BIO-373

Genetics & Genomics

Cancer genetics

Sebastian M. Waszak

School of Life Sciences

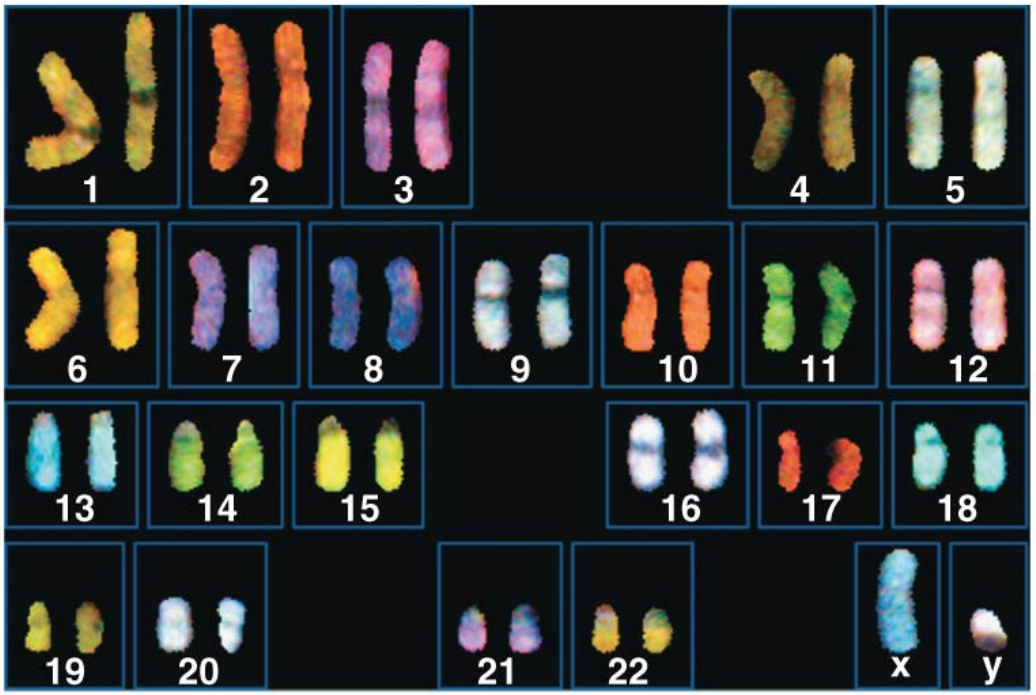
Laboratory of Computational Neuro-Oncology

Plan

1. Cancer is a genetic disease
2. Genetic and epigenetic changes in cancer cells
3. Control of cell cycle
4. Proto-oncogenes and tumor suppressor genes
5. Familial forms of cancer
6. Carcinogens

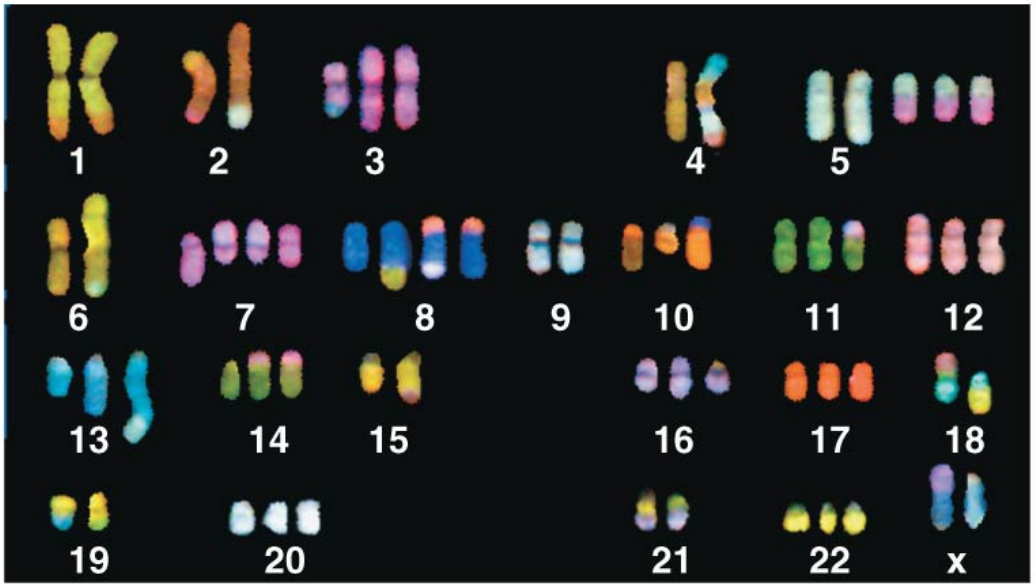
1. Cancer is a genetic disease

(a)



Karyotype of a normal cell

(b)



Karyotype of a cancerous cell, which shows a mix of aneuploidy, deletions and translocations

Malignant tumours (cancer) are genetic diseases

- Malignant tumours are a group of diseases impacting fundamental cellular functions:
  - DNA repair
  - Cellular cycle
  - Apoptosis
  - Cellular differentiation
  - Cell-cell interactions
- Malignant tumours are caused by mutations or chromosomal alterations in **somatic** cells (i.e. genetic disease at the somatic level)
- 0% to 40% of malignant tumours are also associated with pathogenic **germline** variants

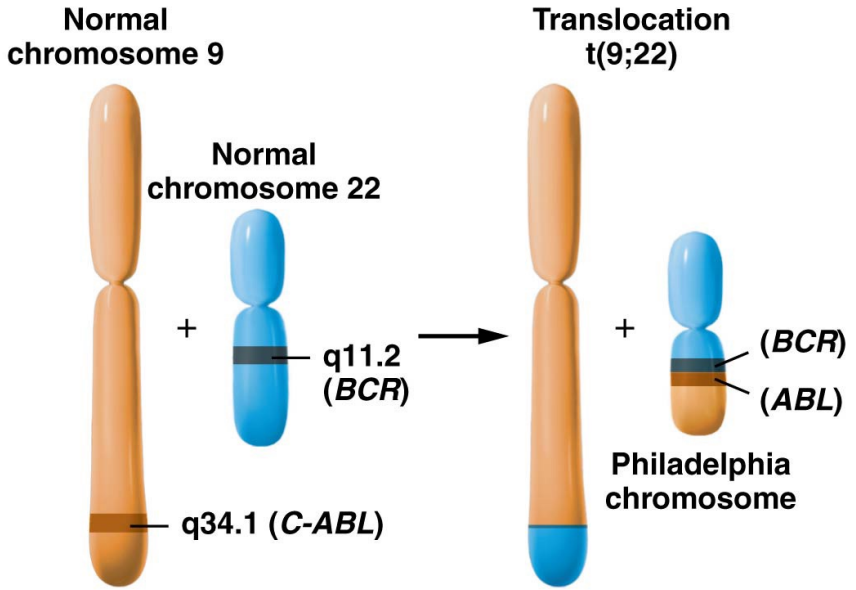
Fundamental properties of cancer

- **Unregulated cell proliferation** due to abnormal cell growth and division
- **Metastatic spread**

- All cancer cells in primary and secondary tumors are **clonal**
  - They all originate from a common ancestral cell that accumulated numerous mutations
  - Proof of clonality of cancer cells:
    - Reciprocal translocations
    - X-inactivation

Proof of cancer cell clonality

- **Reciprocal translocations**
  - Observed in many cancers (eg, leukemia, lymphoma, glioma)
  - Example: **Chronic myelocytic leukemia**:
    - Translocation of *C-ABL* gene from chromosome 9 to 22, close to *BCR* gene
    - Creation of ***BCR-ABL*** fusion gene encoding an abnormal signal transduction protein, which continuously stimulates cell proliferation



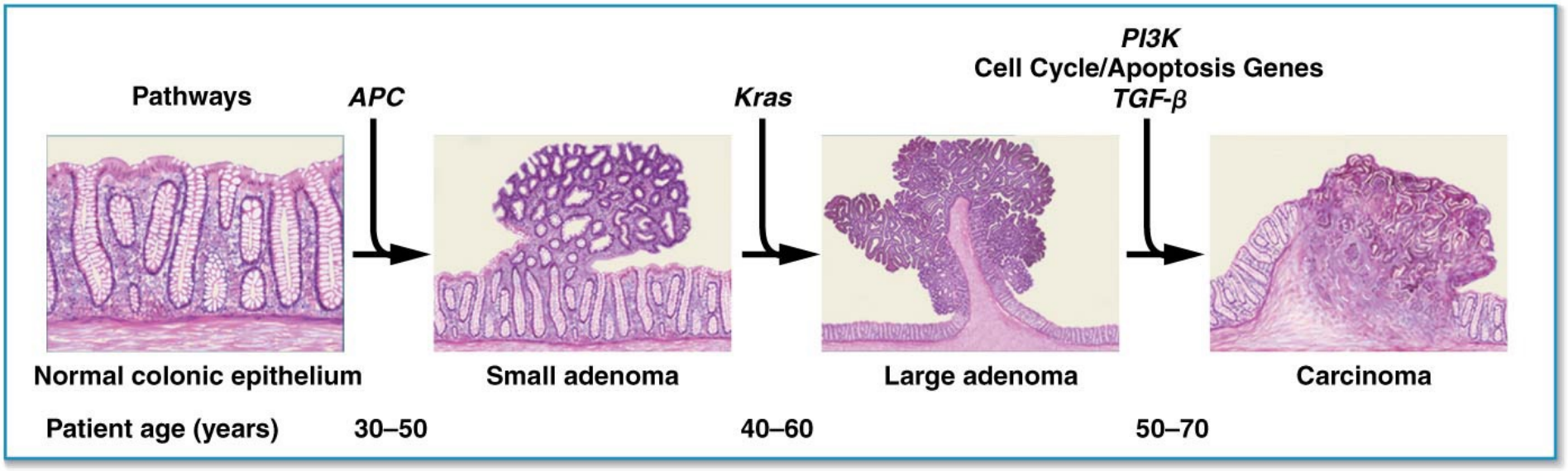
Proof of cancer cell clonality

- **X-chromosome inactivation**
  - Random inactivation of one of the two X chromosomes in each female cell, occurring early in embryonic development
  - All cells from the same cancer in a woman contain the same inactivated X chromosome (both primary and metastatic tumors)

Cancer development is a multistep process

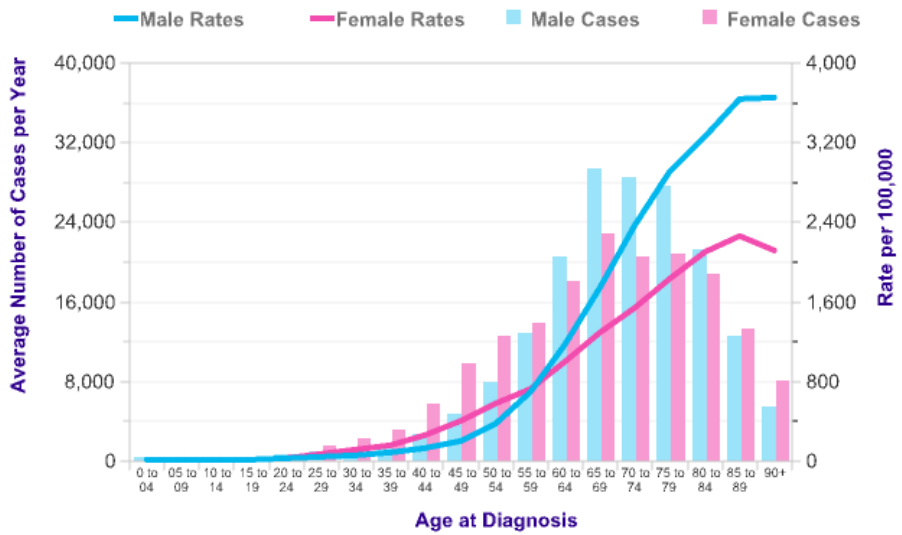
- The steps of cancer development (**carcinogenesis**) are sometimes visible, as here for colon cancer

normal tissue → benign adenoma → carcinoma



Cancer development: age

- The incidence of most cancers increases with age
- Independent random somatic mutations are necessary for the malignant transformation of a cell:
  - Mutation rate in humans: 10^{-6} per gene per cell division
  - Estimated number of cell divisions over 80 years: 10^{16}
  - An individual can thus accumulate up to 10^{10} mutations per gene without developing cancer



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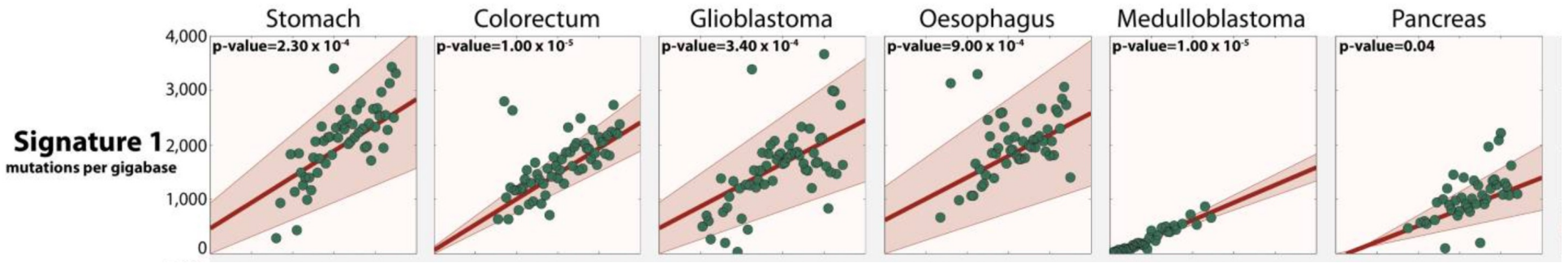
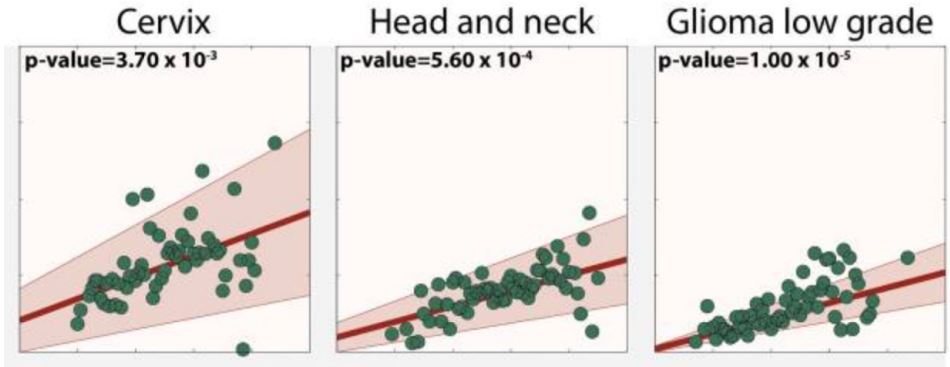
Cancer development: somatic CpG mutations in cancer and ageing





2. Genetic and epigenetic changes in cancer cells

Types of mutations found in tumors

- **Driver mutations** give growth advantage to tumor cells
  - In genes important for **genome stability** and **DNA repair**
  - In genes controlling **cellular cycle** and **apoptosis** (proto-oncogenes and tumor suppressor genes)

- **Passenger mutations** have no direct contribution to cancer phenotype

Genomic instability

- **Genomic instability** in cancer cells, sometimes described as "**mutator phenotype**", manifests in multiple somatic DNA defects such as:
  - Translocations
  - Aneuploidy
  - DNA amplification
  - Deletions

Epigenetic modifications

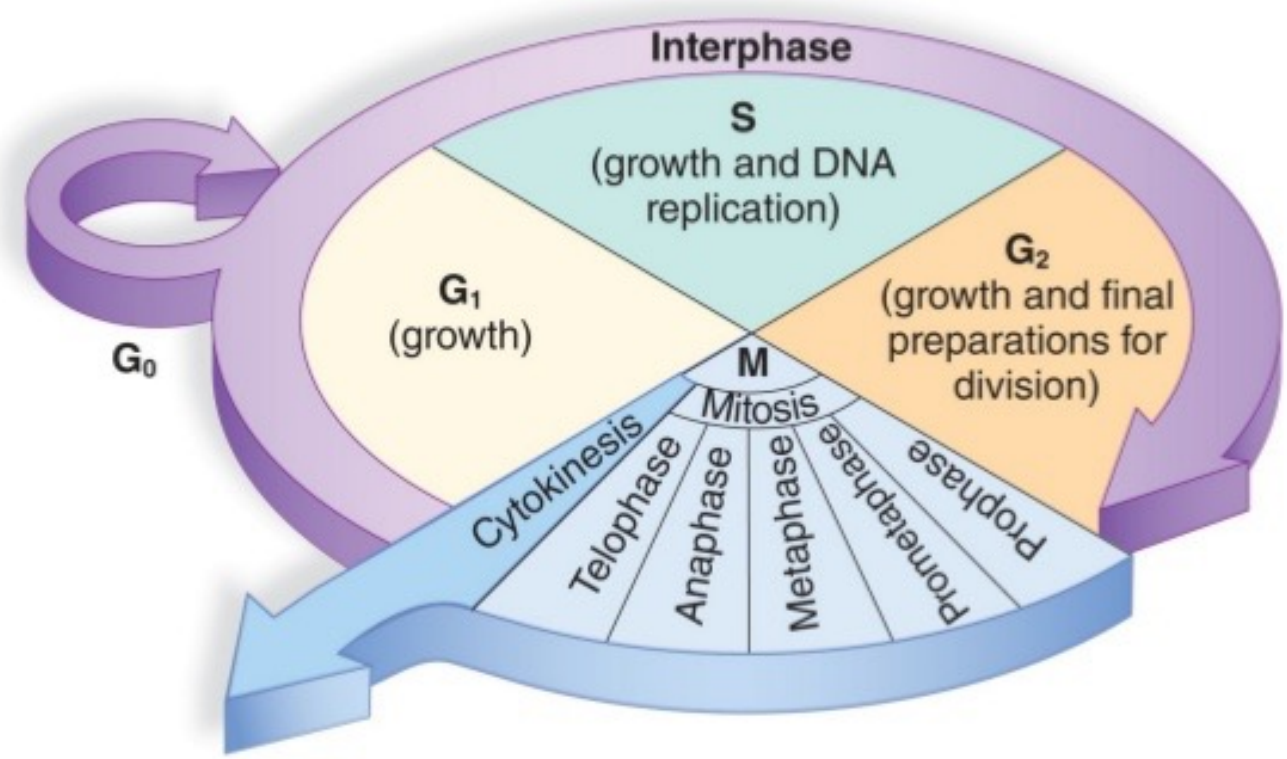
- **Epigenetics** is the study of factors that affect gene expression but do not alter the DNA sequence

- **DNA methylation (5-methylcytosine, 5mC)**
  - Gene silencing (eg, loss of HR due to promoter hypermethylation of *RAD51C* or *BRCA1*)
  - Parental imprinting (eg, loss of IGF2 imprinting in colorectal cancer)
- **Histone modifications**
  - Genes that encode **histone acetylases**, **deacetylases**, **methyltransferases**, and **demethylases** are often mutated or aberrantly expressed in cancer cells

3. Control of cell cycle

Cell cycle

- Cellular events, in sequence, from one division to another
- Phases of cell cycle:



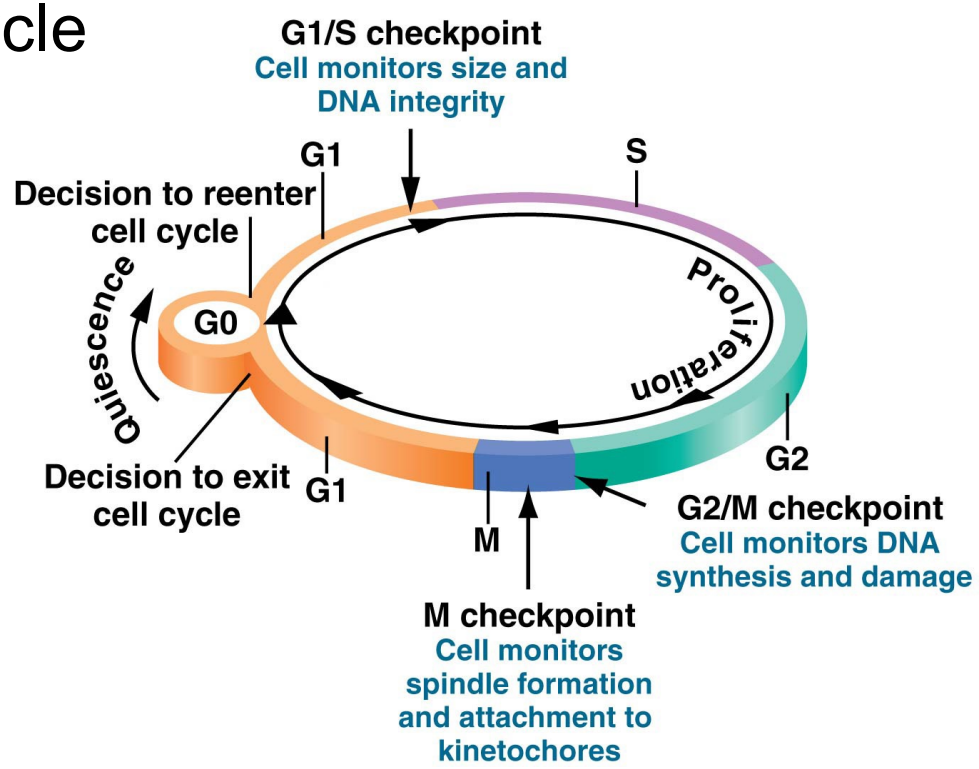
Phases of the cell cycle

- **Mitotic division** (**M** phase)
- **Interphase**
  - Interval during which the cell grows and replicates its DNA (subdivided into **G1**, **S** and **G2** phases)
  - Between mitotic divisions
- **Quiescence** (**G0** phase)
  - Cell does not grow or divide but can be metabolically active

- Cancer cells are unable to enter G0 and cycle continuously

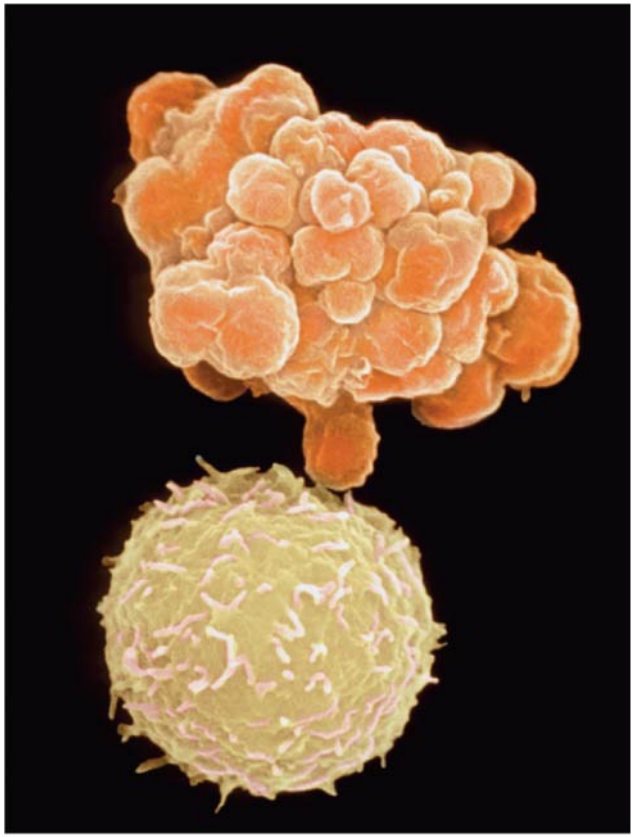
Cell cycle control and checkpoints

- **G1/S**, **G2/M**, and **M checkpoints**
  - Three distinct checkpoints where the cell monitors external signals and internal equilibrium
  - At each checkpoint, the cell decides whether to proceed to the next stage of the cell cycle



Apoptosis

- Programmed cell death
- Occurs when DNA damage is too severe to repair
- Genetically controlled process, essential to avoid cancer
- Different from necrosis: apoptosis does not trigger any inflammatory response



- **Steps of apoptosis**
  - Fragmentation of nuclear envelope
  - Disruption of internal cellular structures
  - Dissolution of cell into small apoptotic bodies
  - Engulfing of apoptotic bodies by phagocytic cells

4. Proto-oncogenes and tumor-suppressor genes

Proto-oncogenes

- Genes whose products promote cell growth and division:
  - **Transcription factors** that stimulate expression of other genes
  - **Signal transduction molecules** that stimulate cell division
  - **Cell-cycle regulators** that move cell through cell cycle

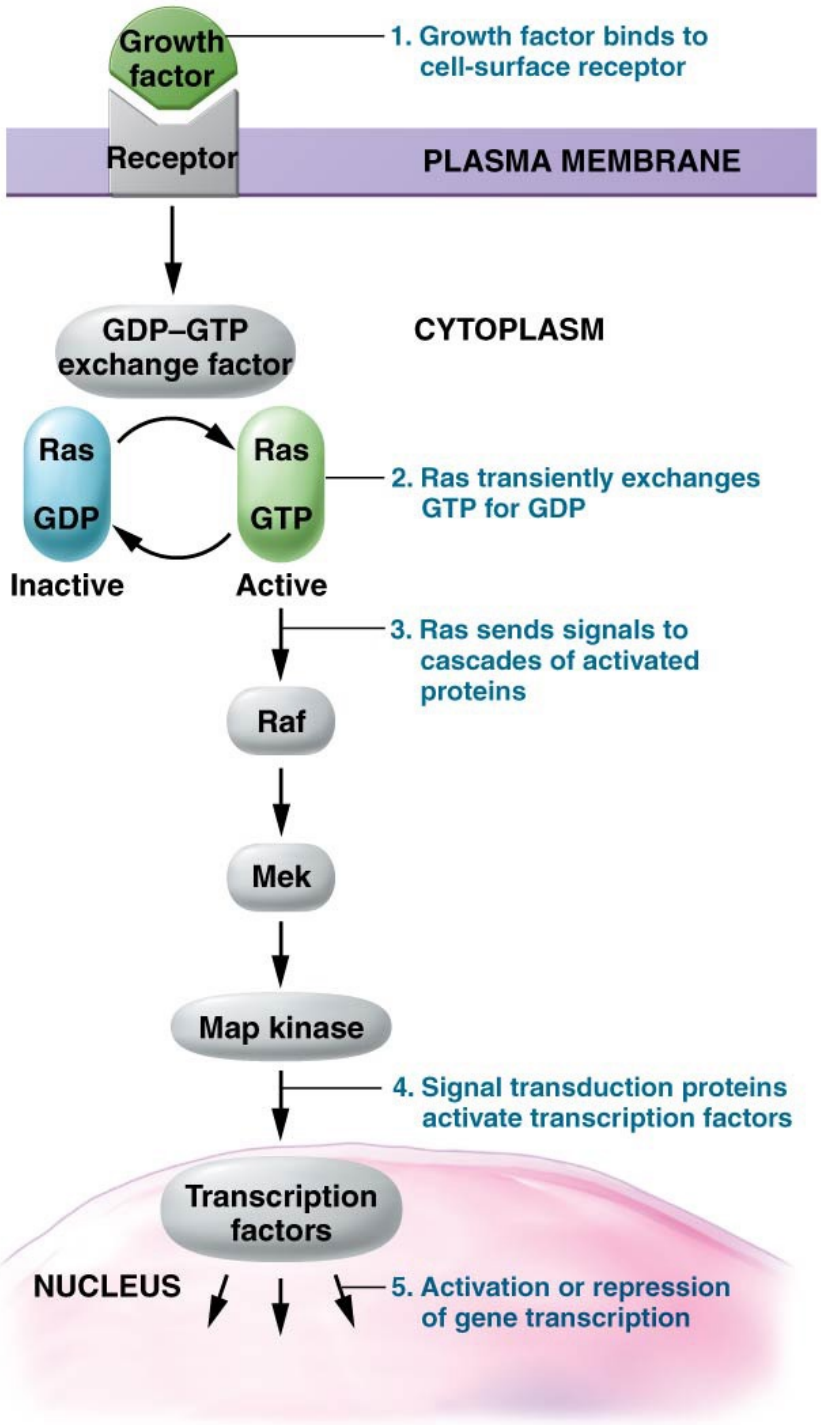
Oncogenes

- Mutated or aberrantly expressed proto-oncogenes
- Result from “gain-of-function” alteration
- **Dominant** phenotype = only one allele needs to be mutated or overexpressed to contribute to cancer

Proto-oncogene	Normal Function	Alteration in Cancer	Associated Cancers
c-myc	Transcription factor, regulates cell cycle, differentiation, apoptosis	Translocation, amplification, point mutations	Lymphomas, leukemias, lung cancer, many types
c-kit	Tyrosine kinase, signal transduction	Mutation	Sarcomas
RARα	Hormone-dependent transcription factor, differentiation	Chromosomal translocations with *PML* gene, fusion product	Acute promyelocytic leukemia
E6	Human papillomavirus encoded oncogene, inactivates p53	HPV infection	Cervical cancer
Cyclins	Bind to CDKs, regulate cell cycle	Gene amplification, overexpression	Lung, esophagus, many types

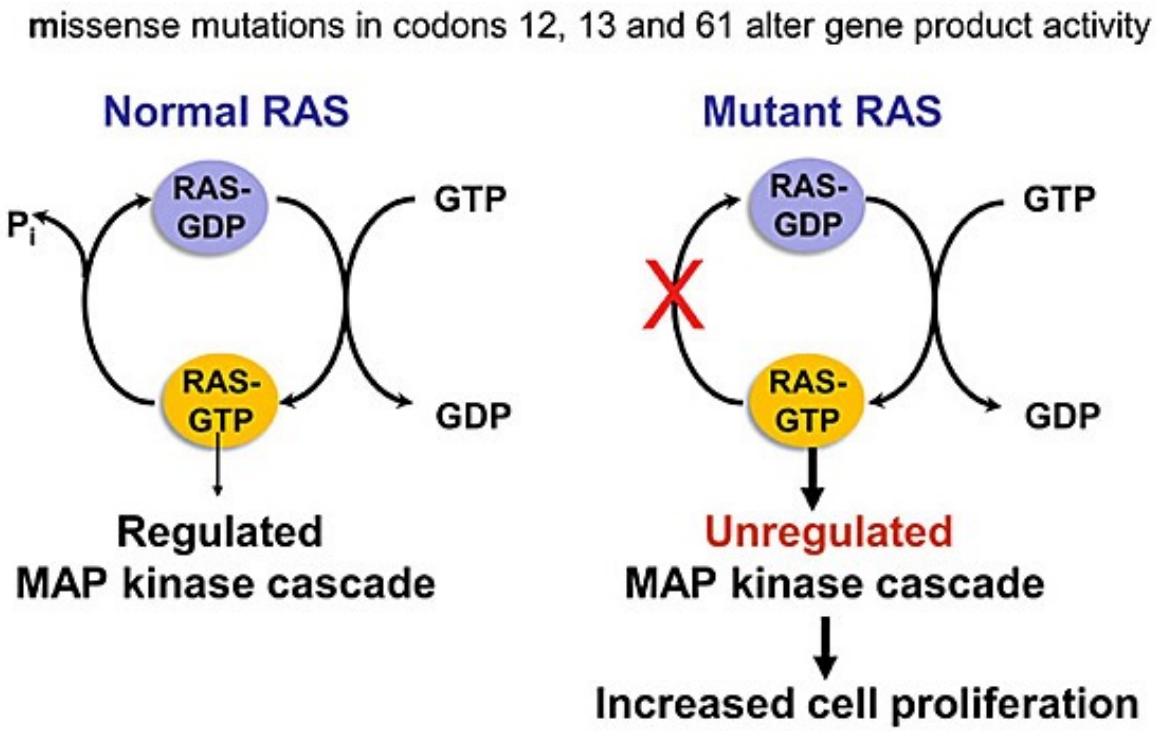
ras proto-oncogenes

- The *ras* genes (*HRAS*, *NRAS* and *KRAS*) are among the most frequently mutated genes in human tumors (about 40% of cancers)
- They encode signal transduction molecules associated with cell membrane
- Normal function = regulation of cell growth and division in response to external stimuli



ras proto-oncogenes

- Mutations (eg, *KRAS G12*) transforming a proto-oncogenic *ras* into an oncogene make the Ras protein unable to hydrolyze GTP into GDP, blocking it in its active form ➔ non-stop stimulation of cell proliferation



Tumor-suppressor genes

- Genes that normally regulate cell-cycle checkpoints and initiate process of apoptosis
- If they are mutated, they are unable to control cell proliferation and apoptosis, which leads to cancer
- **Recessive** phenotype = both alleles must be inactivated for the cell to become cancerous

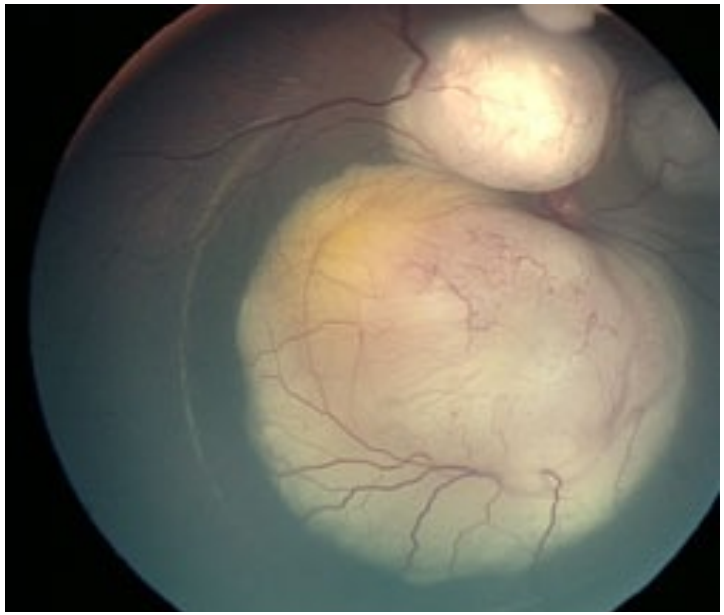
Tumor-Suppressor	Normal Function	Alteration in Cancer	Associated Cancers
RB1	Cell-cycle checkpoints, binds E2F	Mutation, deletion, inactivation by viral oncogene products	Retinoblastoma, osteosarcoma, many types
APC	Cell–cell interaction	Mutation	Colorectal cancers, brain, thyroid
p53	Transcription regulation	Mutation, deletion, viruses	Many types
BRCA1, BRCA2	DNA repair	Point mutations	Breast, ovarian, prostate cancers

p53 tumor-suppressor gene

- Most frequently mutated gene (50% of all cancers)
- *p53* encodes a transcription factor that represses or stimulates transcription of >50 genes
- The p53 protein is continuously synthesized but rapidly degraded, so is present at low levels in normal cells
- DNA damage ➔ rapid increase in p53 levels ➔ cell cycle arrest and initiation of apoptosis
- Due to its central role in genomic integrity, *p53* has been called the “guardian of the genome”

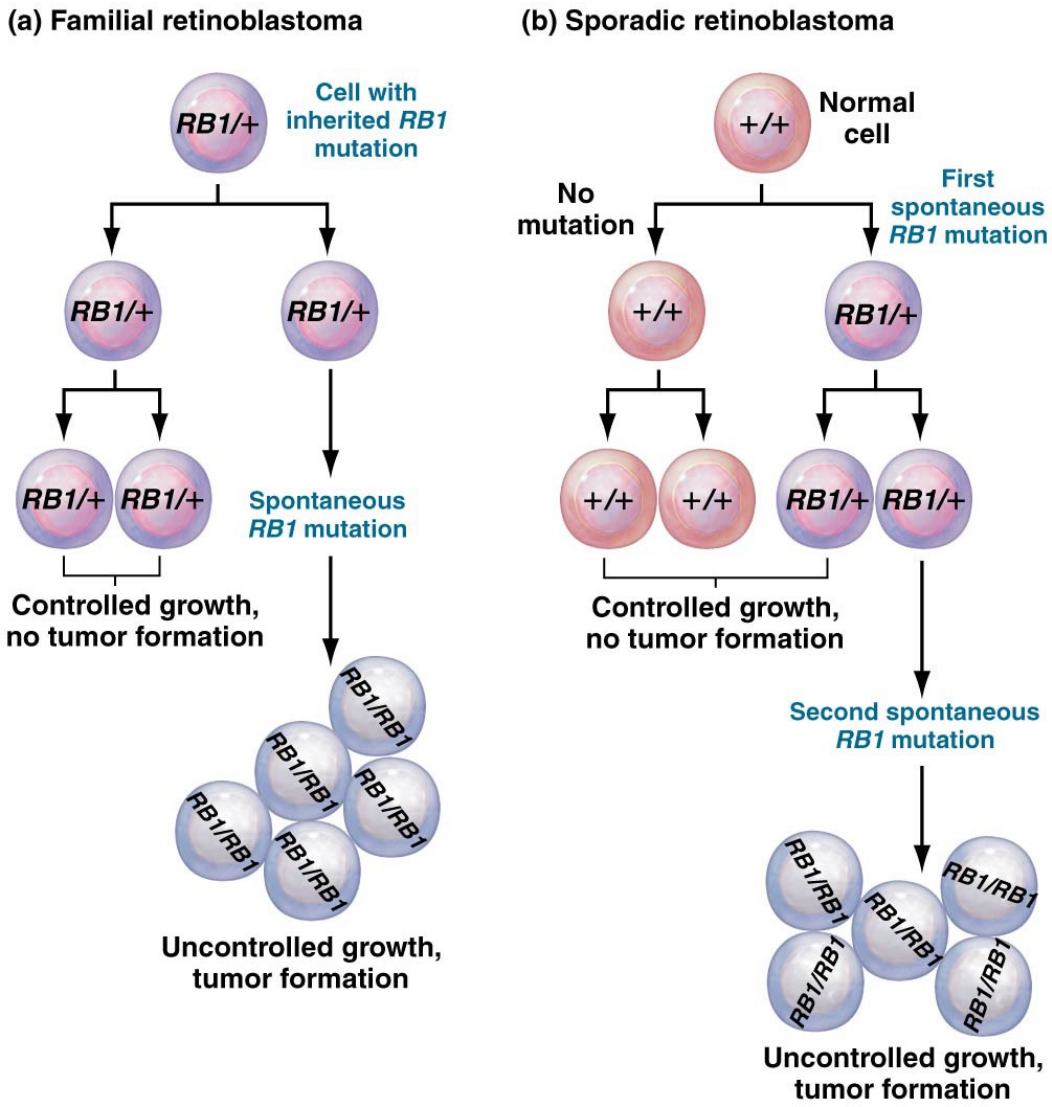
RB1 tumor-suppressor gene

- *RB1* encodes a protein that controls the G1/S checkpoint
- Loss or mutation of *RB1* leads to development of multiple cancers due to unregulated progression through cell cycle
- Originally identified via studies on **retinoblastoma**, an inherited disorder that causes tumor development in eyes of young children



Retinoblastoma

- **Familial** retinoblastoma
  - Mutated *RB1* allele is inherited ➔ 85% risk of retinoblastoma
- **Sporadic** retinoblastoma
  - Requires two independent mutational events of *RB1* within same cell



5. Familial forms of cancer

Hereditary cancers

- Rare germline **genetic factors** confer a markedly increased risk of developing cancer
- 2023 WHO Classification of Tumors includes 90 distinct genetic tumour syndromes
  - Growth factors & signaling pathways
  - Oxidative stress response & metabolism
  - Cell cycle & apoptosis pathways
  - DNA repair & genome instability
  - Telomere maintenance
  - Epigenetic drivers & chromatin remodelling
  - RNA & protein regulation

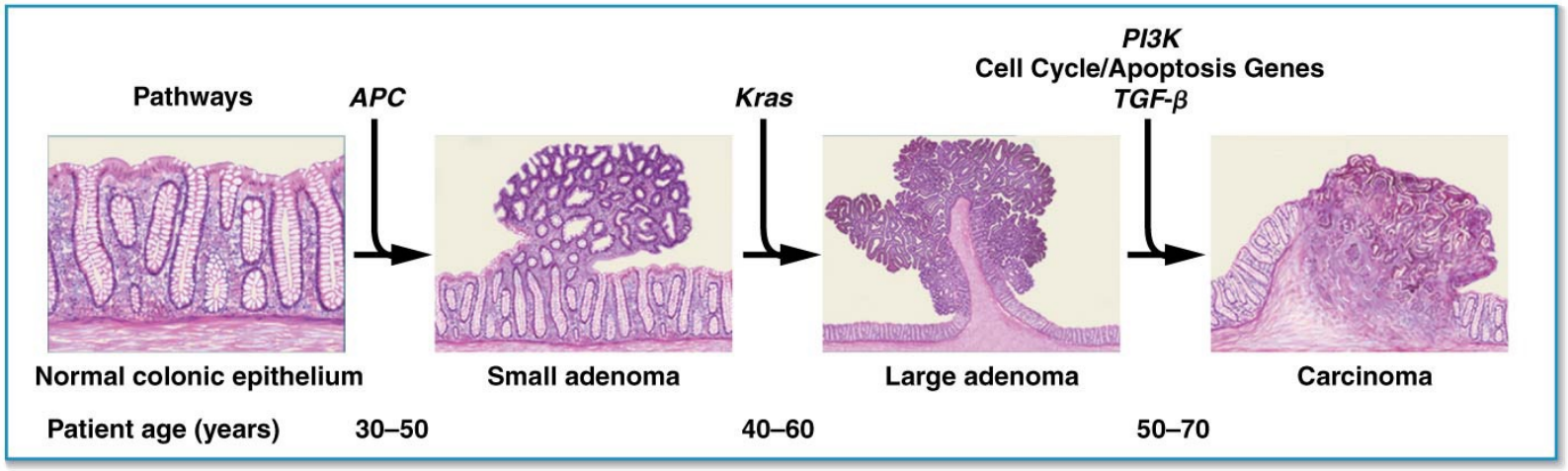
https://whobluebooks.iarc.fr/structures/genetic-tumour-syndromes/

TABLE 19.2 **Some Inherited Predispositions to Cancer**

Tumor Predisposition Syndrome	Chromosome	Gene Affected
Early-onset familial breast cancer	17q	*BRCA1*
Familial adenomatous polyposis	5q	*APC*
Familial melanoma	9p	*CDKN2*
Gorlin syndrome	9q	*PTCH1*
Hereditary nonpolyposis colon cancer	2p	*MSH2, 6*
Li-Fraumeni syndrome	17p	*p53*
Multiple endocrine neoplasia, type 1	11q	*MEN1*
Multiple endocrine neoplasia, type 2	10q	*RET*
Neurofibromatosis, type 1	17q	*NF1*
Neurofibromatosis, type 2	22q	*NF2*
Retinoblastoma	13q	*pRb*
Von Hippel–Lindau syndrome	3p	*VHL*
Wilms tumor	11p	*WT1*

Familial adenomatous polyposis

- Genetic predisposition to cancer (eg, colon cancer, medulloblastoma)
- Due to the presence of **germline heterozygous variants** in ***APC***
  - *APC* (adenomatous polyposis coli) is a tumor suppressor gene encoding a protein that controls the normal differentiation of intestinal cells
  - Several types of deleterious variants can be involved, including deletions, frameshift, and single nucleotide variants
- Formation of many polyps or adenomas in early life
- Other somatic *APC* mutations, followed by mutation in other genes (*ras, DCC, p53, etc…*) lead to the development of an adenocarcinoma

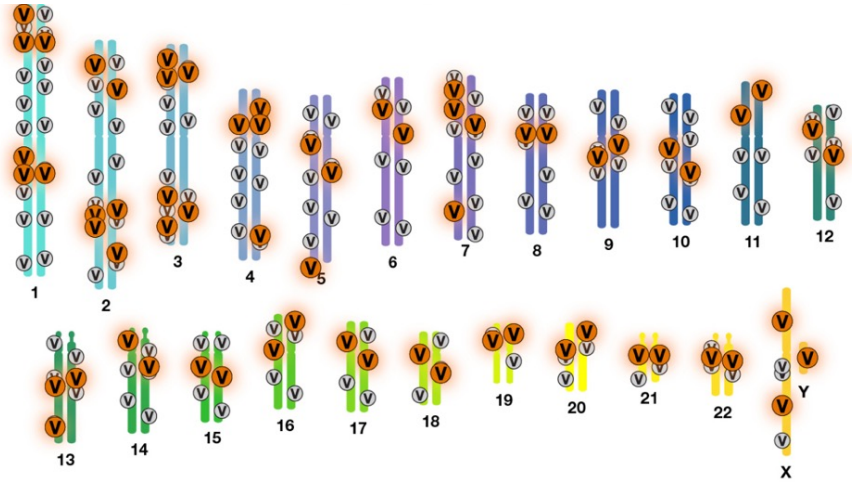


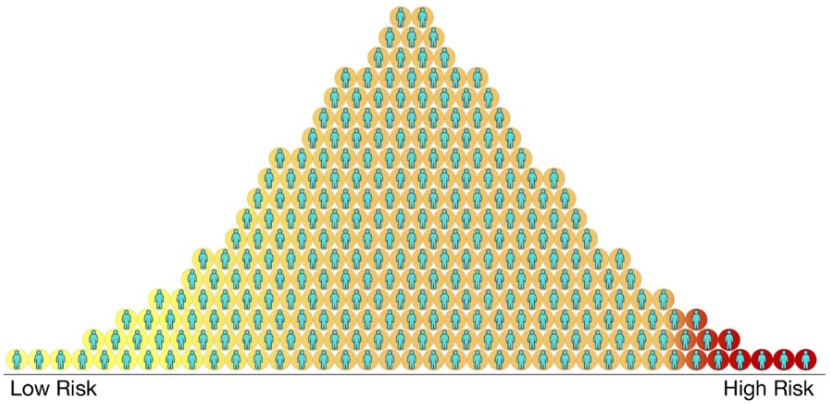
Breast and ovary cancer

- *BRCA1* and *BRCA2* encode proteins that play a role in HR repair
- 2-3/1000 women carry a pathogenic germline variant in *BRCA1/2*
  - Risk of cancer before age 70:
    - Breast: 40-85% vs. 10% in general population
    - Ovary: 10-63% vs. 1% in general population
  - Risk depends on gene:
    - *BRCA1* : breast 65%, ovary 45%
    - *BRCA2* : breast 45%, ovary 11%

Polygenic risk of cancer

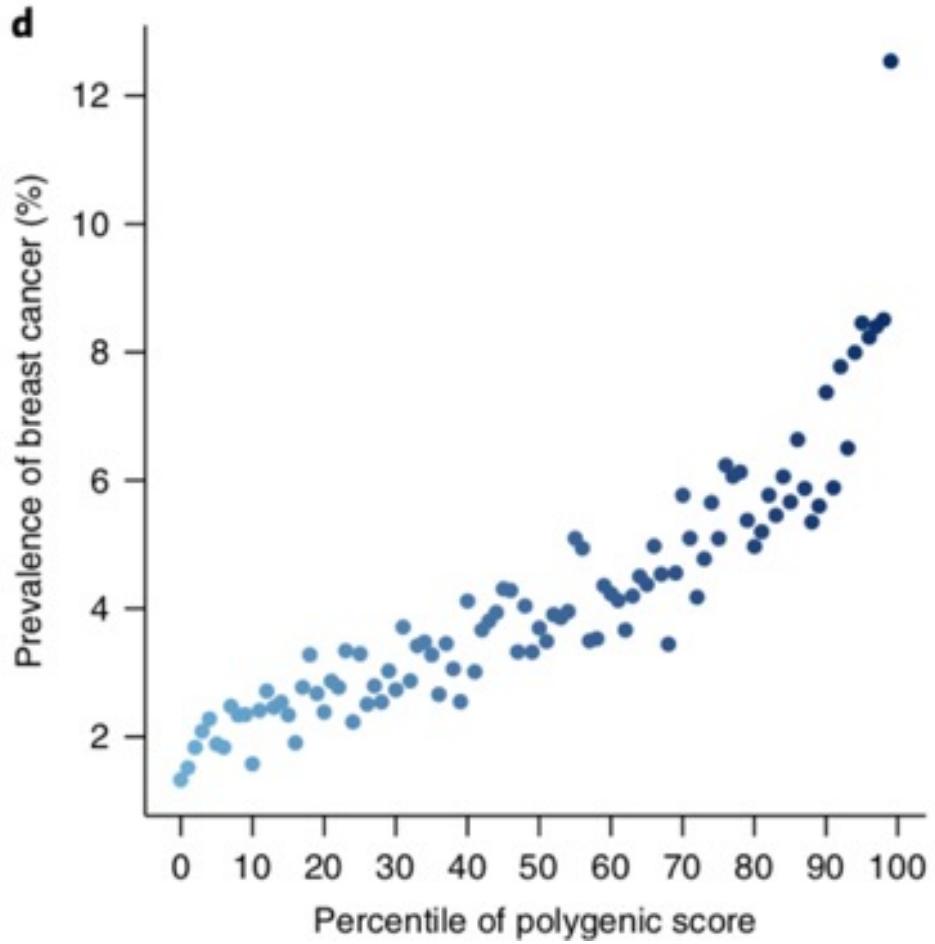
- Polygenic risk scores have recently been developed that allow the estimation of the individual risk of complex diseases, including cancer
- Combined effect of multiple **common genetic variants**





https://www.genome.gov/Health/Genomics-and-Medicine/Polygenic-risk-scores

Polygenic risk of cancer



nature genetics

Letter | Published: 13 August 2018

Genome-wide polygenic scores for common diseases identify individuals with risk equivalent to monogenic mutations

Amit V. Khera, Mark Chaffin, Krishna G. Aragam, Mary E. Haas, Carolina Roselli, Seung Hoan Choi, Pradeep Natarajan, Eric S. Lander, Steven A. Lubitz, Patrick T. Ellinor & Sekar Kathiresan

Nature Genetics **50**, 1219–1224 (2018) | Download Citation

6. Carcinogens

Carcinogens

- Any substance or event that damages DNA and causes mutations to occur in proto-oncogenes or tumor-suppressor genes
  - Chemicals, radiation, some viruses, chronic inflammation, …
- Delay between exposure to a carcinogen and appearance of cancer = reflects the slow accumulation of mutations
  - Radiation exposure in Hiroshima and Nagasaki caused leukemias 5 to 8 years later
  - X-ray treatment for tuberculosis in the 1930’ led to a wave of breast cancers 15 years later

Infectious agents

- Responsible for 15-20% of cancers
- Several causal mechanisms:
  - Integration into host DNA
    - Retroviruses
  - Chronic inflammation
    - Helicobacter pylori → gastric cancer
    - Hepatitis B and C viruses --> liver cancer
    - Schistosomiasis → bladder cancer
  - Modulation of cellular mechanisms
    - Inhibition of p53 and pRB by papillomavirus
  - Immune suppression
    - HIV

TABLE 19.3 **Human Viruses Associated with Cancers**

Virus		Associated Cancers
DNA Viruses		
Epstein-Barr virus	EBV	Burkitt lymphoma, nasopharyngeal carcinoma, Hodgkin lymphoma
Hepatitis B virus	HBV	Hepatocellular carcinoma
Hepatitis C virus	HCV	Hepatocellular carcinoma, non-Hodgkin lymphoma
Human papilloma viruses 16, 18	HPV16, 18	Cervical cancer, ano-genital cancers, oral cancers
Kaposi sarcoma-associated herpesvirus	KSHV	Kaposi sarcoma, primary effusion lymphoma
Retroviruses		
Human T-cell lymphotropic virus type 1	HTLV-1	Adult T-cell leukemia and lymphoma
Human immunodeficiency virus type-1	HIV-1	Immune suppression, leading to cancers caused by other viruses (KSHV, EBV, HPV)

Retroviruses and cancer

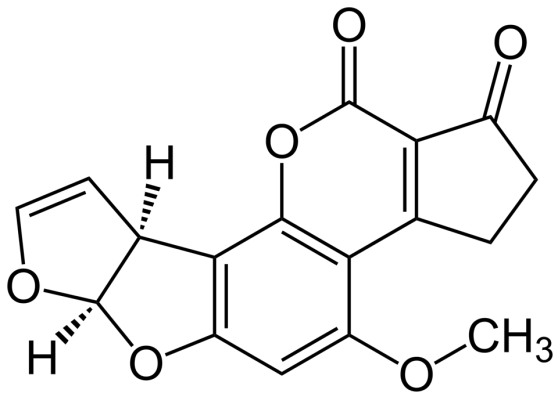
- Retroviruses can cause cancer in two different ways:
  1. **Integration of proviral DNA close to a host proto-oncogene**, leading to its activation
  2. **"Transforming" retroviruses**
     - carry an oncogene in their own genomes
     - Transform the infected cell into a cancerous cell
     - Rous sarcoma virus (RSV)
       - 1st identified retrovirus in 1910
       - Found in transmissible sarcoma of the chicken
     - No known transforming retrovirus in humans

Environmental carcinogens

- **Tobacco smoke**
  - Most significant environmental carcinogen
  - >60 mutagenic chemicals
  - Smokers have a 20-fold increased risk of developing lung cancer
- **Alcohol**
  - may cause liver steatosis (fat accumulation) → chronic liver inflammation → hepatocarcinoma
- **X-rays**
  - Risk is dose-dependent and highest in rapidly dividing tissues and in children
  - Risk of secondary tumours following treatment of primary tumours (eg, radiation-induced gliomas; breast cancer;
- **UV light**
  - UV-B → CPD → unrepaired by NER → C>T and CC>TT mutations in TP53 and BRAF
  - Basal cell carcinoma, squamous cell carcinoma, melanoma

Environmental carcinogens

- **Aflatoxin B_1**
  - Toxin produced by a mold (*Aspergillus*) on stored grains, nuts, and maize in warm/humid conditions
  - Aflatoxin B_1 → liver cells convert it to Aflatoxin B_1-8,9-epoxide (reactive metabolite) → **DNA adduct** on guanine bases → G>T mutations
  - Liver cancer



- **Nitrosamines**
  - organic compounds of the chemical structure $R_2N{-}N{=}O$, where R is usually an alkyl group
  - Converted in the liver by metabolic activation into **alkylating agents** that induce DNA mutations
  - Used as meat preservative
  - Gastic cancer and liver cancer