Basics of Cancer and Cancer Genomics

A series of lectures given at Department of Computer Science, National University of Singapore

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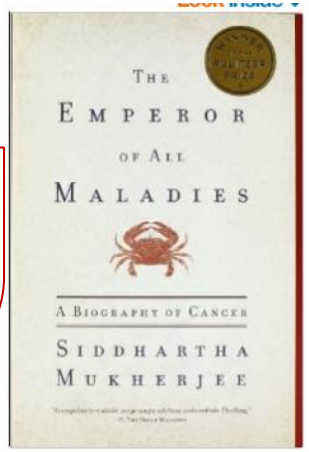
Source: http://www.sriganeshsrihari.net/

Will be in five parts

- Part 1: Basics of cancer (30 May 2014)
  - Mostly in lay-man's language
- Part 2: Lessons from the cancer genome (30 May 2014)
- Part 3: Hallmarks of cancer (05 June 2014)
  - Mechanistic details, tumour microenvironment, cancer stem cells
- Part 4: Breast cancer classification (13 June 2014)
  - Personalized genomics and targeted therapy
- Part 5: Breast cancer therapy (05 June 2014)
  - Focus on synthetic lethalit

Recommended reading!



The Emperor of All Maladies: A Biography of Cancer

by Siddhartha Mukherjee (Author)

★★★★½ 780 customer reviews

Part - 1

BASICS OF CANCER

Cancer: A Genetic Disease

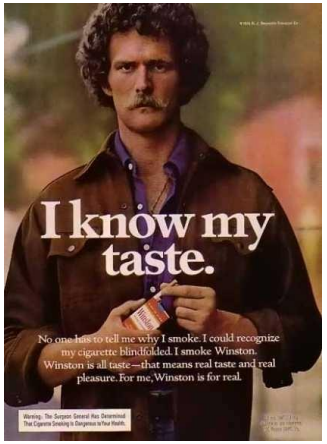
Aberrations in **genes** that control cell growth and division are responsible for cancer.

A common form of aberration is gene mutation

Defective or no gene inherited (*e.g. BRCA1* & *BRCA2*)

Exposure to carcinogens that can cause mutations

(e.g. cigarette smoke)



(1960s-70s)

Caution—cigarette smoking *may* be hazardous to your health (1966, USA)

Cancer: A Genetic Disease

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(e.g. cigarette smoke)

Lung cancer – 224,210 new cases and 159,260 deaths (2013 - 2014)
Breast cancer – 235,030 new cases and 40,000 deaths every year (2013-2014) (Seigel et al, 2014)

Gasoline and chemicals from combustion are the biggest 'mammary carcinogens' -- *Environmental Health Perspectives*, a study by NIH (2014)

Even unfortunate

- Even now the most widely adopted forms of therapy are:
  - Surgery – e.g. in cases lacking/with defects in the BRCA gene, double mastectomy
  - Chemotherapy – bombard the body with cytotoxic chemicals
    - Highly toxic but not selective
    - Severe side-effects

- Tremendous trauma and suffering to patients and their family members

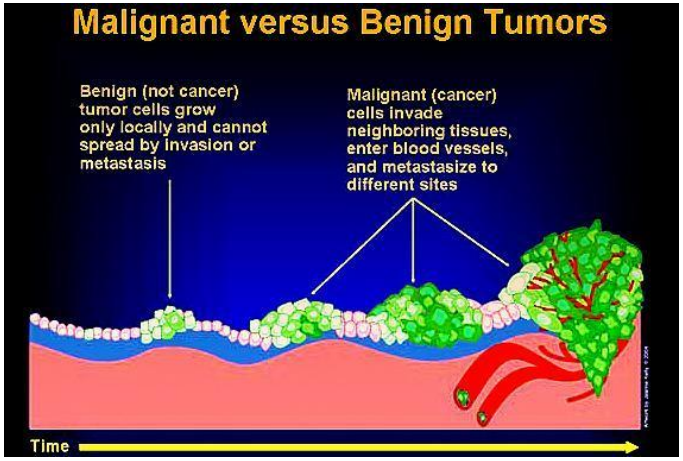
Cancer

- Cancers arise when critical genes are mutated, causing unregulated proliferation of cells.
  - Disturb the 1:1 ratio of cell division to cell death

- These rapidly dividing cells pile up on top of each other to form a **tumour**.

- When cells detach from the tumour and invade surrounding tissues, the tumour is **malignant** and may form secondary tumours at other locations in a process called **metastasis**.

- A tumour whose cells do not invade surrounding tissues is **benign**
  - Usually not considered as cancer



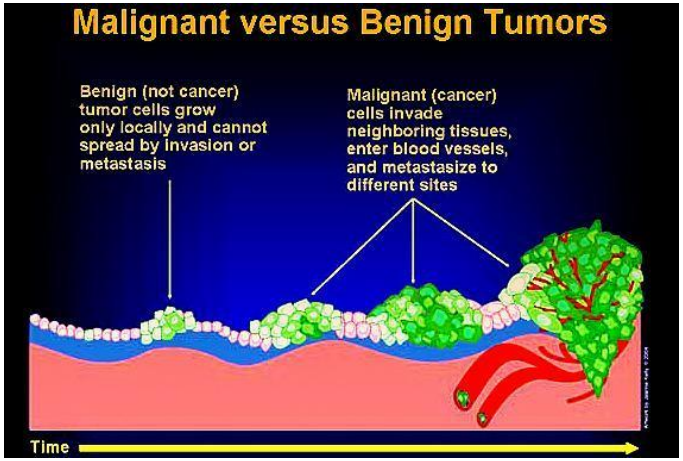
Tumour – is a condition where there is abnormal cellular growth thus forming a lesion or in most cases, a **lump** in some part of your body

- E.g. A wart can also be considered as a tumour

Benign tumour – grows in a confined area

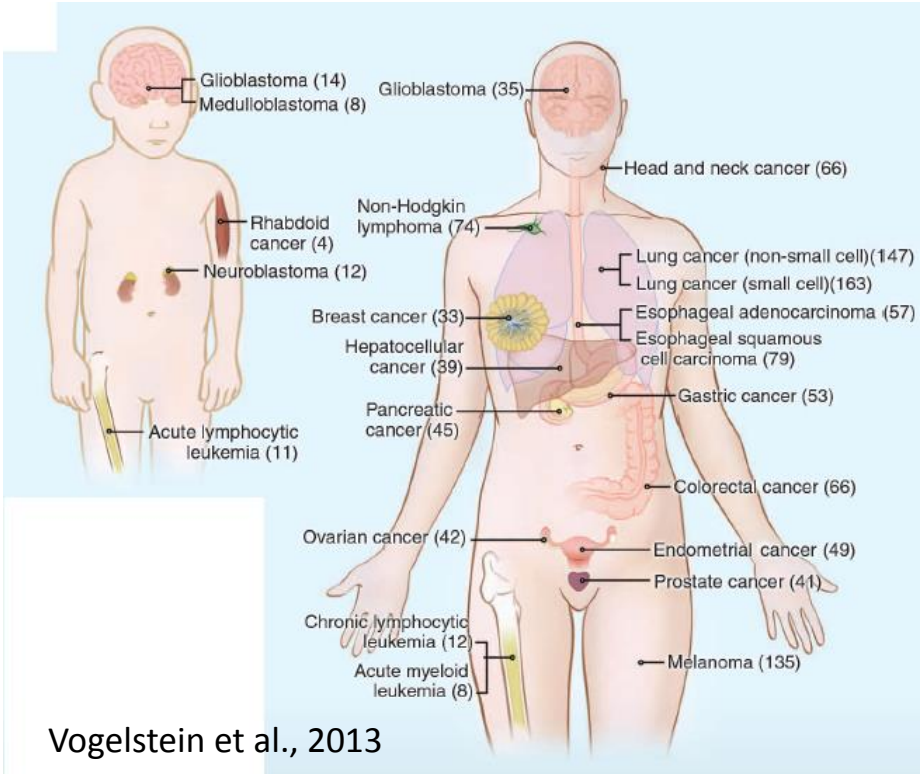
Malignant tumour – capable of **invading** surrounding tissues – becomes a cancer

Cancer – disease condition where there is uncontrolled growing mass of cells capable of invading neighboring tissues and spreading via body fluids to other parts of the body

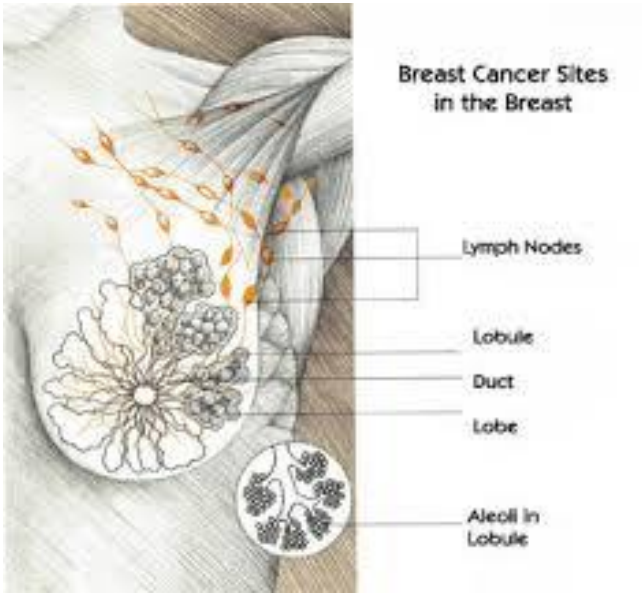


Cancer is a large collection of diseases

- Its not one disease
- Even if it affects the same organ site
  - E.g. breast cancer (at least 5 confirmed subtypes, a recent paper identifying 10 (Curtis et al, Nature 2012))



Vogelstein et al., 2013



Different kinds of breast cancer based on the specific organ site – duct or lobule / basal or luminal

Named for site of origin

Carcinomas – epithelial cells; cover external & internal body surfaces (90%)

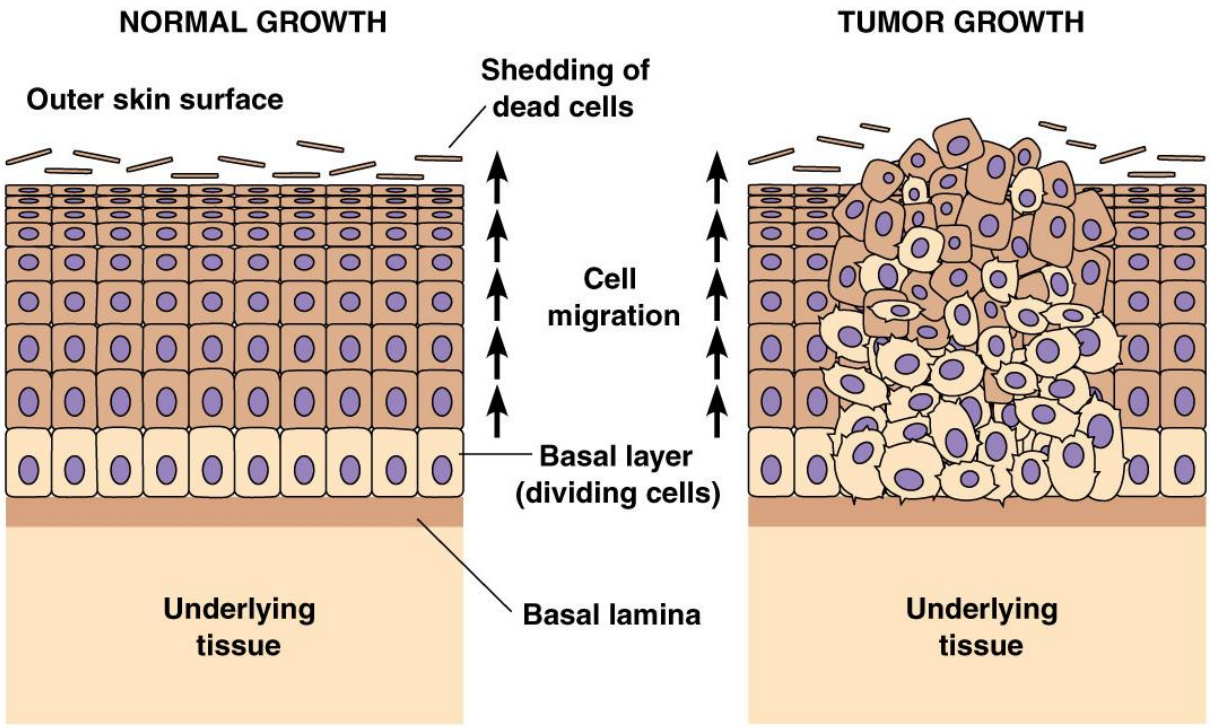
- Adenocarcinoma – involving secretory glands
  - Pancreatic ductal adenocarcinoma
  - Breast invasive carcinoma

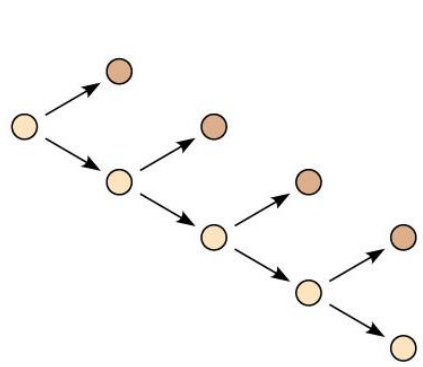
Sarcomas – supporting tissue; bone, cartilage, fat, connective tissue, pancreas, liver.

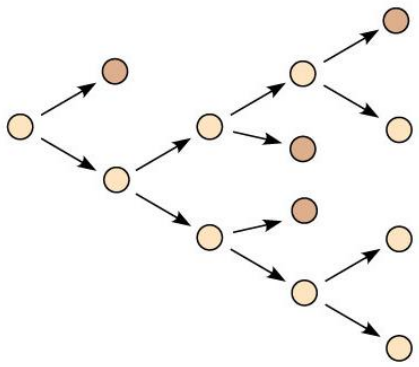
Lymphoma & leukemias – blood & lymphatic tissue

(leukemia reserved for cancers that reside in bloodstream not as solid tissue)

Comparison of Normal and Tumor Growth in the Epithelium of the Skin







Location/distribution

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Normal cells vs. Cancer cells

Normal cell proliferation	**Cancer cell proliferation**
Anchorage dependent	Anchorage independent
Density-dependent inhibition	Can grow on top of one another
Limited number of cell divisions	Immortal
Telomere shortening	**Telomere maintenance**
Proliferation dependent upon extracellular signals	**Constant signal to divide independently**
Checkpoints activated at appropriate times	**Loss of checkpoints**
Apoptosis functional	**Apoptosis inhibited**

Control of the Cell Cycle

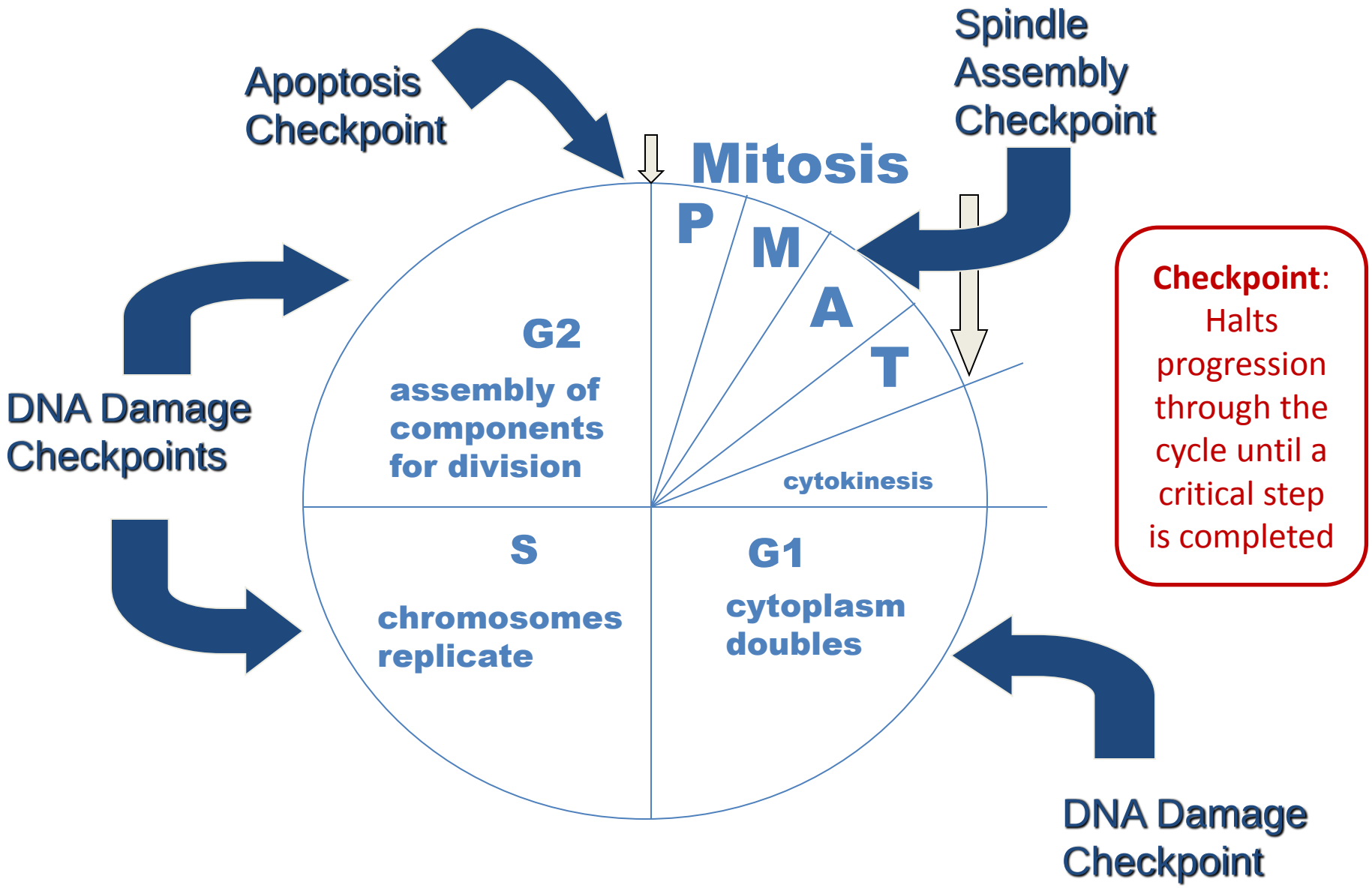
The process through which a cell divides to produce two daughter cells

In eukaryotes, it occurs in four phases

Mechanisms for controlling progress through the cell cycle:

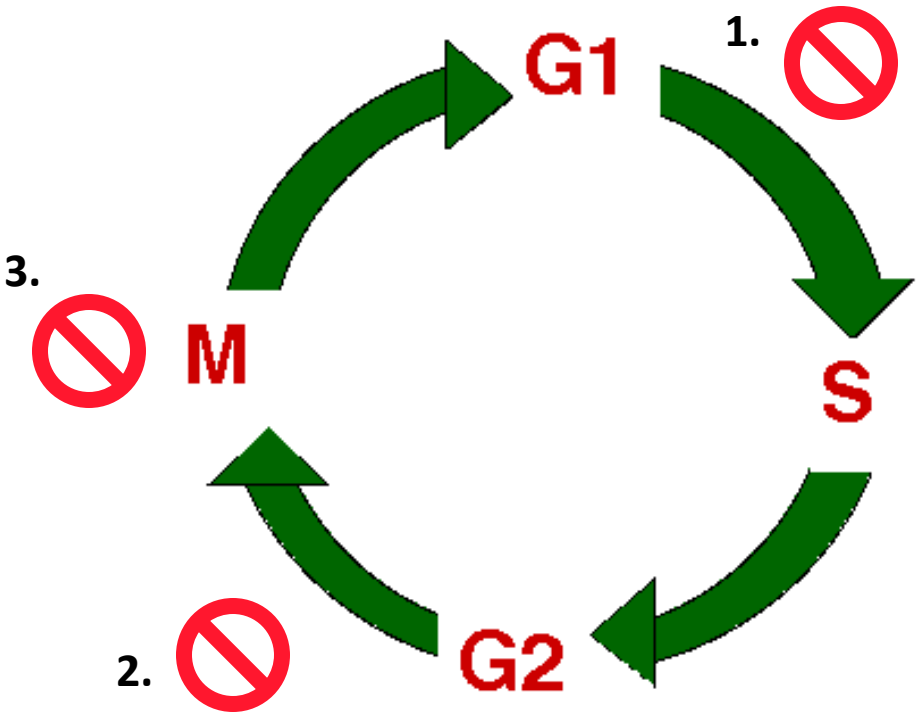
- ✓ Checkpoints
- ✓ Length of telomeres
- ✓ Chemical signals from within and outside the cell

Cell Cycle Checkpoints



Why are these checkpoints important ?

•2001 Nobel Prize was awarded to 3 scientists who studied genes that regulate the cell cycle

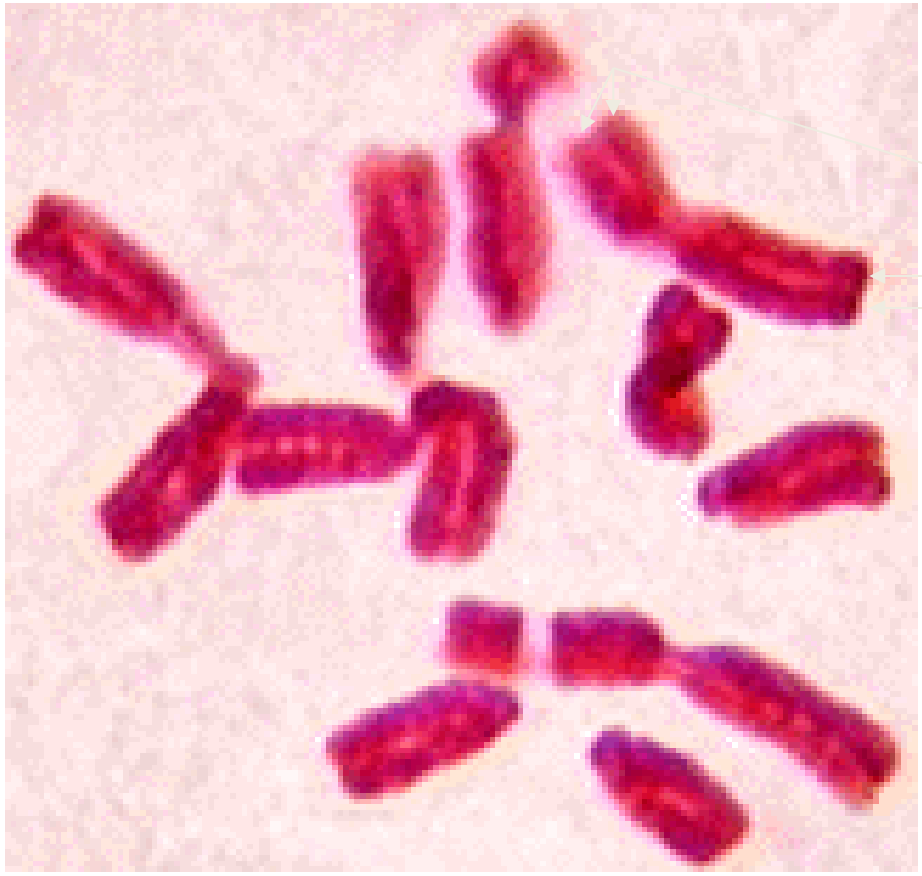


1. Is cell the correct size?
   Is DNA damaged?

2. Is DNA fully replicated?
   Is DNA damage repaired?

3. Have spindle fibers formed?
   Have they attached to chromosomes correctly?

Length of Telomeres



Telomeres are structures at the ends of chromosomes that shorten with each cell division. After 50 divisions, the shortened length of telomeres causes mitosis to stop.

Failure to Stop at Cell Cycle Checkpoints

Mutation in a gene that usually slows the cell cycle	Rate of cell division is accelerated.
Failure to pause for DNA repair	Faulty DNA leads to unregulated cell growth.
Loss of control over telomere length	Cancer cells have telomerase, an enzyme that elongates telomeres. Cells continue to divide after 50 mitoses.

Cancer and Programmed Cell Death

- **Apoptosis** is part of the normal developmental program in animals and is important in the prevention of cancer.
- The **caspases**, a family of proteolytic enzymes, are involved in apoptosis and cleave many target proteins.
- If apoptosis is impaired, a cell that should be killed can survive and proliferate, potentially forming a clone that could become cancerous.

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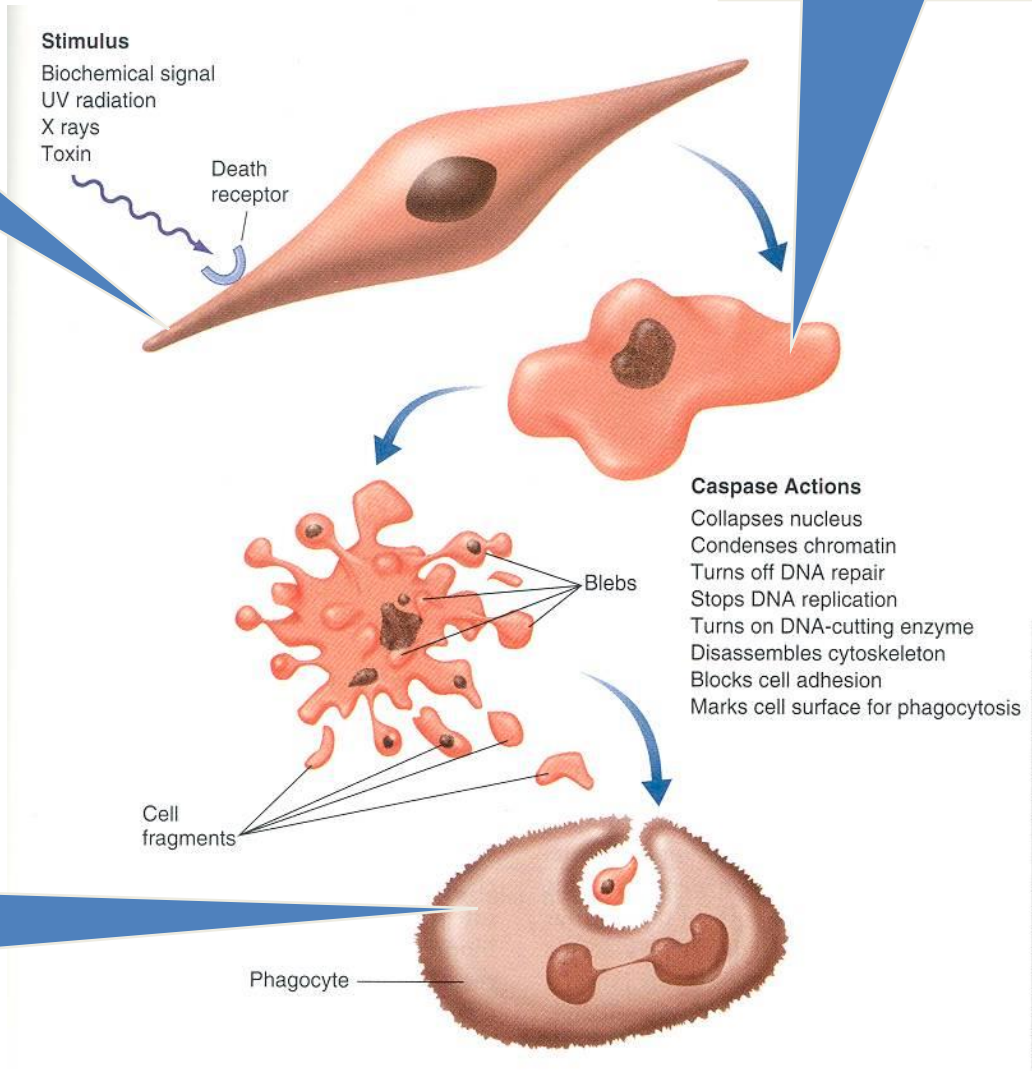
Apoptosis: Cell Death

Caspase enzymes carry out cell destruction

Signal arrives at “death” receptor on cell



White blood cells destroy cell fragments

Accumulation of genomic damage

- Continual cell divisions by evading cell-cycle checkpoints and apoptosis leads to accumulation of genomic damage

- In some cases, this damage is an outcome and contributes to the development/progression, and in some this causes the cancer

- *E.g.* BRCA-mutation carriers have a high chance of developing breast cancer during their lifetimes due to lack of effective repair functionality.
  - > 90% of such tumours show P53 mutations

But to cause to cancer…

- You need the right number and the right combination of genes to be altered
  - Not just any (combination of) gene(s)

  - Its like a game in a casino
  - Often also compared to the jammed accelerators and failed brakes of a car
  - Typically certain accelerating genes (oncogenes) will need to be up-regulated/amplified, and certain breaking genes (tumour suppressors) will need to be down-regulated/lost
  - Alterations in the genes disturb the 1:1 ratio of cell formation: cell death

Oncogenes and tumour suppressor genes

- Oncogene
  - Accelerates the cancer formation or development
  - Usually genes necessary for cell growth and division
    - Growth signals, hormones
    - RTL, RAS and MAPK, PI3K – signalling kinases
- A gene capable of becoming an oncogene is a proto-oncogene
    - All of us have proto-oncogenes
    - A proto-oncogene gone awry can turn into an oncogene
    - By mutation, copy-number amplification or up-regulated
    - Similar to a switch left ON
      - E.g. RTK kinases, CDK

"Hunting of the Sarc" (from *Hunting of the Snark* by Lewis Carroll)

Varmus & Bishop (1979), while setting out to understand the evolutionary history of the *src* gene, stumbled upon the realization that src, a retrovirus gene, is in fact in the genome of every organism, and is a proto-oncogene

Oncogenes and tumour suppressor genes

- Tumour suppressor gene
  - A breaking gene
  - Controls cell growth, maintains genomic integrity
    - Cell-cycle checkpointing genes (Chk1, chk2)
    - DNA-damage repair genes (BRCA1, BRCA2)
    - DNA-damage sensing and apoptosis (P53, ATM)
    - Controlling growth signals (negative feedback – PTEN in PI3K pathway)

Oncogenes and tumour suppressor genes

- But, remember that we have two copies of each gene
- So, an alteration in
    - Oncogene – one copy is sufficient
    - Tumour suppressor – both copies

- Knudson's two-hit hypothesis (Alfred Knudson, 1971)
  - Refers more to tumour suppressor genes
  - Familial vs sporadic cancers

Knudson's discovery
Familial and sporadic cohorts of retinoblastoma patients showed distinct onset times, correlated with *Rb* mutations

- Alterations in these genes alter the 1:1 ratio
  - Even a 0.4% increase, over years can result in a huge mass of billions of cells (compounded increase)

- How do we identify these genes?

Part - 2

LESSONS FROM THE CANCER GENOME

The initial years

- Human Genome Project
  - Launched in 1990
  - Draft sequence completed by 2000
  - Near-complete sequence completed by 2003
- Multi-genomes projects
  - 1000 genome project
  - Genetic variation in population (HapMap)

- Human cancer genome project (NCI) – 2004/2005
  - The Cancer Genome Atlas (TCGA)
  - International Cancer Genome Consortium (ICGC)
    - Gastric and other cancers endemic to Asia – Singapore node
    - We in Australia are sequencing familial breast tumours (kConfab) and pancreatic tumours

16000 tumours have been sequenced (as of 2012)

Enabled by technology revolution

- whole-exome and whole-genome sequencing at much lower costs

Identify genes that are affected by frequent

- Mutations
- Copy-number alterations
- Chromosomal gains or losses
- Gene/chromosomal fusions
- Chromosomal shattering and stitching

Lots of stuffs happening within the cancer genomes…..studying these helps us to understand cancer mechanisms

Table 1. Current Large-Scale Cancer Genome Projects[a]

Anatomic Site	Tumor Type
Brain/Central nervous system	glioblastoma multiforme
	low-grade glioma
	pediatric: medulloblastoma
	pediatric: pilocytic astrocytoma
Head and neck	head/neck squamous cell cancer
	thyroid carcinoma
Thoracic	lung adenocarcinoma
	lung squamous cell carcinoma
Breast	breast lobular carcinoma
	breast ductal carcinoma
	breast triple-negative
	breast HER-2 positive
	breast ER positive vs. negative
Gastrointestinal	esophageal adenocarcinoma
	esophageal squamous carcinoma
	gastric adenocarcinoma
	gastric (intestinal/diffuse)
	hepatocellular (alcohol/adiposity)
	hepatocellular (virus)
	hepatocellular (general)
	pancreatic adenocarcinoma
	colorectal adenocarcinoma
	colon cancer (non-Western)
Gynecologic	ovarian serous cystadenocarcinoma
	endometrial carcinoma
	cervical cancer (squamous + adeno)
Urologic	renal: clear cell carcinoma
	renal: papillary carcinoma
	renal: chromophobe carcinoma
	bladder cancer
	prostate adenocarcinoma
	prostate adenocarcinoma, early onset
Skin	melanoma, cutaneous
Soft tissue (Sarcoma)	solitary fibrous tumors
	desmoid tumors
	angiorsarcomas
	leiomyosarcomas
	extraskeletal myxoid chondrosarcomas
Hematologic	acute myeloid leukemia
	lymphoma: chronic lymphocytic leuk.
	lymphoma: germinal B cell
	lymphoma: diffuse large B cell
	chronic myeloid disorders

[a]In conjunction with The Cancer Genome Atlas, International Cancer

How many genes are mutated?

- How many genes are mutated in a typical cancer?
  - Breast, colon, pancreas and brain – 30 to 66 genes on average
  - Melanomas and lung ~200 (indication that mutagens like UV and cigarette smoke accelerate the mutations)
  - Pediatric tumours and leukemias have on average 9.6 mutations

- The age factor!
  - A 90-year old colorectal cancer patient has nearly twice as many mutations as a 45-year old patient

- The tissue or organ site
  - Pancreatic vs colorectal

- So, these mutations that accumulate provide a "evolutionary clock"

- Which or how many genes or alterations are required to initiate cancer or contribute in its development?
  - *That's a difficult question!*
  - Bert Vogelstein suggested that cancer progresses as discrete steps (mutational clonal sweeps)

Drivers vs Passengers

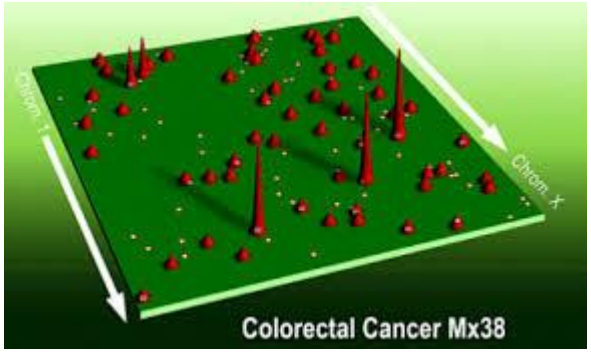
- Abundant mutations accumulate in fast-dividing cancer cells
- But, not these mutations all contribute to cancer
- In fact mutations accumulate in normal cells as well due to replication errors, errors during repair or division (triggering apoptosis)
- Only a very small and specific fraction lead or contribute to cancer
- These are called driver mutations

- The remaining vast majority are passenger mutations

- Formally, driver mutations confer a selective growth advantage to cells
- Passenger mutations are essentially background mutations

- Fishing out the driver mutations from the background passenger mutations is very tricky!
  - Variable background mutation rates
  - Long-tail
  - A subject of considerable computational research

- Driver genes and passenger genes



Vogelstein et al., Wood et al. 2008

Some disambiguation

- Driver genes
  - Mut-driver genes and Epi-driver genes
  - Mut-drivers: frequently mutated/genomically altered
  - Epi-drivers: Up-regulated due to other reasons – usually by changes in DNA methylation or chromatin modification

- Driver mutations
  - Needs to hit the region of a gene which results in a selective growth advantage
  - So, a (driver) gene can be hit by non-driver mutations as well

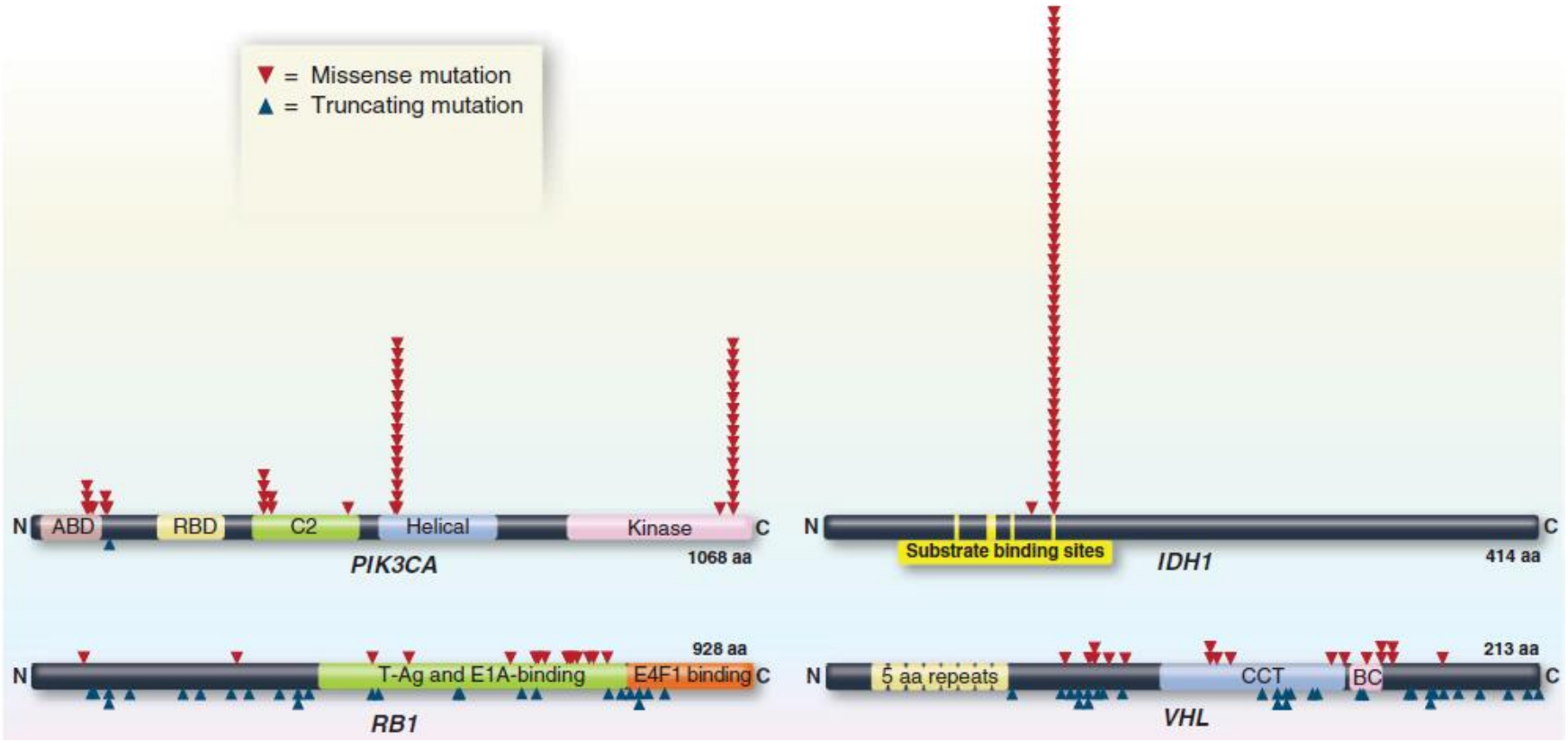
Patterns of driver mutations

- Can affect both oncogenes as well tumour suppressor genes

- You would have guessed
  - Mutations in oncogenes are activating mutations
  - While those in tumour suppressor genes are deactivating mutations

- A characteristic pattern of driver mutations
  - Highly characteristic and non-random
  - Oncogenes – recurrently mutated leading to changes at the same amino acid positions – missense mutation
  - TSG – cause protein-truncations all throughout the length
  - A simple 20/20 % rule (Vogelstein)

Patterns of driver mutations

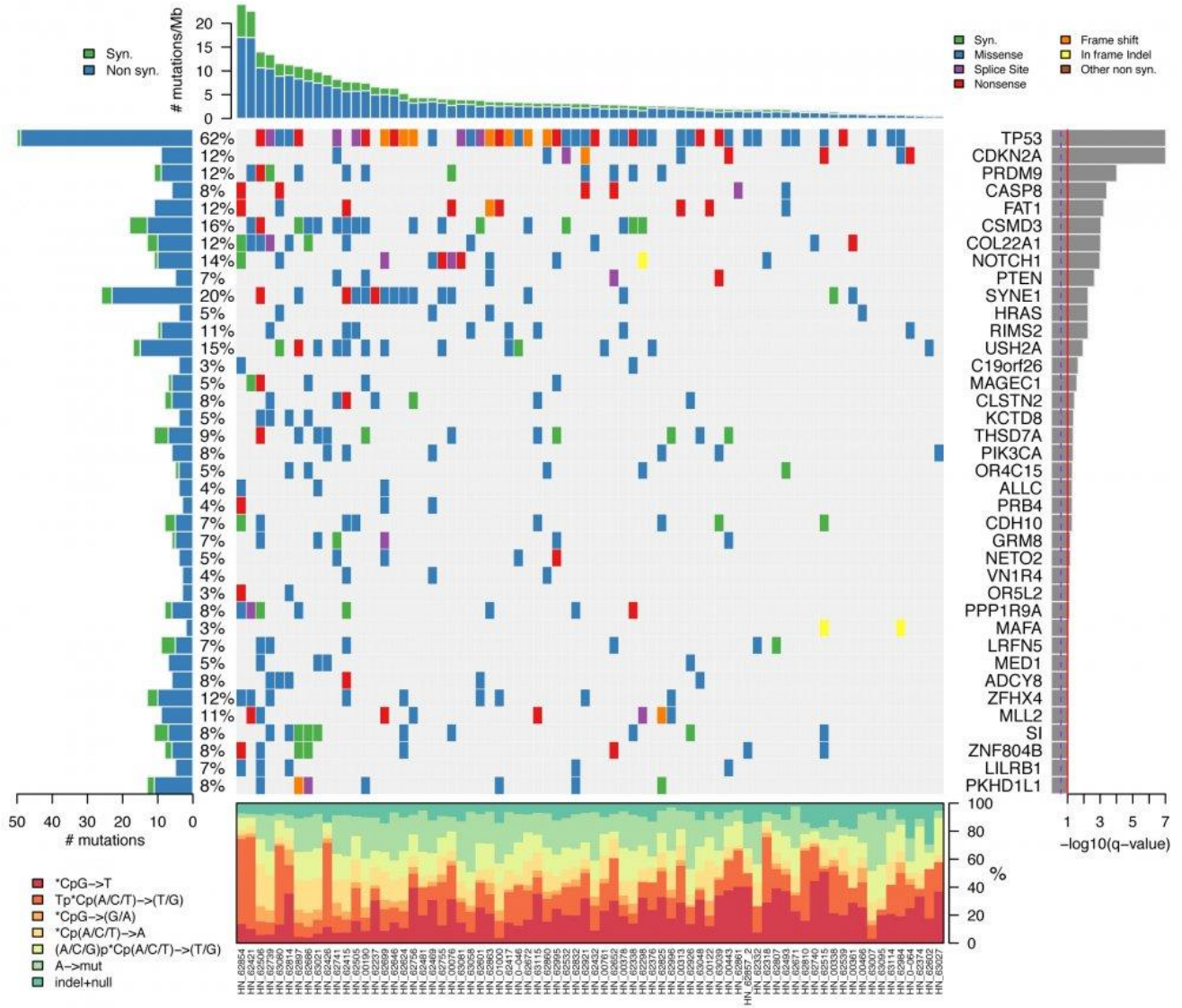


Vogelstein et al 2013

How many Mut-driver genes exist / known

- About 20, 000 genes have been evaluated in about 3500 tumours
- Identified 125 mut-driver genes through mutations
  - 71 are tumour suppressor genes
  - 54 are oncogenes
- 13 mut-driver genes through amplification or deletion
- Total 138 mut-driver genes

Mut-driver genes

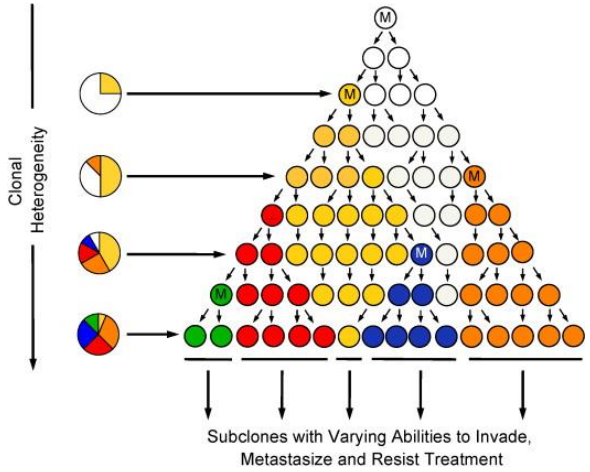


Genetic heterogeneity

- Intra-tumoural
  - Waves of mutations sweeping across the cells (Vogelstein)
  - Clonal populations
  - Depends on the sequence of mutations
  - Spatially distinct cells display more differences

- Intra- and Inter-metastatic
  - Depends on the site of metastatis
  - Some tumours prefer certain sites

- Inter-patient
  - Clinical relevance



Genetic heterogeneity

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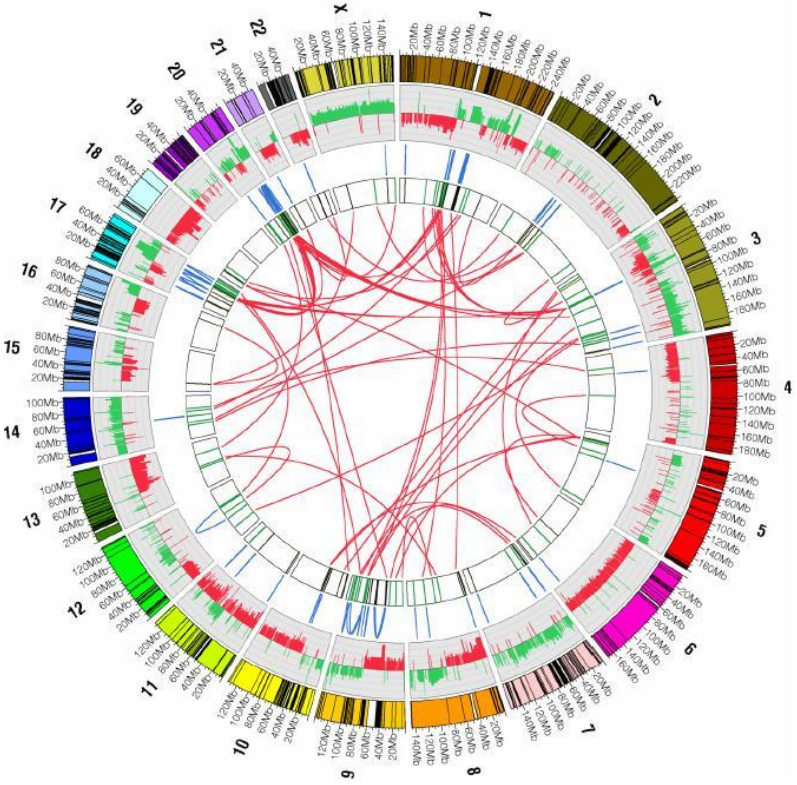
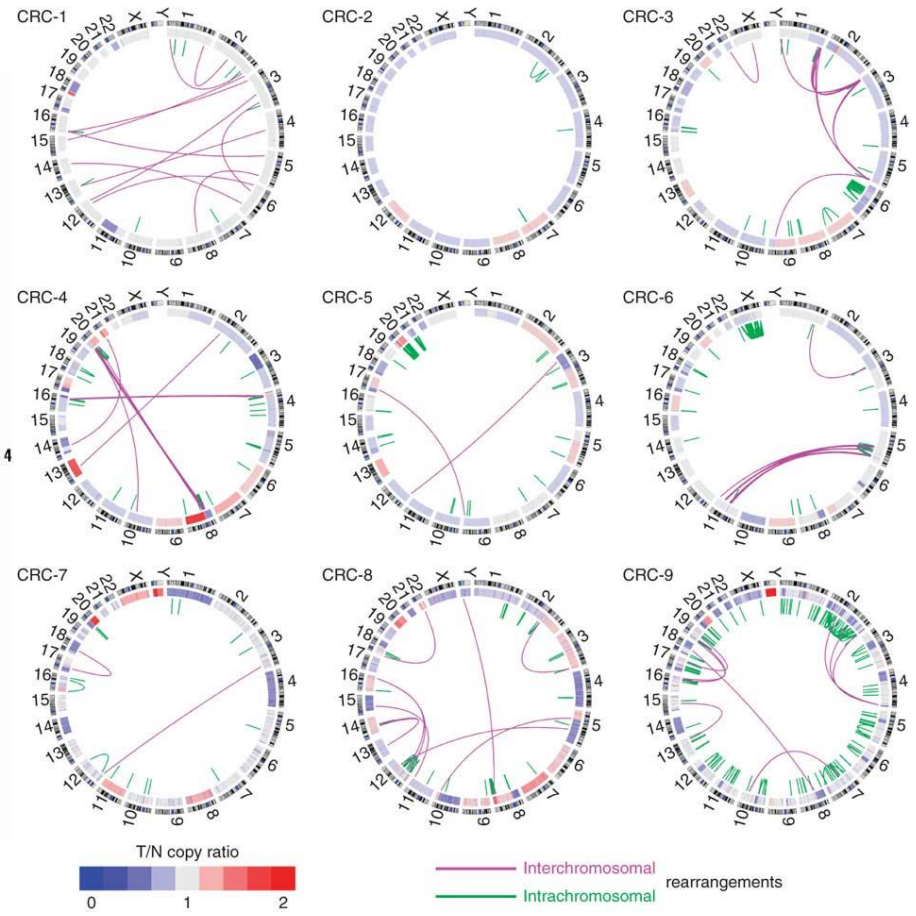




Signalling pathways in tumours

138 potential driver genes

Genetic heterogeneity

How do we comprehend the impact of all these?

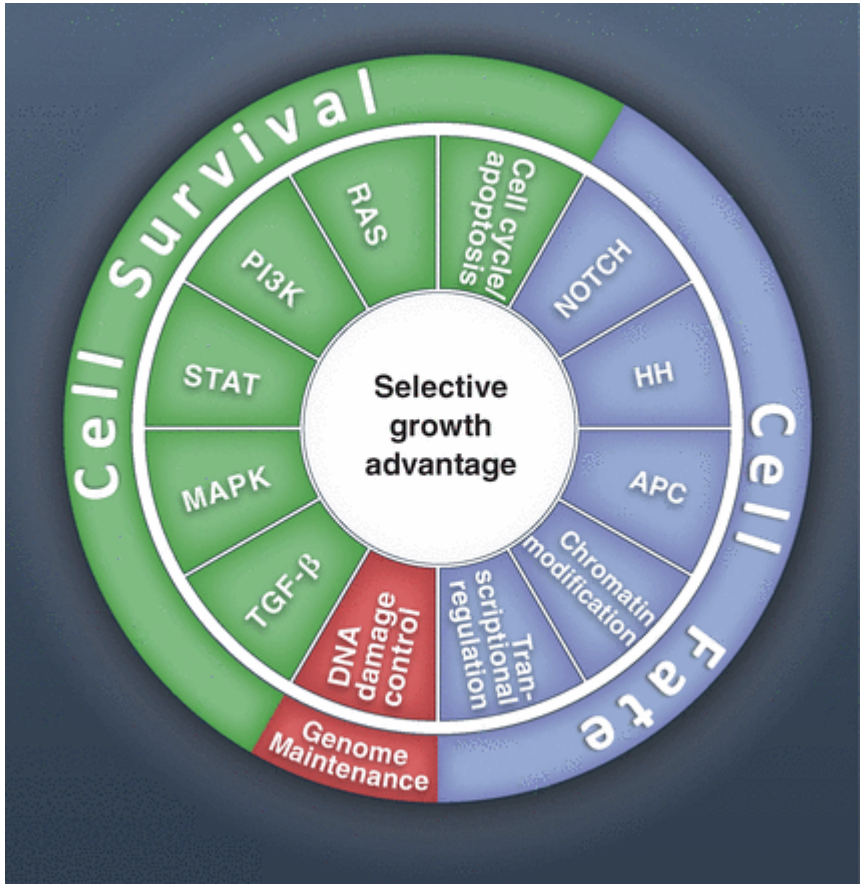
What clinical implications do these have?

The key is to see the forest instead of only the trees

- There's an order in cancer too!
- Summarize and study the impact as pathways

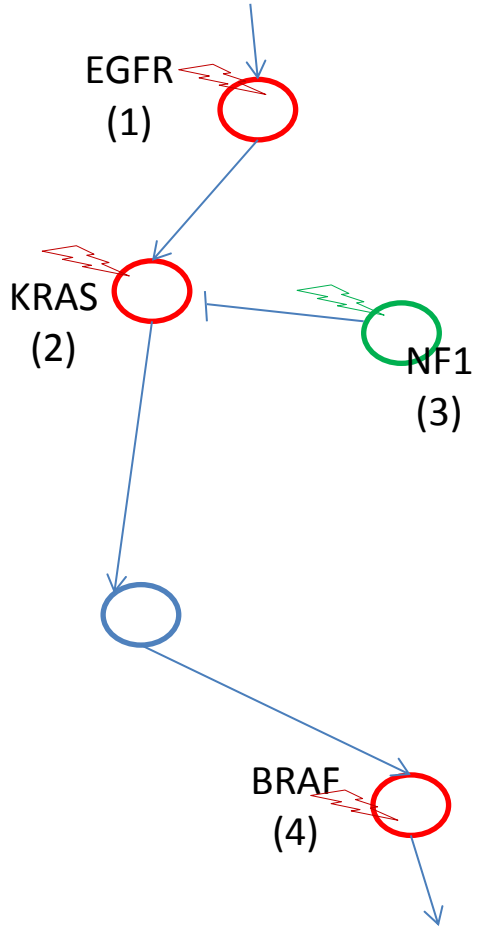
Signalling pathways in tumours

- All of the known driver genes can be classified into one or more of the 12 pathways



Source: Cancer genome landscapes, Vogelstein et al., Science 2013

A simplistic example: Four lung cancers



Even though these four lung cancer patients have mutations in different genes, all these genes are in the **same pathway**. So, appropriate targeting of this pathway could be a common therapeutic strategy to treat all these four patients.

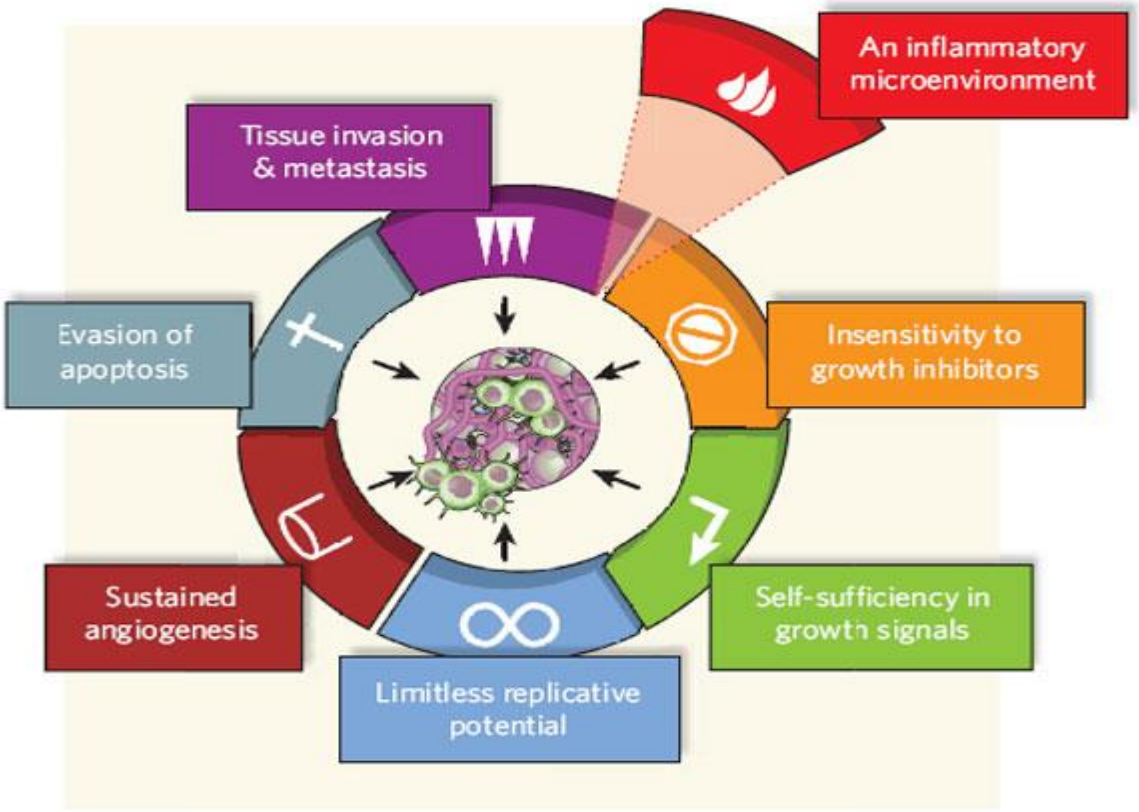
But, having said that, I will be very surprised if genomic heterogeneity between the patients did not have any role in determining therapeutic response.

Part - 3

HALLMARKS OF CANCER

Hallmarks of Cancer

Hanahan D, Weinberg RA (*Cell* 2000)



Comprise of six biological capabilities acquired by cancer cells in a multistep fashion

- Distinctive and complementary
- Provide a logical and solid foundation for the understanding of cancer

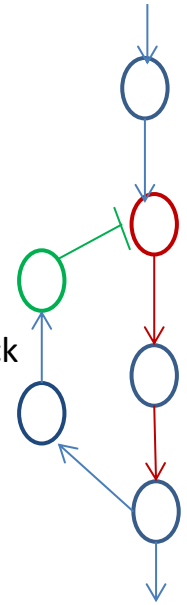
Source: Hanahan and Weinberg, *Cell* 2000

Hallmark 1: Sustained proliferative signalling

- Normal tissues carefully control the production and release of growth-promoting signals that instruct entry into and progression through the cell growth-and-division cycle

- Cancer cells dsyregulate these signals and become masters of their own destinies.

- Enabled through somatic mutations in growth-factor and transmitted genes
  - BRAF
    - MAP-kinase pathway
  - PI3K
    - PI3-kinase pathway
    - Akt/PKB pathway
- Disruptions in negative feedback mechanisms
  - Mutations in ras genes compromise Ras GTPase activity, which operates negative feedback
  - Loss of PTEN phosphotase amplifies PI3K signalling
  - mTOR activation inhibits PI3K signalling via negative feedback in some cancer cells



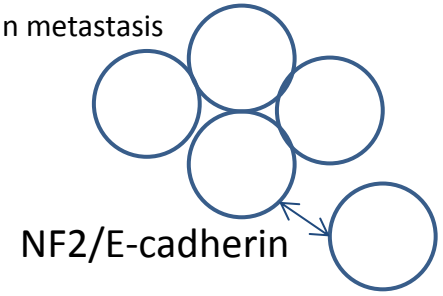
Hallmark 2: Evading growth suppressors

- There are two bona fide tumour suppressors – RB (retinoblastoma-associated) and TP53 proteins
- Both are essentially decision makers

- RB proteins integrate diverse extra and intra cellular signals and decide whether the cells should proceed through its growth-and-division cycle

- TP53 monitors genomic integrity, glucose and oxygenation, etc. and can trigger apoptosis in the face of alarm signals

- **Contact inhibition and evasion**
  - Important role in metastasis



Hallmark 3: Resisting cell death

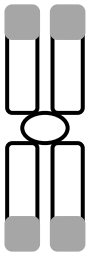
- Apoptosis -- programmed cell death
  - Very tightly regulated mechanism
  - Culminates in a set of caspases which are protolytic enzymes, which progressively disassemble and consume the cell, both by the neighbours as well as professional phagocytic cells
  - TP53 – upon DNA damage
  - Pro- and anti-apoptotic members of the Bcl-2 family

- Autophagy – mediates tumour cell survival and death
  - Catabolic mechanism involves cell degradation of cellular organelles through lysosomes
  - Down regulation of PI3K, ART, mTOR kinase pathways triggers autophagy

- Necrosis has tumour promoting potential
  - Cells become bloated and explode
  - Capable of releasing key growth hormones and nutrients to surrounding tumour cells
  - *Necropotosis* – programmed necrosis

Hallmark 4: Enabling replicative immortality

- Two barriers to proliferation "hardwired" into cells
  - Senescence: a non-proliferative but viable state
  - Apoptosis: usually in times of crisis, e.g. DNA damage



- Coming out of these two and achieving unlimited replicative potential is called immortalization of cells

- Telomeres protecting the end of chromosomes act as "clocking devices" to control proliferation

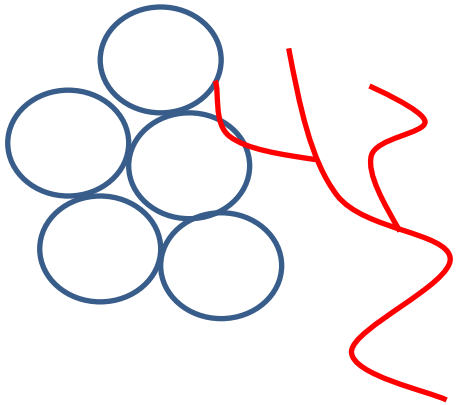
- In cancer cells, telomerase/TERT, a specialized DNA polymerase that adds telomere repeat segments, is highly upregulated (almost absent in normal cells).

- Fusion of chromosomes

Hallmark 5: Inducing angiogenesis

- Vasculature usually present as part of embryogenesis

- In adult, usually quiescent except during wound healing and female reproductive cycle
  - Angiogenic switch temporally turned on

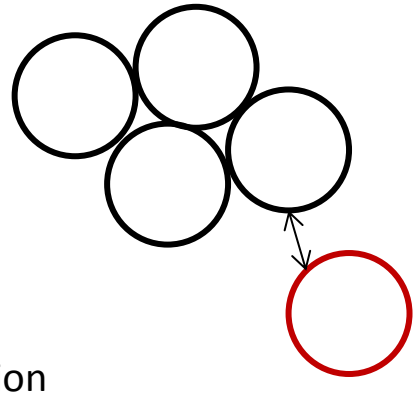


- In cancer cells, this switch is left on
  - VEGF-A (vascular endothelial growth factor – A)
  - Thrombospondin (TSP-1)

- Neovasculature
  - Capillary sprouting
  - Convoluted and excessive vessel branching
  - Distorted and enlarged vessels
  - Erratic blood flow
  - Hemorrhaging, leakiness

Hallmark 6: Activating invasion and metastasis

- E-cadherin
  - Forms adhesive junctions between cells
  - Helps to maintain quiescence of cells
  - Frequently down-regulated in invasive cells



- N-cadherin
  - Enables cell migration during embryogenesis and inflamation
  - Expressed in migrating neurons and mesenchymal cells during organogenesis
  - Frequently up-regulated in invasive cells

- Epithelial-Mesenchymal Transition (EMT)
  - Process by which epithelial cells lose their cell-cell adhesion and gain migratory and invasive properties to become mesenchymal stem cells.
  - These are multipotent stromal cells capable of differentiating into a variety of cell types

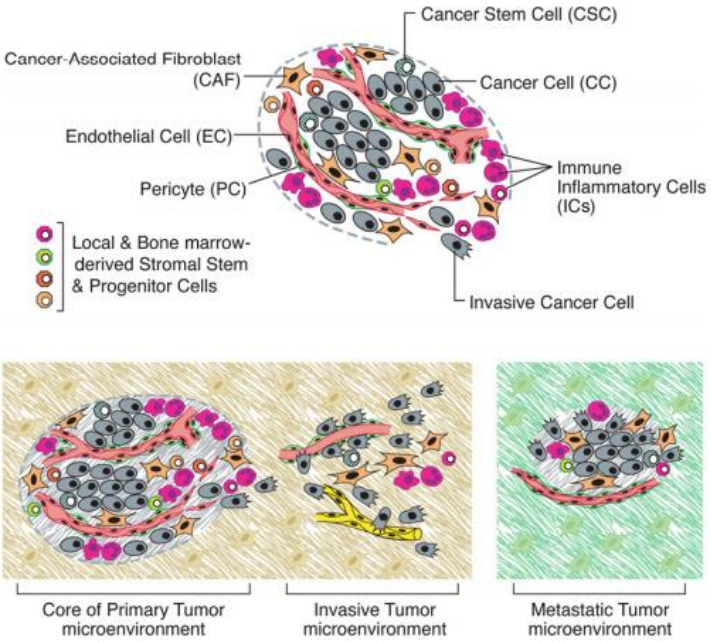
Thiery, J.P. (2002) Epithelial-mesenchymal transitions in tumor progression. *Nature Reviews Cancer*, 2 : 442-454.

Tumour microenvironment

- Traditional view: a tumour is a reasonably homogeneous cell population that grows, passing on the mutations

- An assemblage of distinct cell types, distributed into distinct clonal subpopulations
  - Clonal heterogeneity
  - Regions are demarcated by varying degrees of differentiation, proliferation, vascularity, inflammation and invasiveness

- Forms a "tumour microenvironment"
  - Cancer cells
  - Invasive cancer cells
  - Immune inflammatory cells
    - pro and anti-tumour
  - Cancer Stem Cells
  - Stromal cells



Hanahan & Weingberg 2013

Cancer Stem Cells

- Operational definition: Capable of seeding new tumours upon inoculation into recipient host (mice).
- Show similar expression profiles as normal stem cells, motivating their stem-like designation.

- Emerging theories suggest CSCs to be responsible for repopulation and hence relapse of tumours

- These are typically not targeted by conventional therapies

- Have self-renewal and differentiating capabilities (multi-potent) – hence stem-like

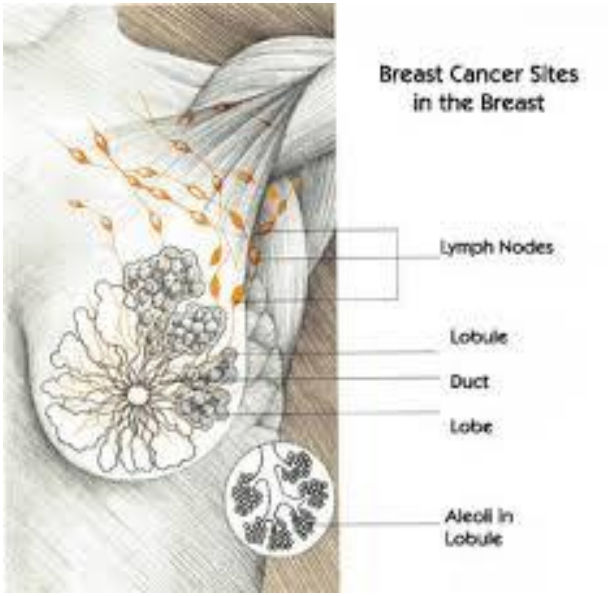
Part - 4

BREAST CANCER CLASSIFICATION

Breast Cancer

- Cancer affecting female and male breast tissues

- Most common malignancy among women
  - Estimated 1.38 million cases per year
  - 458,000 would die



- One of the first cancers documented
  - Egyptian queen Atossa got her breasts removed

- Considerable research breakthrough to improve survival over the last 30 years

Heterogeneity of breast cancer

- Traditionally perceived as a single disease
  - Just had varying degrees of severity
- Now understood and studied as a collection of diseases
  - Mainly enabled by large-scale gene expression and genomic profiling
- Affect the same organ site but have different
  - Histopathological features (as seen under the microscope)
  - Responses to treatment
  - Survival outcomes

Heterogeneity of breast cancer

- This poses a severe challenge
- Difficulty in
  - Estimating the type and dosage of treatment
  - Requires proper patient stratification

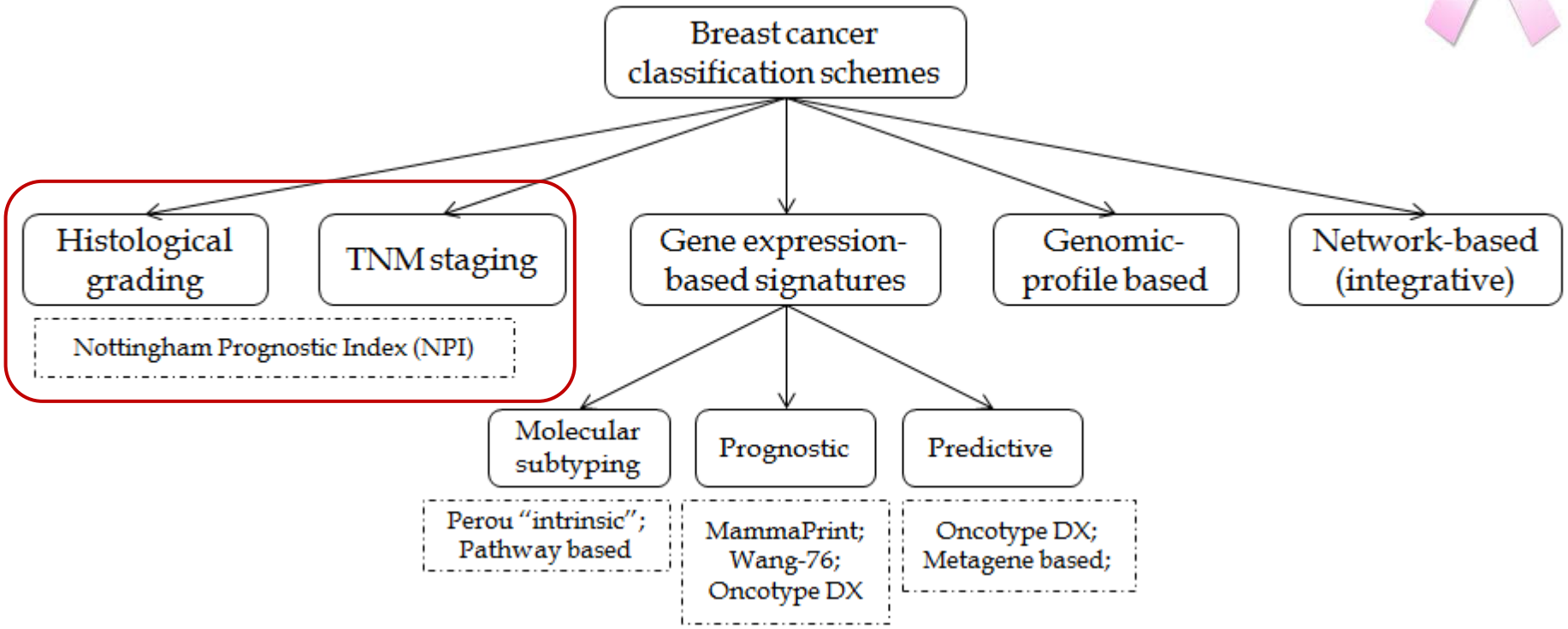
- A retrospective study of patients mammography screened between 1976 – 2008 in the US found (*NEJM 2012*)
  - ~1.3 million women were over-diagnosed and over-treated
  - Side-effects would be significant

Breast Cancer Classification

- Very important to
  1. Stratify patients for type and dosage of treatment
  2. Differentiate the diseases based on their molecular mechanisms and histopathological characteristics

Link the two – allow better decision making in the clinic

Breast Cancer Classification Schemes



Taherian, Srihari and Ragan, *Brief Bioinf* (2014)

Histological grading

- Description of the tumour based on the abnormality of tumour cells *vis-à-vis* normal cells
  - By looking at the cells under a microscope

- Four grades are assigned
  - Grade 1: highly similar to normal cells, grow slowly, low-grade tumours
  - Grade 3 – 4: highly dissimilar, grow rapidly, high-grade tumours

TNM System

- Tumour size, Lymph node invasion and Metastatic spread

- T (0-4) – size and location of tumour
  - T0 – no evidence of tumour
  - T1 – invasive part ≤ 20 mm, *in situ* (ducts and lobules)
  - T2 – invasive part 20 – 50 mm
  - T3 – invasive part ≥ 50 mm
  - T4 – tumour grown into the chest wall and skin, inflammation

- N (0-3): lymph node invasion
  - N0 – no cancer cells found in lymph nodes
  - N1 – cancer cells spread to three nodes
  - N2 – cancer cells spread to four-nine nodes
  - N3 – cancer cells spread to ≥ 10 nodes

E.g. T3 N2 M3

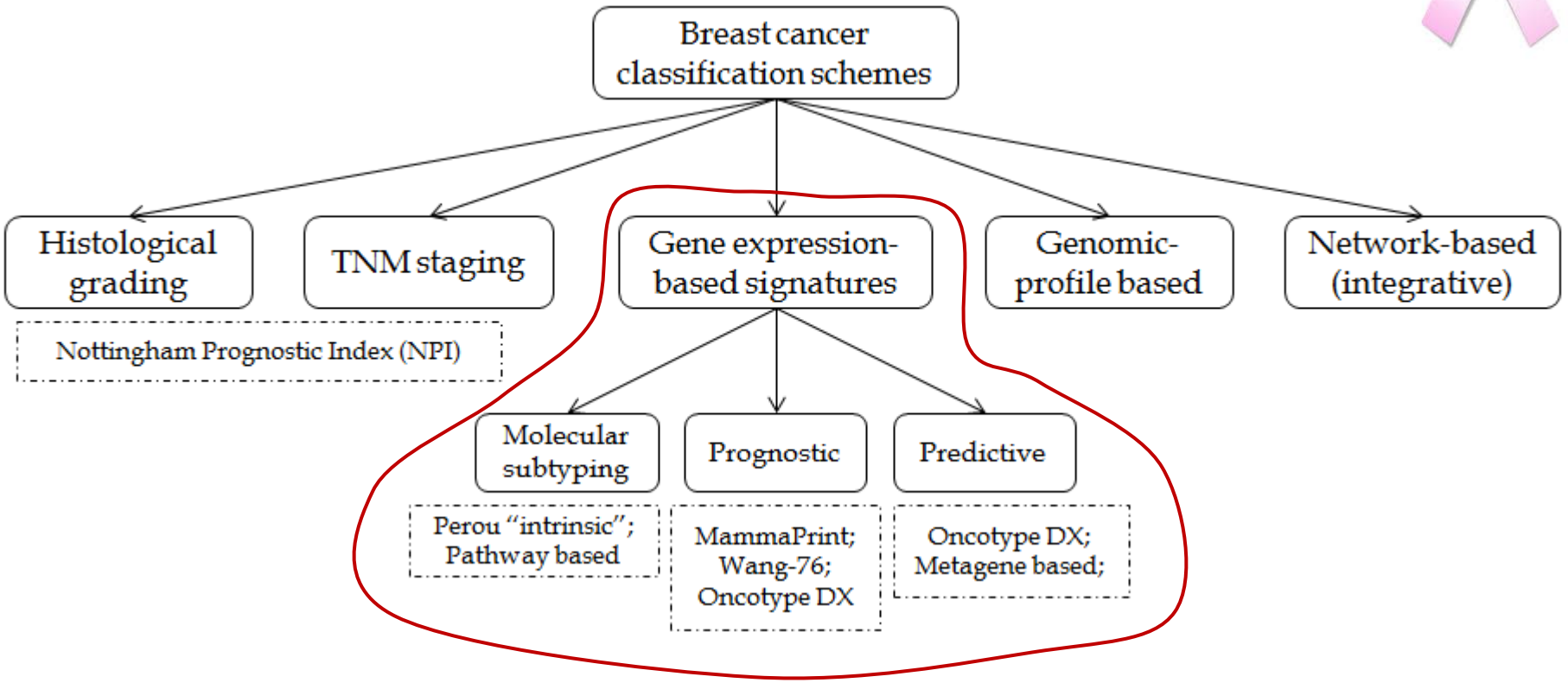
- M (0-4): metastatic spread
  - M0: no signs of metastatic spread
  - M1-M4: invasive *in situ* carcinoma

Nottingham Prognostic Index (NPI)

- Histological grading combined with TNM staging
  - E.g. G3 T3 N2 M3

- Approved and being used in clinics in the UK
  - Determine treatment options for patients

Breast Cancer Classification Schemes



Taherian, Srihari and Ragan, *Brief Bioinf* (2014)

Gene-expression signatures

- Based on landmark gene-expression profiling studies of 100's of patients : Perou *et al*., 2000; Sorlie *et al.,* 2001; van't Veer *et al*., 2002

- Gene-expression signatures
  - Molecular biomarkers
  - Could be used to determine disease prognosis
    - Severity of the disease, survival outcomes
    - Response to treatment

- Most importantly, identified first links between molecular mechanisms and disease prognosis

Molecular or “intrinsic” subtypes

- 85 tumour patients, 8102 genes measured
- 456 “intrinsic” genes by clustering
  - Genes that varied more in expression between clusters than repeated samples of the same cluster
- Robust when tested on an independent dataset of 78 breast cancers

- Four intrinsic subtypes:
  - Basal-like
  - luminal-A,
  - luminal-B
  - HER2+

ER-/PR- -- Basal-like
ER+/PR+ -- Luminal-A
ER+/PR- -- Luminal-B

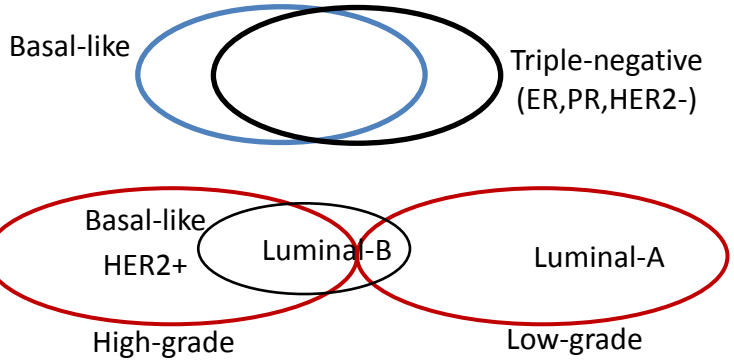
HER2+ -- HER2+ type
(mostly ER/PR+)

Receptor status

Estrogen Receptor (ER) +/-

Progesterone Receptor (PR) +/-

HER2+ -- ERBB2 gene over-expressed



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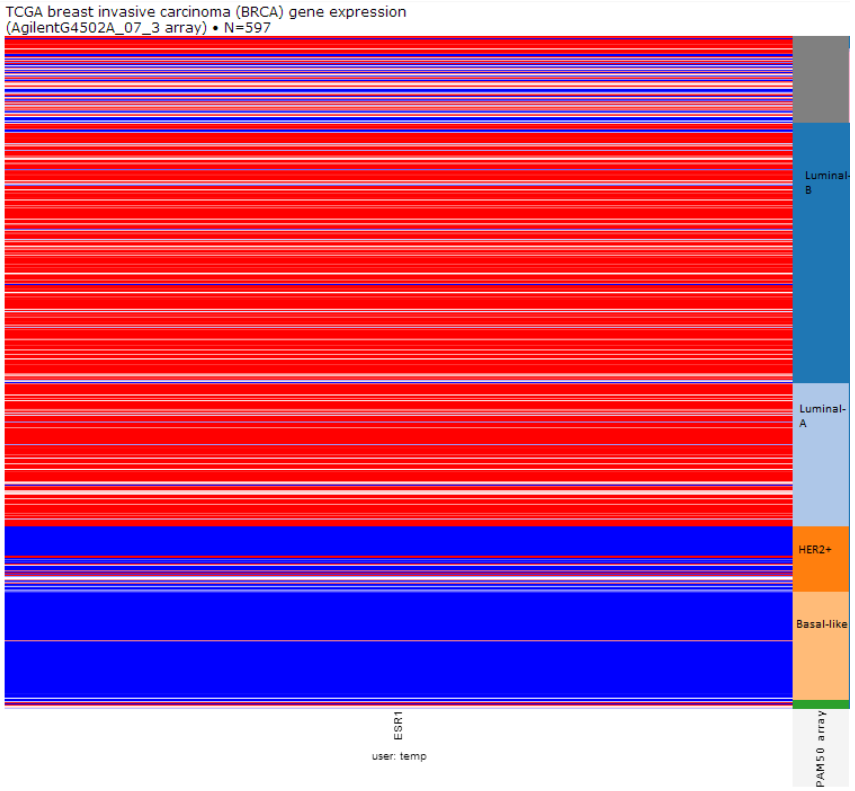
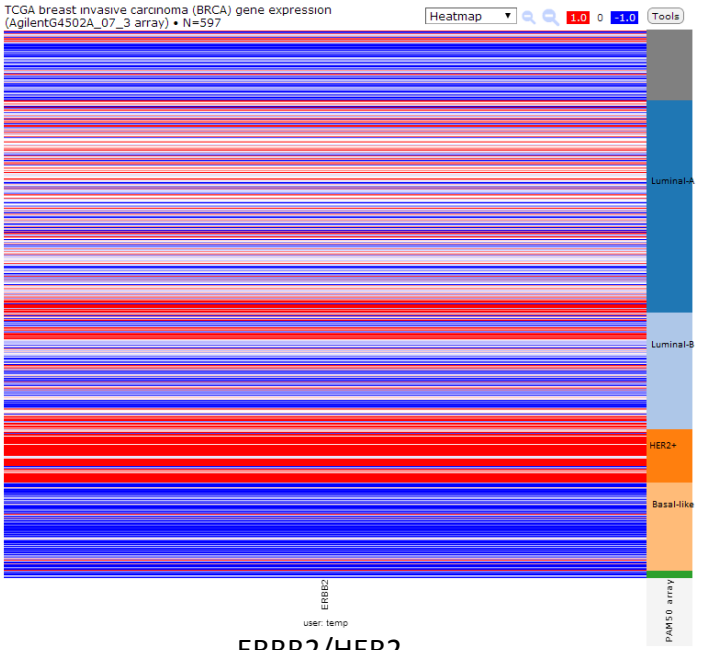


ESR1



ERBB2/HER2

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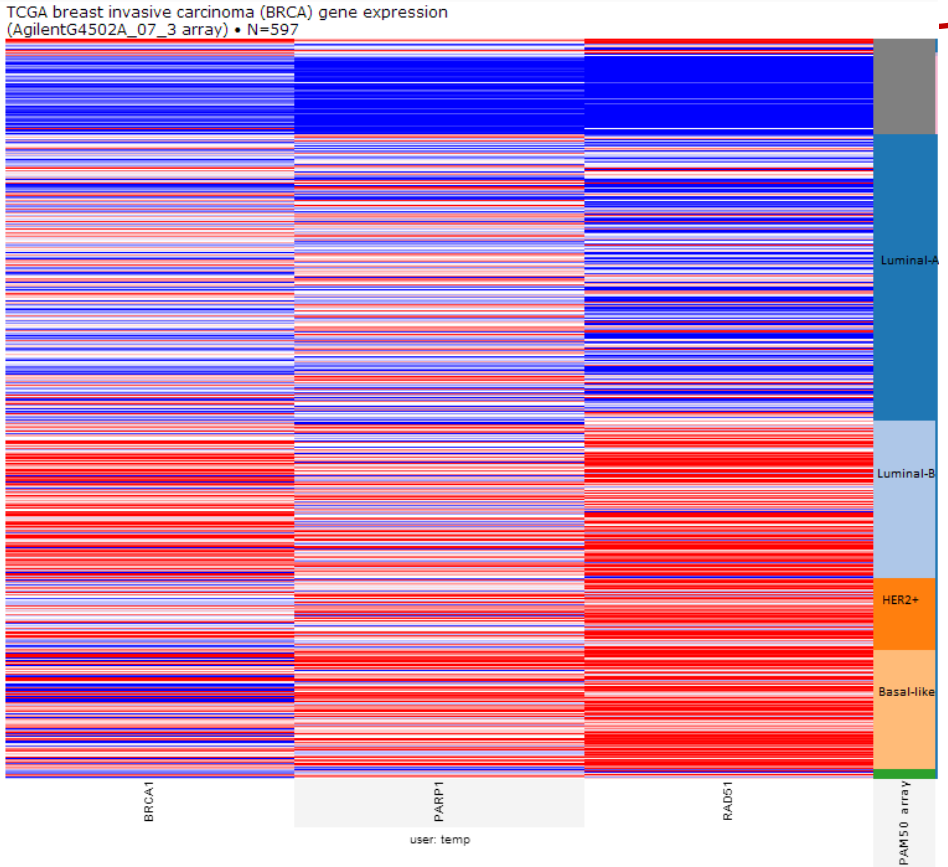

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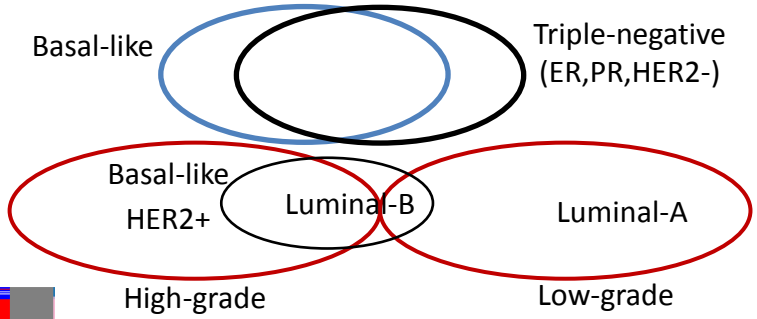

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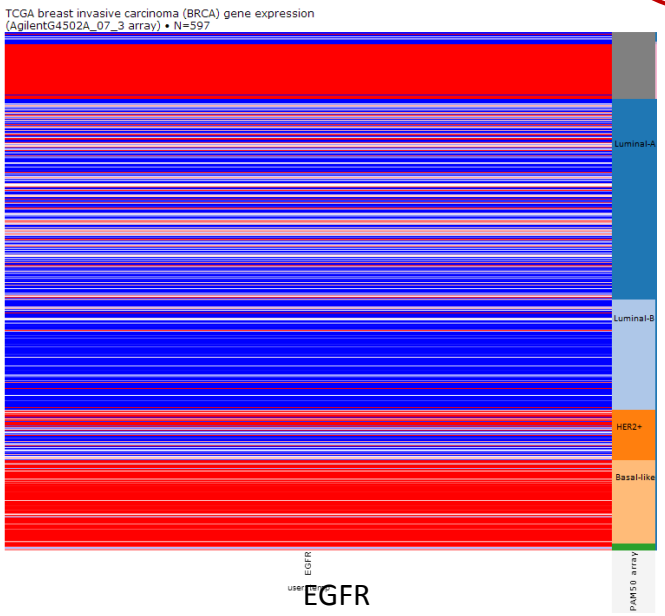
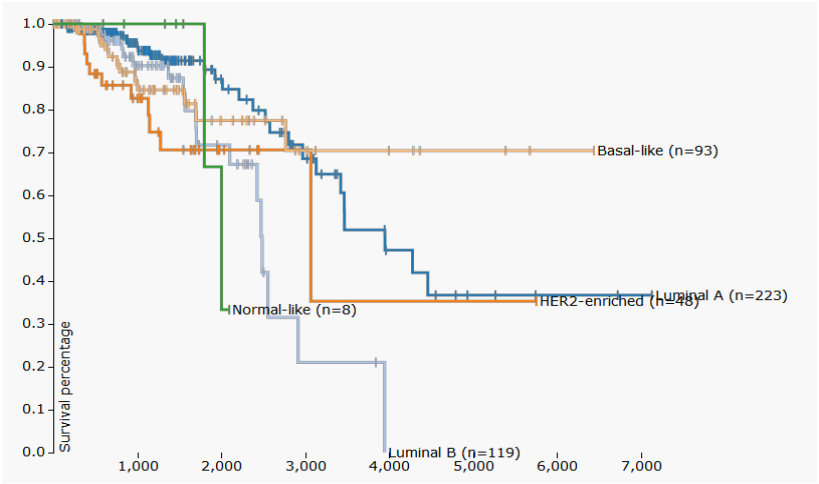


EGFR

Kaplan-Meier Curves



(days to death)

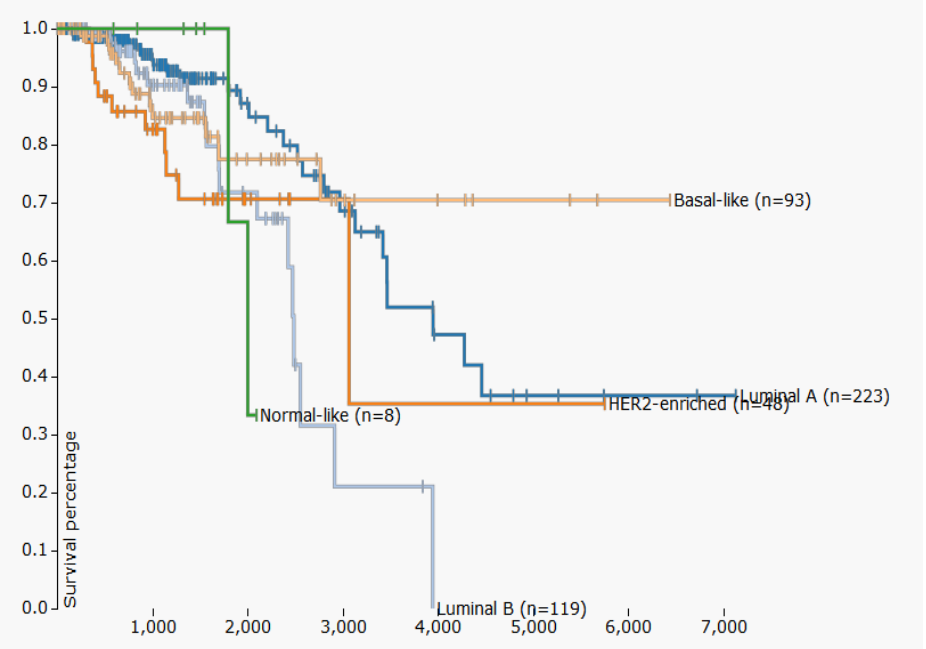
PAM 50

- Predictive Analysis of Microarray 50

- Parker *et al.,* 2009 found 50 genes which correlate well with these 456 intrinsic genes.

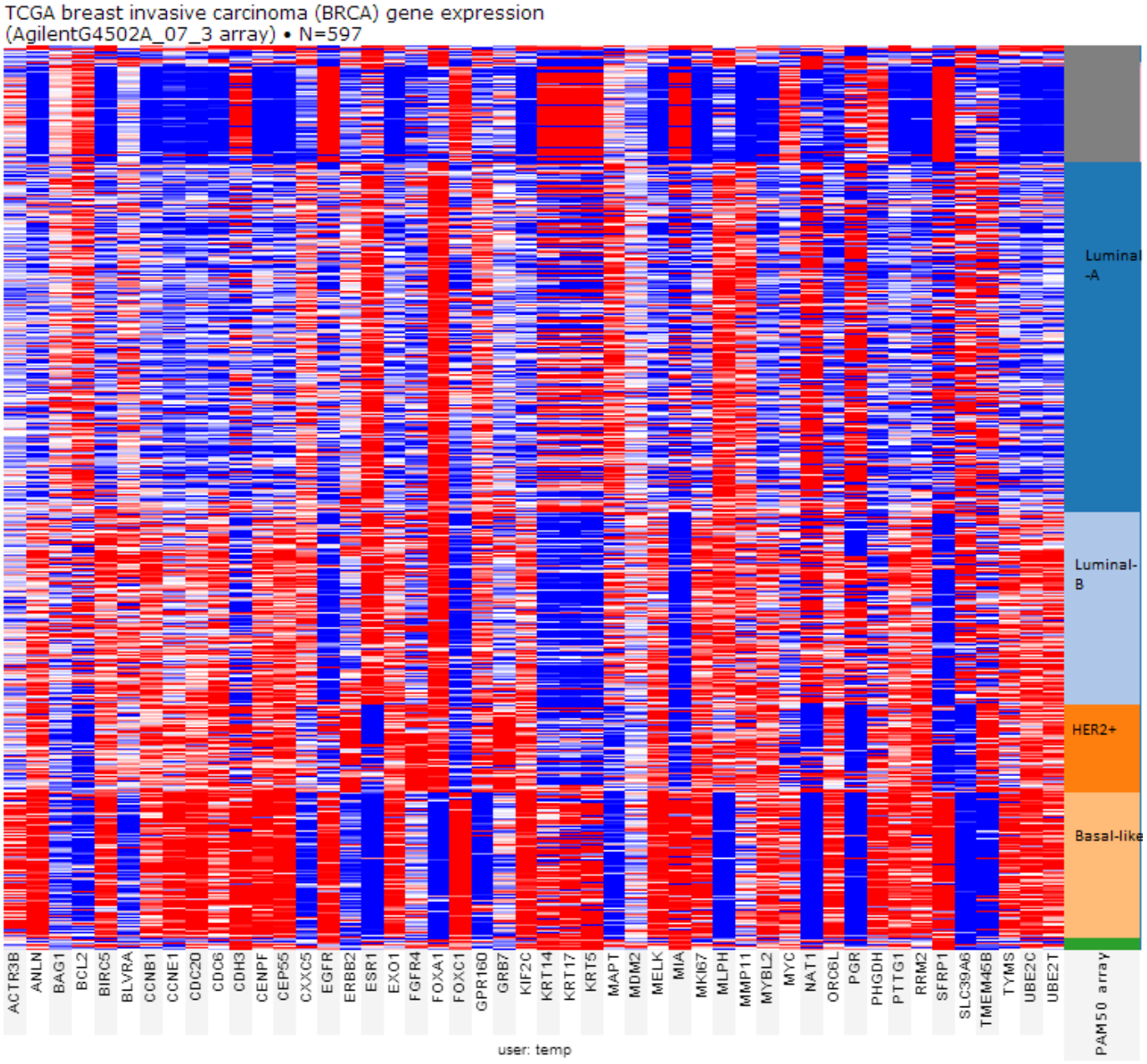
- PAM50 is more well-known now
- In the clinic
  - Just measure the ER, PR and HER2 status
  - Also, measure cytokeratins

Kaplan-Meier Curves



(days to death)

Expression pattern of PAM50 genes



First-generation signatures

- 70-gene signature (van’t Veer et al., 2002)
- 76-gene signature (Wang et al., 2005)
- 21-gene signature (Paik et al., 2004)

70-gene signature (van't Veer et al., 2002)

- Amterdam-70 / MammaPrint (Agendia, Amsterdam)
  - Approved by the US Food and Drug Administration

- TNM stage 1-2, node-negative, size ≤ 50mm
  - Predict prognosis in these patients
  - Chemotherapy sensitivity

- Found from 78 such patients, probing 25000 genes
- Tested on independent dataset of 295 patients

- Poor in differentiating within ER-negative tumours

76-gene signature (Wang et al., 2005)

- 115 cancers were analysed
- ER+ and ER- were treated separately
- ER-positive: 60 genes
- ER-negative: 16 genes

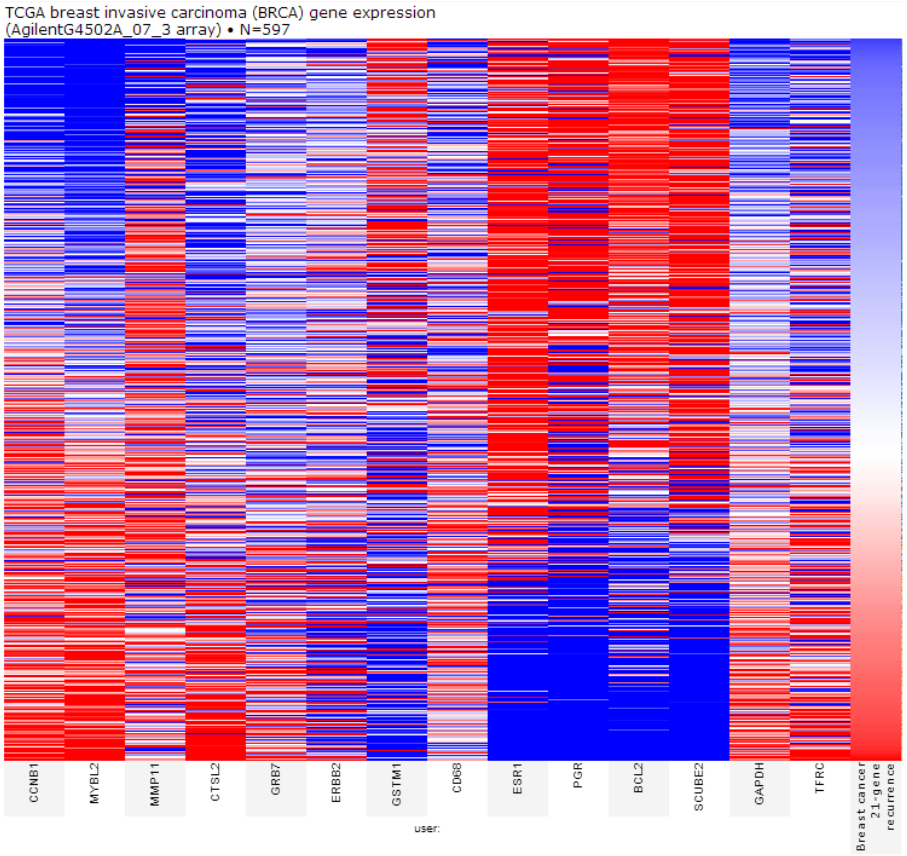
- Validated on an independent dataset of 171 patients
- Again, 16 genes not effective on ER-negative tumours

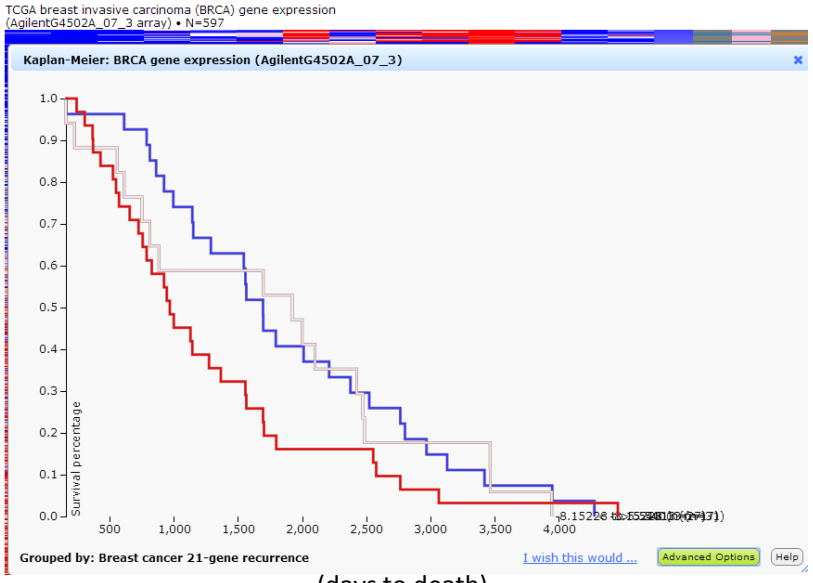
Oncotype DX (Paik et al., 2004)

- Genomic Health, USA
- FDA approved

- 21-gene signature
- Recurrence Score (RS) – predict distant relapse at 10 years for ER-positive, node-negative tumours
- Tested retrospectively on 668 tamoxifen-treated patients (Habel et al., 2006)
- 651 patients with adjuvant chemotherapy (Paik et al., 2006)

Oncotype DX expression pattern and survival plots





(days to death)

Second-generation signatures

- First-generation signatures fail at predicting outcomes for ER-negative tumours

- These signatures contain proliferation genes
- ER-negative tumours are highly proliferative

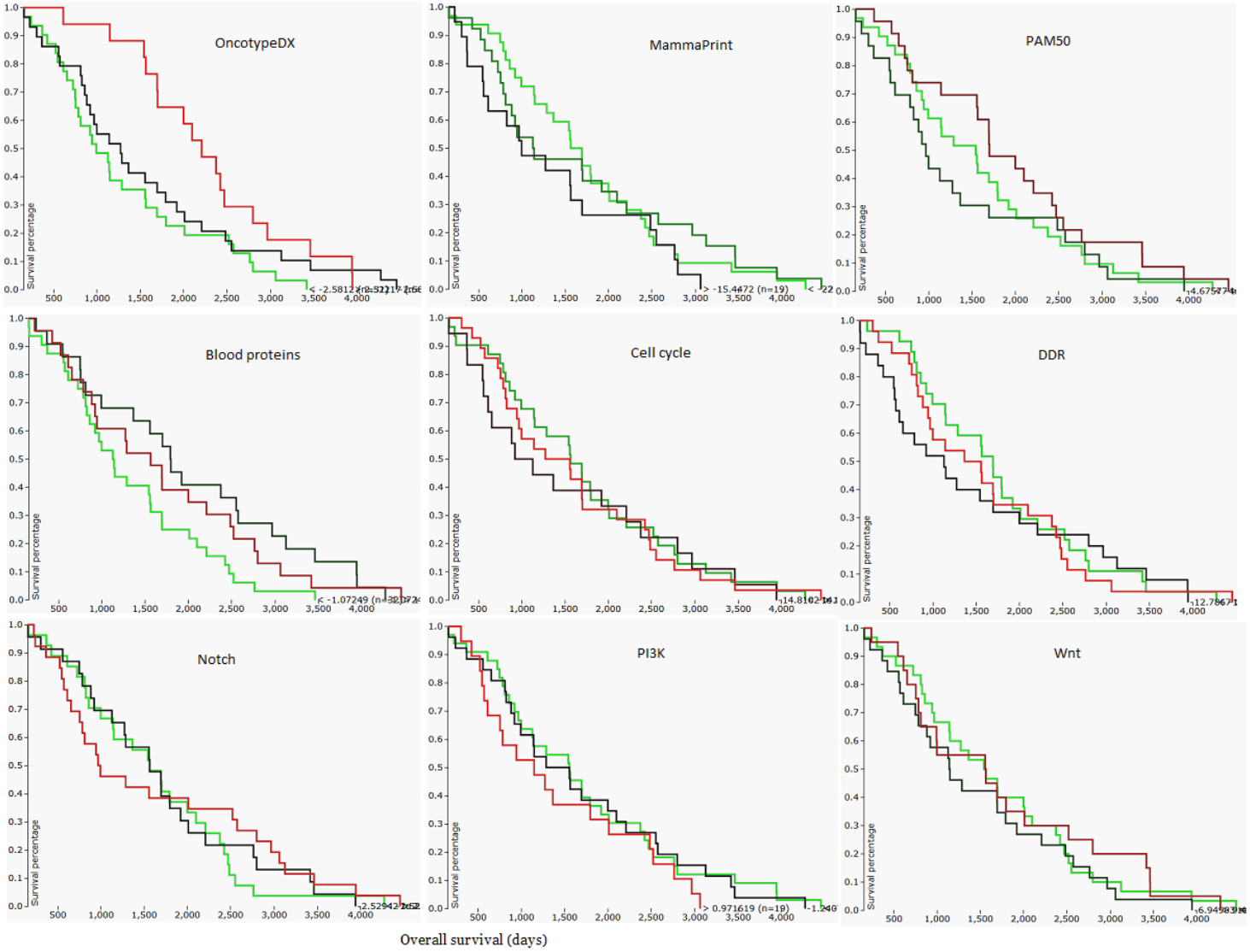
- Other genes are required such as immune response, stromal compartment, etc.
- Bianchini et al., 2010; Nagalla et al., 2013

Predictive signatures

- Signatures that could predict the response to therapies
- Oncotype DX has been shown to be beneficial for
  - Adjuvant chemotherapy
  - Neoadjuvant chemotherapy

- A recent study identified ‘metagenes’, ie, groups of co-expressed genes (Farmer et al., 2009)
  - Neoadjuvant chemotherapy
  - 82 breast cancer patients
  - Tested on independent 51 patients

Pathway-based signatures

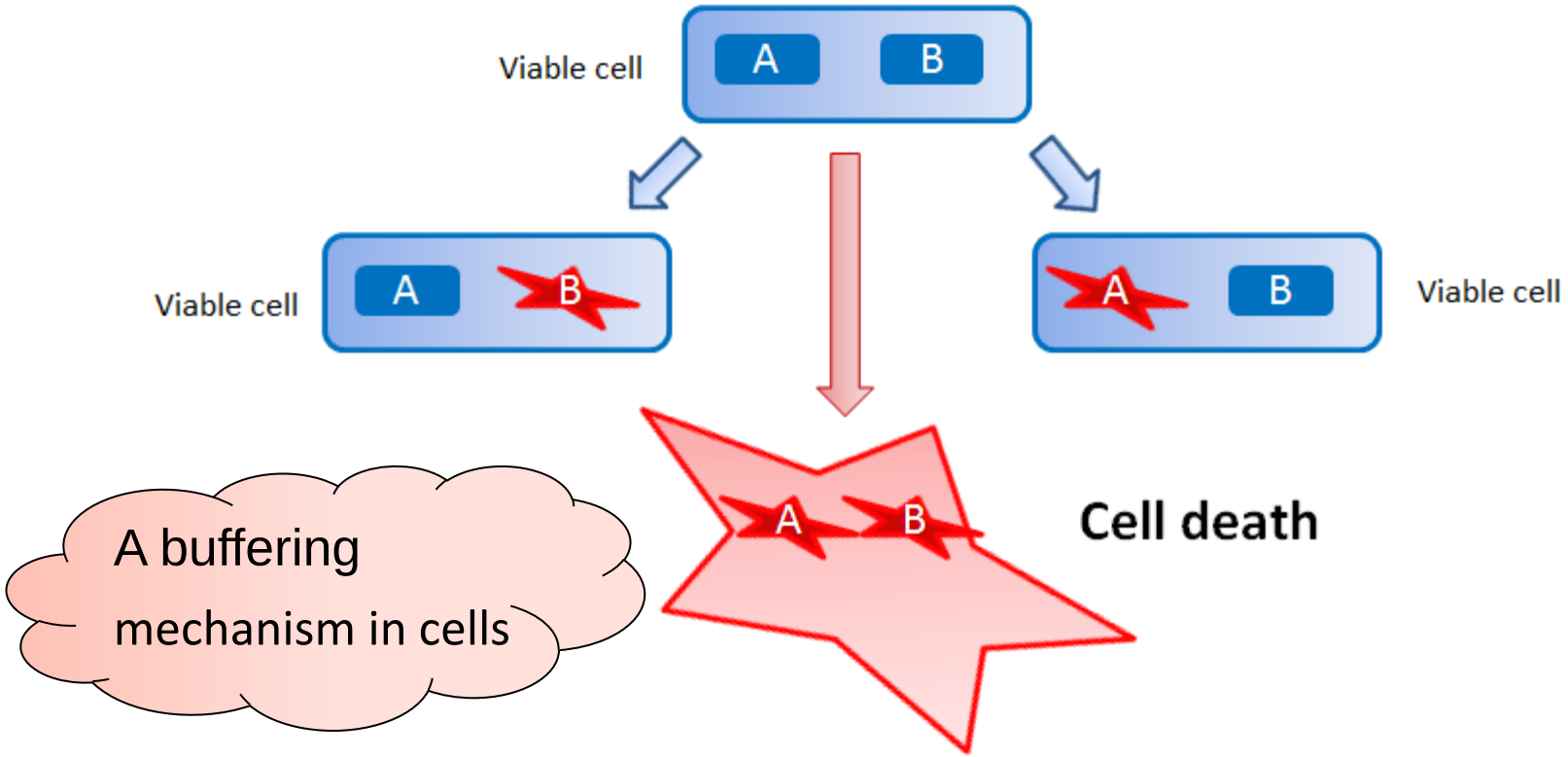


Taherian, Srihari and Ragan, *Brief Bioinf* (2014)

Part - 5

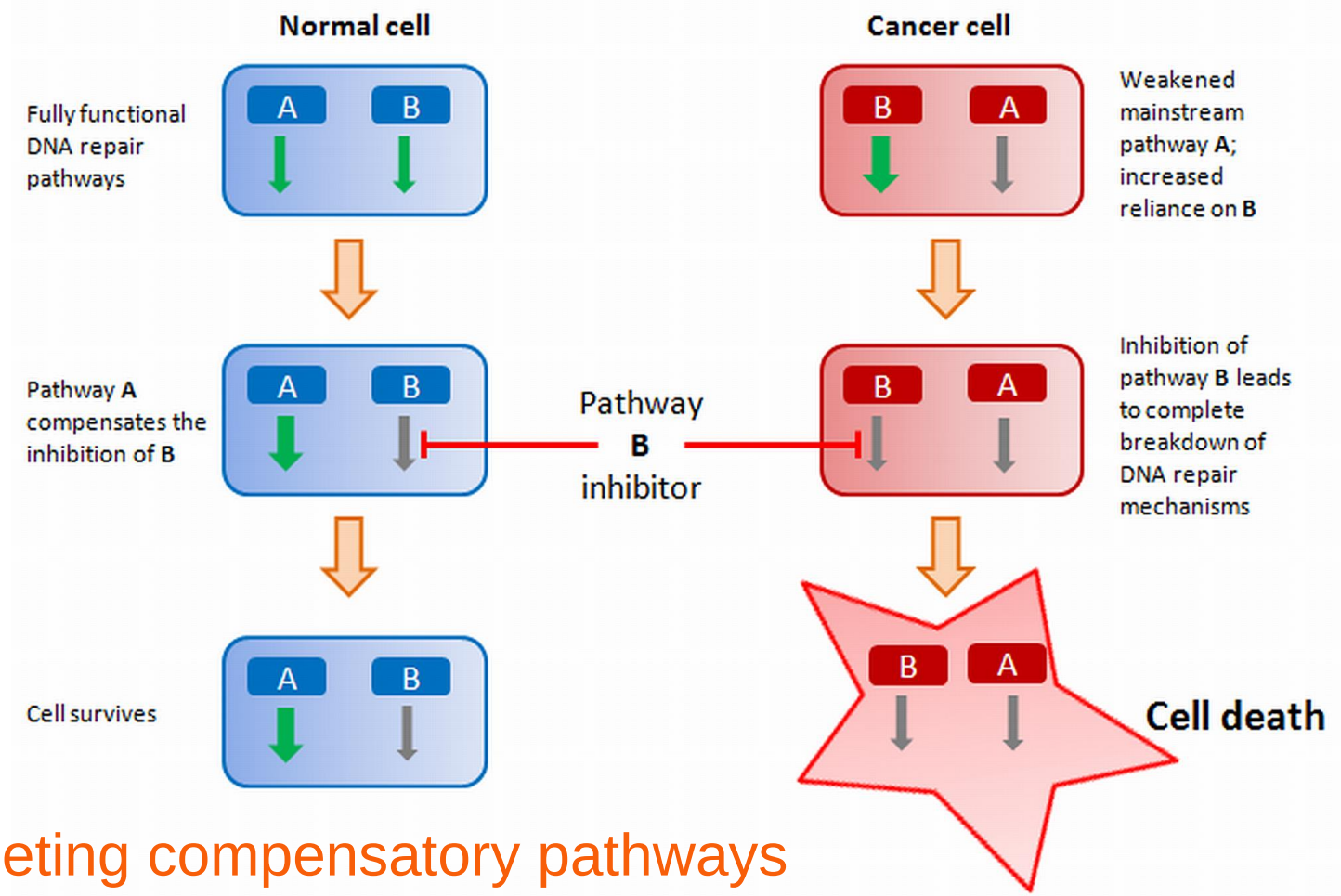
CANCER THERAPY – FOCUS ON SYNTHETIC LETHALITY-BASED THERAPY

Synthetic lethality: a new promise for selective killing of cancer cells



Targeting the **synthetic lethal partners** of lost genes (tumour suppressors) can selectively kill cancer cells.

SL-based therapy: how does it work?

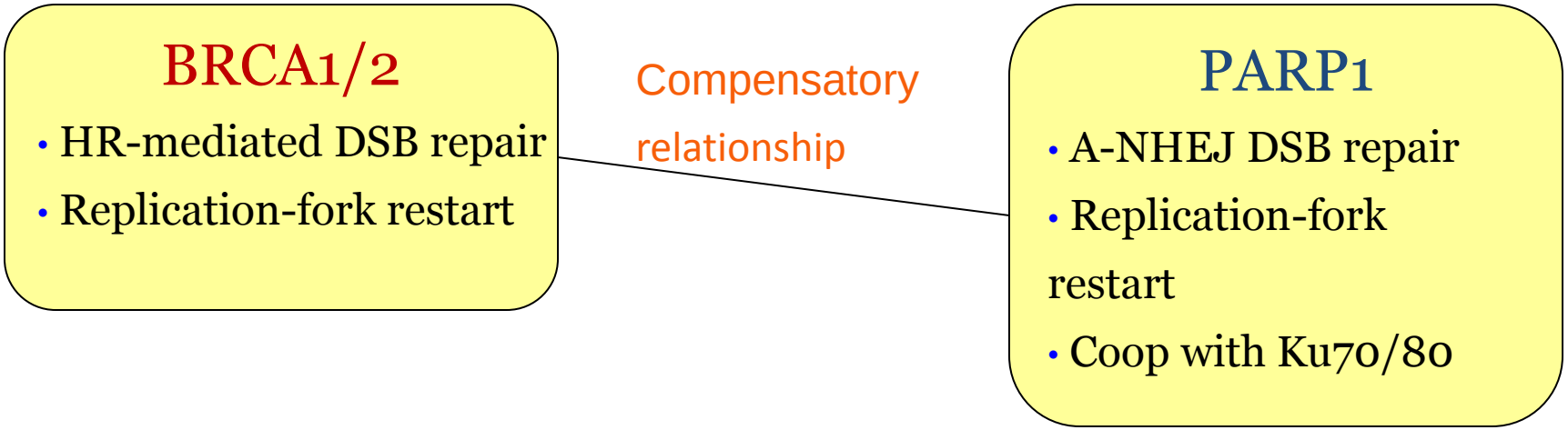


Targeting compensatory pathways

Liu & Srihari *et al.*, *Nucleic Acids Research* 2014 (Review)

BRCA1-PARP1: a clinically relevant relationship

- BRCA1/BRCA2-deficient tumours are sensitive to PARP inhibition
  - Such cancer cells die upon PARP inhibition
  - In general, PARP is SL with several other HR genes
  - Explored as a therapeutic target in “BRCAness” tumours



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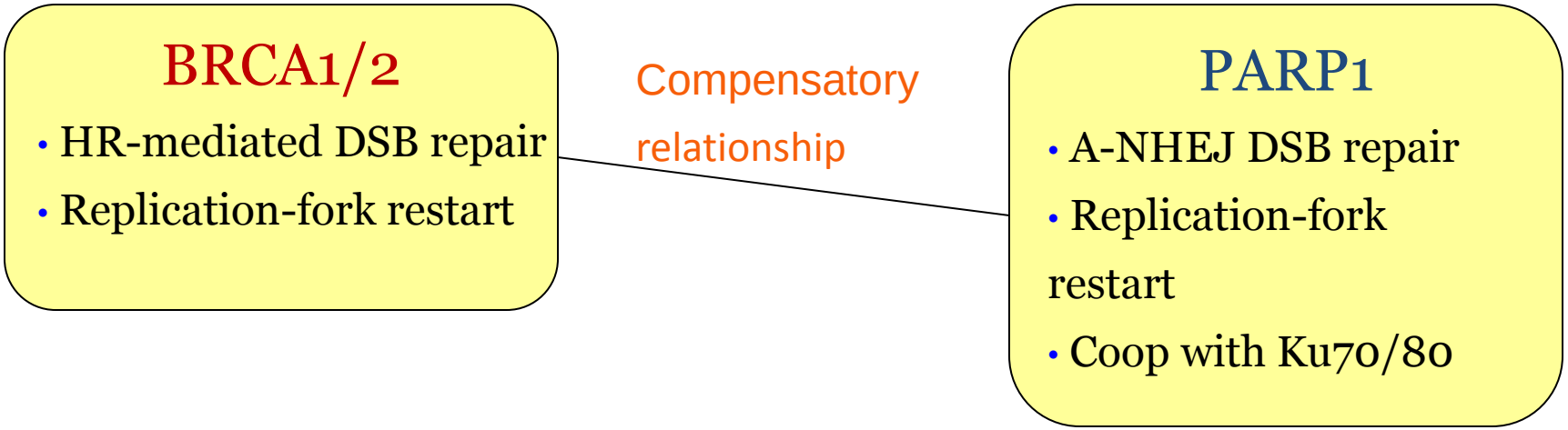

- PARP1-/- mice are viable and fertile; PARP1-*ko* is not lethal.
- Cancer cells undergo significant replicative stress
- Generates considerable lesions through replication fork stalling
- PARP is required to facilitate fork restart and enable HR-dependent repair of lesions
- In the event of deficient HR, further inhibition of PARP is lethal to cancer cells

Thank You!

Source: http://www.sriganeshsrihari.net/