319fs*1
TPS3 splice site 559+1G>A

§ Refer to appendix for limitation statements related to detection of any copy number alterations, gene rearrangements, BRCA1/2 alterations, LOH, MSI, or TMB results in this section.

Please refer to appendix for Explanation of Clinical Significance Classification and for variants of unknown significance (VUS).

TABLE 1 COMPANION DIAGNOSTIC INDICATIONS

INDICATION	BIOMARKER	THERAPY
Non-small cell lung cancer (NSCLC)	*EGFR* exon 19 deletions and *EGFR* exon 21 L858R alterations	Gilotrif® (Afatinib), Iressa® (Gefitinib), Tagrisso® (Osimertinib), or Tarceva® (Erlotinib)
	EGFR exon 20 T790M alterations	Tagrisso® (Osimertinib)
	ALK rearrangements	Alecensa® (Alectinib), Xalkori® (Crizotinib), or Zykadia® (Ceritinib)
Melanoma	*BRAF* V600E	Tafinlar® (Dabrafenib) in combination with Mekinist® (Trametinib)
	BRAF V600E	Tafinlar® (Dabrafenib) or Zelboraf® (Vemurafenib)
	BRAF V600E and V600K	Mekinist® (Trametinib) or Cotellic® (Cobimetinib) in combination with Zelboraf® (Vemurafenib)
Breast cancer	*ERBB2* (*HER2*) amplification	Herceptin® (Trastuzumab), Kadcyla® (Ado-trastuzumab emtansine), or Perjeta® (Pertuzumab)
	PIK3CA C420R, E542K, E545A, E545D (G/D/G>T only), E545G, E545K, Q546R, Q546E, H1047L, H1047R, and H1047Y alterations	Piqray® (Alpelisib)
Colorectal cancer	*KRAS* wild-type (absence of mutation in codons 12 and 13)	Erbitux® (Cetuximab)
	KRAS wild-type (absence of mutations in exons 2, 3, and 4) and *NRAS* wild-type (absence of mutations in exons 2, 3, and 4)	Vectibix® (Panitumumab)
Ovarian cancer	*BRCA1/2* alterations	Lynparza® (Olaparib) or Rubraca® (Rucaparib)

ABOUT THE TEST FoundationOne®CDx is the first FDA-approved broad companion diagnostic for solid tumors.

Electronically signed by Matthew Hiemenz, M.D. |
Julia Elvin, M.D., Ph.D., Laboratory Director CLIA: 22D2027531
Shakti Ramkissoon, M.D., Ph.D., M.M. Sc, Laboratory Director CLIA: 34D2044309
Foundation Medicine, Inc. | 1.888.988.3639

Sample Preparation: 150 Second St., 1st Floor, Cambridge, MA 02141 - CLIA: 22D2027531
Sample Analysis: 150 Second St., 1st Floor, Cambridge, MA 02141 - CLIA: 22D2027531
Post-Sequencing Analysis: 150 Second St., 1st Floor, Cambridge, MA 02141 - CLIA: 22D2027531

Note: The intended use (IU) statement and claims made on this sample report may not be up to date. For the latest version of the FoundationOne CDx claims and IU, please see the current label: www.foundationmedicine.com/f1cdx

FDA APPROVED CLAIMS - PAGE 1 of 1

FOUNDATIONONE® CDx

PATIENT

TUMOR TYPE
Breast carcinoma (NOS)
COUNTRY CODE

REPORT DATE
ORDERED TEST #

ABOUT THE TEST FoundationOne®CDx is the first and only FDA-Approved comprehensive companion diagnostic for all solid tumors.

Interpretive content on this page and subsequent pages is provided as a professional service, and is not reviewed or approved by the FDA.

PATIENT
DISEASE Breast carcinoma (NOS)
NAME
DATE OF BIRTH
SEX
MEDICAL RECORD #

PHYSICIAN
ORDERING PHYSICIAN
MEDICAL FACILITY
ADDITIONAL RECIPIENT
MEDICAL FACILITY ID
PATHOLOGIST

SPECIMEN
SPECIMEN SITE
SPECIMEN ID
SPECIMEN TYPE
DATE OF COLLECTION
SPECIMEN RECEIVED

Biomarker Findings

Microsatellite status - MS-Stable
Tumor Mutational Burden - 5 Muts/Mb

Genomic Findings

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

CDK4 amplification
ESR1 Y537S
PIK3CA E542K
PTEN T319fs*1
FGFR2 amplification
TP53 splice site 559+1G>A

3 Disease relevant genes with no reportable alterations: ***BRCA1, BRCA2, ERBB2***

8 Therapies with Clinical Benefit
3 Therapies with Lack of Response
35 Clinical Trials

BIOMARKER FINDINGS	ACTIONABILITY
Microsatellite status - MS-Stable	**No therapies or clinical trials.** see Biomarker Findings section
Tumor Mutational Burden - 5 Muts/Mb	**No therapies or clinical trials.** see Biomarker Findings section

The content provided as a professional service by Foundation Medicine, Inc., has not been reviewed or approved by the FDA.

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PROFESSIONAL SERVICES - PAGE 1 of 26

Άλλα γενετικά τεστ: FoundationOne

FOUNDATIONONE® CDx

PATIENT

TUMOR TYPE
Breast carcinoma (NOS)
COUNTRY CODE

REPORT DATE
ORDERED TEST #

GENOMIC FINDINGS	THERAPIES WITH CLINICAL BENEFIT (IN PATIENT'S TUMOR TYPE)		THERAPIES WITH CLINICAL BENEFIT (IN OTHER TUMOR TYPE)
CDK4 - amplification	Palbociclib	1	none
10 Trials see p. 17	Ribociclib	1	
ESR1 - Y537S	Fulvestrant	1	none
	▲ Anastrozole[1]		
	▲ Exemestane[1]		
10 Trials see p. 19	▲ Letrozole[1]		
PIK3CA - E542K	Alpelisib	1	Temsirolimus
10 Trials see p. 23	Everolimus	2A	
PTEN - T319fs*1	Everolimus	2A	Temsirolimus
10 Trials see p. 25			
FGFR2 - amplification	none		Erdafitinib
9 Trials see p. 21			Pazopanib

▲ 1. Patient may be resistant to indicated therapy

☐ NCCN category

GENOMIC FINDINGS WITH NO REPORTABLE THERAPEUTIC OR CLINICAL TRIAL OPTIONS

For more information regarding biological and clinical significance, including prognostic, diagnostic, germline, and potential chemosensitivity implications, see the Genomic Findings section.

TP53 - splice site 559+1G>A ..... p. 8

NOTE Genomic alterations detected may be associated with activity of certain FDA-approved drugs; however, the agents listed in this report may have varied clinical evidence in the patient's tumor type.

Neither the therapeutic agents nor the trials enrolled are ranked in order of potential clinical efficacy for this patient, nor are they ranked in order of level of evidence for this patient's tumor type.

Electronically signed by Matthew Hawtrey, M.D. |
Julia Elvin, M.D., Ph.D., Laboratory Director CLIA: 22D2027531
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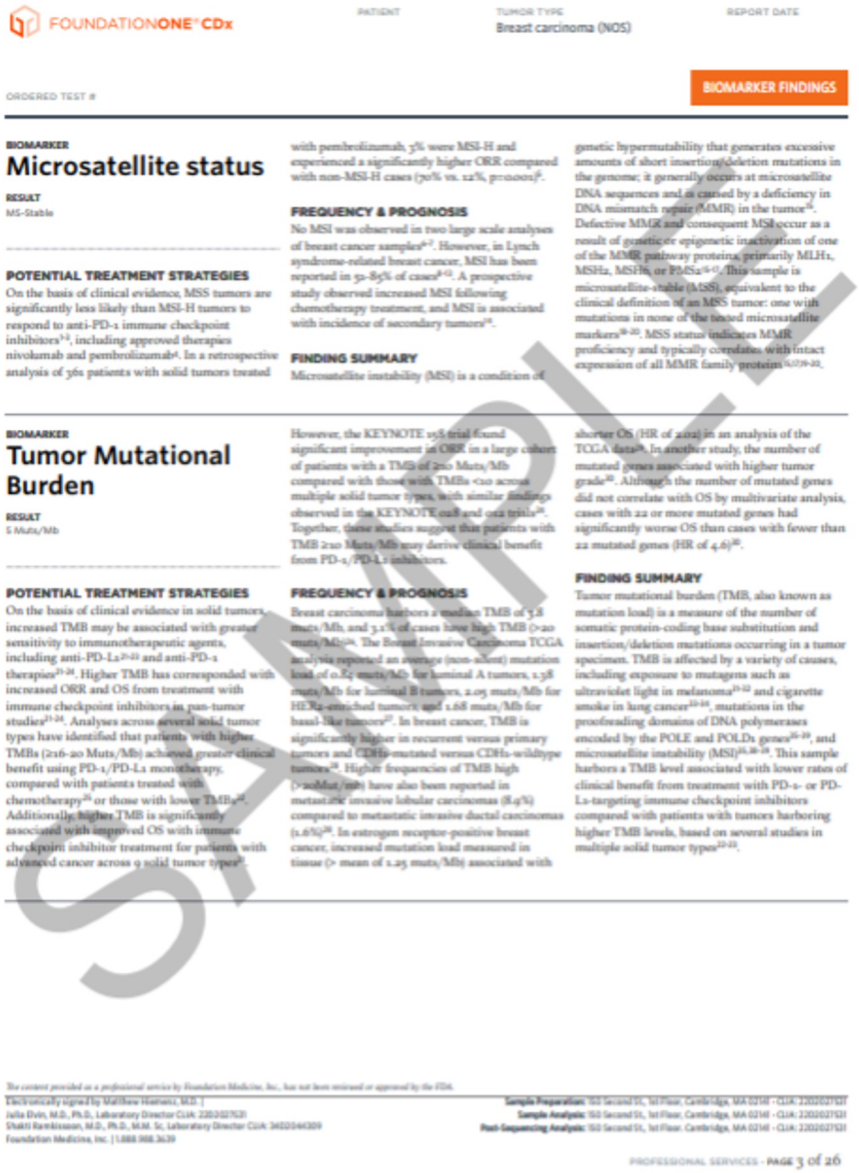

FOUNDATIONONE® CDx

PATIENT

TUMOR TYPE
Breast carcinoma (NOS)

REPORT DATE

ORDERED TEST #

BIOMARKER FINDINGS

BIOMARKER
Microsatellite status

RESULT
MS-Stable

POTENTIAL TREATMENT STRATEGIES
On the basis of clinical evidence, MSS tumors are significantly less likely than MSI-H tumors to respond to anti-PD-1 immune checkpoint inhibitors[1-3], including approved therapies nivolumab and pembrolizumab[4]. In a retrospective analysis of 361 patients with solid tumors treated with pembrolizumab, 3% were MSI-H and experienced a significantly higher ORR compared with non-MSI-H cases (70% vs. 12%, $p<0.001$)[5].

FREQUENCY & PROGNOSIS
No MSI was observed in two large scale analyses of breast cancer samples[6,7]. However, in Lynch syndrome-related breast cancer, MSI has been reported in 52-85% of cases[8-12]. A prospective study observed increased MSI following chemotherapy treatment, and MSI is associated with incidence of secondary tumors[13].

FINDING SUMMARY
Microsatellite instability (MSI) is a condition of genetic hypermutability that generates excessive amounts of short insertion/deletion mutations in the genome; it generally occurs at microsatellite DNA sequences and is caused by a deficiency in DNA mismatch repair (MMR) in the tumor[14]. Defective MMR and consequent MSI occur as a result of genetic or epigenetic inactivation of one of the MMR pathway proteins, primarily MLH1, MSH2, MSH6, or PMS2[15-17]. This sample is microsatellite-stable (MSS), equivalent to the clinical definition of an MSS tumor: one with mutations in none of the tested microsatellite markers[18-20]. MSS status indicates MMR proficiency and typically correlates with intact expression of all MMR family proteins[6,17,19-21].

BIOMARKER
Tumor Mutational Burden

RESULT
5 Muts/Mb

POTENTIAL TREATMENT STRATEGIES
On the basis of clinical evidence in solid tumors, increased TMB may be associated with greater sensitivity to immunotherapeutic agents, including anti-PD-L1[22-23] and anti-PD-1 therapies[23-24]. Higher TMB has corresponded with increased ORR and OS from treatment with immune checkpoint inhibitors in pan-tumor studies[23-24]. Analyses across several solid tumor types have identified that patients with higher TMBs (≥16-20 Muts/Mb) achieved greater clinical benefit using PD-1/PD-L1 monotherapy, compared with patients treated with chemotherapy[24] or those with lower TMBs[18]. Additionally, higher TMB is significantly associated with improved OS with immune checkpoint inhibitor treatment for patients with advanced cancer across 9 solid tumor types[18].

FREQUENCY & PROGNOSIS
Breast carcinoma harbors a median TMB of 3.8 muts/Mb, and 3.1% of cases have high TMB (>20 muts/Mb)[25]. The Breast Invasive Carcinoma TCGA analysis reported an average (non-silent) mutation load of 0.84 muts/Mb for luminal A tumors, 1.38 muts/Mb for luminal B tumors, 2.05 muts/Mb for HER2-enriched tumors, and 1.68 muts/Mb for basal-like tumors[26]. In breast cancer, TMB is significantly higher in recurrent versus primary tumors and CDH1-mutated versus CDH1-wildtype tumors[26]. Higher frequencies of TMB high (>20Mut/Mb) have also been reported in metastatic invasive lobular carcinomas (8.4%) compared to metastatic invasive ductal carcinomas (1.6%)[26]. In estrogen receptor-positive breast cancer, increased mutation load measured in tissue (> mean of 1.25 muts/Mb) associated with however, the KEYNOTE 158 trial found significant improvement in ORR in a large cohort of patients with a TMB of ≥10 Muts/Mb compared with those with TMBs <10 across multiple solid tumor types, with similar findings observed in the KEYNOTE 028 and 012 trials[24]. Together, these studies suggest that patients with TMB ≥10 Muts/Mb may derive clinical benefit from PD-1/PD-L1 inhibitors.

shorter OS (HR of 2.02) in an analysis of the TCGA data[26]. In another study, the number of mutated genes associated with higher tumor grade[20]. Although the number of mutated genes did not correlate with OS by multivariate analysis, cases with 22 or more mutated genes had significantly worse OS than cases with fewer than 22 mutated genes (HR of 4.6)[20].

FINDING SUMMARY
Tumor mutational burden (TMB, also known as mutation load) is a measure of the number of somatic protein-coding base substitution and insertion/deletion mutations occurring in a tumor specimen. TMB is affected by a variety of causes, including exposure to mutagens such as ultraviolet light in melanoma[27-32] and cigarette smoke in lung cancer[33-35], mutations in the proofreading domains of DNA polymerases encoded by the POLE and POLD1 genes[36-39], and microsatellite instability (MSI)[35,38-39]. This sample harbors a TMB level associated with lower rates of clinical benefit from treatment with PD-1- or PD-L1-targeting immune checkpoint inhibitors compared with patients with tumors harboring higher TMB levels, based on several studies in multiple solid tumor types[22-23].

Electronically signed by Matthew Hawtrey, M.D. |
Julia Elvin, M.D., Ph.D., Laboratory Director CLIA: 22D2027531
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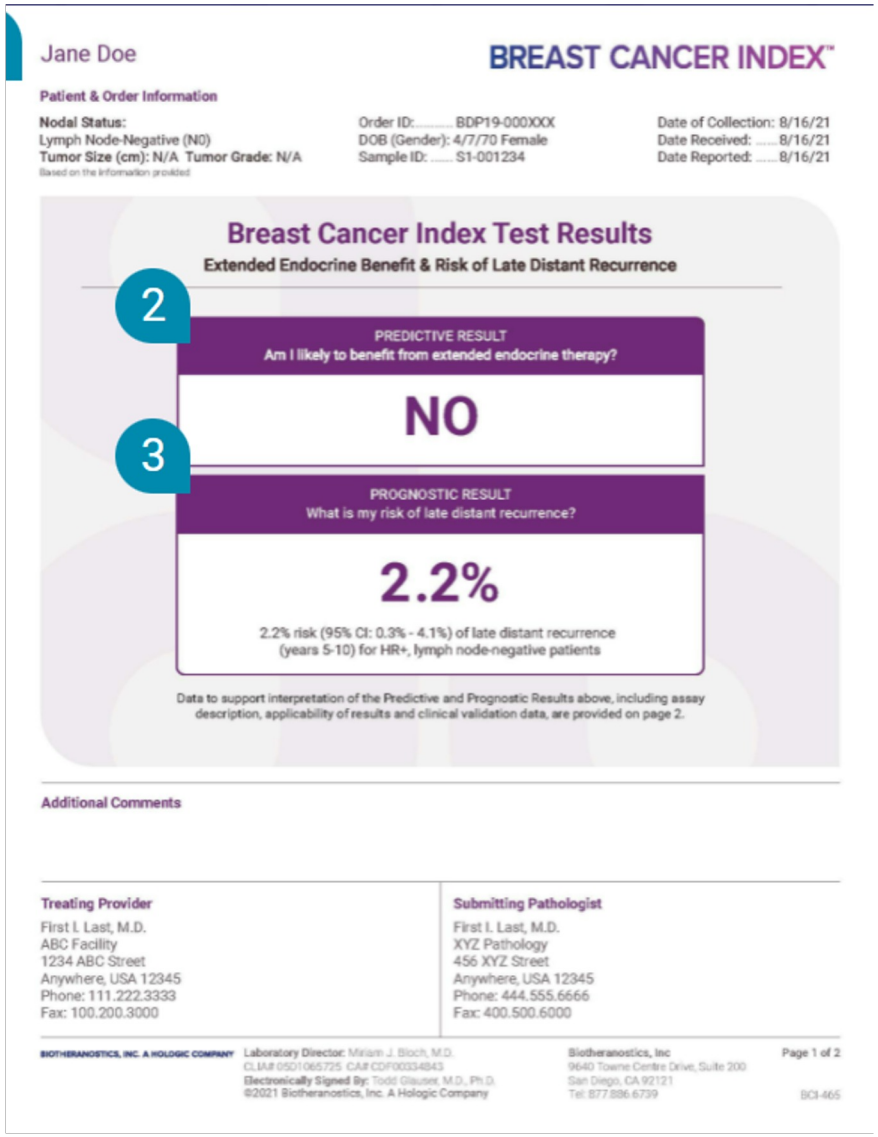
Άλλα γενετικά τεστ: bioTheranostics Breast Cancer Index

- Γενετικό τεστ που σχετίζεται με το ρίσκο επανεμφάνισης καρκίνου 5 χρόνια μετά από τη anti-estrogen θεραπεία (το οποίο εμφανίζεται με ένα ποσοστό), καθώς και το πόσο αυτή μπορεί να ωφελήσει μετά από τόσο μεγάλο χρονικό διάστημα..
- Το δείγμα που εξετάζεται είναι αυτό που αφαιρέθηκε μετά τη διάγνωση.
- Ασθενείς που γίνεται να εξεταστούν με αυτό το τεστ είναι άτομα με καρκίνο σταδίου από I ως IIIA, έχει αφαιρεθεί ο όγκος. Επιπλέον, ήταν ER θετικοί ή/και PGR θετικοί και έχουν υποβληθεί σε ορμονική θεραπεία.
- Το τεστ χρησιμοποιεί 11 γονίδια, μεταξύ αυτών και τα γονίδια HOXB13 και IL17BR για την αναλογία HOXB13/IL17BR..

Άλλα γενετικά τεστ: bioTheranostics Breast Cancer Index



Jane Doe

BREAST CANCER INDEX™

Patient & Order Information

Nodal Status:
Lymph Node-Negative (N0)
Tumor Size (cm): N/A **Tumor Grade:** N/A
Based on the information provided

Order ID: BDP19-000XXX
DOB (Gender): 4/7/70 Female
Sample ID: S1-001234

Date of Collection: 8/16/21
Date Received: 8/16/21
Date Reported: 8/16/21

Breast Cancer Index Test Results

Extended Endocrine Benefit & Risk of Late Distant Recurrence

2

PREDICTIVE RESULT
Am I likely to benefit from extended endocrine therapy?

NO

3

PROGNOSTIC RESULT
What is my risk of late distant recurrence?

2.2%

2.2% risk (95% CI: 0.3% - 4.1%) of late distant recurrence (years 5-10) for HR+, lymph node-negative patients

Data to support interpretation of the Predictive and Prognostic Results above, including assay description, applicability of results and clinical validation data, are provided on page 2.

Additional Comments

Treating Provider
First I. Last, M.D.
ABC Facility
1234 ABC Street
Anywhere, USA 12345
Phone: 111.222.3333
Fax: 100.200.3000

Submitting Pathologist
First I. Last, M.D.
XYZ Pathology
456 XYZ Street
Anywhere, USA 12345
Phone: 444.555.6666
Fax: 400.500.6000

BIOTHERANOSTICS, INC. A HOLOGIC COMPANY

Laboratory Director: Miriam J. Bloch, M.D.
CLIA# 05D1065725. CAP CDF00334843
Electronically Signed By: Todd Glauser, M.D., Ph.D.
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Biotheranostics, Inc
9640 Towne Centre Drive, Suite 200
San Diego, CA 92121
Tel: 877.886.6739

Page 1 of 2

BCI-465

Άλλα γενετικά τεστ:

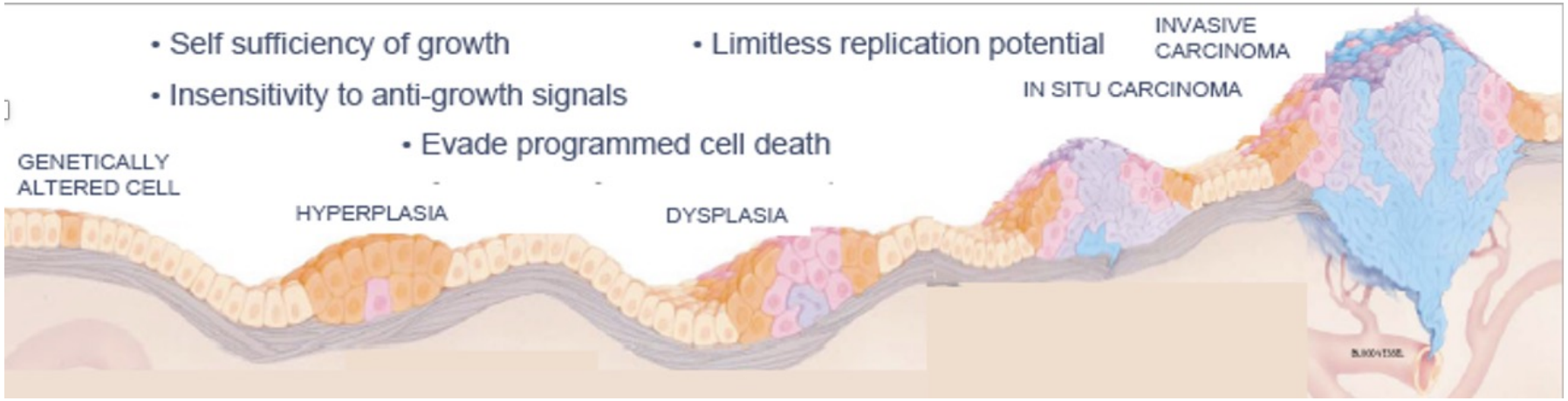
Γενετικός έλεγχος BRCA1 και BRCA2: Αυτές οι δοκιμές επικεντρώνονται
ειδικά σε μεταλλάξεις στα γονίδια BRCA1 και BRCA2,
που σχετίζονται με το σύνδρομο κληρονομικού καρκίνου του μαστού και των ωοθηκών.

Άλλα γενετικά τεστ: EndoPredict

- Endopredict: Της Myriad genetics. Για την εξέταση τόσο node positve όσο και node negative περιστατικών.
- Μαζί με κλινικά ευρήματα υπολογίζεται το ρίσκο μακρινής επανεμφάνισης μετά από 10 χρόνια.
- Βοηθάει τους ασθενείς με χαμηλό ρίσκο επανεμφάνισης να αποφύγουν τη χημειοθεραπεία, καθώς και υποδεικνύει ποιοι ασθενείς έχουν πολύ υψηλό ρίσκο επανεμφάνισης ώστε να χορηγηθεί συνδυαστική θεραπεία.
- Χρησιμοποιεί 12 γονίδια συνολικά (8 target genes, 3 normalization genes & 1 control gene).
- Τα 8 γονίδια που ελέγχει: *AZGP1*, *BIRC5*, *DHCR7*, *IL6ST*, *MGP*, *RBBP8*, *STC2*, *UBE2C*.
- Τα 3 γονίδια για κανονικοποίηση: *CALM2*, *OAZ1* and *RPL37A*.

Cancer Genomics:

What Does It Mean for You?



Cancer is a disease of the genome

What I mean by this is that all known cancers carry somatic DNA alterations that make it possible for the cells to grow without the normal limits.

Cancer risk can be familial, due to inherited mutations that are present in every cell.

Cancer is a disease of the genome

- **Therefore, if we precisely define the cancer genome, we will understand and cure cancer**
  - Why we must be cautious about such statements
- **Definitions**
- **Founder mutations-first genomic mutation**
  - These are often lesions that lead to genomic/chromosomal instability (p53, RB, etc.) and are often not fully transforming
- **Driver mutations-these are mutations that are required for expression of fully transformed phenotype**

Cancer is a disease of the genome (cont)

- Driver mutations are the mutations we would like to target and inhibit their function

- **Passenger mutations-these mutations are "collateral damage" resulting from genomic instability and are not required for maintaining the transformed phenotype, therefore are "noise" in the system**
- **Since most cancers are rapidly evolving biologic entities, it is a major task to sort out "drivers" from "passengers", and these may change over time**

Signaling pathways are not 1-way

- **Driver mutations in signaling pathways (kinases) are components of highly integrated "wiring" that is not a one way flow of information**
  - Because these are critically important for normal cell functions, these are **highly regulated** pathways
- **Perturbation of a single component of will lead to activation of other components due to feedback activation or loss of feedback repression**

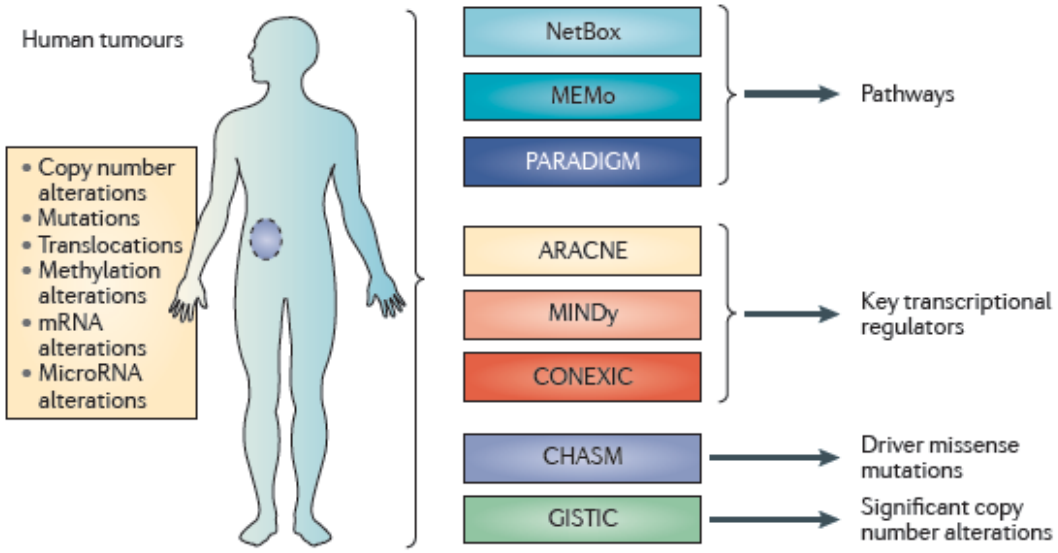
How many of these changes are meaningful?

How can we exploit these massive data sets to yield new targets for cancer therapy?

From cancer genomes to oncogenic drivers

emerging approaches

computational approaches



NetBox, MEMo, PARADIGM ,each seek to identify pathways that are deregulated in cancer and that are therefore likely to contain significant driver genes

ARACNE , MINDy, aim to identify the key transcriptional regulators of oncogenic programs and and CONEXIC also uses DNA copy number alterations to predict key transcriptional regulators.

CHASM attempts to predict which missense mutations are likely to drive tumorigenesis

GISTIC analyses copy number variations across tumour samples to predict which regions might contain driver genes

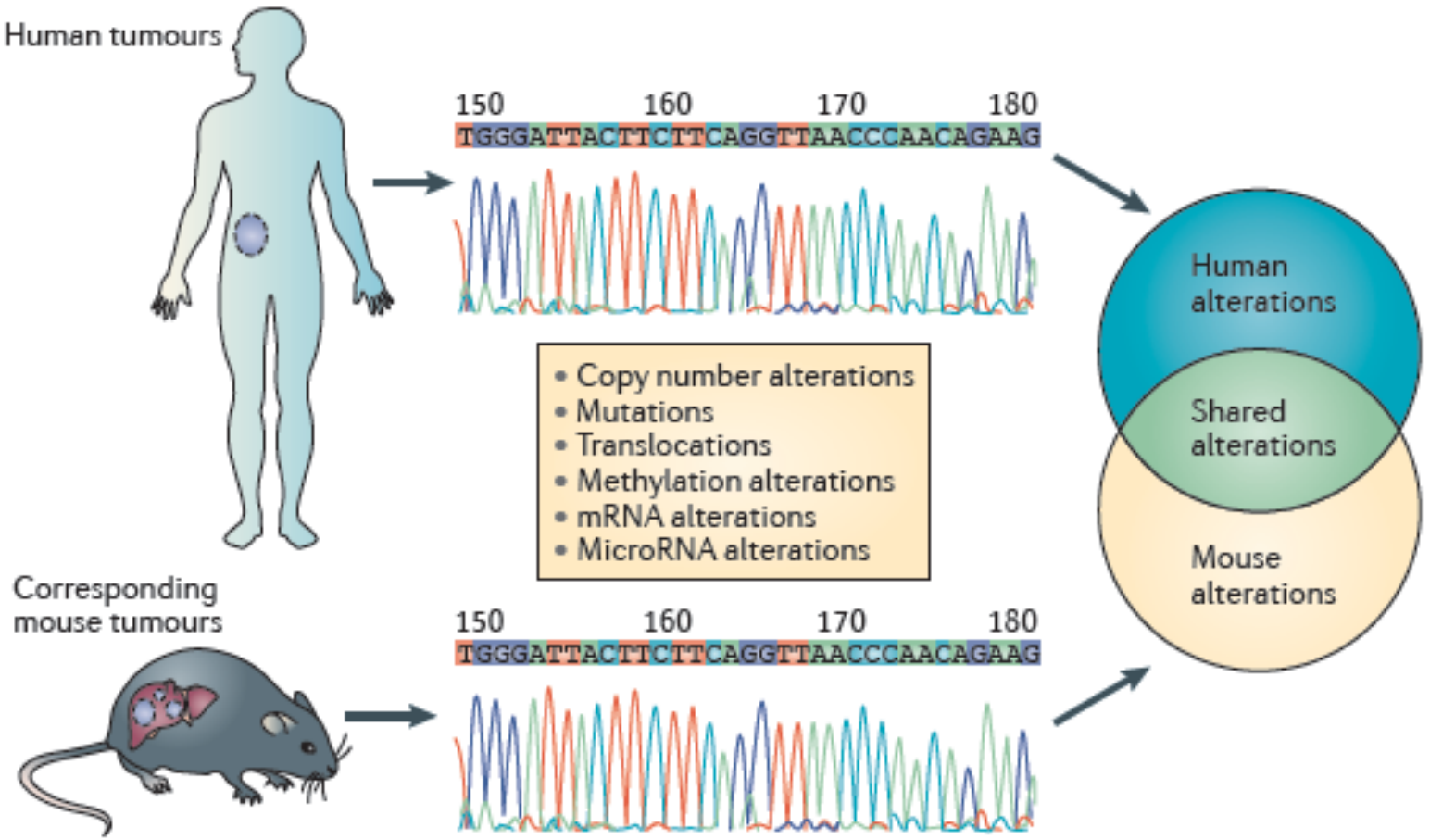
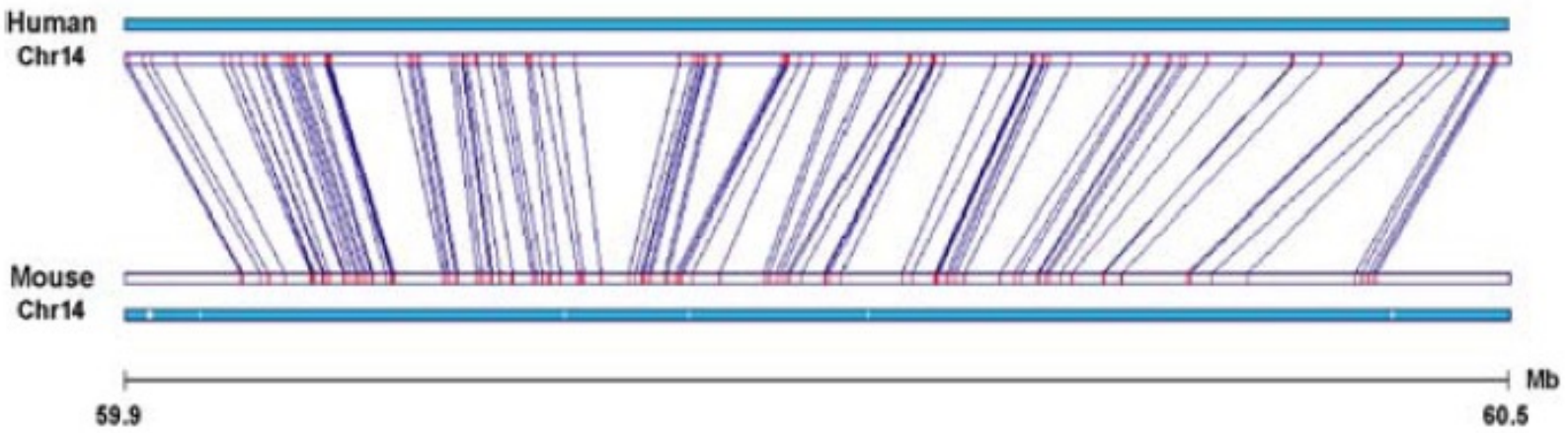


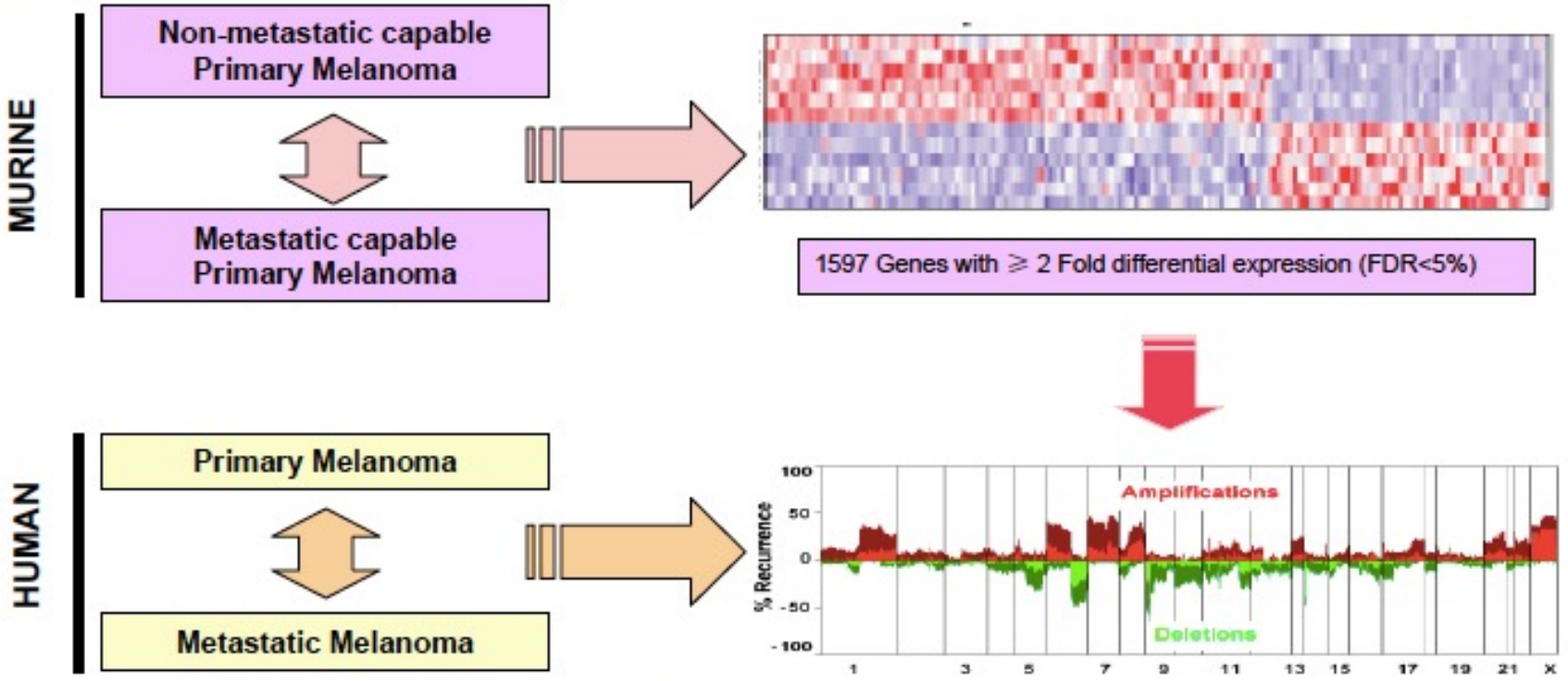
Figure 2 | **Cross-species comparative genomic approaches.** The central idea behind this approach is to improve the signal-to-noise ratio by comparing the genetic alterations that occur in a specific human cancer type with the genetic alterations that occur in a corresponding mouse model, be it genetically engineered or spontaneous. Passenger mutations are assumed to be randomly selected and not as likely to be found altered in both human and mouse tumours, whereas driver mutations will be selected for in both species and are therefore more likely to be shared. This approach has been used to compare point mutations, copy number alterations and expression changes.


The Mouse Genome
nature
Mouse Genome Sequencing Consortium, <date>

Regions of conserved synteny: ~95% of genome



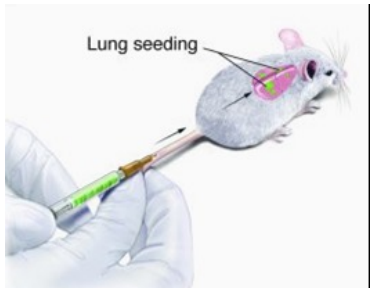
Triangulation across species and genome dimensions



Metastasis Determinants Signature

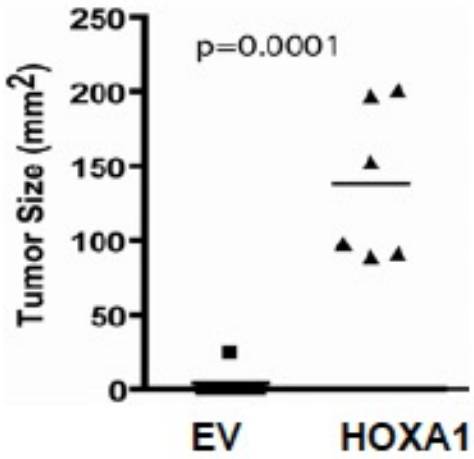
295 up-regulated/amplified **65** down-regulated/deleted

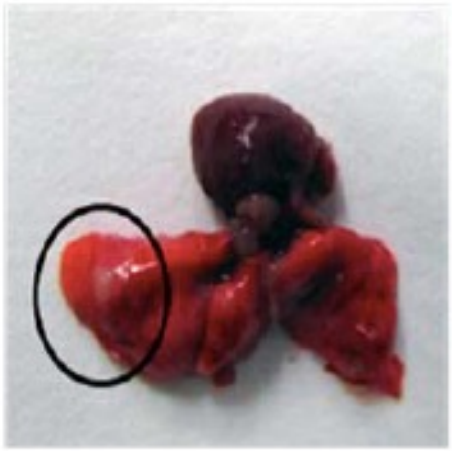
Validation of Metastasis Determinants

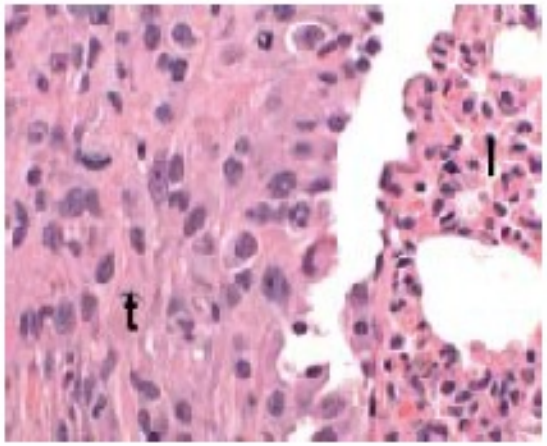


HOXA1 drives metastasis in vivo

Tail-vein

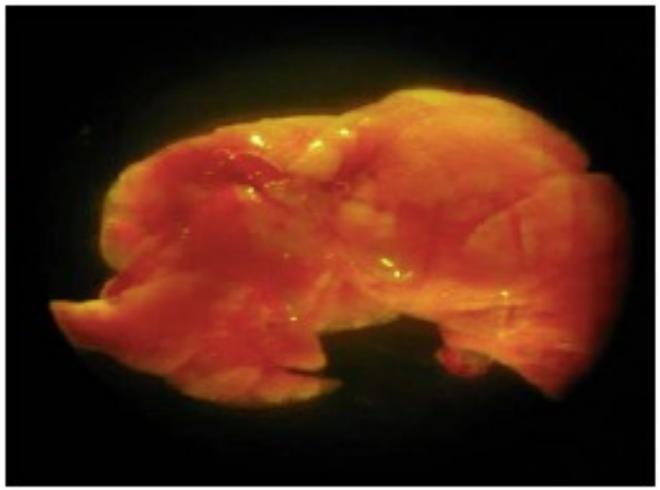




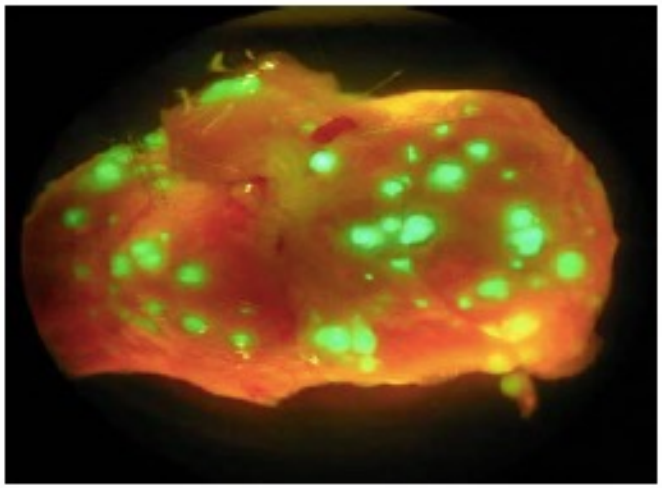


Distal Met from orthotopic site

EV

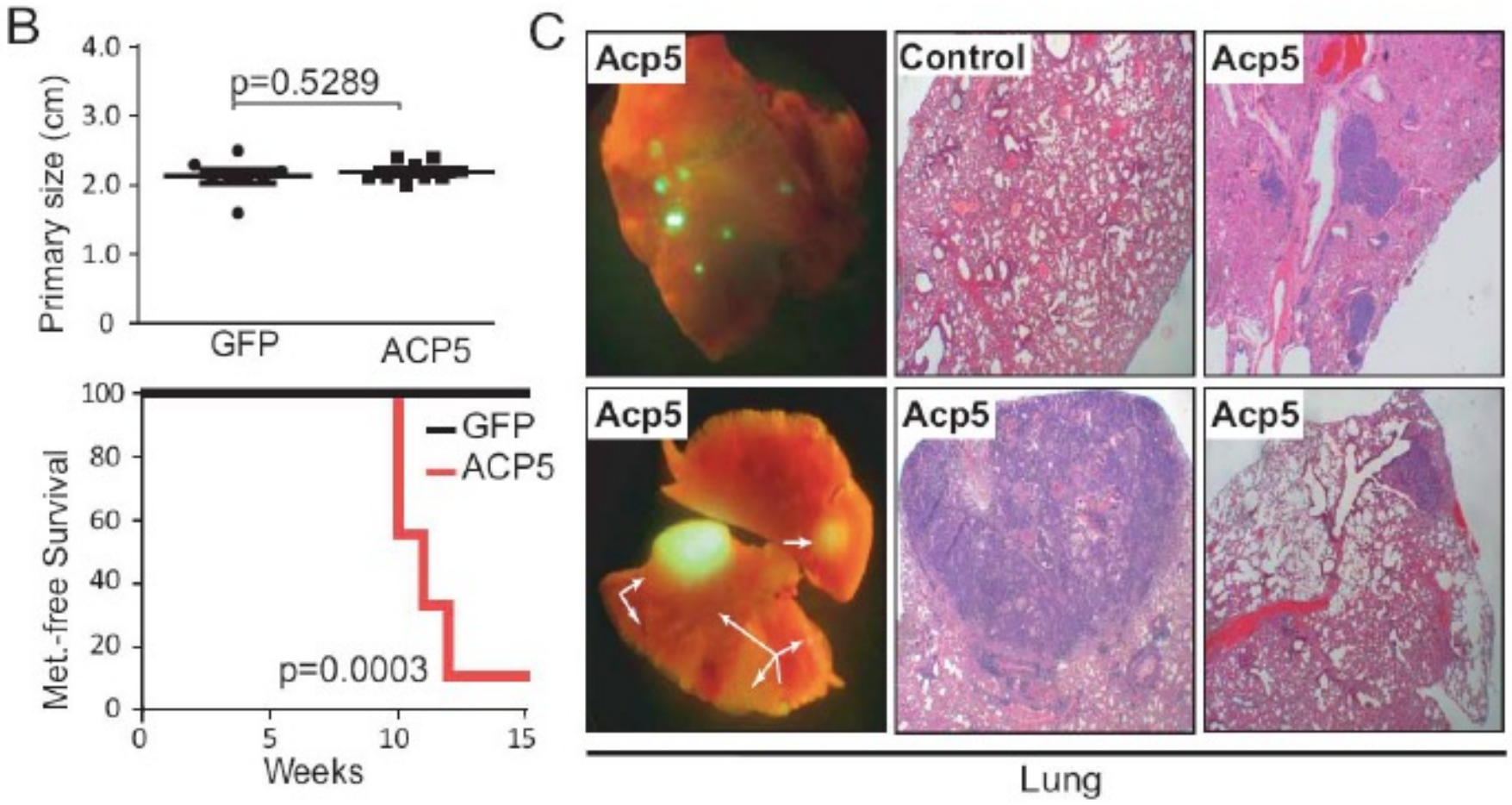


HOXA1



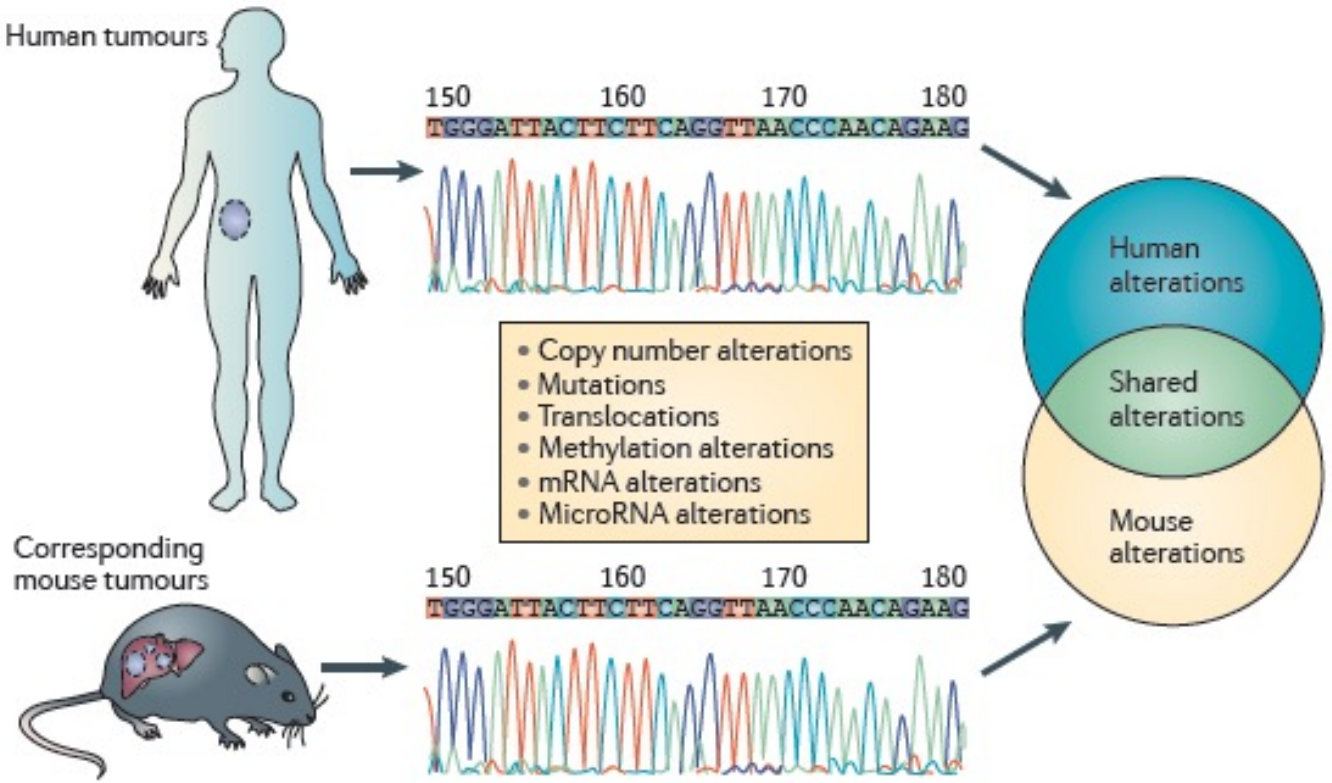
Kenneth Scott

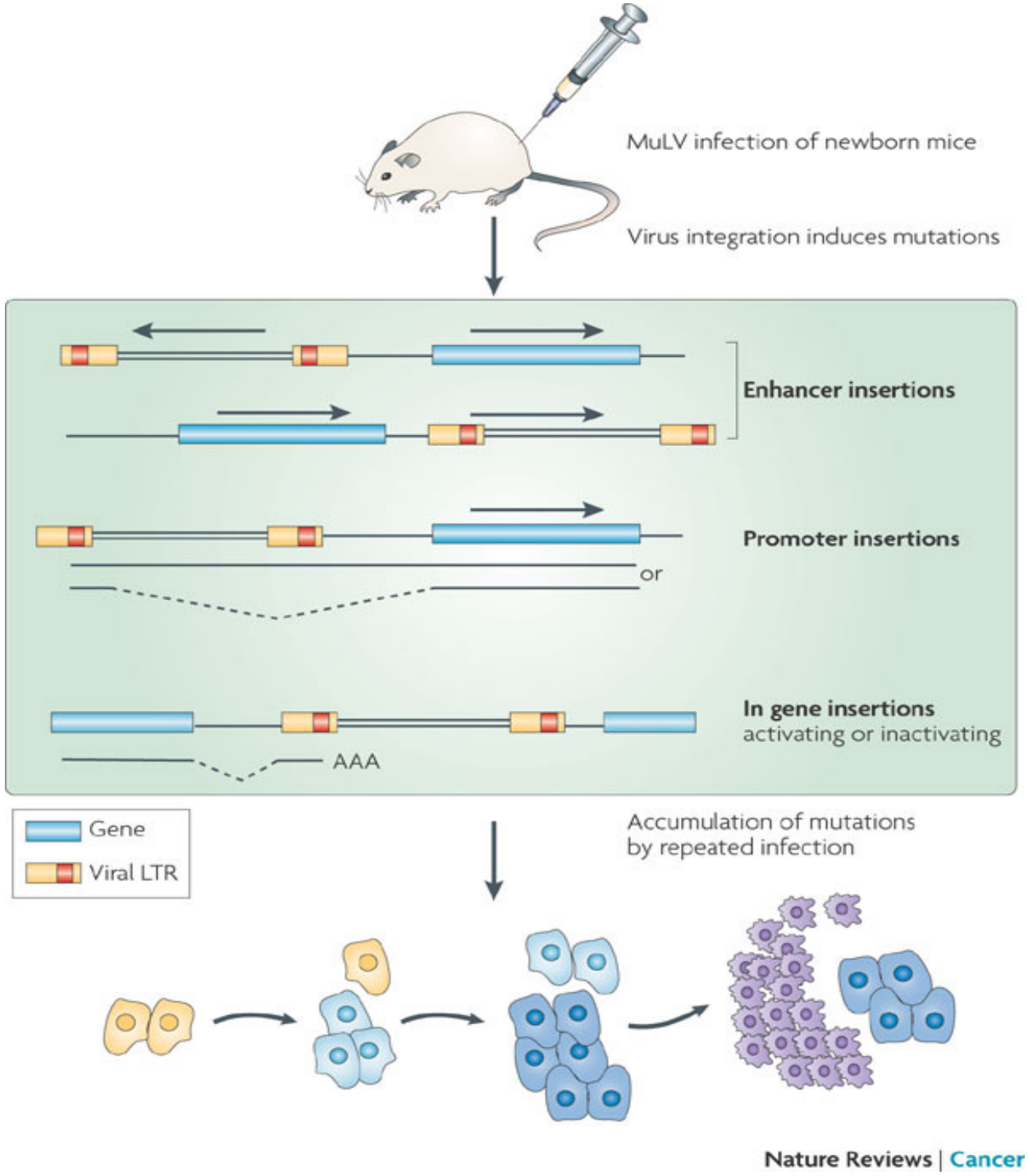
ACP5 drives metastasis in vivo

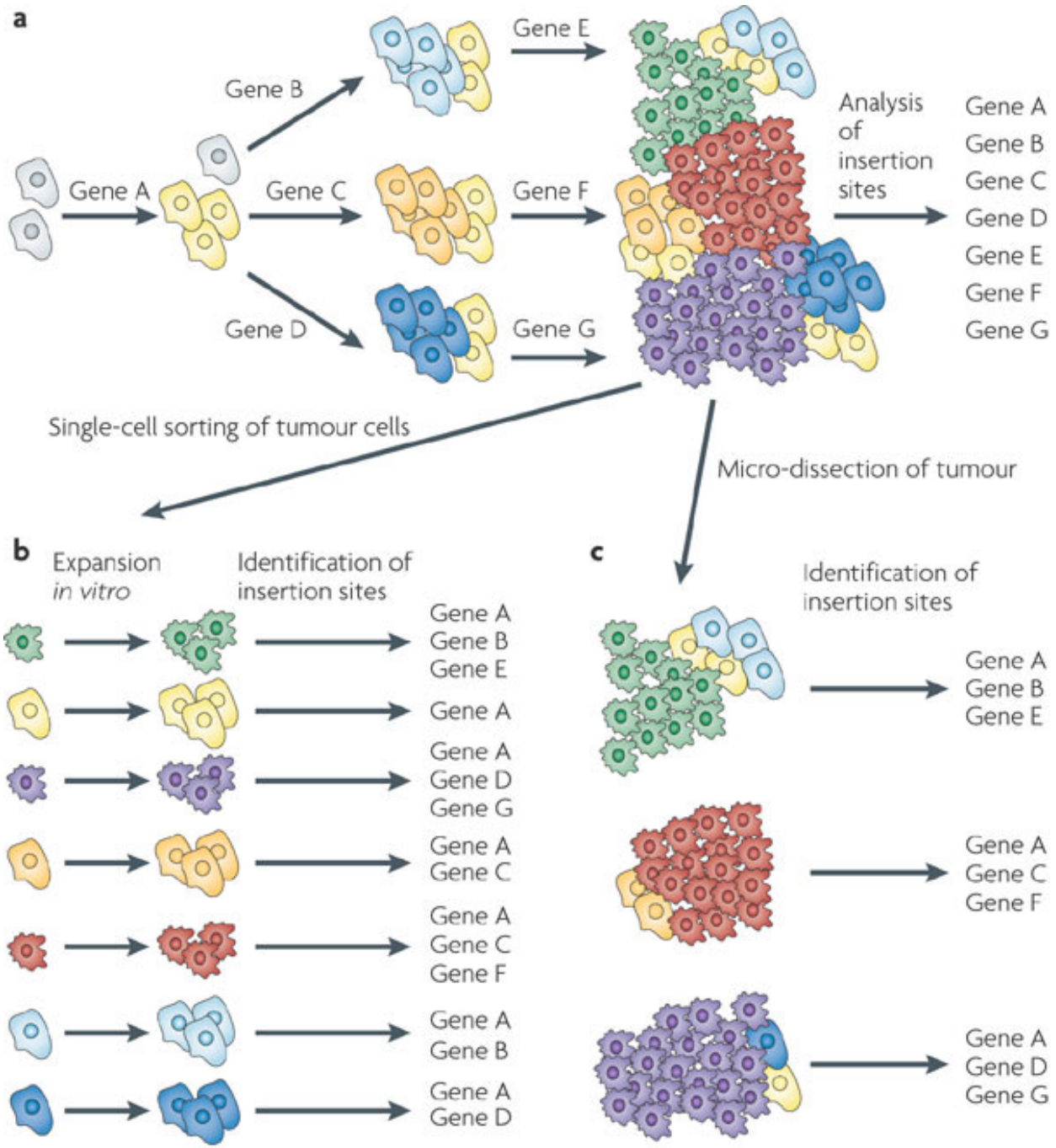


Kenneth Scott; Chengyin Min

insertional mutagenesis screens




MuLV infection of newborn mice
Virus integration induces mutations
Enhancer insertions
Promoter insertions
or
In gene insertions
activating or inactivating
AAA
Gene
Viral LTR
Accumulation of mutations
by repeated infection


a
Gene A
Gene B
Gene C
Gene D
Gene E
Gene F
Gene G
Analysis of insertion sites
Gene A
Gene B
Gene C
Gene D
Gene E
Gene F
Gene G
Single-cell sorting of tumour cells
Micro-dissection of tumour
b
Expansion in vitro
Identification of insertion sites
Gene A
Gene B
Gene E
Gene A
Gene A
Gene D
Gene G
Gene A
Gene C
Gene A
Gene C
Gene F
Gene A
Gene B
Gene A
Gene D
c
Identification of insertion sites
Gene A
Gene B
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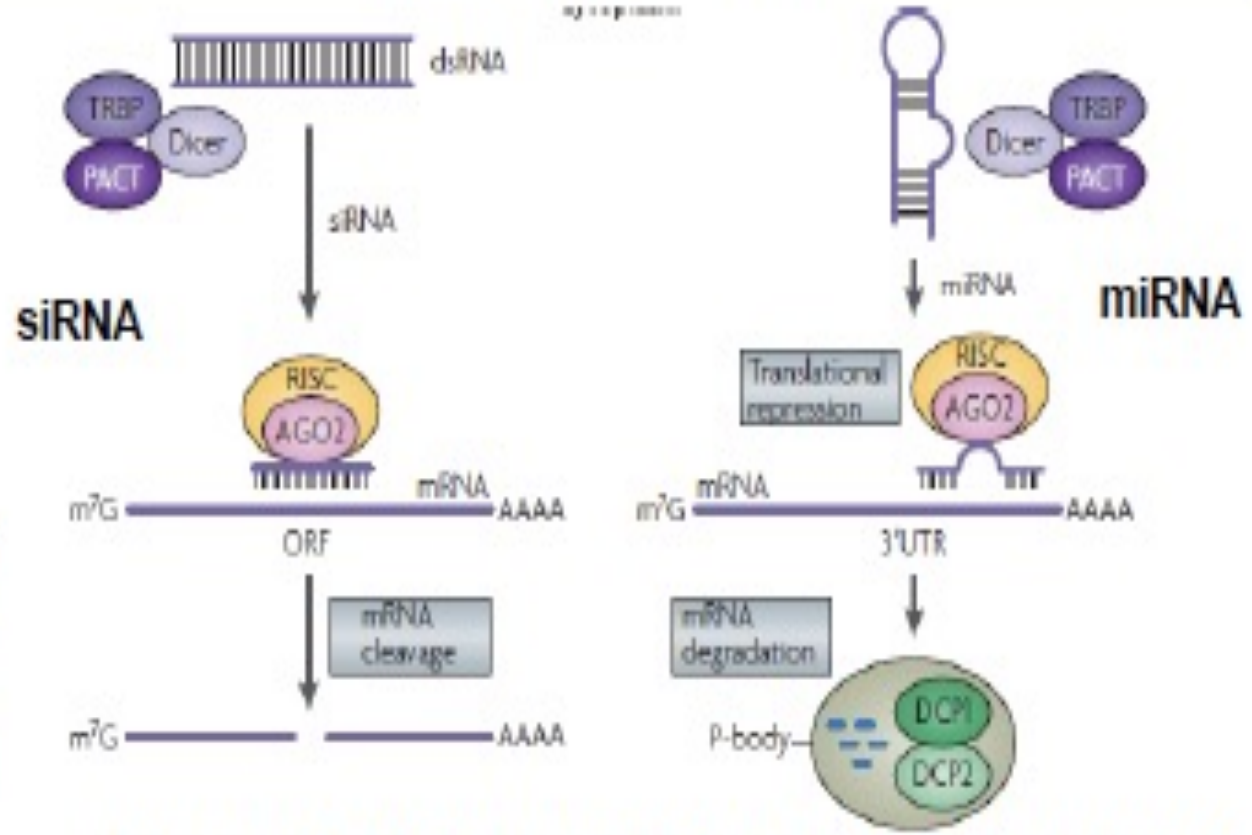
Gene A
Gene C
Gene F
Gene A
Gene D
Gene G

Loss-of-function techniques

Mutagenesis

RNAi

RNA transcripts can modify gene expression

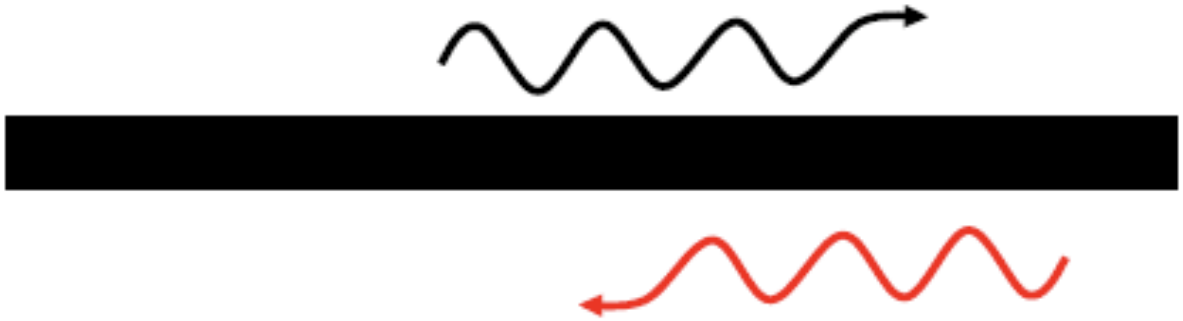


Kim and Rossi, Nature Rev Genet 2007

Processed double stranded RNAs (siRNA) hybridize to target genes and downregulate expression

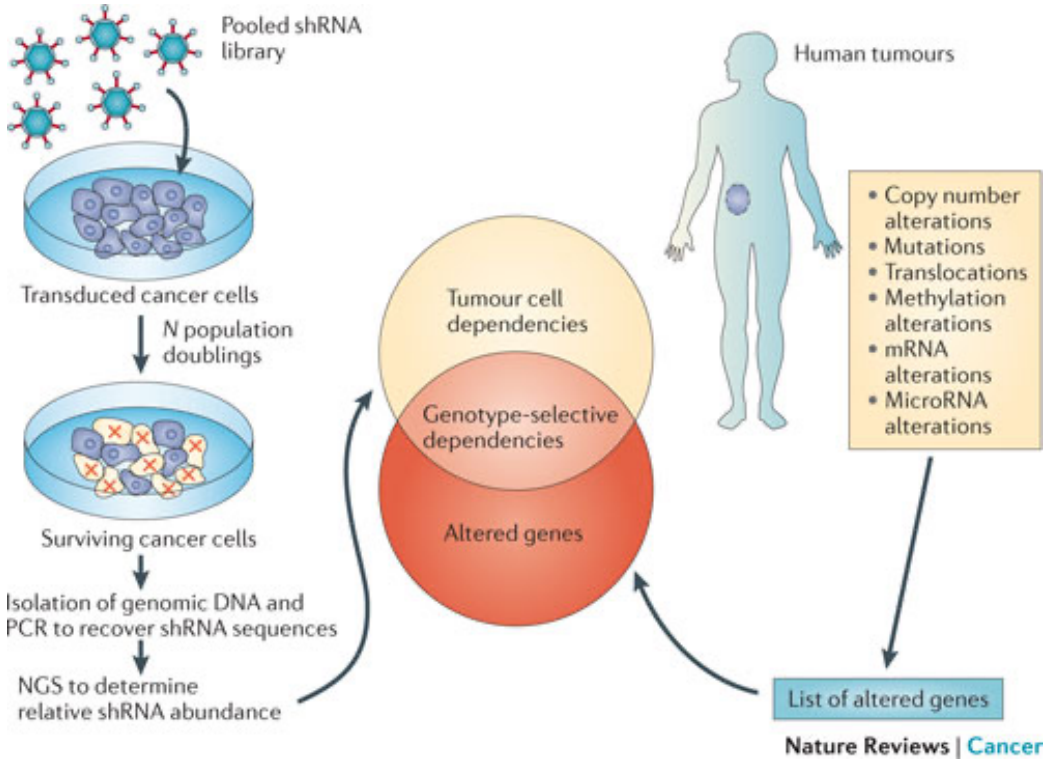
MicroRNAs important in development

RNA transcripts can modify gene expression

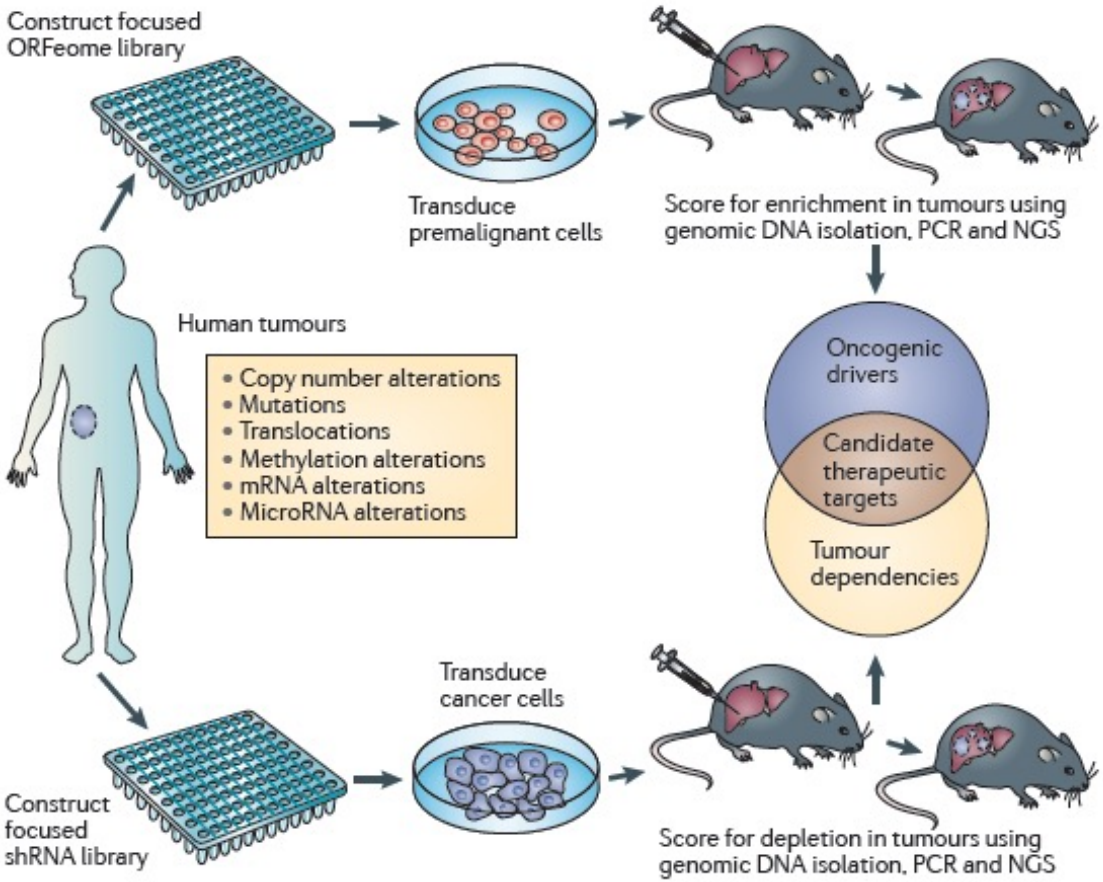


Antisense RNA

Whole-genome RNA interference screens



cancer-genome-focused screening



Exploration often gives a different perspective

Earthrise from Apollo 11, 1969

CANCER GENOMICS

A panoramic view of cancer

The Pan-Cancer Initiative of The Cancer Genome Atlas (TCGA) has now taken the next step — comparative genomic analyses across the 12 cancer types for which genomic data have so far been generated.

In a coordinately published set of papers in Nature, Nature Genetics and other journals, the Pan-Cancer group has analysed up to 5,000 individual cancers, including cancers of the breast, uterus, ovaries, lung, brain, head and neck, colon and rectum, bladder, kidney and blood. Owing to the large sample sizes, the analyses are impressively highly powered and provide a range of insights.

focused on point mutations and small insertions and deletions (indels) from 3,281 tumours across the 12 tumour types to identify 127 significantly mutated genes. These genes are involved in a wide range of cellular processes.

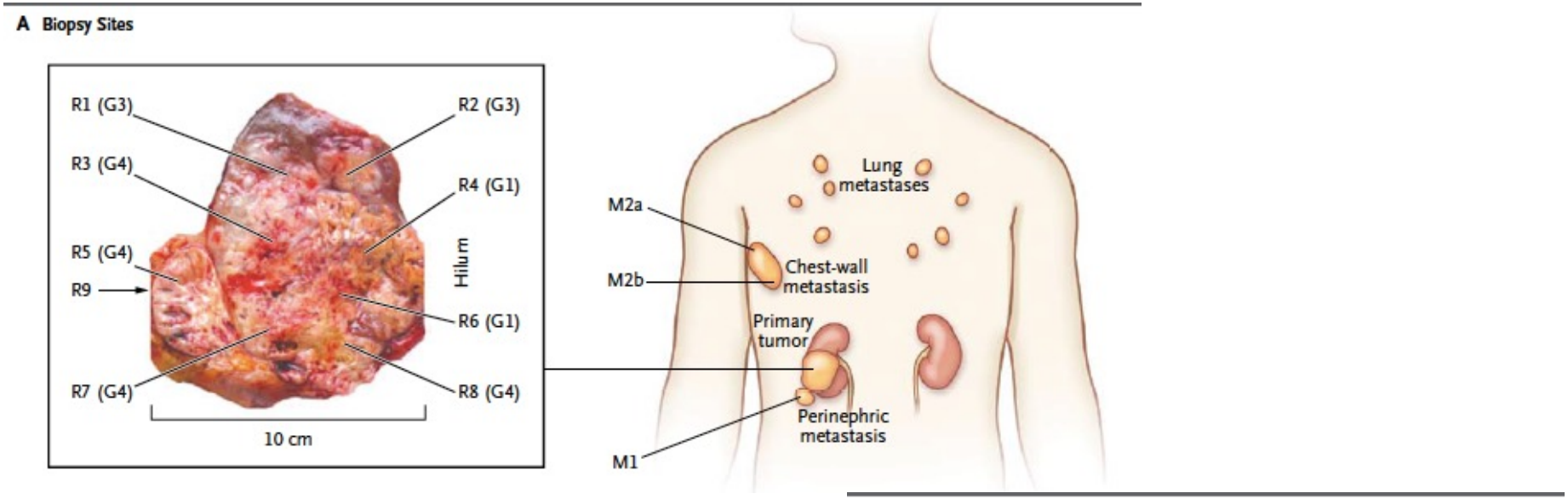
Human Cancer Genome Project

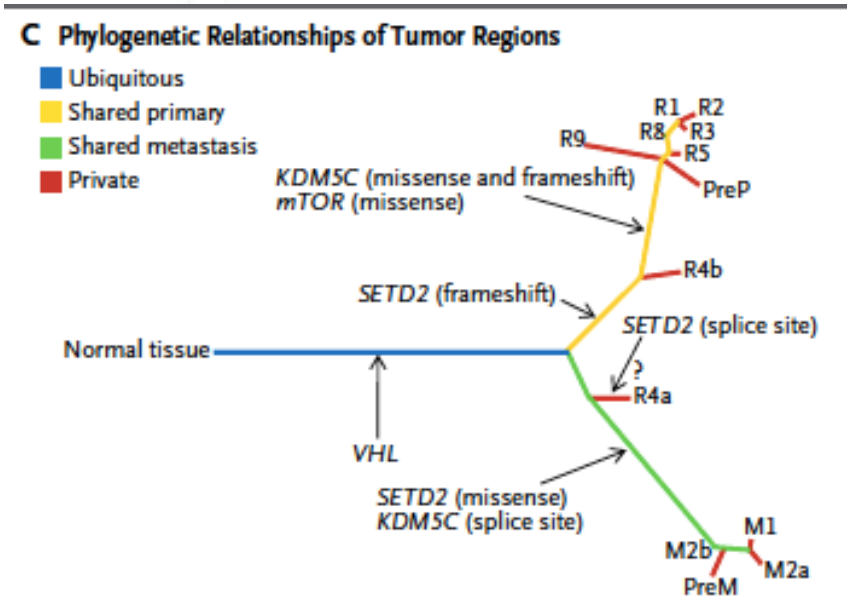
NCI Task Force

Find *all* genomic alterations *significantly associated* (5%) with *all* major types of cancer

- Genomic loss and amplification analysis
- Mutation detection in all human genes
- Chromosomal rearrangements
- Epigenomic analysis
- For sensitivity and specificity, ~250 tumors per cancer

Cancer Genomics informs on clonal evolution





Vol 463|14 January 2010|doi:10.1038/nature08658

nature

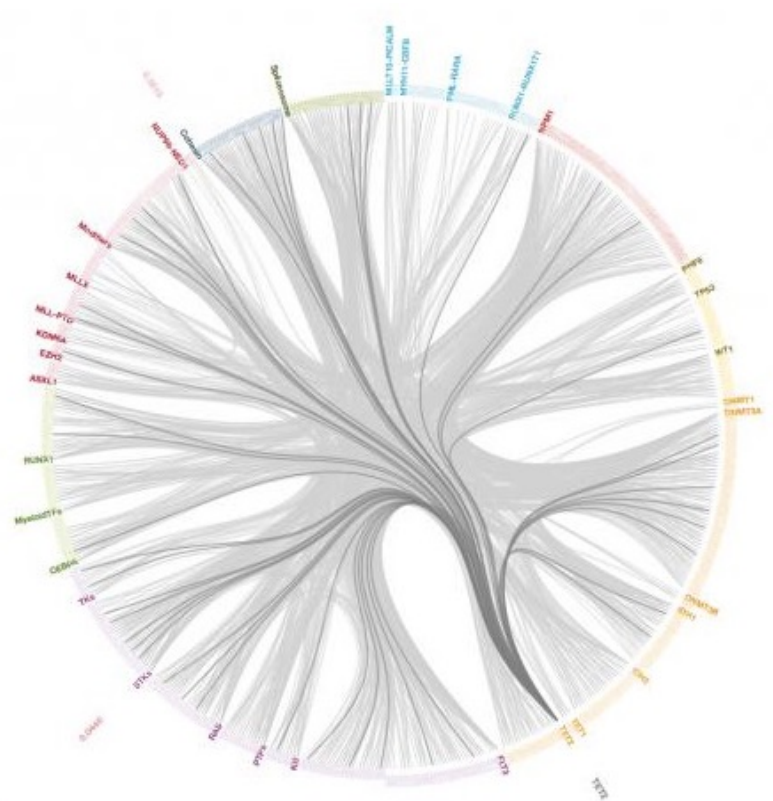
ARTICLES

A comprehensive catalogue of somatic mutations from a human cancer genome

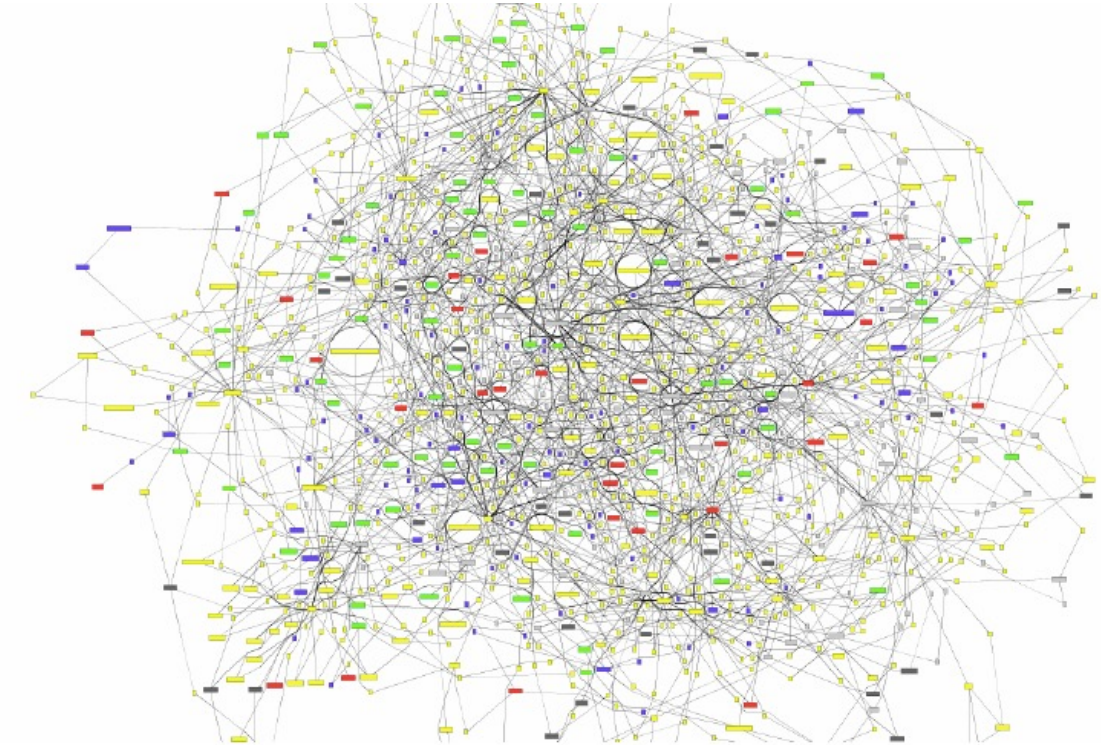
Erin D. Pleasance[1]*, R. Keira Cheetham[2]*, Philip J. Stephens[1], David J. McBride[1], Sean J. Humphray[2], Chris D. Greenman[1], Ignacio Varela[1], Meng-Lay Lin[1], Gonzalo R. Ordóñez[1], Graham R. Bignell[1], Kai Ye[3], Julie Alipaz[4], Markus J. Bauer[2], David Beare[1], Adam Butler[1], Richard J. Carter[2], Lina Chen[1], Anthony J. Cox[2], Sarah Edkins[1], Paula I. Kokko-Gonzales[2], Niall A. Gormley[2], Russell J. Grocock[2], Christian D. Haudenschild[5], Matthew M. Hims[2], Terena James[2], Mingming Jia[1], Zoya Kingsbury[2], Catherine Leroy[1], John Marshall[1], Andrew Menzies[1], Laura J. Mudie[1], Zemin Ning[1], Tom Royce[4], Ole B. Schulz-Trieglaff[2], Anastassia Spiridou[2], Lucy A. Stebbings[1], Lukasz Szajkowski[2], Jon Teague[1], David Williamson[5], Lynda Chin[6], Mark T. Ross[2], Peter J. Campbell[1], David R. Bentley[2], P. Andrew Futreal[1] & Michael R. Stratton[1,7]

All cancers carry somatic mutations. A subset of these somatic alterations, termed driver mutations, confer selective growth advantage and are implicated in cancer development, whereas the remainder are passengers. Here we have sequenced the genomes of a malignant melanoma and a lymphoblastoid cell line from the same person, providing the first comprehensive catalogue of somatic mutations from an individual cancer. The catalogue provides remarkable insights into the forces that have shaped this cancer genome. The dominant mutational signature reflects DNA damage due to ultraviolet light exposure, a known risk factor for malignant melanoma, whereas the uneven distribution of mutations across the genome, with a lower prevalence in gene footprints, indicates that DNA repair has been preferentially deployed towards transcribed regions. The results illustrate the power of a cancer genome sequence to reveal traces of the DNA damage, repair, mutation and selection processes that were operative years before the cancer became symptomatic.

Cancer possess myriad mutations that cooperate to maintain tumor survival

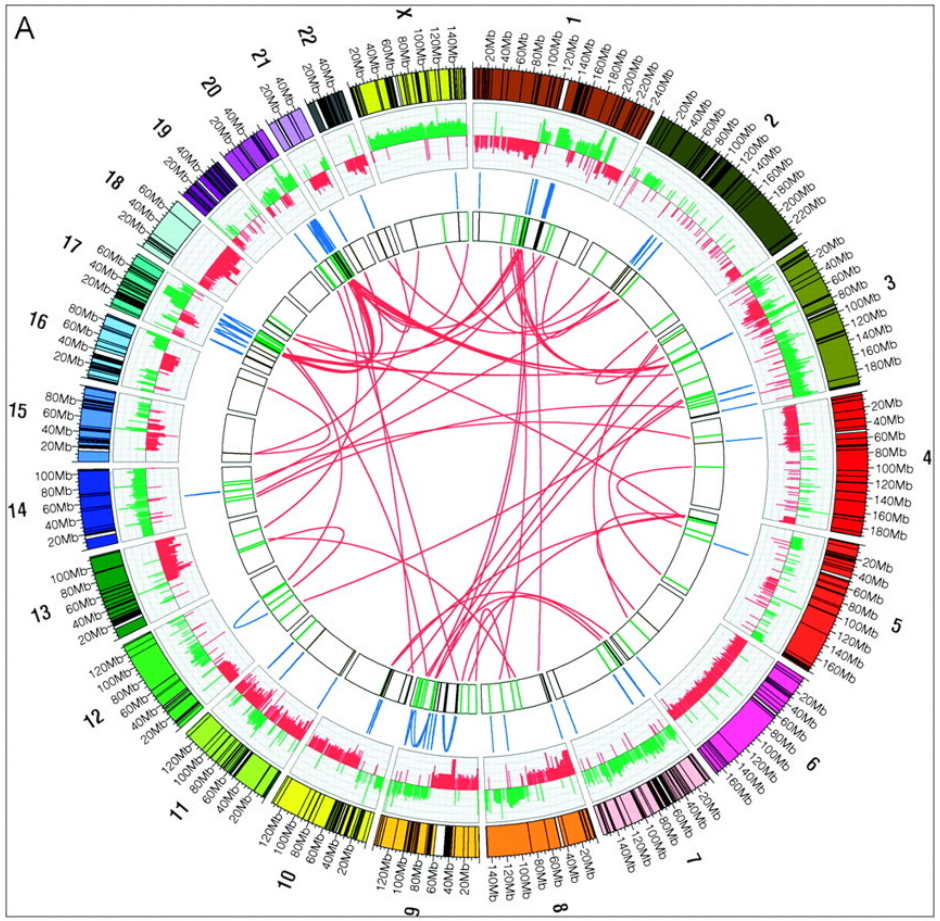


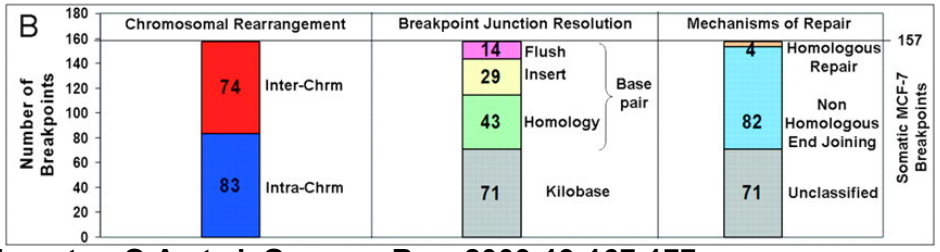
An interactive catalog of genetic mutations
The genetic profile of a given cancer can involve mutations of different genes in different patients. Dendrix, a powerful algorithm, can search enormous datasets for associations of genetic mutations, any one of might cause disease.
Credit: Department of Computer Science

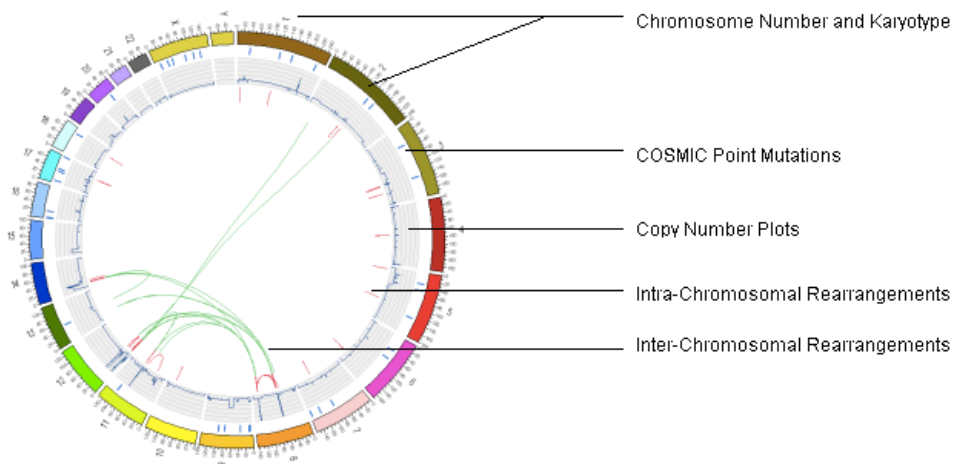


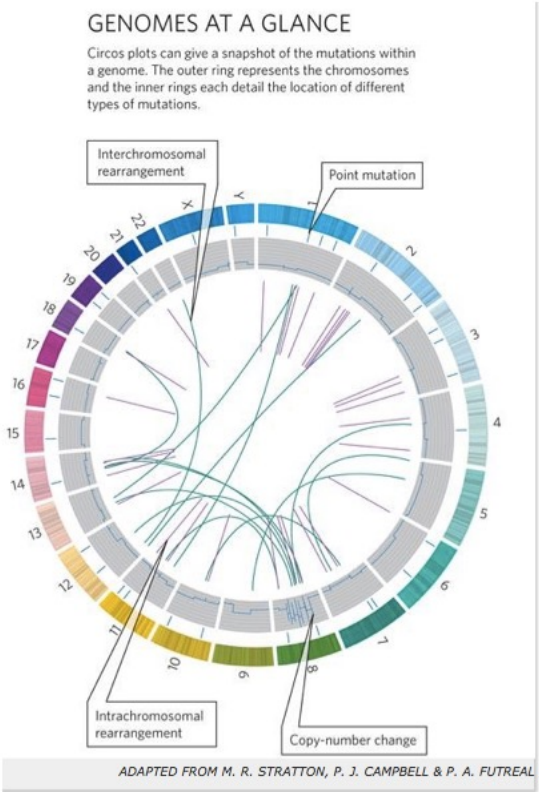
The Cancer Genome Atlas Research Network
N Engl J Med 2013; 368:2059-2074 | May 30, 2013 |

(A) Circular visualization of the MCF-7 genome obtained using Circos software.









Hampton O A et al. Genome Res. 2009;19:167-177

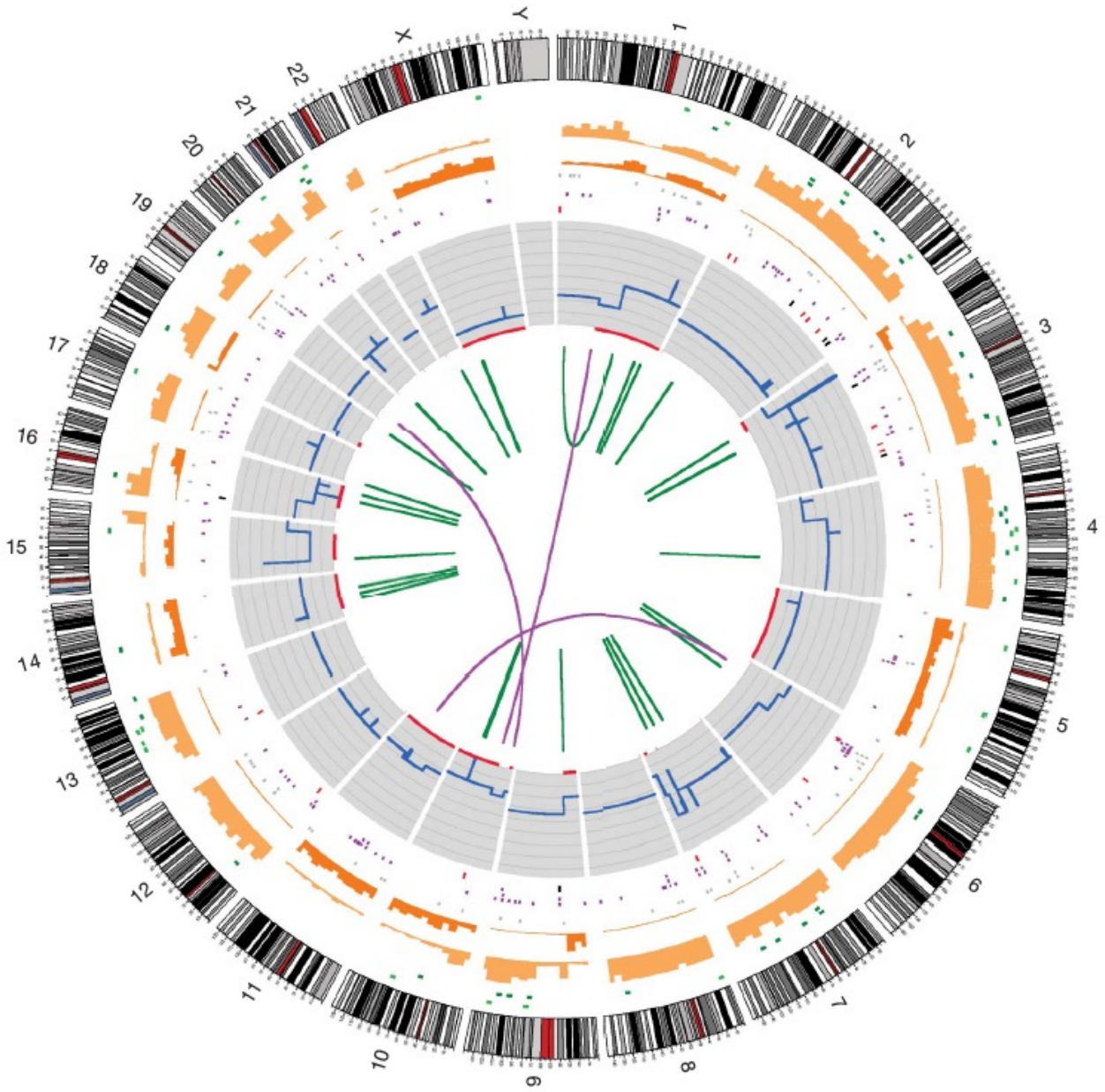


Figure 1 | The catalogue of somatic mutations in COLO-829. Chromosome ideograms are shown around the outer ring and are oriented pter–qter in a clockwise direction with centromeres indicated in red. Other tracks contain somatic alterations (from outside to inside): validated insertions (light-green rectangles); validated deletions (dark-green rectangles); heterozygous (light-orange bars) and homozygous (dark-orange bars) substitutions shown by density per 10 megabases; coding substitutions (coloured squares: silent in grey, missense in purple, nonsense in red and splice site in black); copy number (blue lines); regions of LOH (red lines); validated intrachromosomal rearrangements (green lines); validated interchromosomal rearrangements (purple lines).

Table 2. Selection of databases commonly used in our workflows.

Database	Entities	Properties
Ensembl	Genes, proteins, transcripts, regulatory regions, variants	Genomic positions, relationships between them, identifiers in different formats, GO terms, PFAM domains
Entrez	Genes, articles	Articles for genes, abstracts of articles, links to full text
UniProt	Proteins	PDBs, known variants
KEGG, Reactome, Biocarta, Gene Ontology	Genes	Pathways, processes, function, cell location
TFacts	Genes	Transcription regulation
Barcode	Genes	Expression by tissue
PINA, HPRD, STRING	Proteins	Interactions
PharmaGKB	Drugs, proteins, variants	Drug targets, pharmacogenetics
STITCH, Matador	Drugs, proteins	Drug targets
Drug clinical trials	Investigational drugs	Diseases or conditions in they are being tested
GEO, ArrayExpress	Genes (microarray probes)	Expression values
ICGC, TCGA	Cancer Genomes	Point mutations, methylation, CNV, structural variants
dbSNP, 1000 genomes	Germline variations	Association with diseases or conditions
COSMIC	Somatic variations	Association with cancer types

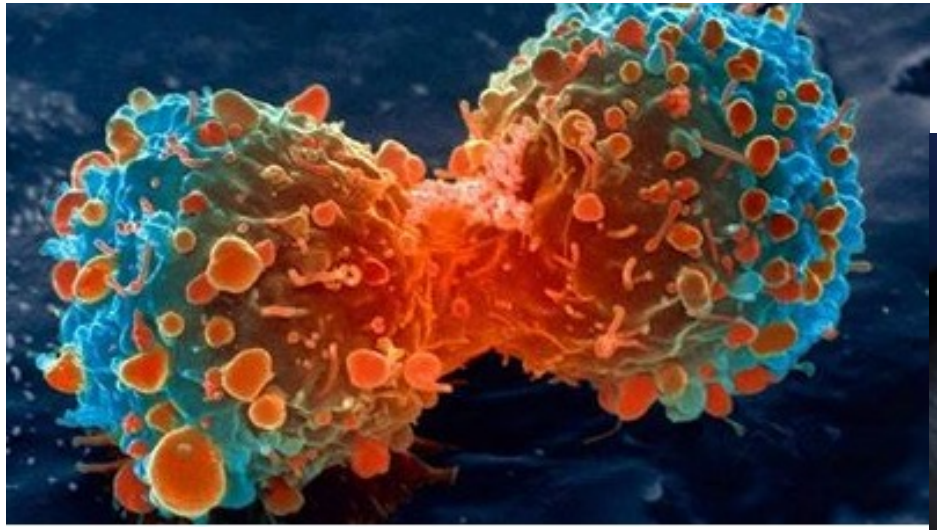
doi:10.1371/journal.pcbi.1002824.t002

Vazquez M, de la Torre V, Valencia A (2012) Chapter 14: Cancer Genome Analysis. PLOS Computational Biology 8(12): e1002824. https://doi.org/10.1371/journal.pcbi.1002824
http://journals.plos.org/ploscompbiol/article?id=10.1371/journal.pcbi.1002824

National Cancer Institute

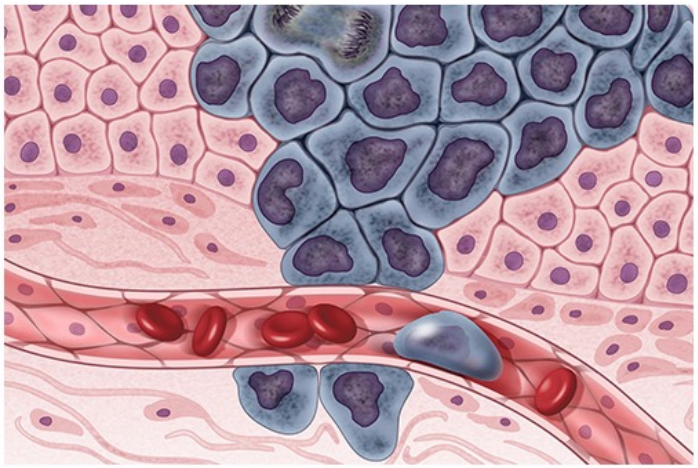
The Cancer Genome Atlas

Understanding genomics to improve cancer care



THE CANCER GENOME ATLAS

The Cancer Genome Atlas (TCGA)



Growing cancer cells (in purple) are surrounded by healthy cells (in pink), illustrating a primary tumor spreading to other parts of the body through the circulatory system. Image credit: Darryl Leja, NHGRI.]

The Molecular Characteristics of Breast Cancer

33 cancer types, catalogs all the key genomic changes - the modifications in DNA, RNA and proteins that cause the uncontrolled cell growth that is the hallmark of malignant tumors.

Today, the TCGA network published a paper analyzing the molecular characteristics of breast cancer by African and European Ancestry. This paper identifies molecular differences by genetic ancestry, most of which can be captured by known breast cancer subtypes.

International Cancer Genome Consortium

ICGC

Cancer Genomic Projects

International Cancer Genome Consortium

The Cancer Genome Atlas 

OBJECTIVE:

Obtain full catalog of genetic alterations in 500 tumors from 50 tumor types

- Somatic mutations
- Copy Number Alterations
- Abnormal expression of genes
- Translocations
- Epigenetic modifications
- etc.

Childhood Cancer Genomics (PDQ®)

There are examples of genomic lesions that have provided immediate therapeutic direction, including the following:

NPM-ALK fusion genes associated with anaplastic large cell lymphoma cases.

ALK point mutations associated with a subset of neuroblastoma cases.

BRAF and other kinase genomic alterations associated with subsets of pediatric glioma cases.

Hedgehog pathway mutations associated with a subset of medulloblastoma cases.

ABL family genes activated by translocation in a subset of acute lymphoblastic leukemia (ALL) cases.

Acute Lymphoblastic Leukemia (ALL)
Genomics of childhood ALL

The genomics of childhood ALL has been extensively investigated, and multiple distinctive subtypes have been defined on the basis of cytogenetic and molecular characterizations, each with its own pattern of clinical and prognostic characteristics. Figure 1 illustrates the distribution of ALL cases by cytogenetic/molecular subtype

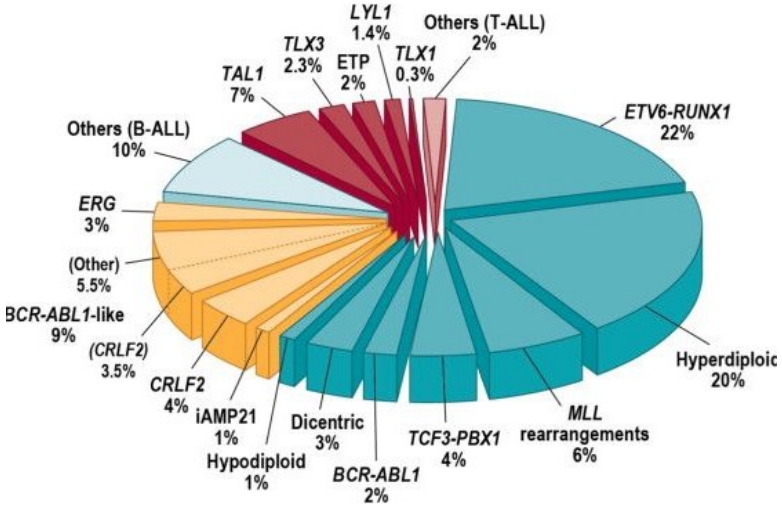
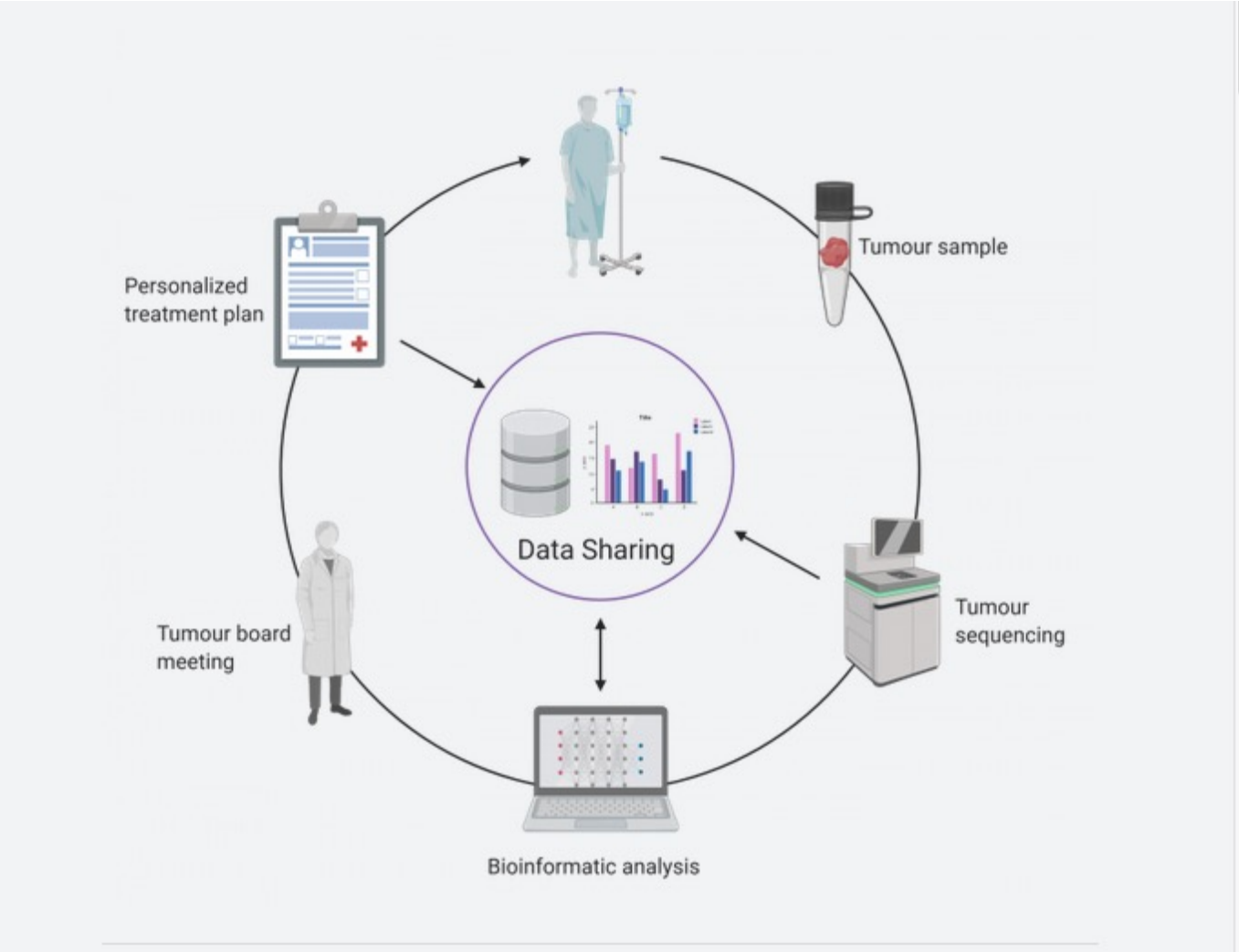
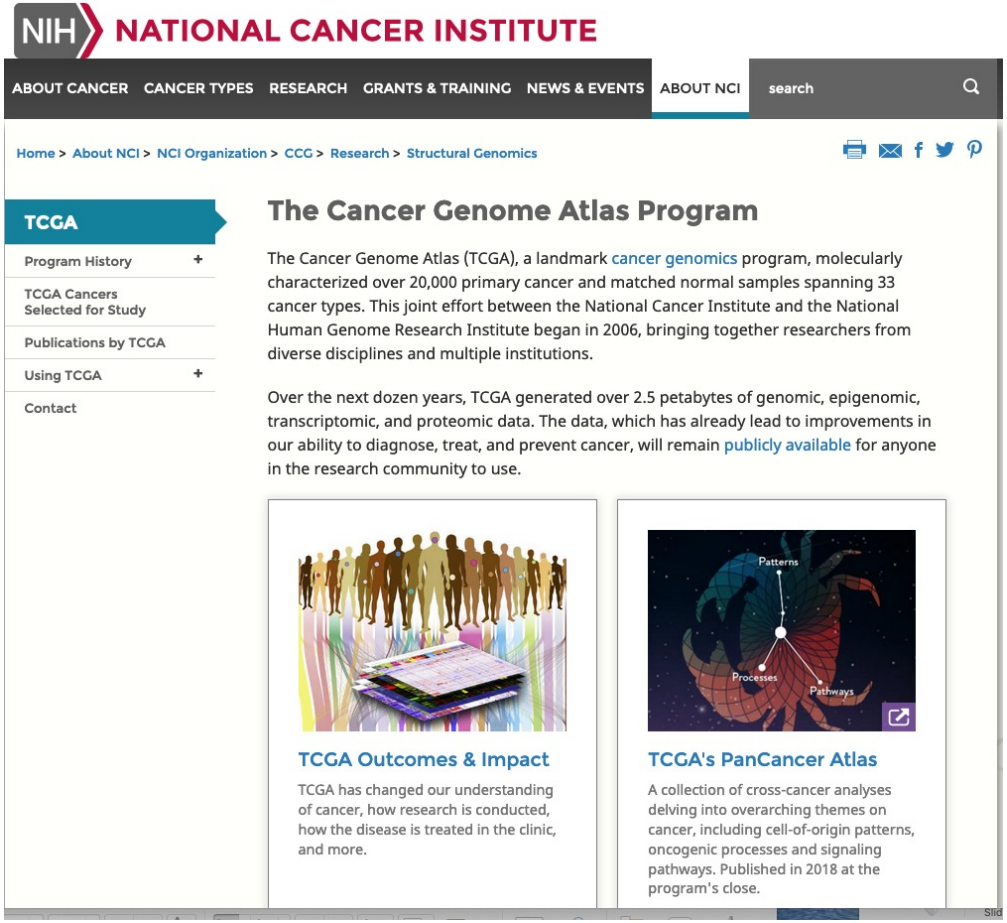


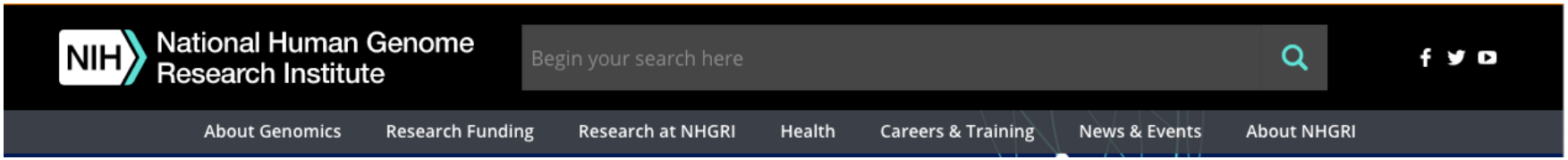
Figure 1. Subclassification of childhood ALL. Blue wedges refer to B-progenitor ALL, yellow to recently identified subtypes of B-ALL, and red wedges to T-lineage ALL. Reprinted from Seminars in Hematology, Volume 50, Charles G. Mullighan, Genomic Characterization of Childhood Acute Lymphoblastic Leukemia, Pages 314–324, Copyright (2013), with permission from Elsevier.

The Personalized OncoGenomics Program



Characterizing Cancer Genomes





TARGET's Resources have been reorganized into TARGET Resources and TARGET Tutorials. Links to TARGET-related resources have been moved to Helpful Links.

TARGET: Therapeutically Applicable Research To Generate Effective Treatments

The Therapeutically Applicable Research to Generate Effective Treatments (TARGET) program applies a comprehensive genomic approach to determine molecular changes that drive childhood cancers. The goal of the program is to use data to guide the development of effective, less toxic therapies. TARGET is organized into a collaborative network of disease-specific project teams.

VIEW TARGET DATA MATRIX

The TARGET Data Matrix enables the cancer research community to search and download data generated by the Initiative.

View Using TARGET Data Page

CCG

Cancer Genomics Overview

Research

Structural Genomics

Functional Genomics

Computational Genomics

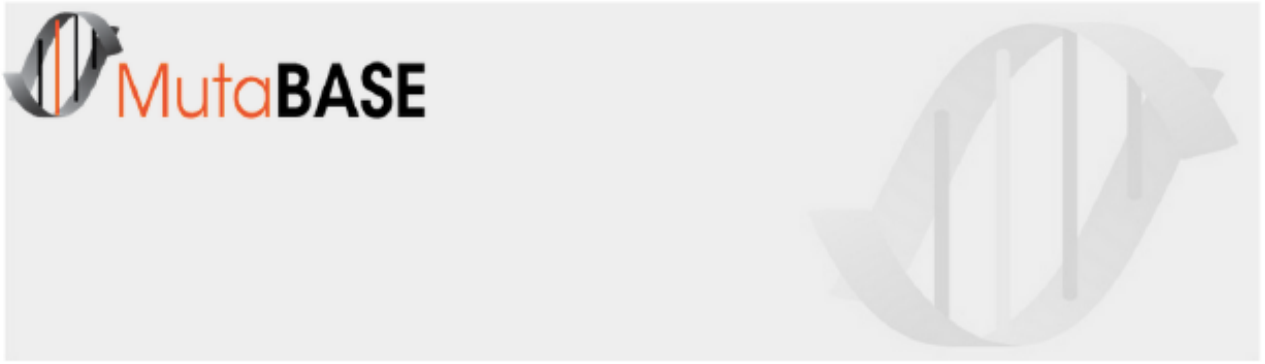
Genome Characterization Pipeline

Funding Opportunities

NCI's Genome Characterization Pipeline

NCI's Center for Cancer Genomics (CCG) coordinates research teams across the United States and Canada to produce rich cancer genomic and clinical datasets for the cancer research community. CCG implements this collaborative effort through an efficient and standardized workflow called the Genome Characterization Pipeline. Learn more about how it works below.

ON THIS PAGE



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MutaREPORTER
- launch platform

MutaCIRCLES
- launch platform

MutaBASE is a bioinformatics company focusing on human molecular genetics.

MutaBASE produces MutaREPORTER, a software package to retrieve, define and archive DNA variations in human DNA leading to genetic disease.

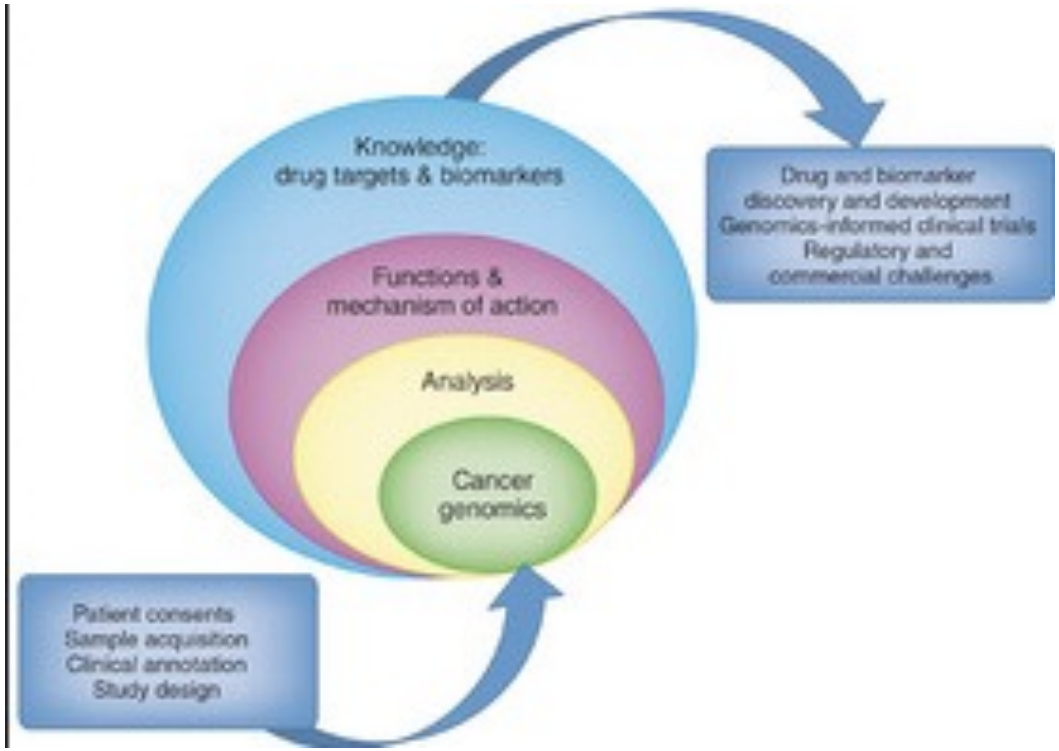
MutaBASE organises MutaCIRCLES, groups of genetic labs offering a particular gene test that share information regarding that gene test.

Latest News

Apr 7, 2011: Announcing the public release of the Beta version of MutaREPORTER

The Beta version of the MutaREPORTER software, a software package to retrieve, define and archive DNA variations in human DNA leading to genetic disease, has been released to a small number of try-out diagnostic labs. These labs will help to improve MutaREPORTER and start populating MutaDATABASE with DNA variants. The release of the commercial version of the MutaREPORTER software is expected for June.

Translating the Cancer Genome:



The genome will inform the right target and the right patients for the right drugs, ONLY when interpreted in context of the biology

HOW HAS THIS CHANGED CANCER TREATMENT?

How personalized medicine can change breast cancer treatment

Landmarks in breast cancer history:

2013: - Deep genomic analysis identified several genetic mutations that might make promising targets for new therapies. Every month a new possible target mutation

Combination of drugs approved for other types of cancers were tried and found effective to breast cancer

- there is a movement towards re-classification of cancers based on molecular profile vs traditional classification based on tissue of origin

How personalized medicine can change breast cancer treatment

Conclusions:

breast cancer patients

armed with genomic information, may now feel comfortable forgoing certain treatments that might not be right for their unique case

How personalized medicine can change breast cancer treatment

Questions that researchers need to answer the years to come include:

Why do some cancers react to certain drugs and not to others?

Why might a cancer become resistant to a drug?

How can we predict these outcomes and changes?

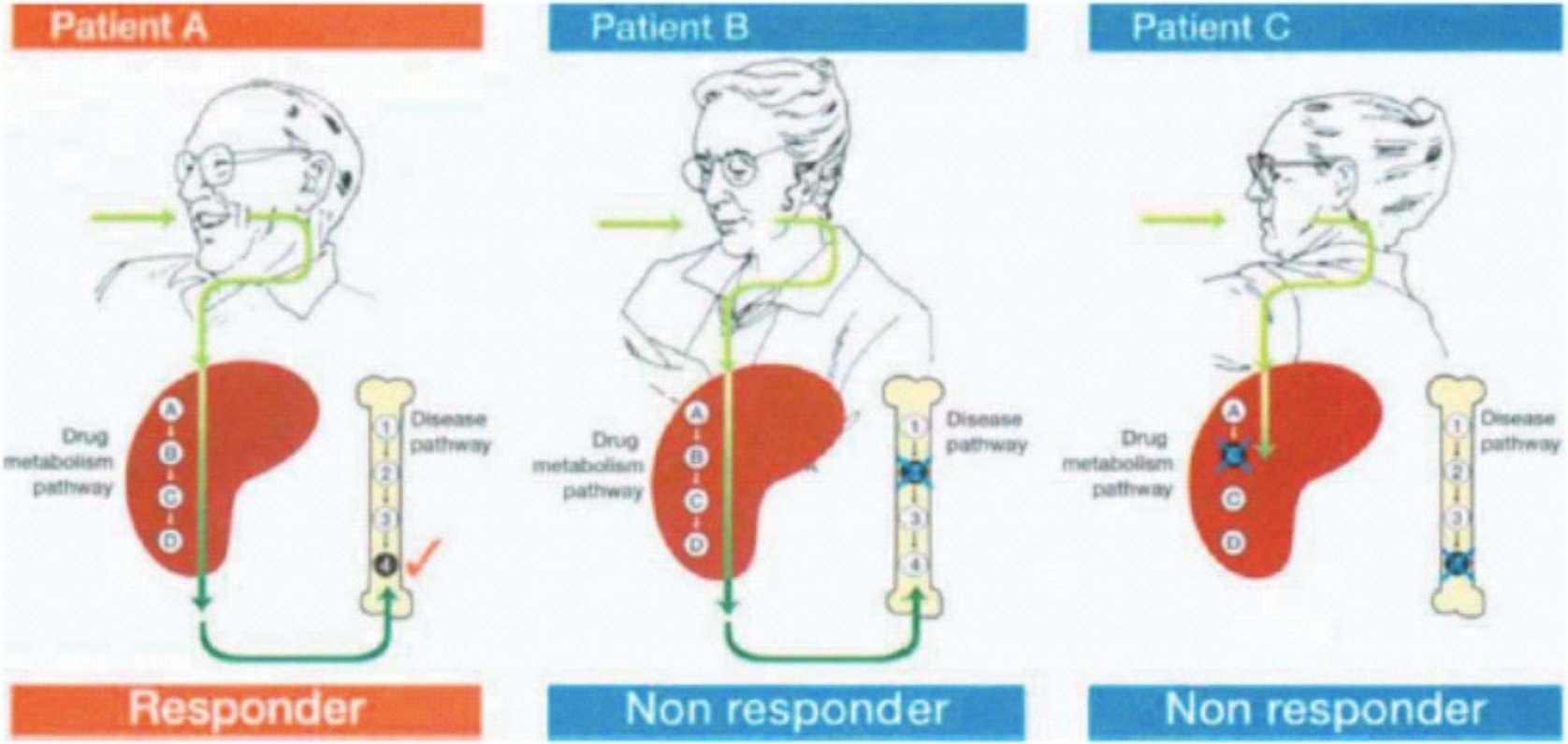
How do we choose a therapy that is just right for a particular patient?

What's next?

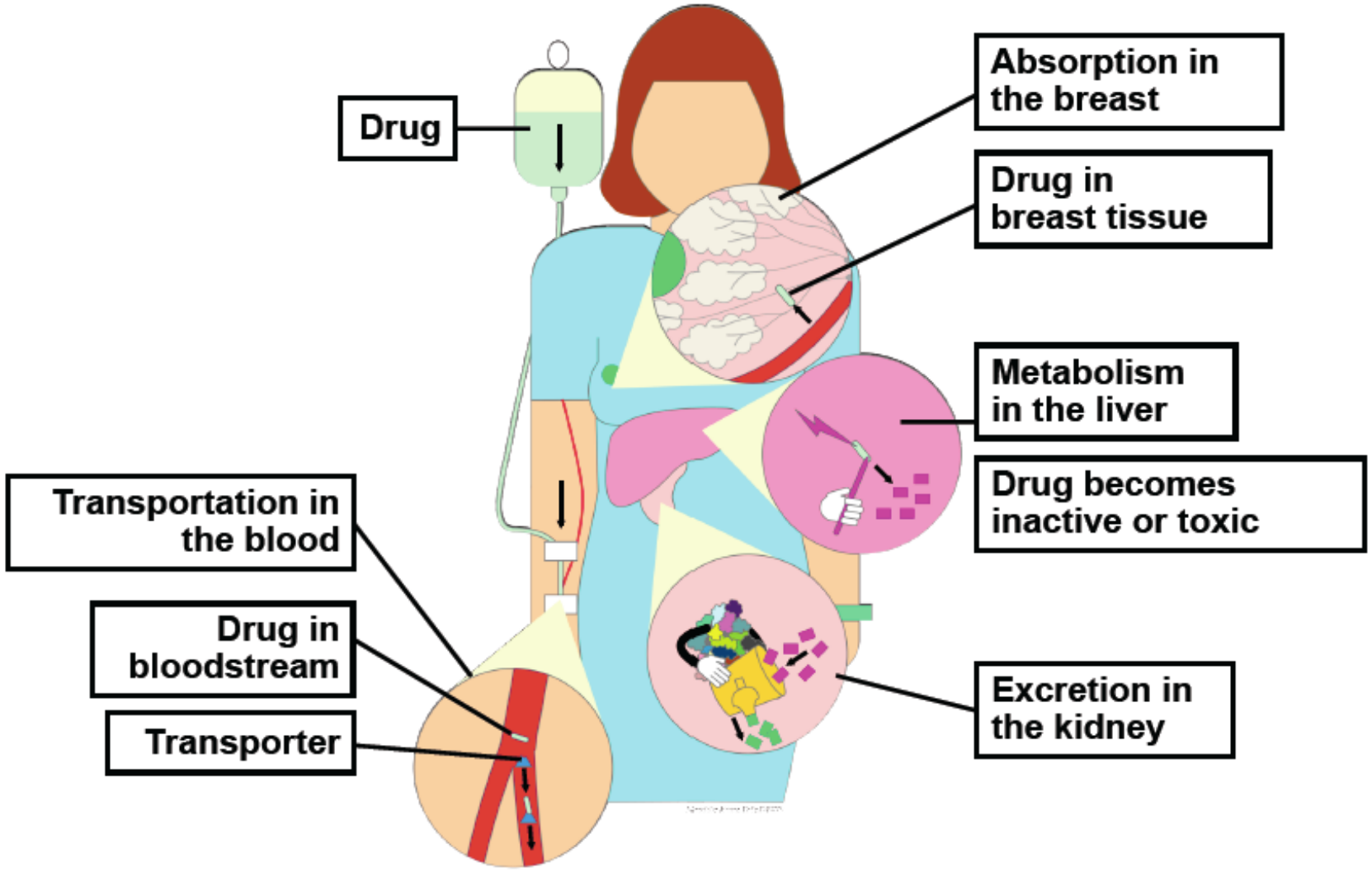
Patient A: metabolizes the drug correctly and has a form of osteoporosis caused by the disease gene targeted by drug

Patient B: a different disease gene which is insensitive to the drug causes osteoporosis

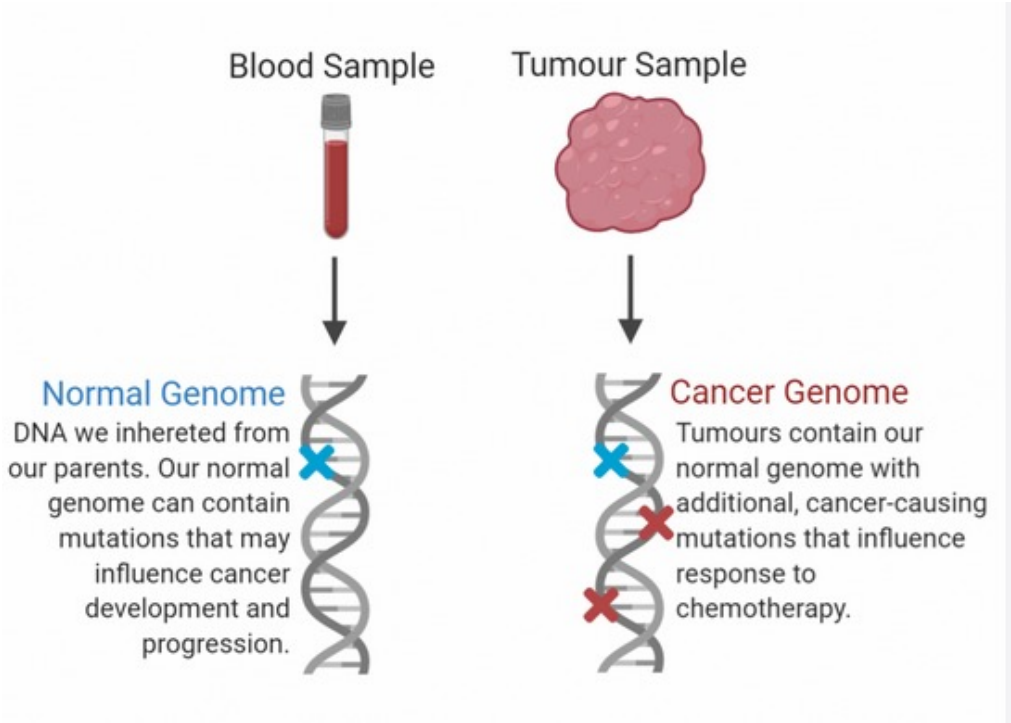
Patient C: defective metabolism pathway prevents conversion of the drug into its active form



SNPs and Drug Interactions



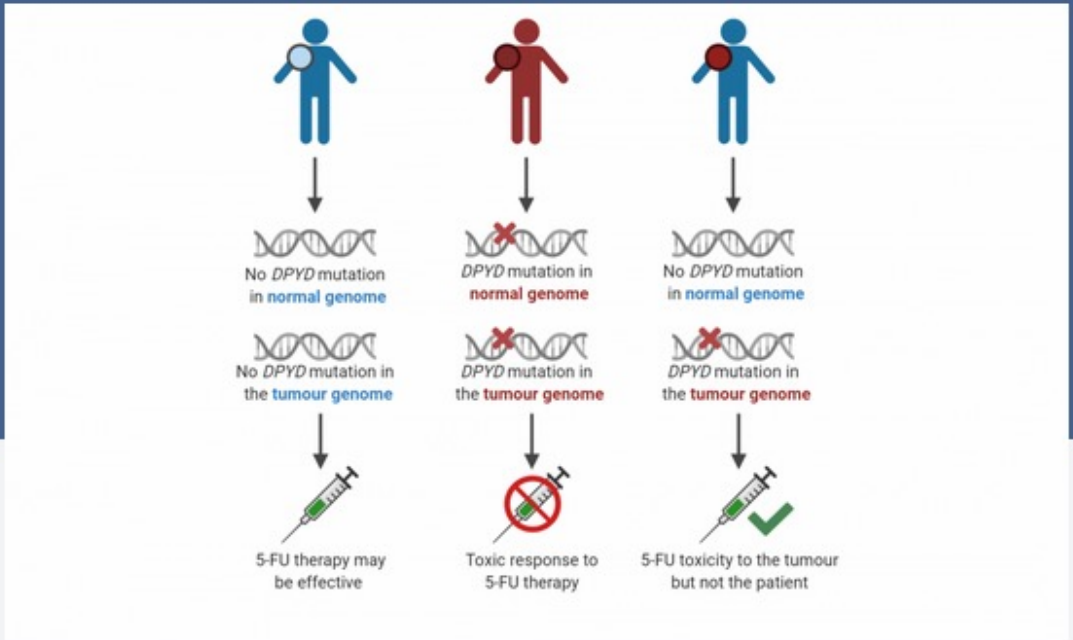
Genome sequencing helps prioritize cancer treatment options



Cancer Genomics

Jan 10, 2020

Genome sequencing helps prioritize cancer treatment options



5-Fluorouracil, commonly known as 5-FU, is a drug used for the treatment of multiple cancer types. For some patients, 5-FU can lead to toxicity. But by using the power of whole genome sequencing, scientists have shown that for other patients, 5-FU may be a potent double-edged sword for the treatment of cancer.

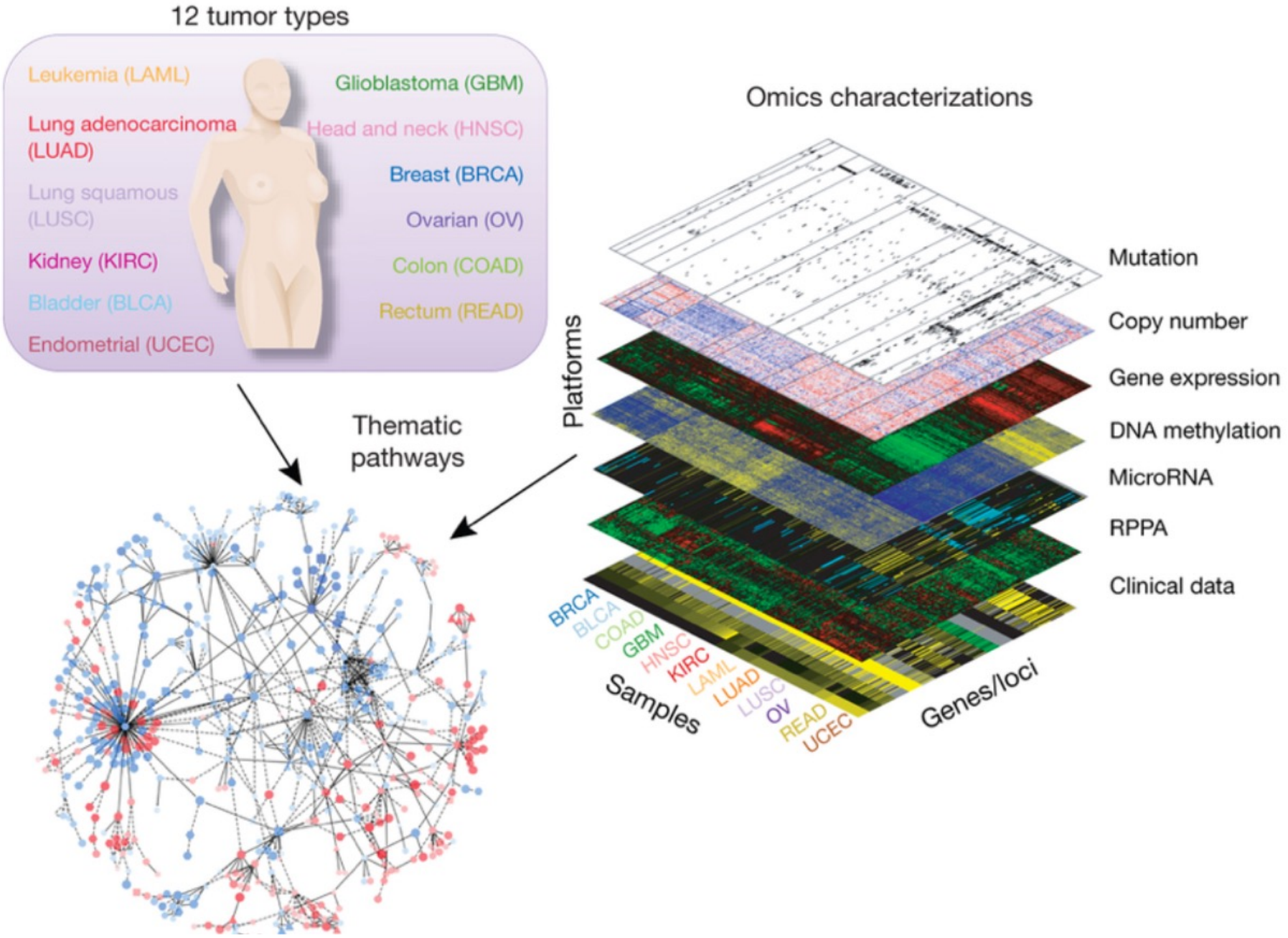
Conclusions

- **The ability to obtain full genomic data on a given tumor will allow us to make rational choices for therapy**
- **Functional genomics may provide help in choosing combination therapy**
  - Combinations will not be easy due to enhanced toxicities
- **Cancer as a chronic disease is not a bad thing as long as we recognize rapid development of resistance and clonal evolution**

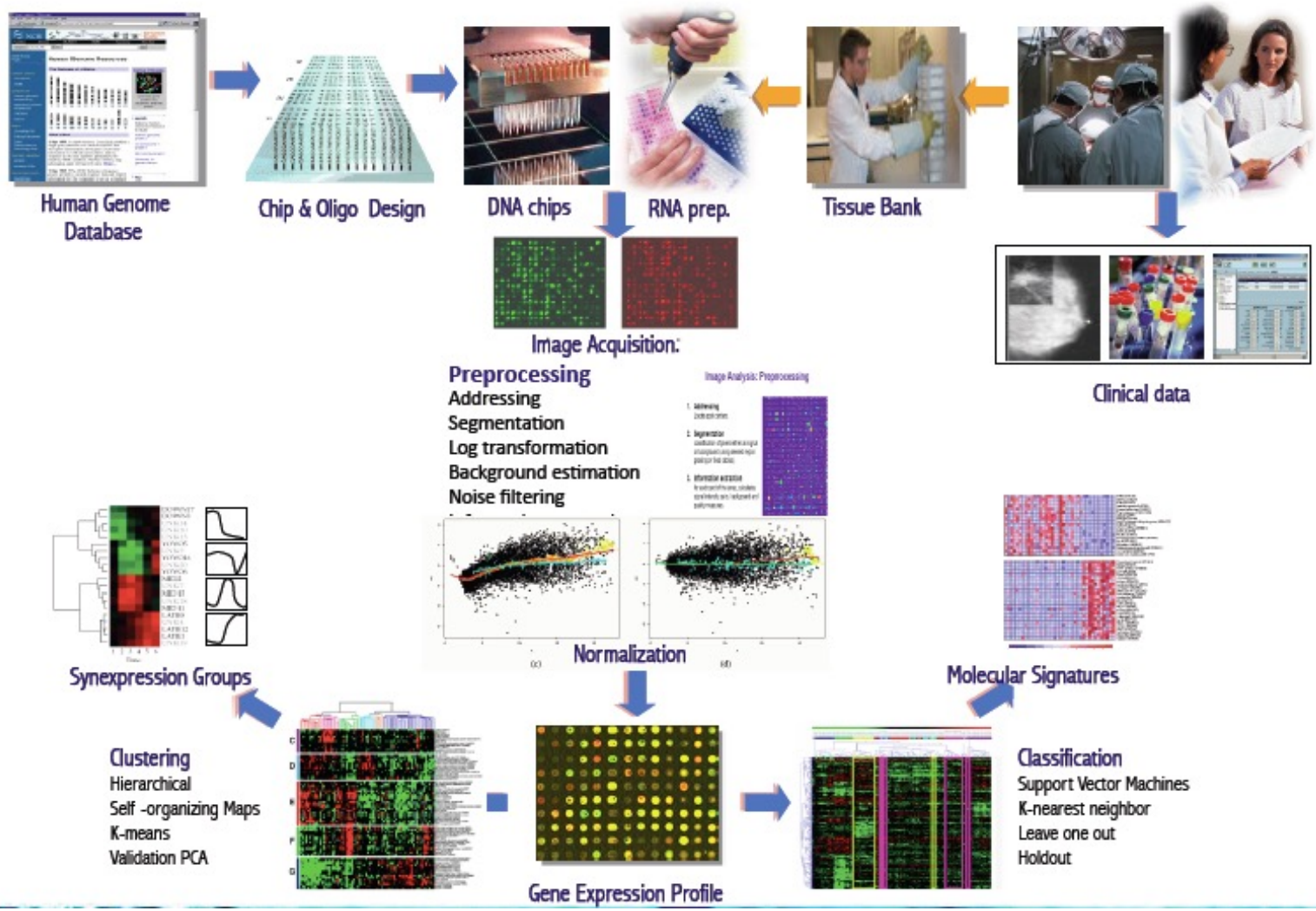
Approaching System Biology

Integration of multilevel genomic data information

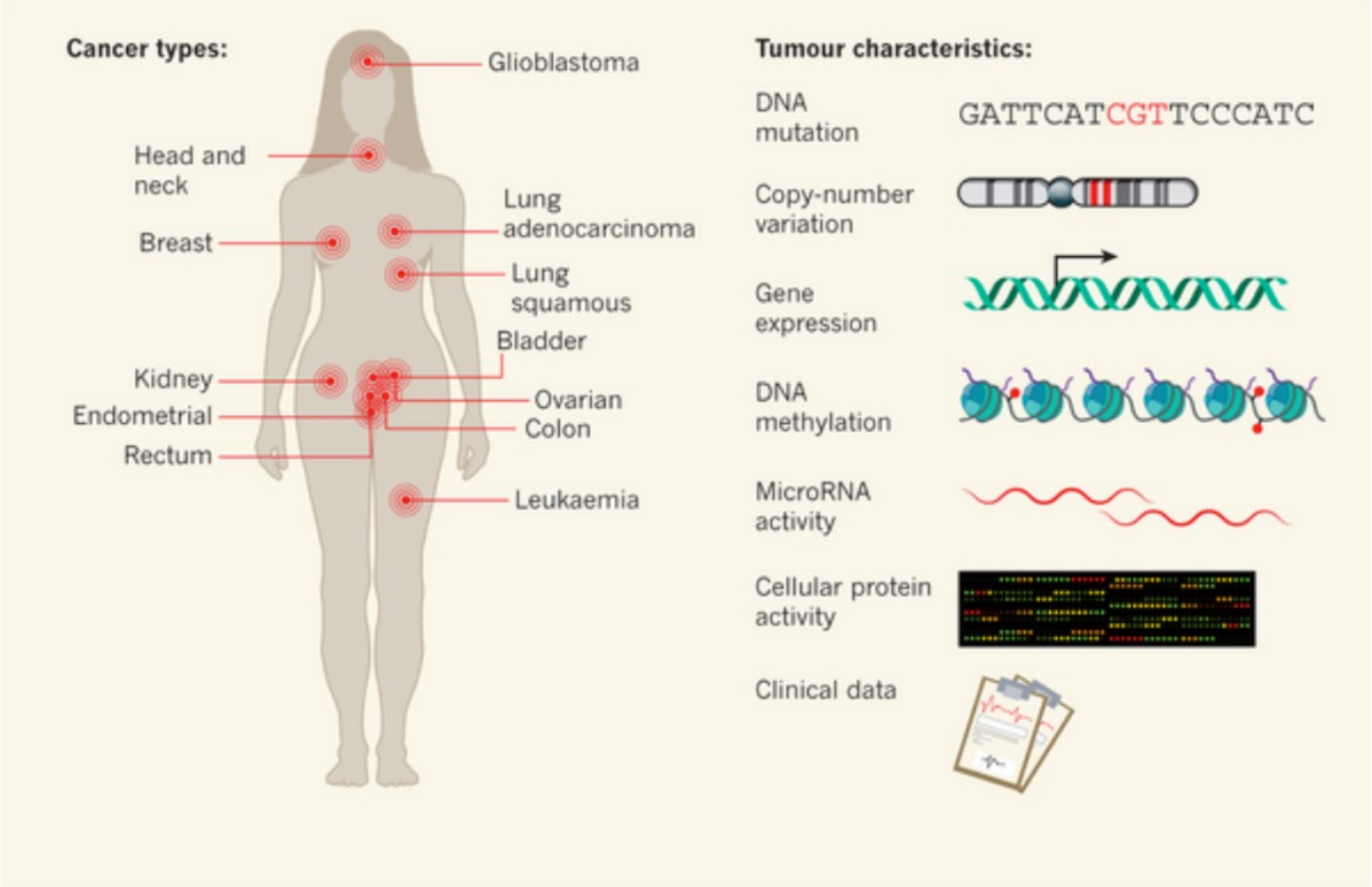
TCGA Pan-Cancer project



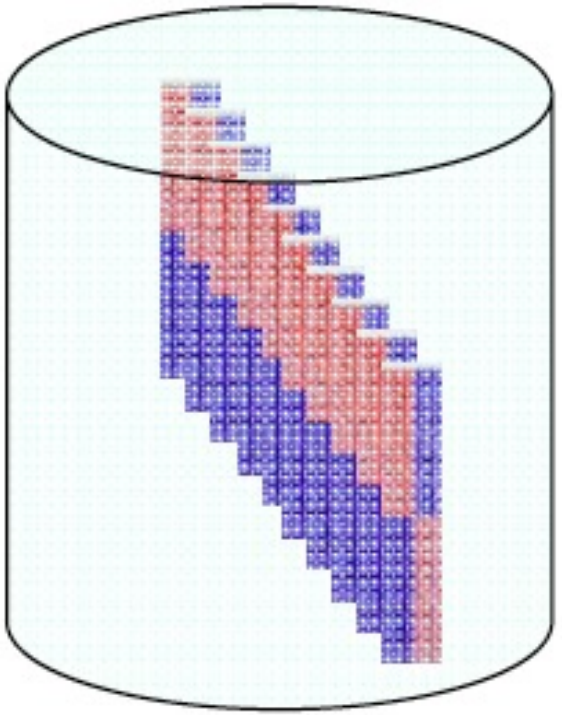
Integration of clinico-genomic data: Approaching Individualized Medicine



Cancer Genomics Projects

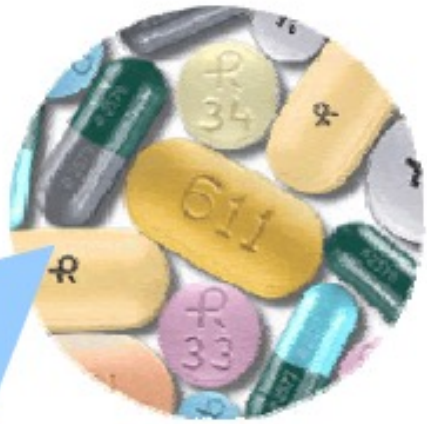


Connectivity Map



diseases

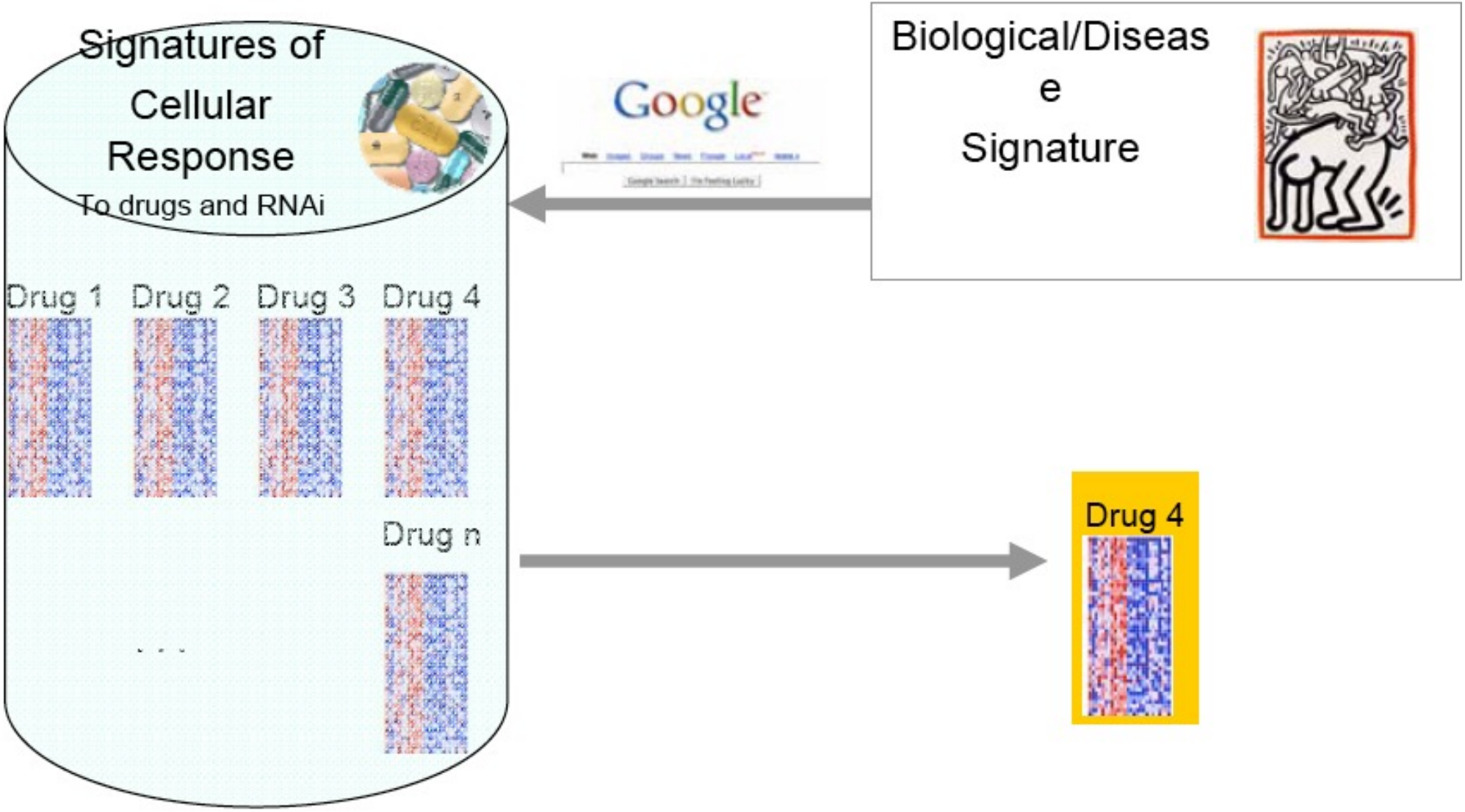
connect
genes with diseases
and the drugs that
treat them



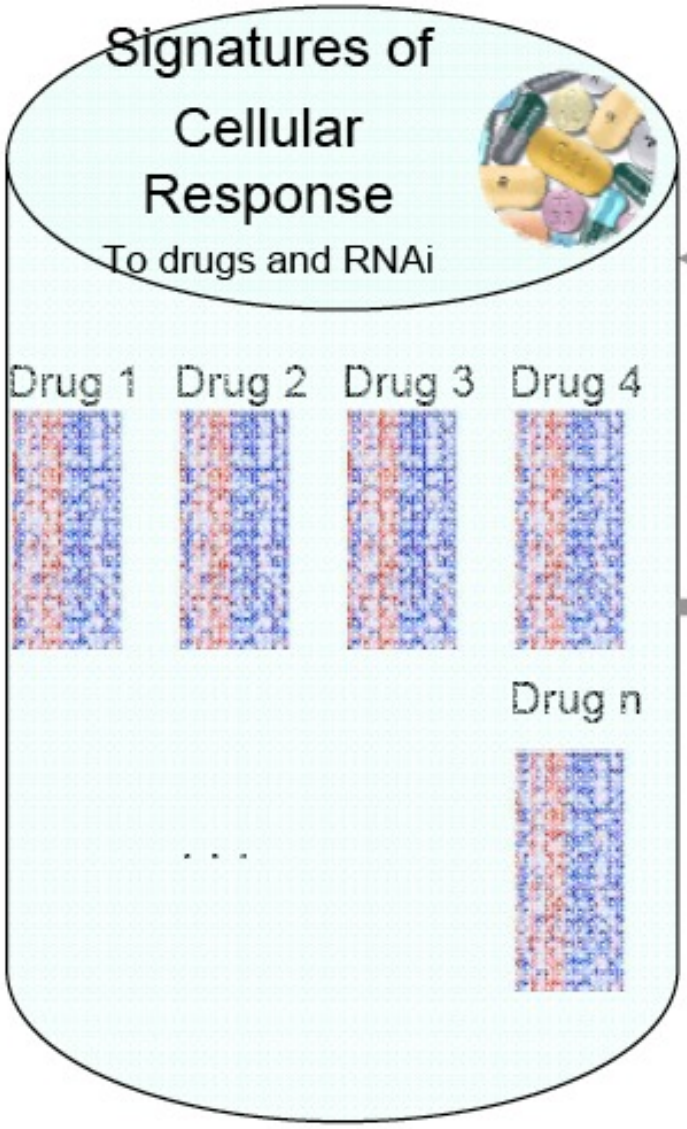
drugs

genes

Connectivity Map: Database of Signatures

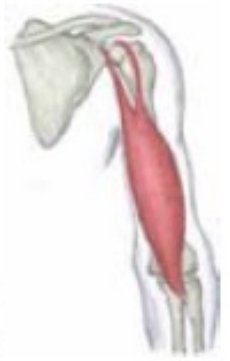


C-map: Type 2 Diabetes



Signature:

Diabetic Muscle



55 genes in
oxidative phosphorylation

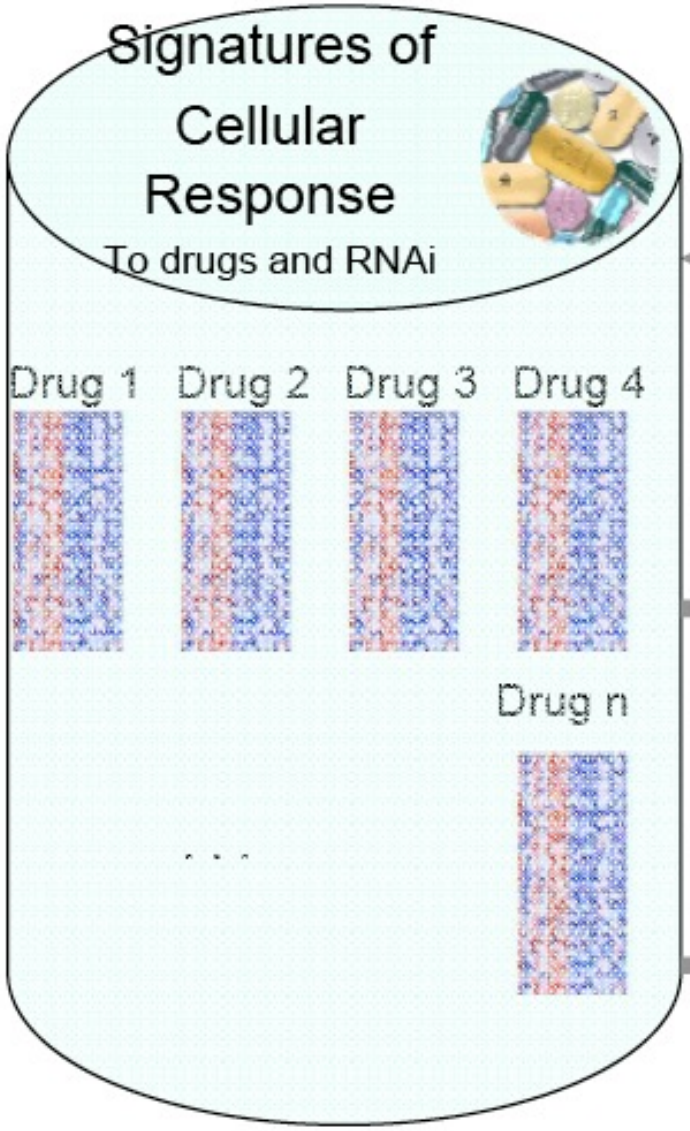
(Mootha *et al.* 2003)

Positive
Signature

Metformin
(*Glucophage®*)

Sodium Valproate ??
(*Anti-convulsant*)

C-map: Estrogen response



Signature:

Estrogen treatment in rat

48 up- and 5 down- regulated genes

(Rhen *et al.* 2003)

Positive Signature

+ Known estrogen analogs

+ Nordihydrogualaretic acid

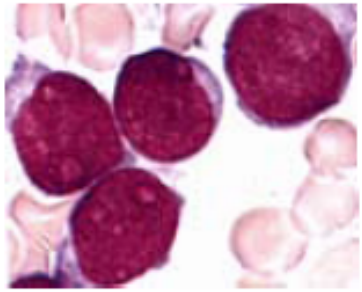
(Fujimoto *et al.* 2004)

Negative Signature

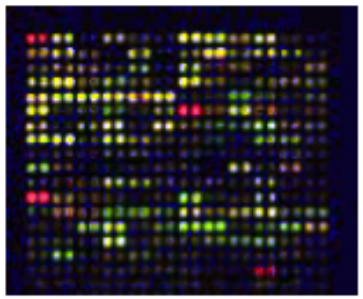
- Tamoxifens/SERMs

Drug-resistant ALL

Scott Armstrong, DFCI
Todd Golub, DFCI

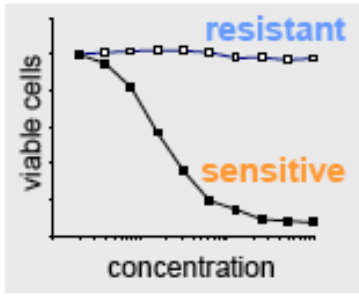


acute lymphoblastic leukemia (*n*=36)

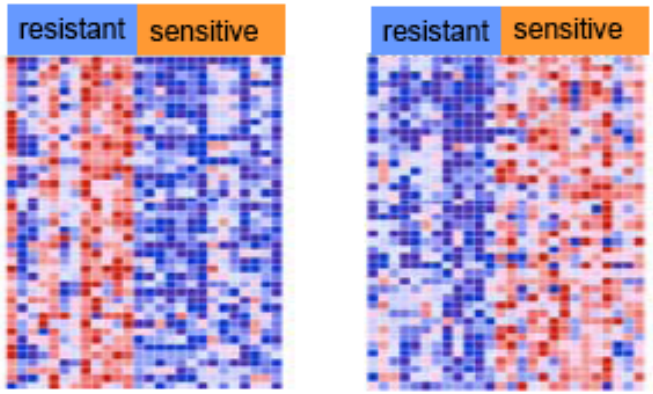


transcriptional analysis

marker selection

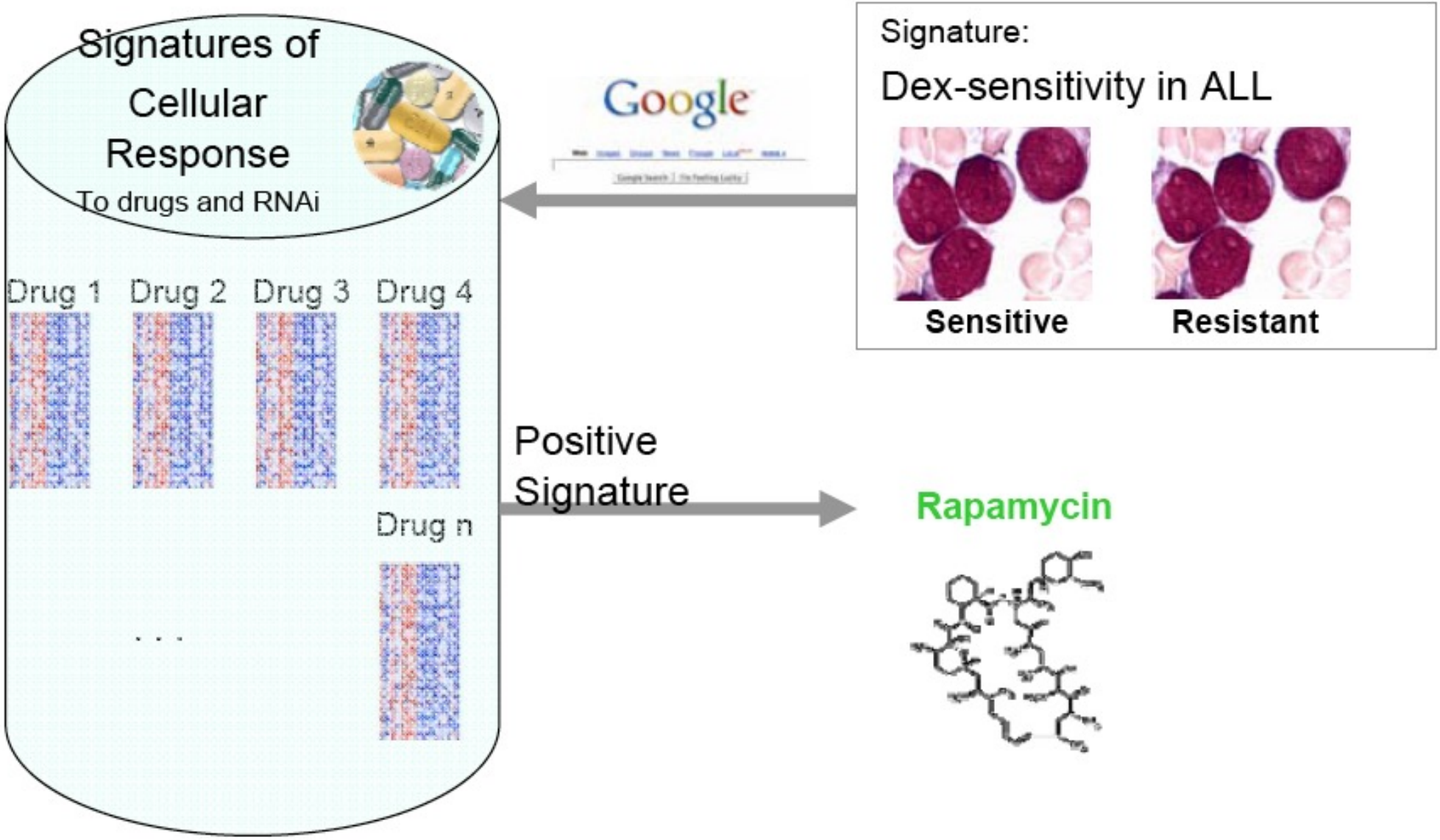


ex vivo steroid response

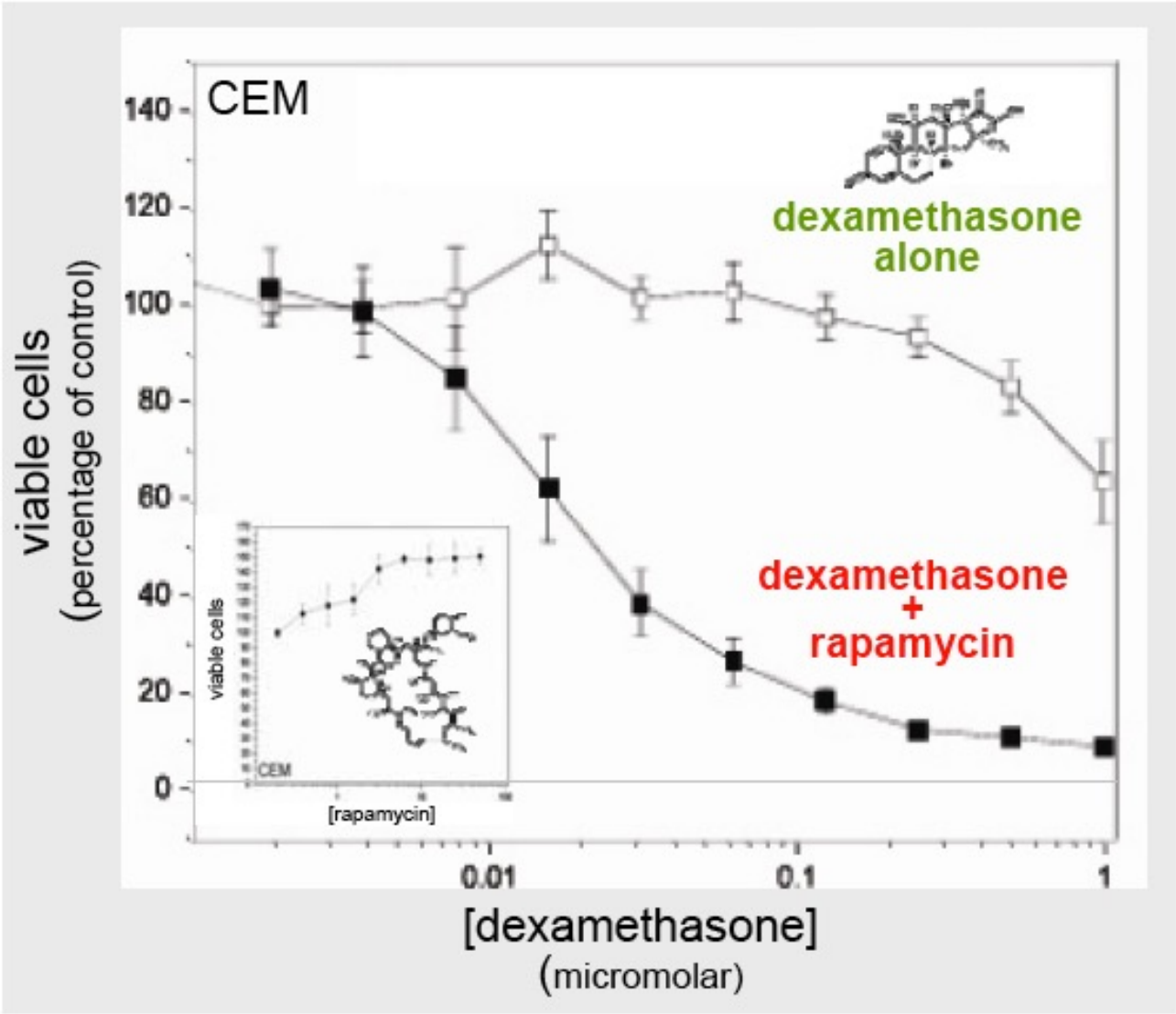


gene expression signature

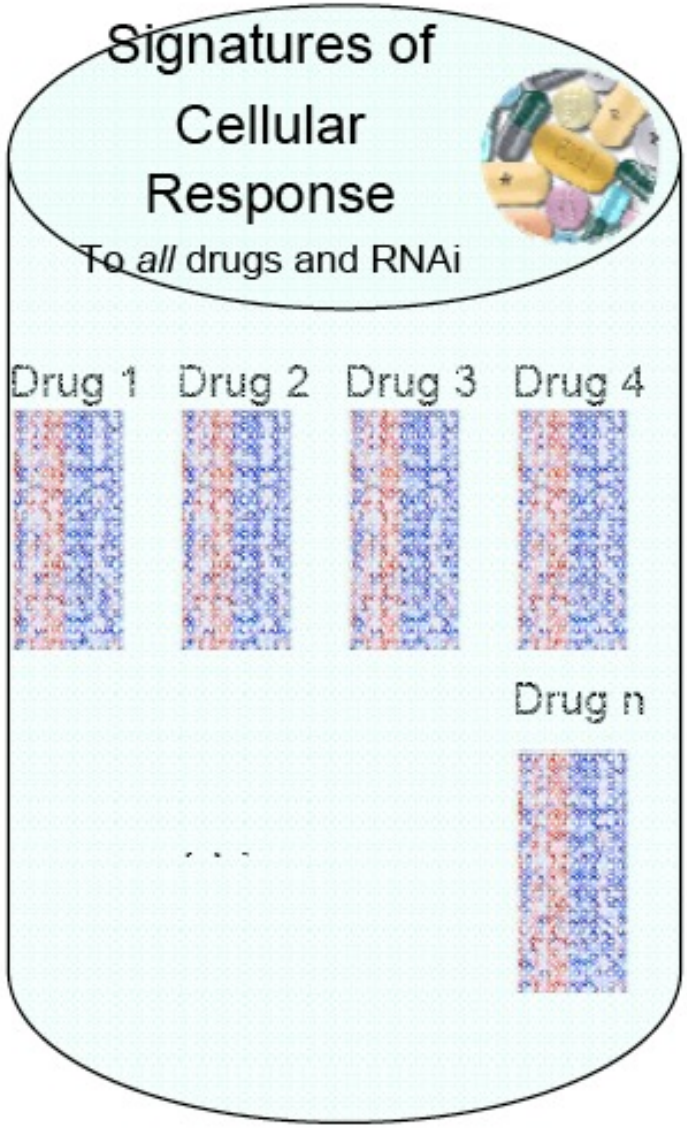
Cmap: Drug-resistance ALL



Rapamycin induces dex sensitivity



Next Step: Full Connectivity Map



Signatures for:

- All FDA-approved drugs
- Many bio-actives
- Many RNAi

Freely available on web
with good search tools

Table 2. Selection of databases commonly used in our workflows.

Database	Entities	Properties
Ensembl	Genes, proteins, transcripts, regulatory regions, variants	Genomic positions, relationships between them, identifiers in different formats, GO terms, PFAM domains
Entrez	Genes, articles	Articles for genes, abstracts of articles, links to full text
UniProt	Proteins	PDBs, known variants
KEGG, Reactome, Biocarta, Gene Ontology	Genes	Pathways, processes, function, cell location
TFacts	Genes	Transcription regulation
Barcode	Genes	Expression by tissue
PINA, HPRD, STRING	Proteins	Interactions
PharmaGKB	Drugs, proteins, variants	Drug targets, pharmacogenetics
STITCH, Matador	Drugs, proteins	Drug targets
Drug clinical trials	Investigational drugs	Diseases or conditions in they are being tested
GEO, ArrayExpress	Genes (microarray probes)	Expression values
ICGC, TCGA	Cancer Genomes	Point mutations, methylation, CNV, structural variants
dbSNP, 1000 genomes	Germline variations	Association with diseases or conditions
COSMIC	Somatic variations	Association with cancer types

doi:10.1371/journal.pcbi.1002824.t002

Vazquez M, de la Torre V, Valencia A (2012) Chapter 14: Cancer Genome Analysis. PLOS Computational Biology 8(12): e1002824. https://doi.org/10.1371/journal.pcbi.1002824
http://journals.plos.org/ploscompbiol/article?id=10.1371/journal.pcbi.1002824

Cancer Genomic Databases

Table 2. *Databases for cancer genomics data*

Database	Link	Data type	Type of information	Access
ICGC	http://dcc.icgc.org/	Levels I–IV	Copy number, rearrangement, expression, and mutation data	Open and controlled
TCGA	http://cancergenome.nih.gov/dataportal	Levels I–III	Copy number, expression (mRNA and miRNA), promoter methylation, and mutation sequencing	Open and controlled
NCBI dbGAP	http://www.ncbi.nlm.nih.gov/gap	Levels I–II	Raw sequencing traces; second-generation sequencing BAM files by TCGA	Controlled
COSMIC	http://www.sanger.ac.uk/genetics/CGP/cosmic	Levels III–IV	Somatic mutations and copy number alterations by gene: amino acid position, tumor type, literature references	Open
Cancer Gene Census	http://www.sanger.ac.uk/genetics/CGP/Census	Level IV	Annotation of mutated or genomically altered genes	Open
WTSI CGP	http://www.sanger.ac.uk/genetics/CGP/Archive	Levels I–II	First-generation trace archive; SNP genotype profiles	Controlled
EGA	http://www.ebi.ac.uk/ega	Levels I–II	Second-generation sequencing BAM files generated by WTSI CGP	Controlled
Tumorscape	http://www.broadinstitute.org/tumorscape	Levels I–IV	Browsable, searchable cancer copy number viewer using SNP array data	Open
Oncomine	http://www.oncomine.org	Level IV	Gene expression and copy number data in readily searchable and comparable fashion	Password-protected
GEO	http://ncbi.nlm.nih.gov/geo	Level I	Gene expression data	Password-protected
caArray	http://caarray.nci.nih.gov	Level I	Gene expression data	Password-protected
UCSC Cancer Genome Browser	https://genome-cancer.soe.ucsc.edu	Levels III–IV	Browsable viewer for cancer copy number and expression data	Open
The cBio Cancer Genomics Portal	http://cbioportal.org	Levels III–IV	Browsable and searchable viewer for cancer copy number and expression data	Open
OMIM	http://www.ncbi.nlm.nih.gov/omim		Inherited syndromes and causative genes for cancer and other diseases, with extensive literature review	Open
Mitelman	http://cgap.nci.nih.gov/Chromosomes/Mitelman		Copy number alterations and translocations based on cytogenetic data	Open

(Level I) Raw; (Level II) normalized/processed; (Level III) interpreted; (Level IV) summarized.

Chin *et al*, Genes. Dev. 2011 March 15; 25(6): 534-555
http://www.ncbi.nlm.nih.gov/pubmed/?term=21406553