Towards High-quality Clinical Trials and
Implementation of Genomic Medicine

ATLAS Training Program

Course : Cancer Genome-based Medicine Course
Lecture Title : Cancer Biology and Molecular Biology
Speaker : Rieko Ohki

ASIAN CLINICAL TRIALS NETWORK FOR CANCERS PROJECT

ICRweb: https://www.icrweb.jp/icr_index.php?lang=en

RIEKO OHKI

Laboratory of Fundamental Oncology, National Cancer Center Research Institute

■ Education

1994 M.S., Faculty of Sciences, University of Tokyo (Japan)
1997 Ph.D., Dept. of Biochemistry and Biophysics, Faculty of Sciences, University of Tokyo (Japan)

■ Work Experience

2002–2011	Research Associate, National Cancer Center Research Institute
2011–2017	Group leader, National Cancer Center Research Institute
2017–present	Laboratory head, Laboratory of Fundamental Oncology, National Cancer Center Research Institute

■ Specialty and Research Field of Interest

Molecular biology of cancer
Identification and analysis of genes involved in tumorigenesis

Characteristics of Cancer Cells : Differences from Non-cancer Cells

Cancer

The cell cycle surveillance mechanism (checkpoints) fails, and cell division is uncontrolled.

Cells proliferate independently of external proliferation signals.

Activation of cell proliferation signals

Failure of genome stabilization mechanism.

Failure of apoptosis control mechanism.

Unlimited number of cell divisions; cells are immortalized.

Activation of telomerase
(Prevents telomere shortening)

Cells have excess angiogenesis potential.

Cells invade and metastasize beyond the original tissue.

Reduced function of cell adhesion factors

Non-cancer

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Genome stabilization mechanism is functional.

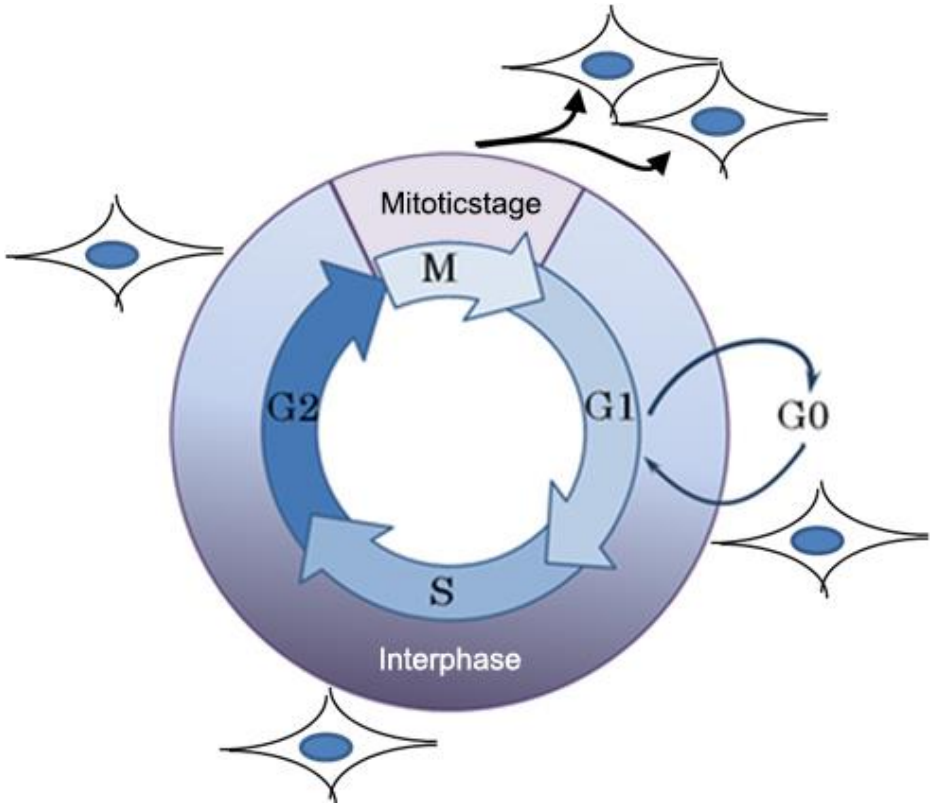
Apoptosis control mechanism is functional.

Number of cell divisions is limited. Cell division has a finite number, and cells are unable to undergo more than the inherent number of divisions, resulting in cellular senescence.

Angiogenesis is controlled.

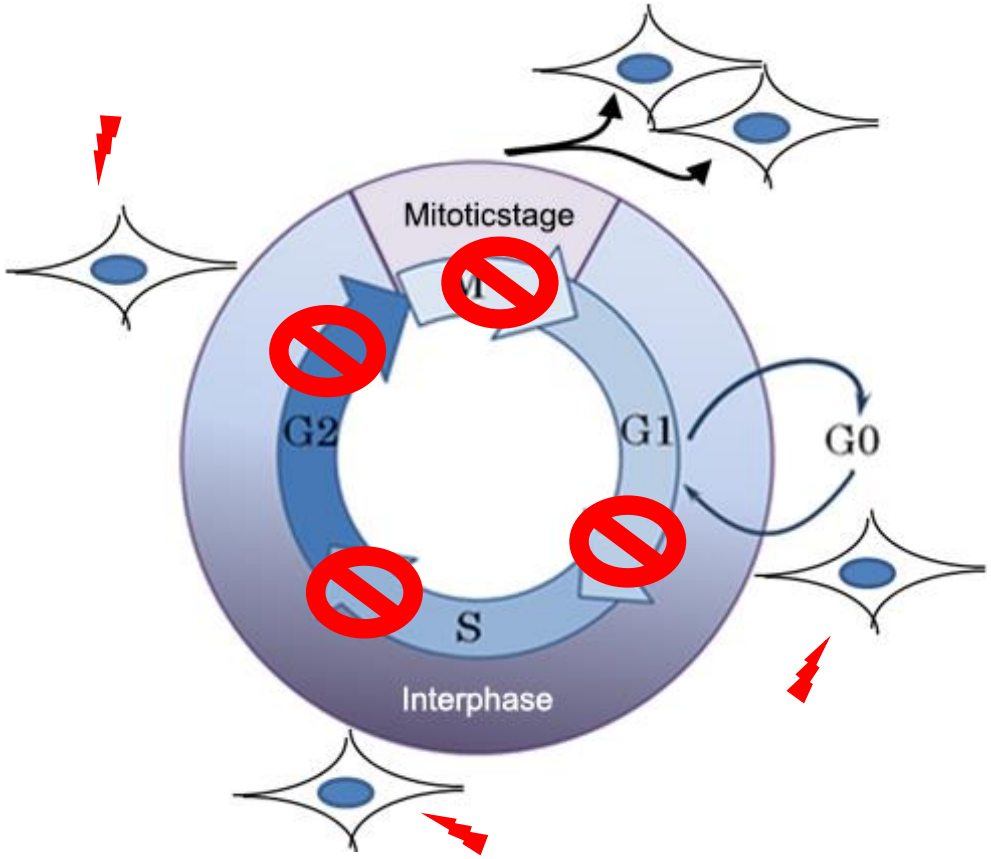
Cells remain in the original tissue and do not become established in other locations.

Failure of Cell Cycle Control



Factors that positively control the cell cycle: oncogenes (cyclin D1, CDK, etc.)
Factors that negatively control the cell cycle: tumor-suppressor genes (CDK inhibitor, etc.)

Cell Cycle And Checkpoint Abnormalities



Checkpoints

Interference

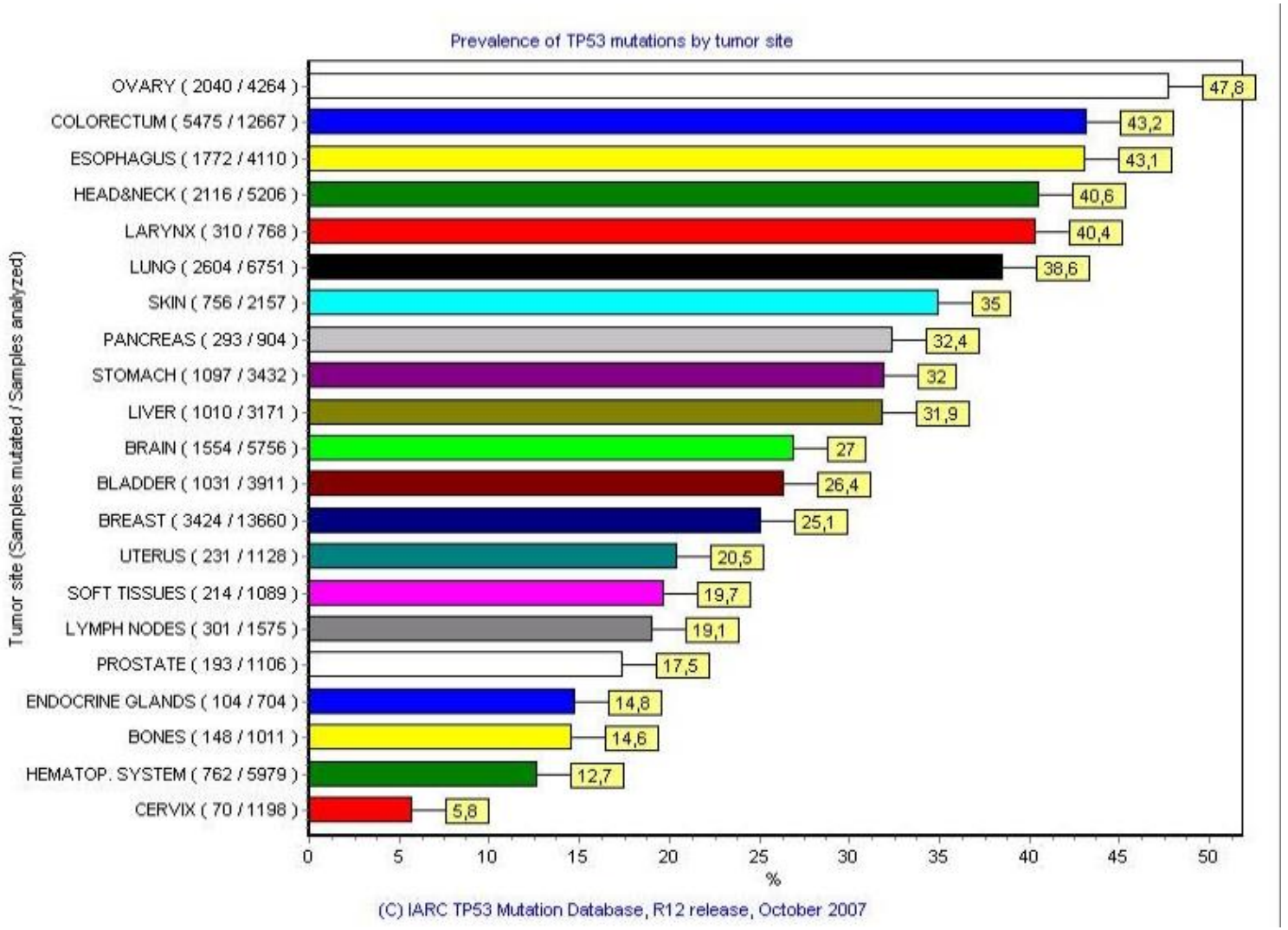
Cell cycle arrest is induced in response to cell damage.

However, in cancer cells, this checkpoint function fails!

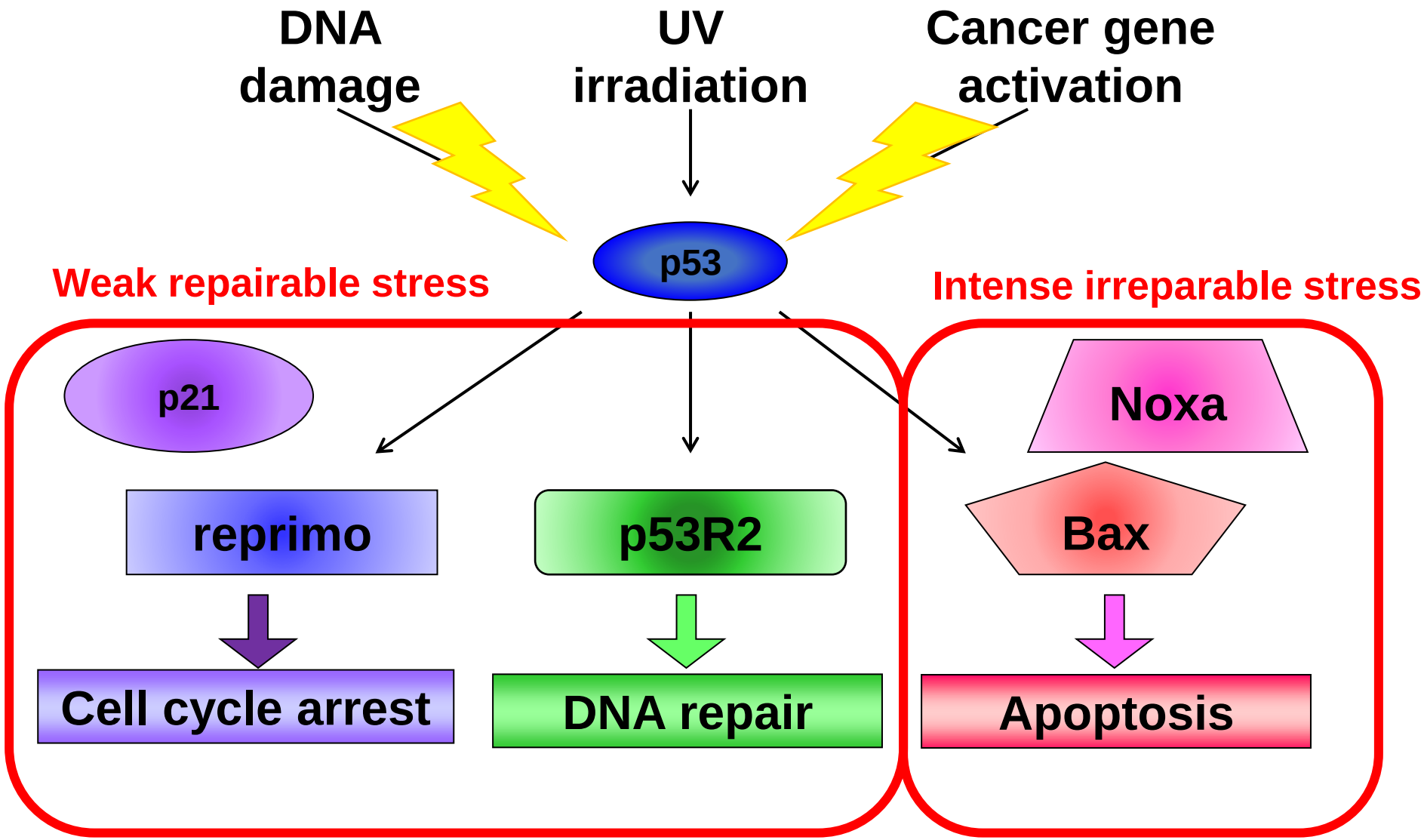
(because of the inactivation of the tumor-suppressor gene p53, which controls the checkpoint)

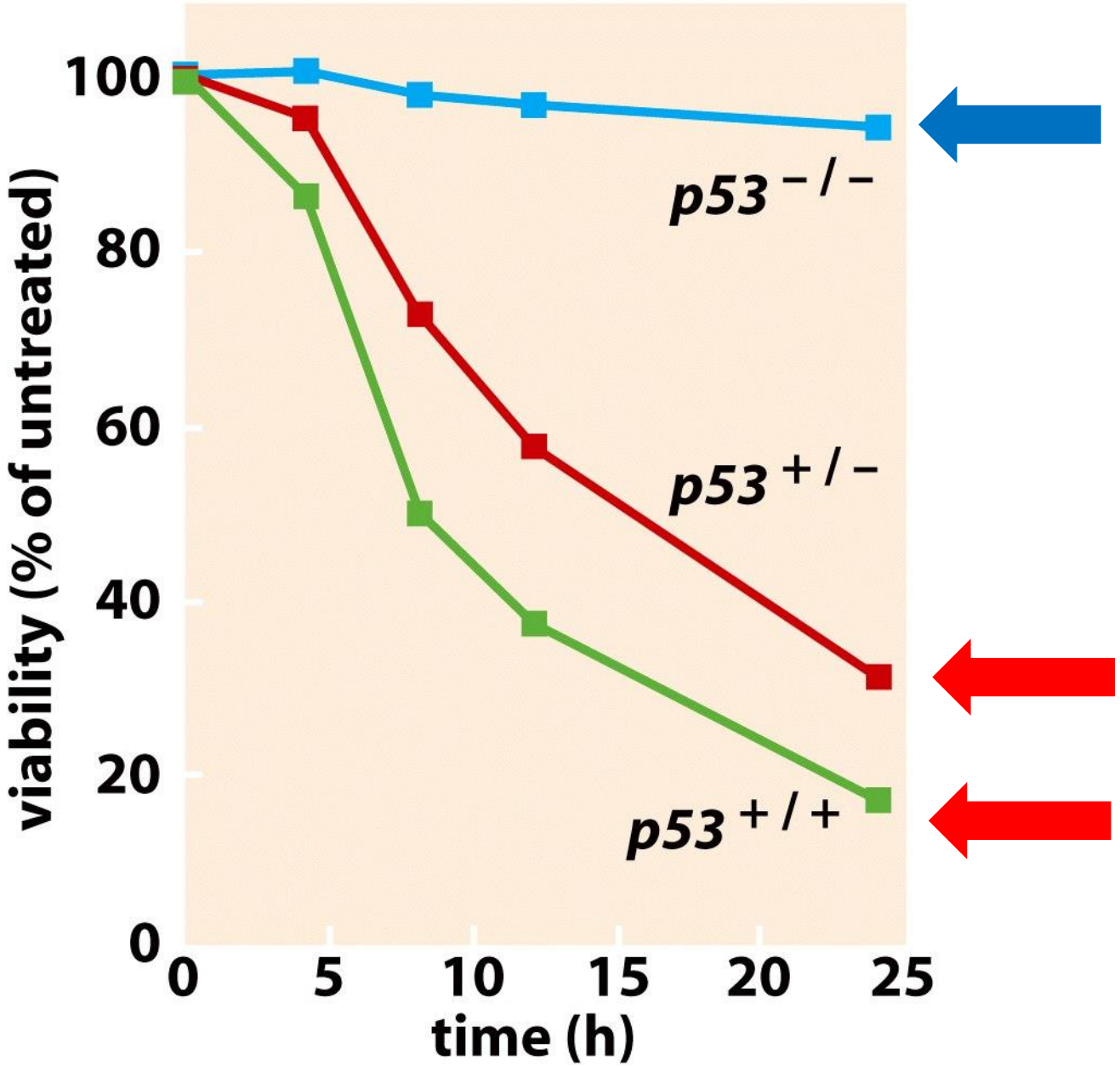
→Leading to the proliferation of cells with genetic abnormalities

P53 is the most frequently mutated gene in human cancer



Approximately one in two cancers have mutated p53





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Cancer	Non-cancer
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Activation of cell proliferation signals	
Failure of genome stabilization mechanism.	Genome stabilization mechanism is functional.
Failure of apoptosis control mechanism.	Apoptosis control mechanism is functional.
Unlimited number of cell divisions; cells are immortalized.	Number of cell divisions is limited. Cell division has a finite number, and cells are unable to undergo more than the inherent number of divisions, resulting in cellular senescence.
Activation of telomerase (Prevents telomere shortening)	
Cells have excess angiogenesis potential.	Angiogenesis is controlled.
Cells invade and metastasize beyond the original tissue.	Cells remain in the original tissue and do not become established in other locations.
Reduced function of cell adhesion factors	

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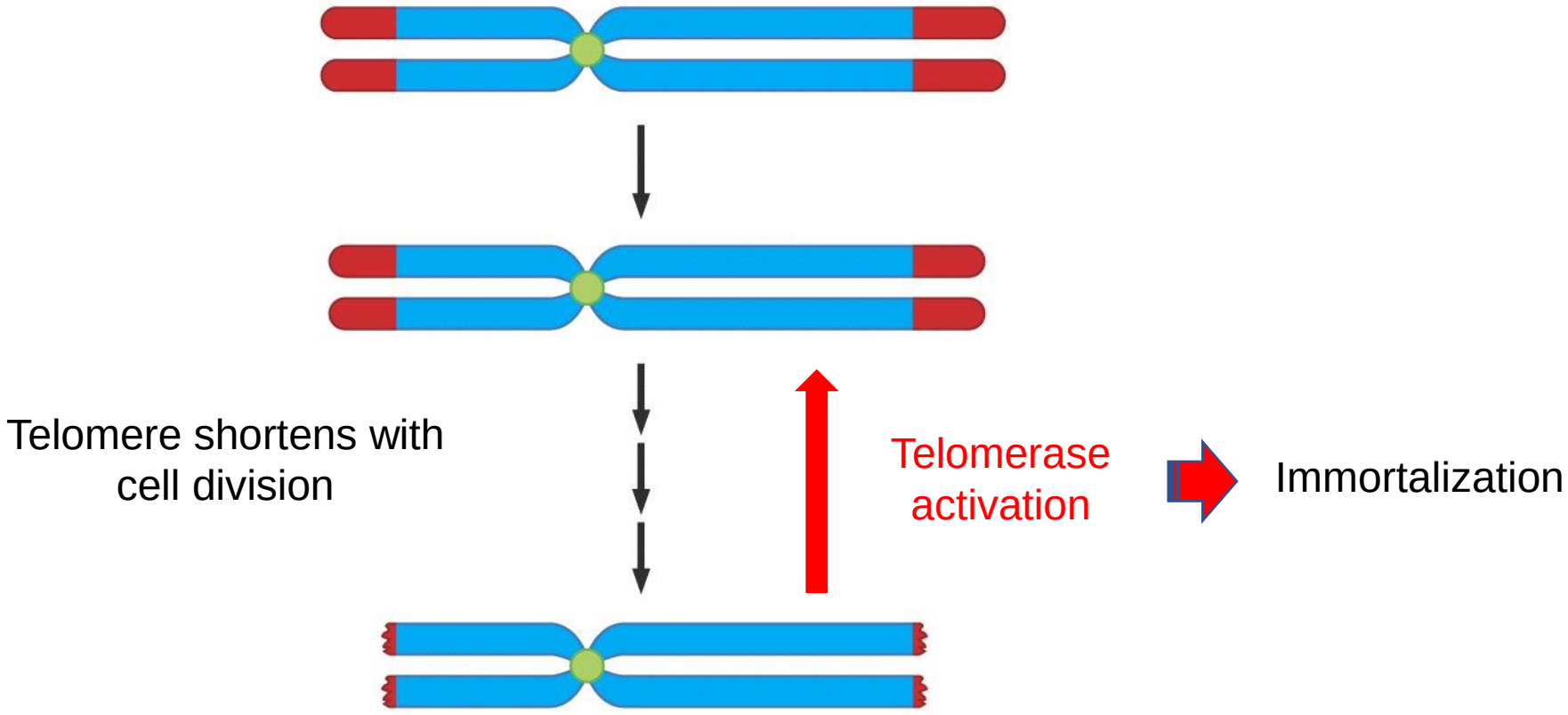
In Cancer Cells, Telomere Shortening is Prevented by Activation of Telomerase



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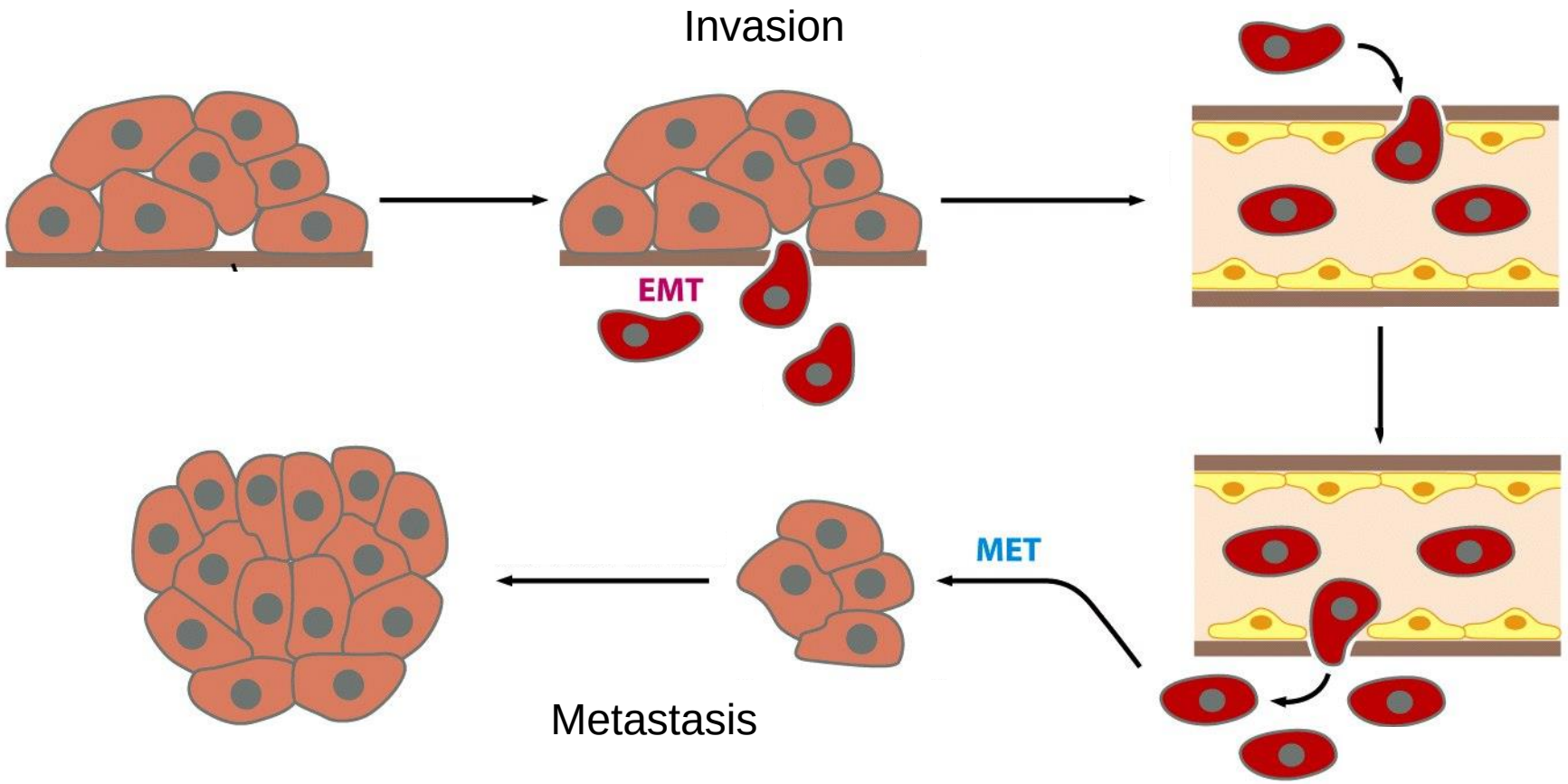
Invasion and metastasis occur in cancer cells because of reduced adhesion factor function and acquisition of anchorage-independent growth



From The Biology of Cancer

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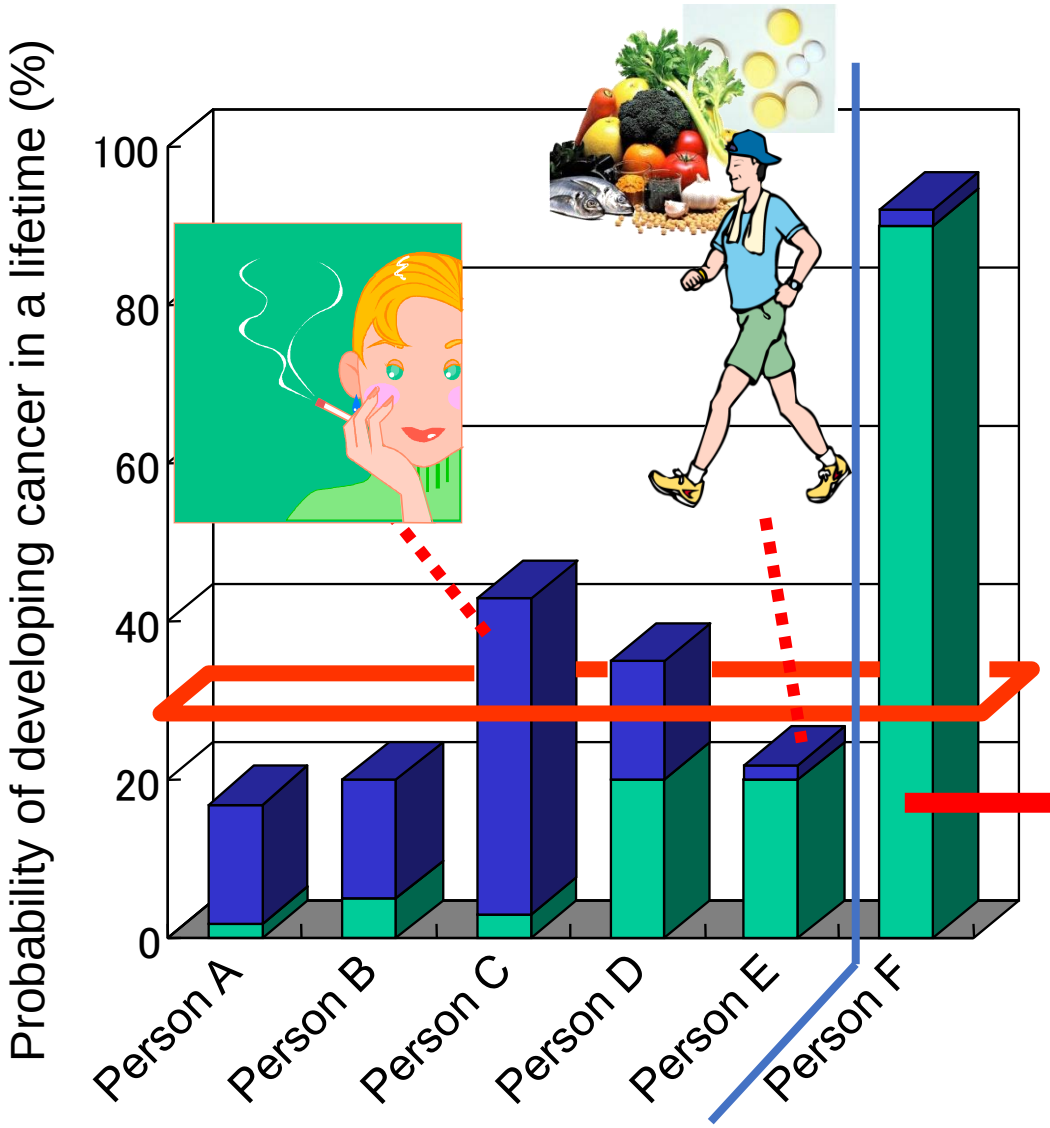
Cancer Risk: External Factors and Internal Factors



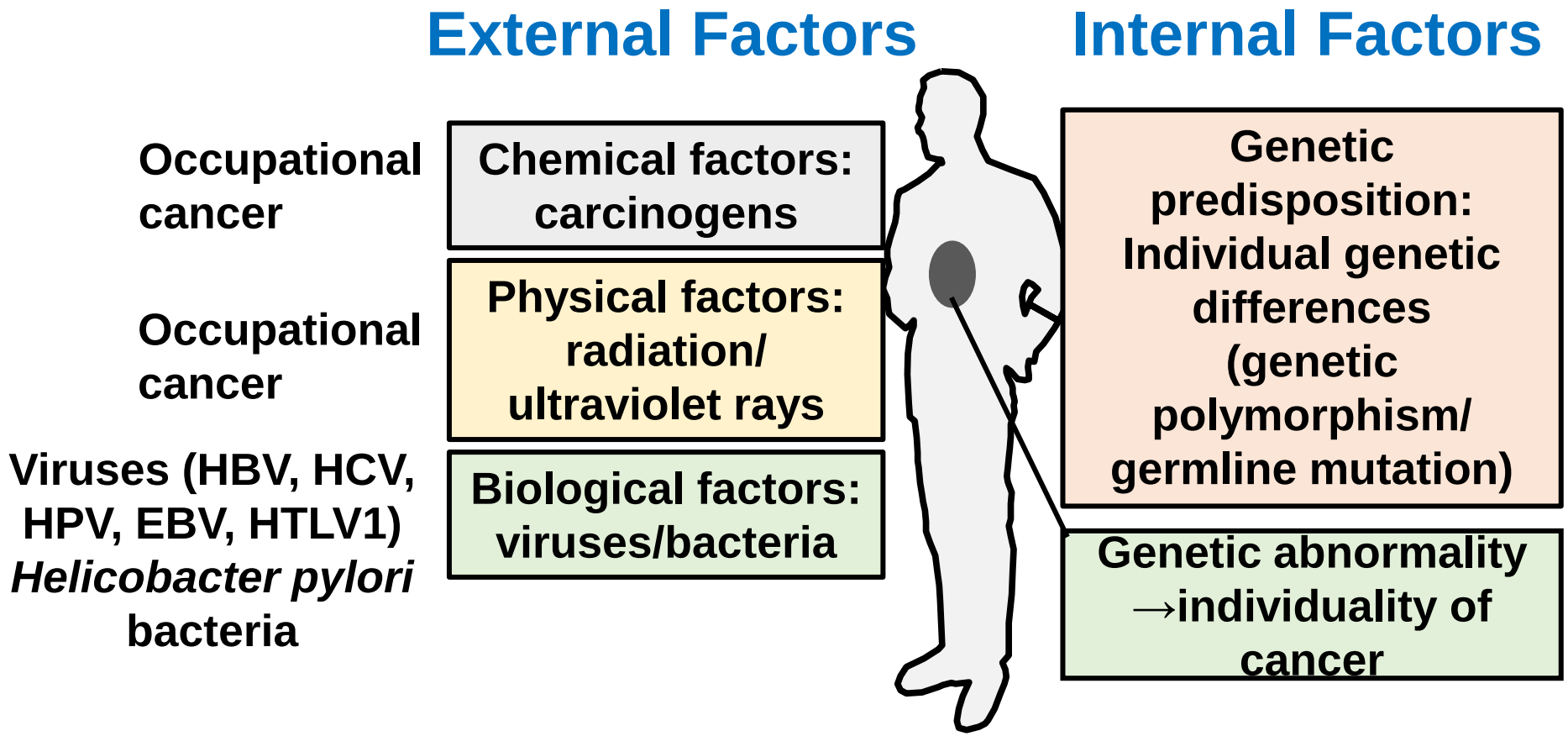
Internal factors

External factors

Genetic predisposition:
Individual genetic differences
(germline mutation)

Familial (hereditary) tumors

Cancer Risk Factors

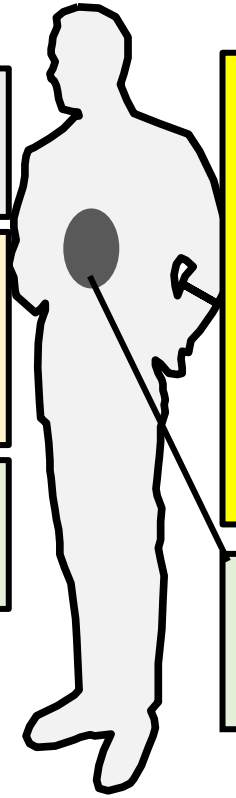


Cancer risk factors include external factors that affect cells from the outside world and internal factors present in the body, and the combination of these factors leads to cancer.

Cancer Risk Factors

External Factors

Internal Factors



Occupational cancer

Chemical factors: carcinogens

Occupational cancer

Physical factors: radiation/ ultraviolet rays

Viruses (HBV, HCV, HPV, EBV, HTLV1)
Helicobacter pylori bacteria

Biological factors: viruses/bacteria

Genetic predisposition: Individual genetic differences (genetic polymorphism/ germline mutation)

Genetic abnormality →individuality of cancer

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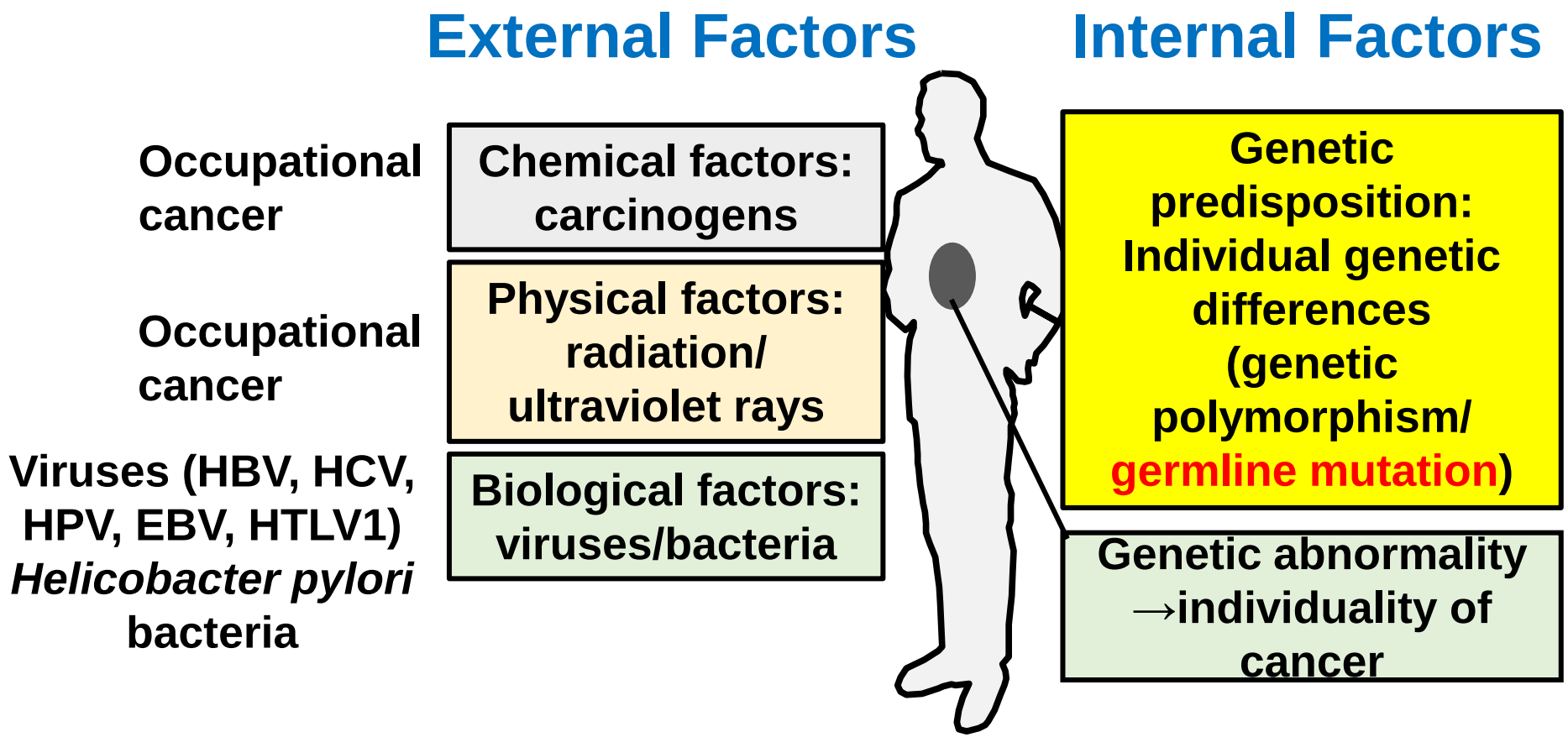
Cancer Risk Factors



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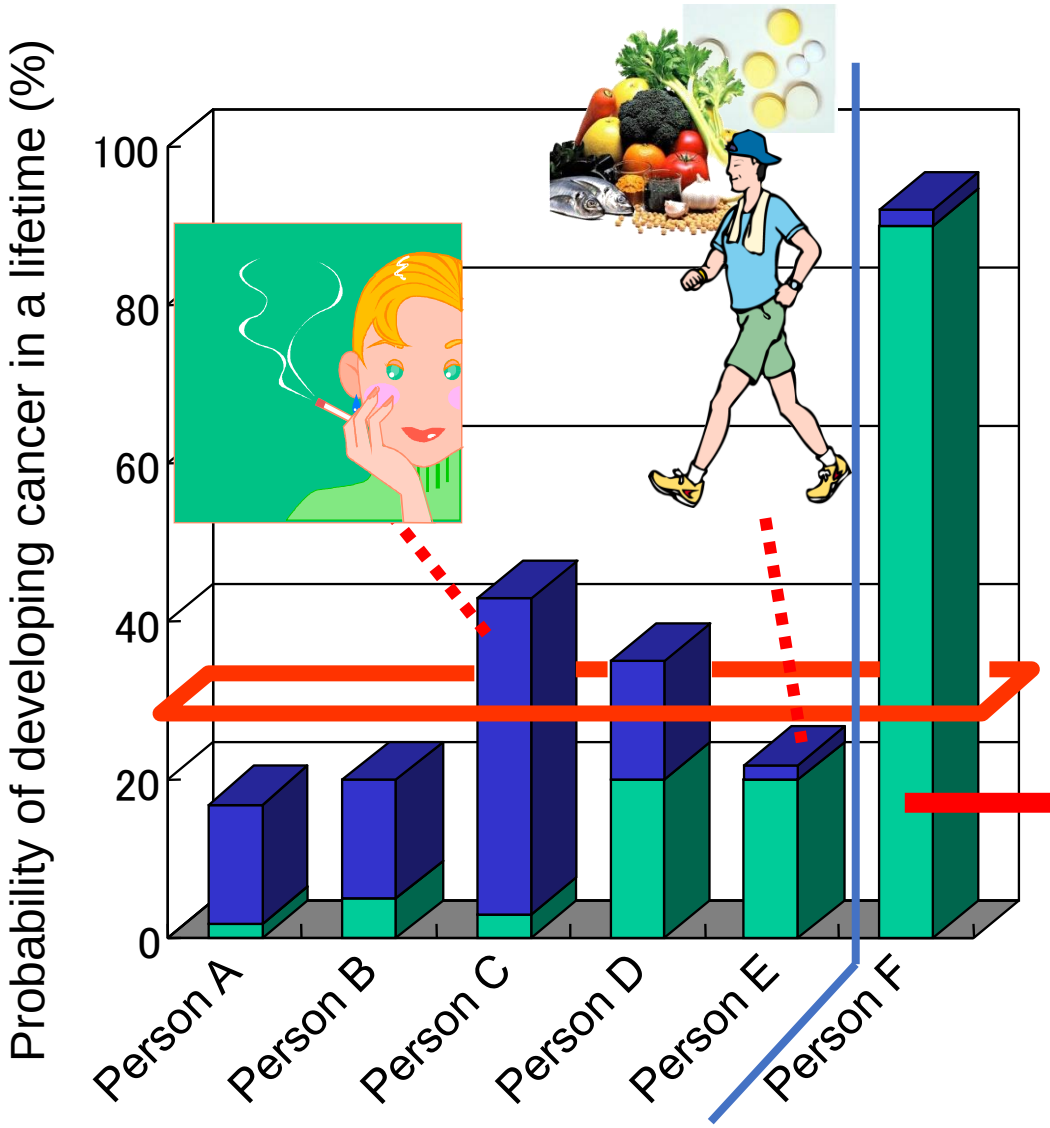

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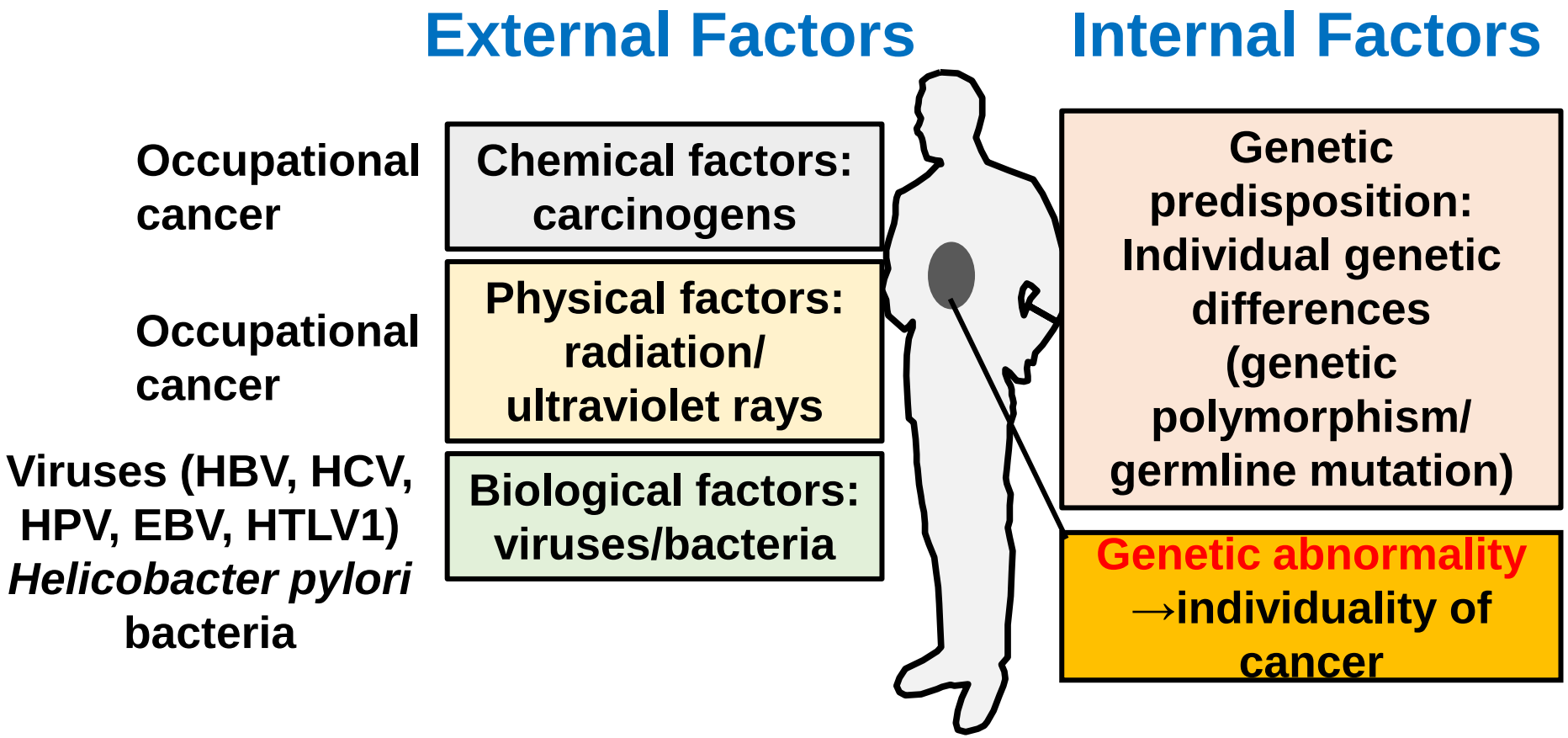
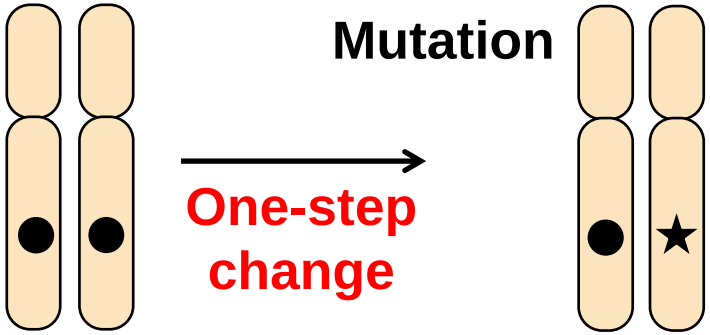


Genetic abnormalities either activate oncogenes or inactivate tumor-suppressor genes

Activation of Oncogenes

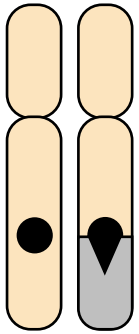


One-step change

Typical example

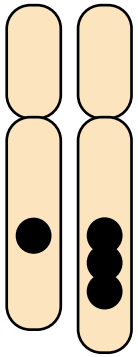
Mutation

EGFR
(lung adenocarcinoma)
RAS (various cancers)



Translocation

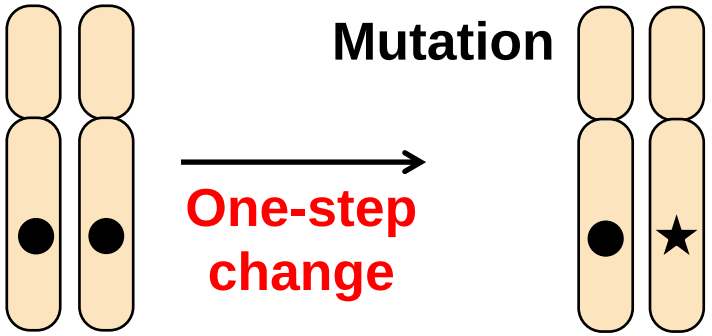
ABL
(leukemia)
ALK
(lung adenocarcinoma)



Amplification

ERBB2
(breast cancer, stomach cancer)
N-MYC
(neuroblastoma)

Activation of Oncogenes



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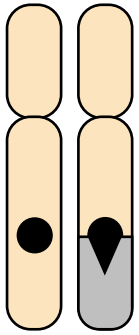

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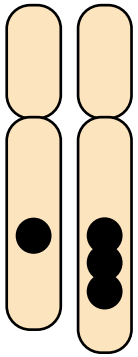

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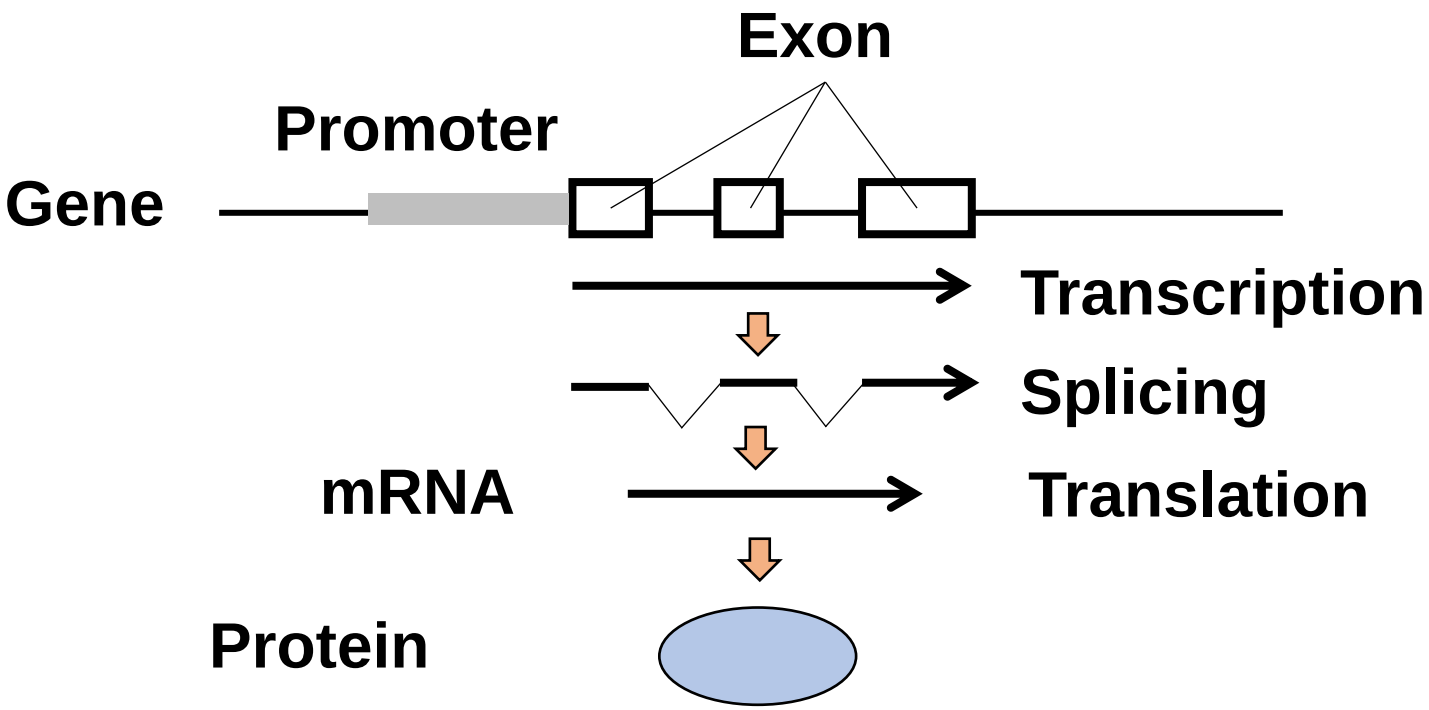
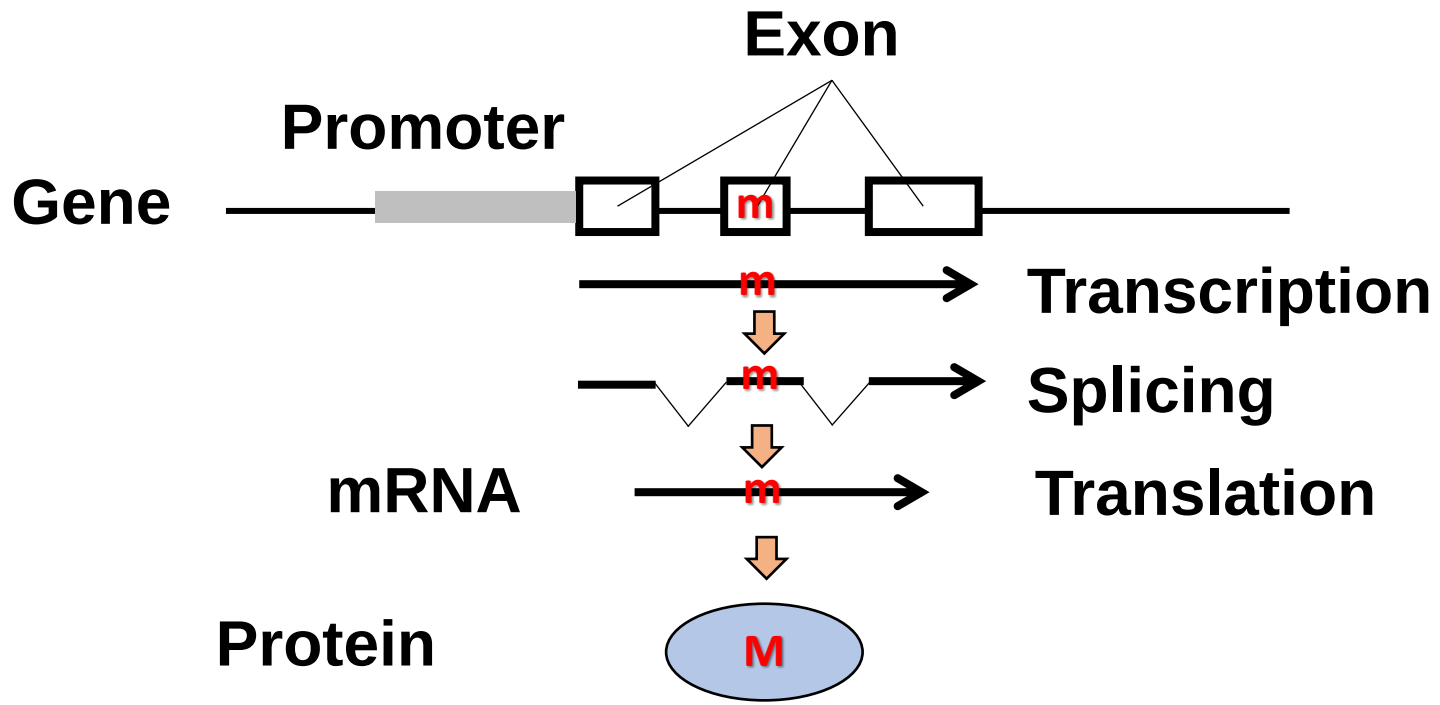
Gene Structure and Transcription/Translation



There are approximately 30,000 human genes.
Genes specifically expressed in tissues and cells are named as housekeeping genes.
In protein non-coding RNAs such as microRNA, the RNA itself has functions.

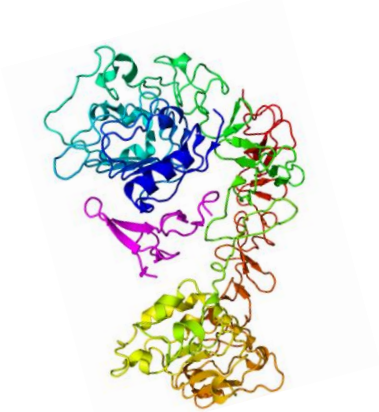
Gene Structure and Transcription/Translation



Activation of Oncogenes: 1. Mutation

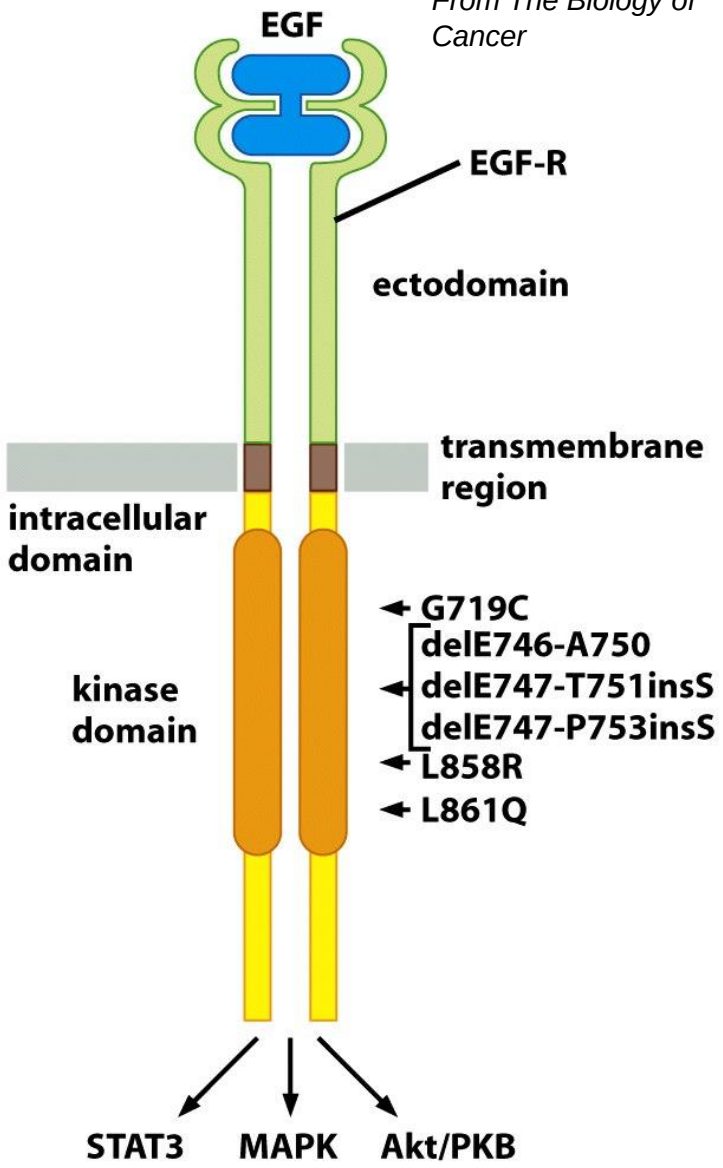
L858R mutation：
Transmits a strong proliferation signal independently of the ligand EGF

Gefitinib　　Erlotinib



Mutated EGFR protein
(continues to release signals instructing cells to proliferate)

From The Biology of Cancer

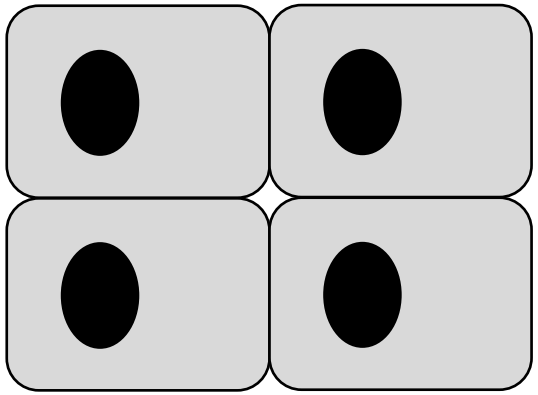


Individualized treatment of lung adenocarcinoma:

EGFR (Epidermal Growth Factor Receptor) Gene Mutation

Toxicity of Oncogenes and Molecular Targeted Therapy

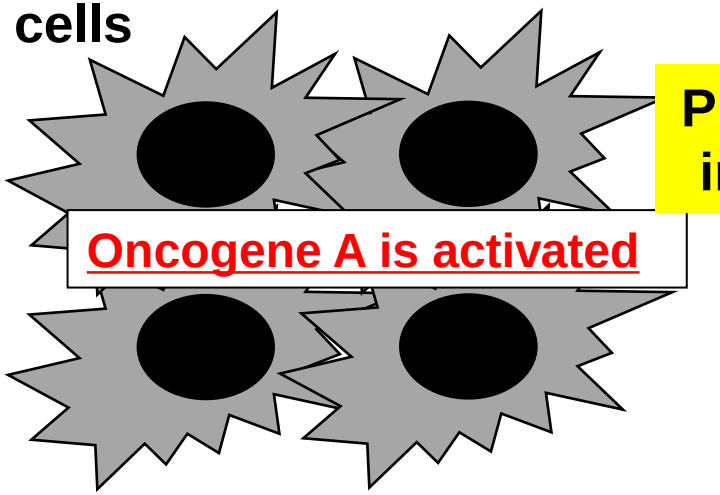
Normal cells



Protein A inhibitor

Can survive without protein A function → Few side effects

Cancer cells



Protein A inhibitor

Cannot survive without protein A function (Addicted state) → Inhibits growth/ cell death

Activation of Oncogenes: 1. Mutation

From The Biology of Cancer

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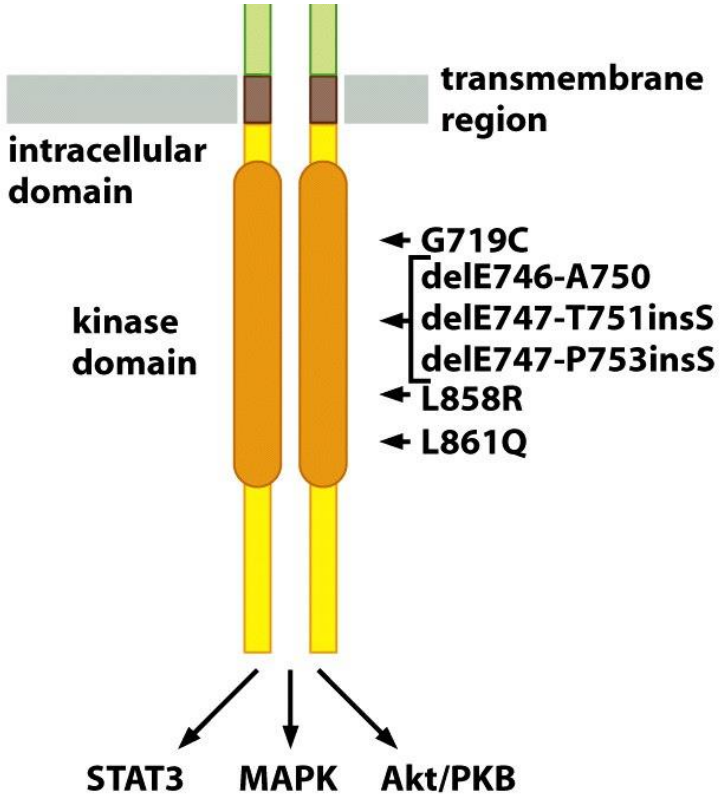

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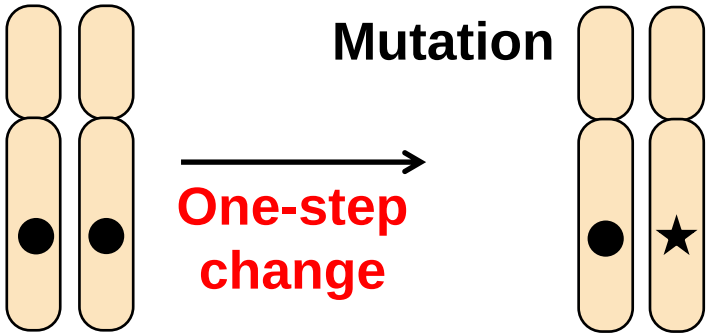

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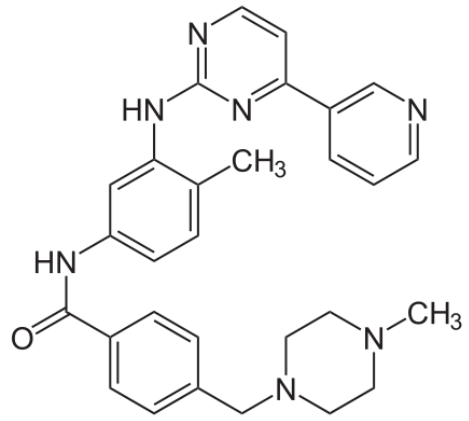
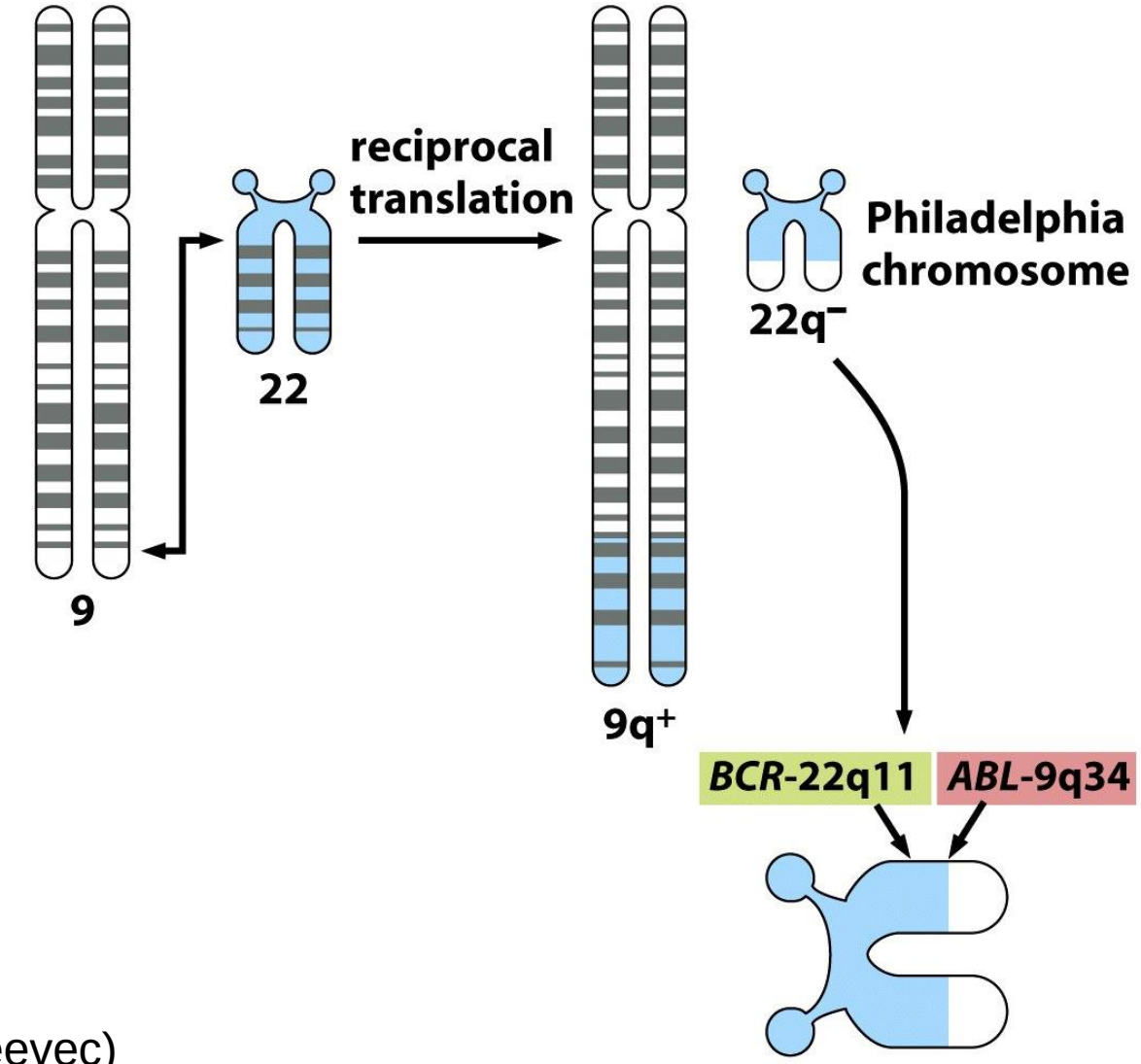
Activation of Oncogenes: 2. Translocation



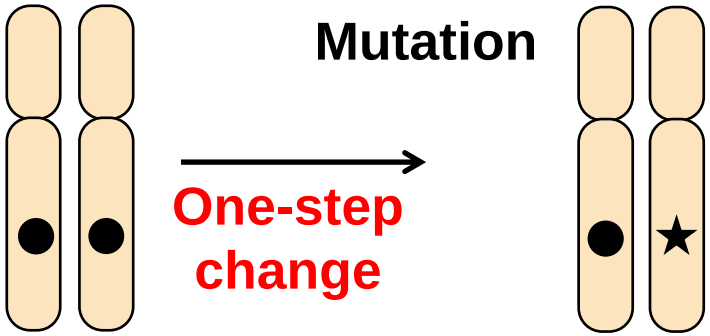
Imatinib (Product name: Gleevec)



From The Biology of Cancer

Individualized treatment for leukemia: BCR-ABL Translocation (Chronic Myelogenous Leukemia)

Activation Of Oncogenes



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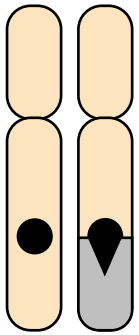

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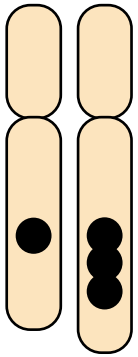

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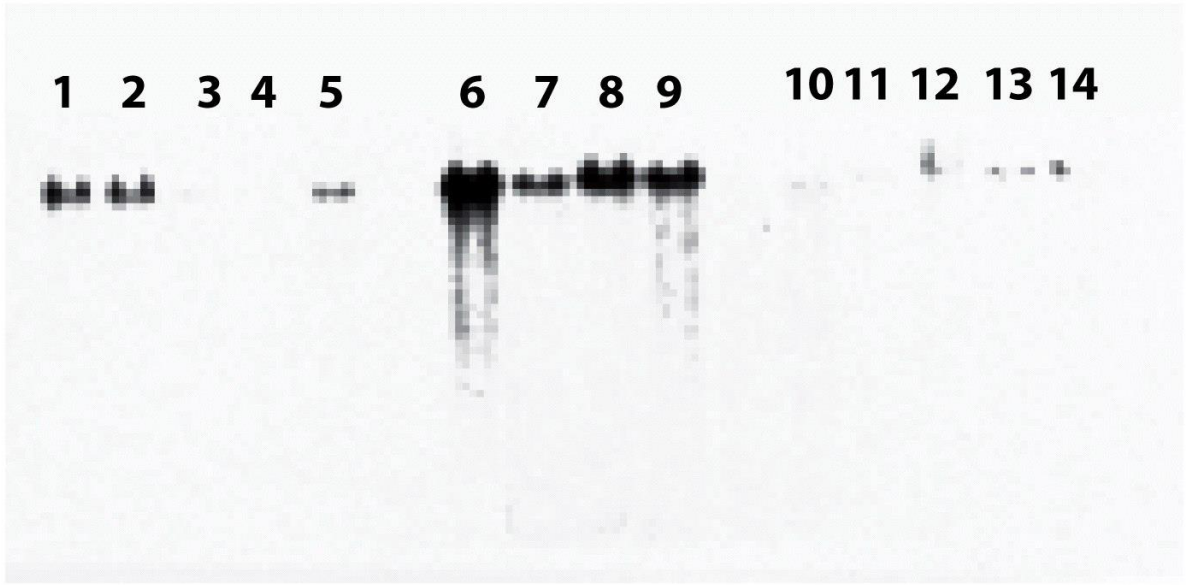
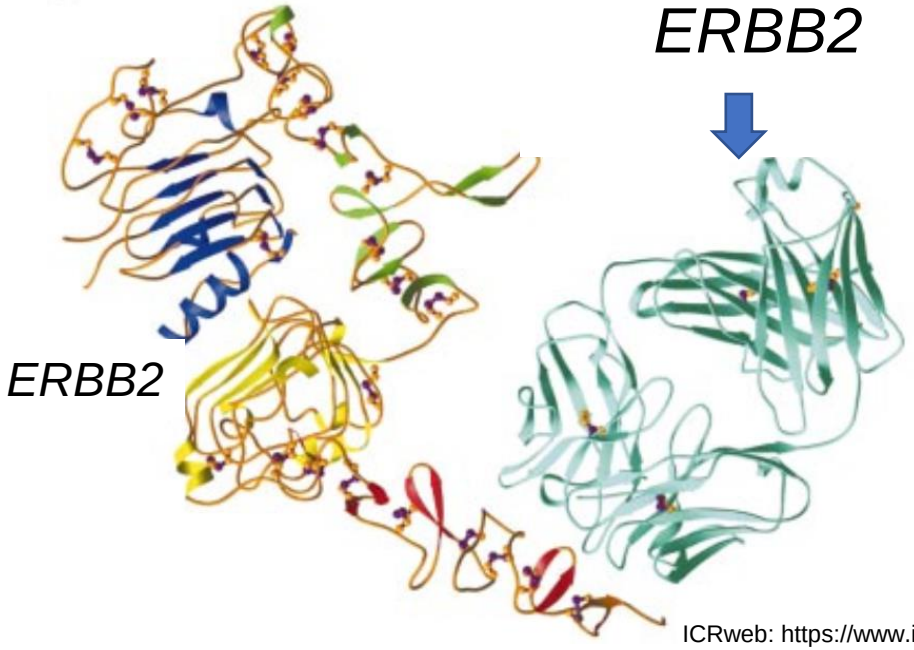
Activation of Oncogenes: 3. Amplification



From The Biology of Cancer

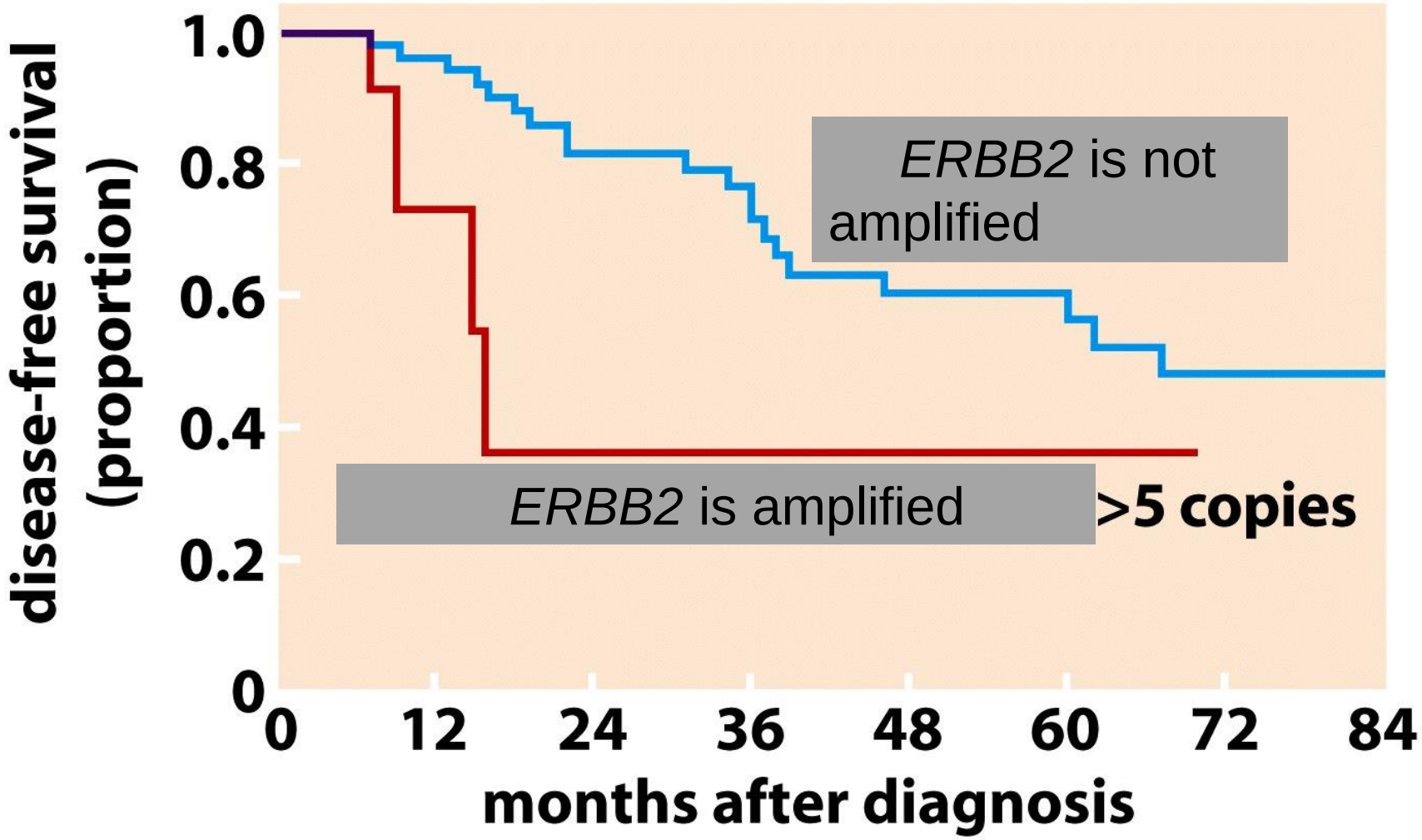
ERBB2 is amplified in breast cancer

Trastuzumab is an antibody for *ERBB2*



Individualized treatment for breast cancer: *ERBB2* Amplification

Individualized treatment for breast cancer: *ERBB2 A*mplification



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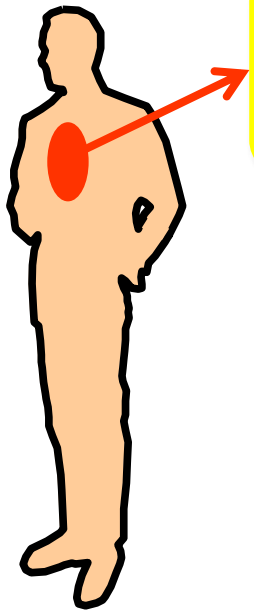
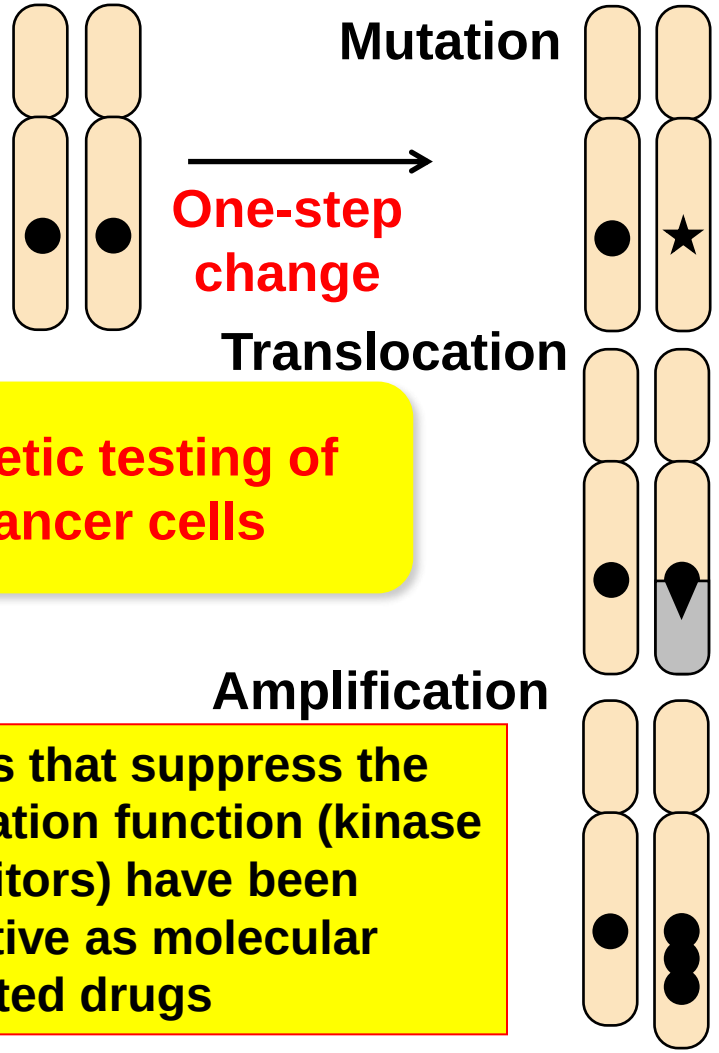


Genetic testing of cancer cells

Drugs that suppress the activation function (kinase inhibitors) have been effective as molecular targeted drugs



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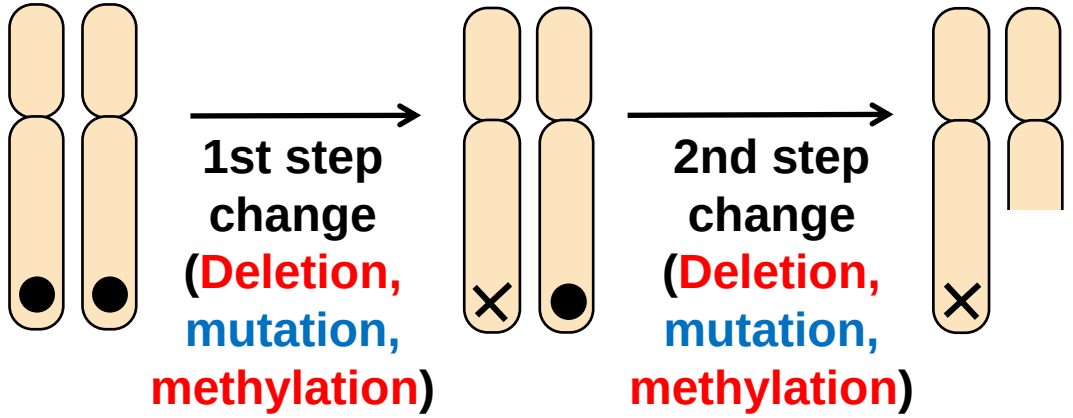
It is important to provide individualized treatment based on the genetic testing of the cancer tissue.

Inactivation of Tumor-suppressor Gene

Occurs through single allele loss (loss of heterozygosity: LOH), mutation, and methylation

Non-hereditary tumor

Normal cancer requires a two-step change



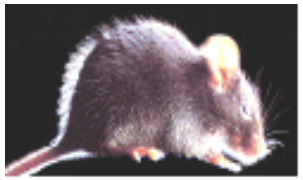
Inactivation of the tumor-suppressor gene requires abnormalities in both homologous genes.

p53-deficient mice frequently develop cancer

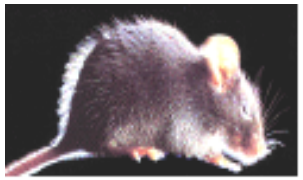
Only 1% of normal mice with p53 developed cancer after 18 months.

75% of mice completely deficient in p53 developed cancer by 6 months.

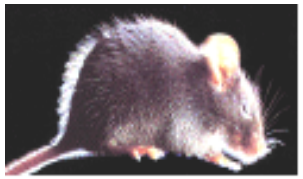
% mice with tumor



$p53^{+/+}$ → **1% at 18 months**



$p53^{+/-}$ → **2% at 9 months**



$p53^{-/-}$ → **75% at 6 months**

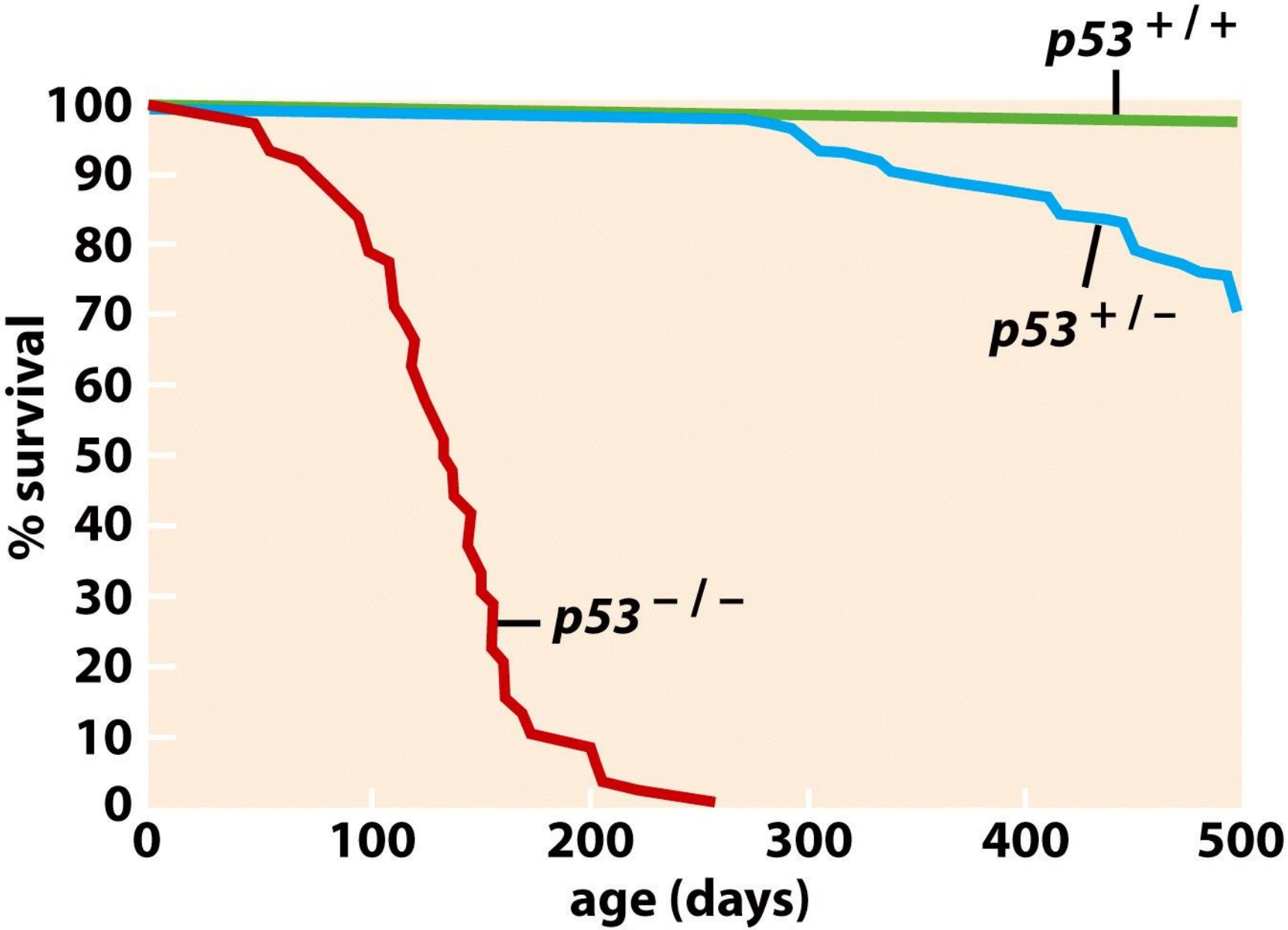
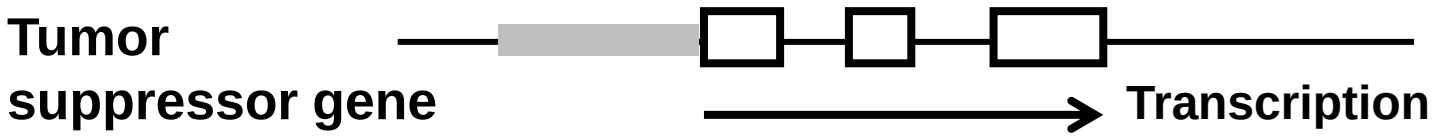


Figure 9.5 *The Biology of Cancer* (© Garland Science 2007)

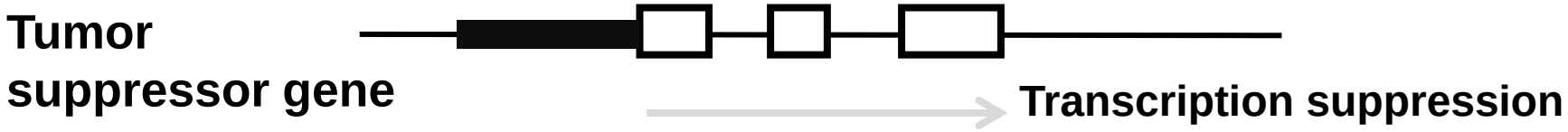
Inactivation of Tumor-suppressor Gene by Methylation of the Gene Promoter Region

Normal cells

Non-methylated promoter



Cancer cells **Methylated promoter**

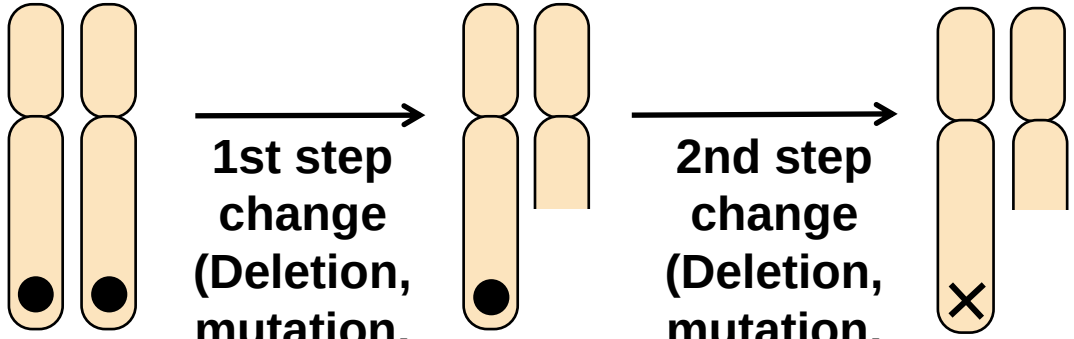


Epigenetic abnormality (no change in the gene sequence)

Tumor-suppressor genes: Include genes that cause familial (hereditary) tumors

Non-hereditary tumors

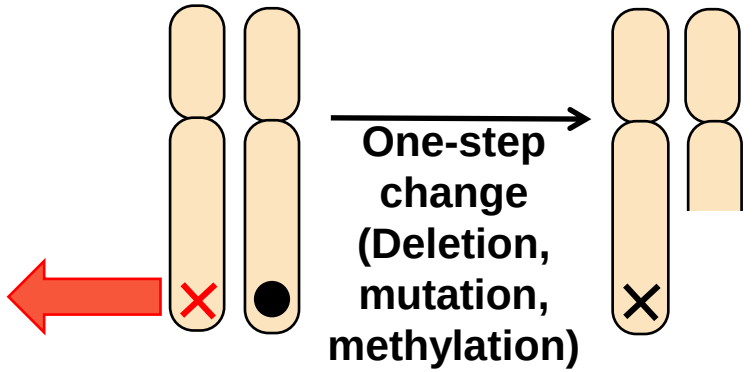
Normal cancer requires a two-step change



Familial (hereditary tumor)

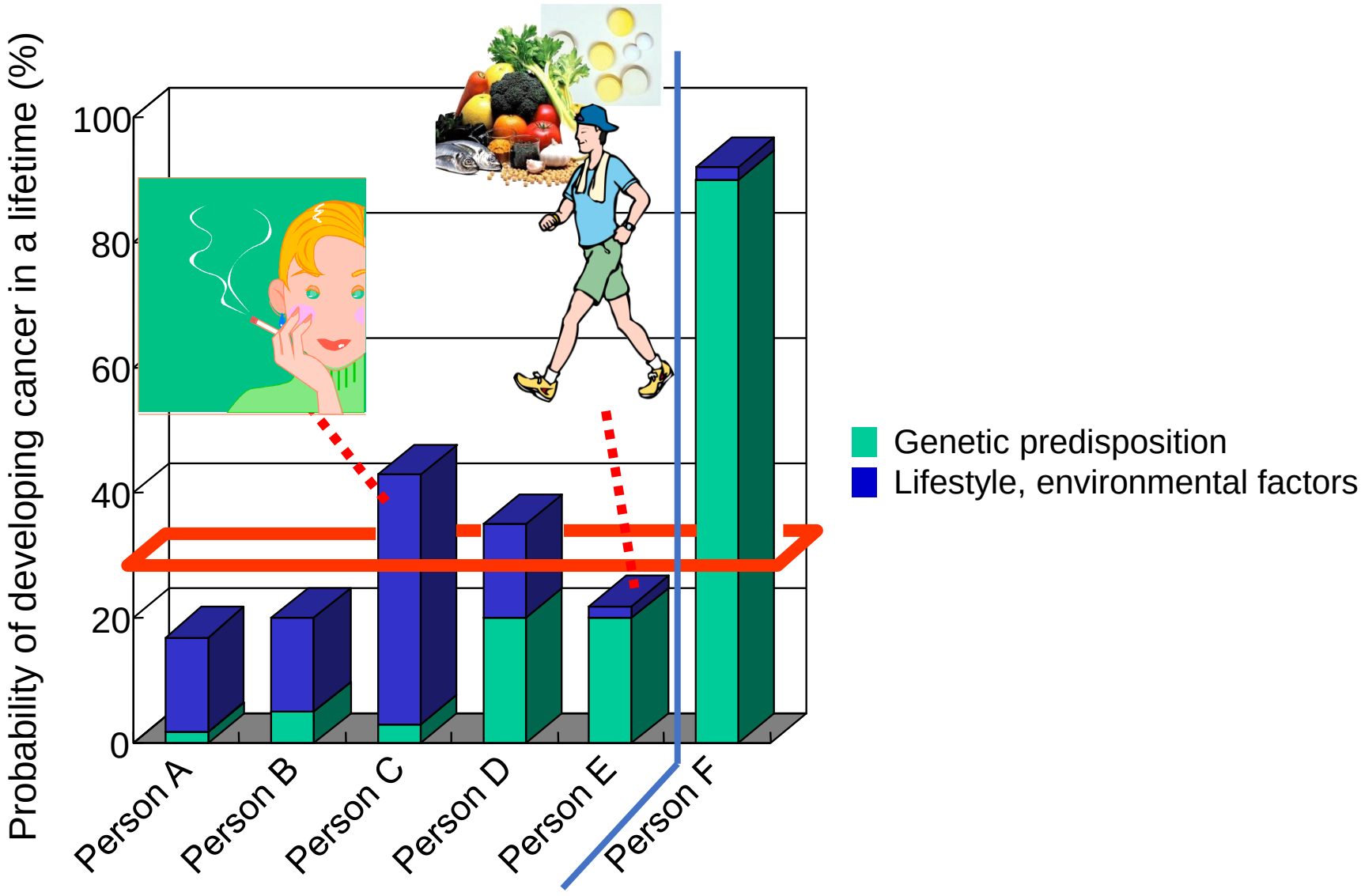
Only requires one-step change

Genetic predisposition: Individual genetic differences (germline mutation)



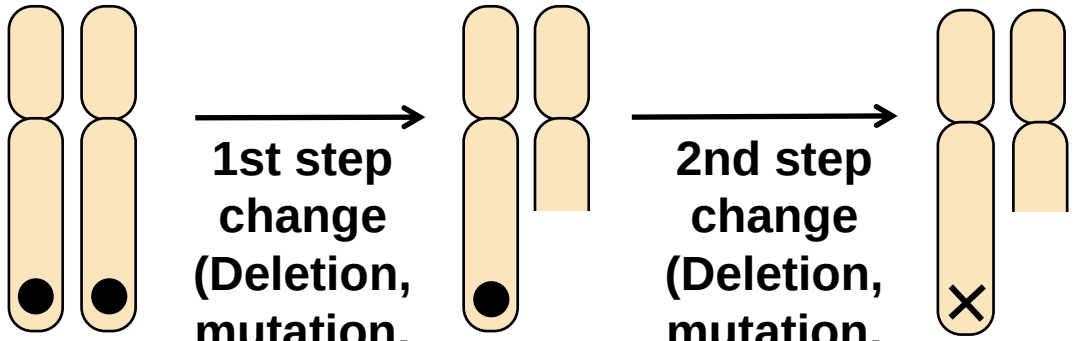
BRCA1, BRCA2 (hereditary breast cancer)
APC (familial adenomatous polyposis)
TP53 (Li-Fraumeni syndrome)

Cancer Risk: External Factors and Internal Factors



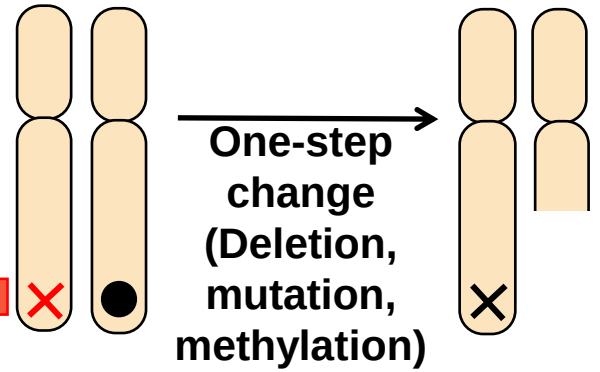
Non-hereditary tumors

Normal cancer requires a two-step change



Familial (hereditary tumor)

Only requires one-step change



Genetic predisposition: Individual genetic differences (germline mutation)

BRCA1, BRCA2 (hereditary breast cancer)
APC (familial adenomatous polyposis)
TP53 (Li-Fraumeni syndrome)

Hereditary breast cancer:

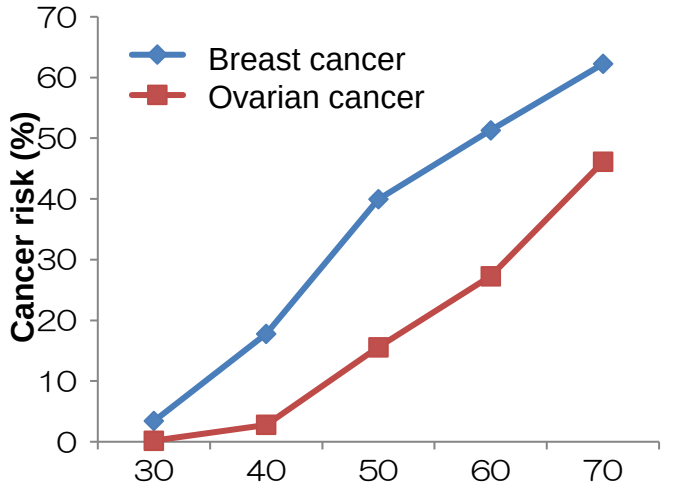
Accounts for 5–10% of all breast cancers

(Hereditary breast and ovarian cancer: HBOC)

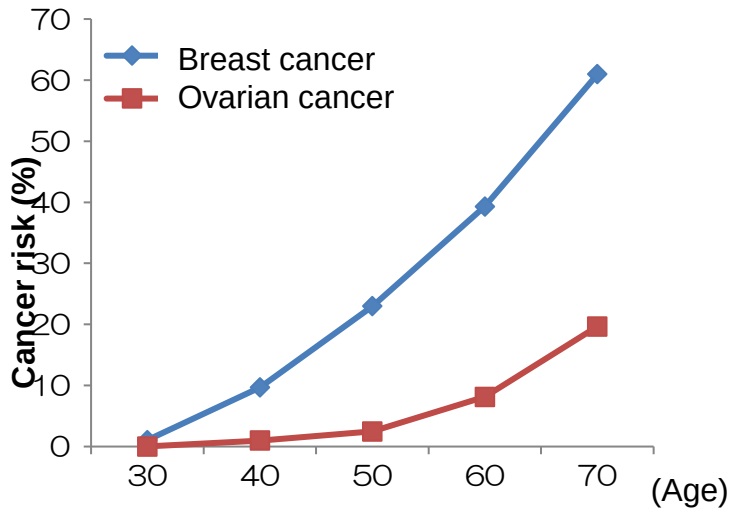
- BRCA1 (Breast Cancer Susceptibility Gene I)
  Identified in 1994 (Science 266: 66, 1994)
- BRCA2 identified in 1995 (Nature 378: 789, 1995) 13q12
- Has an important role in signal transduction when DNA is damaged

Of 260 Japanese patients with breast cancer with a family history of breast cancer, 17.7% have the BRCA1 mutation and 13.5% have the BRCA2 mutation

Hereditary Breast Cancer and Ovarian Cancer Syndrome



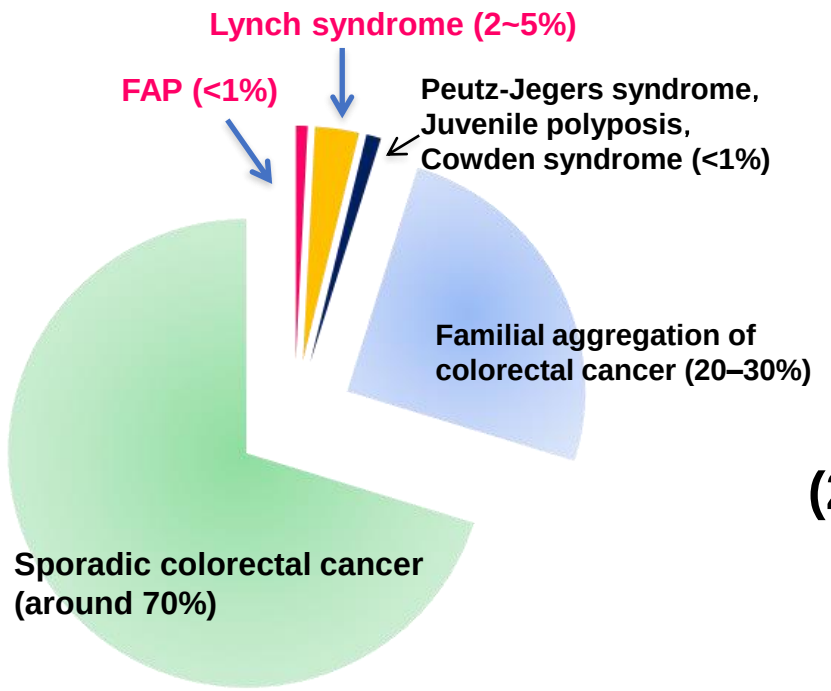
BRCA1 positive breast cancer



BRCA2 positive breast cancer

(Chen S, J Clin Oncol 24:863-71, 2006; Nakamura S, Breast Cancer 2013)

Hereditary colorectal cancer



(1) Familial adenomatous polyposis (FAP)

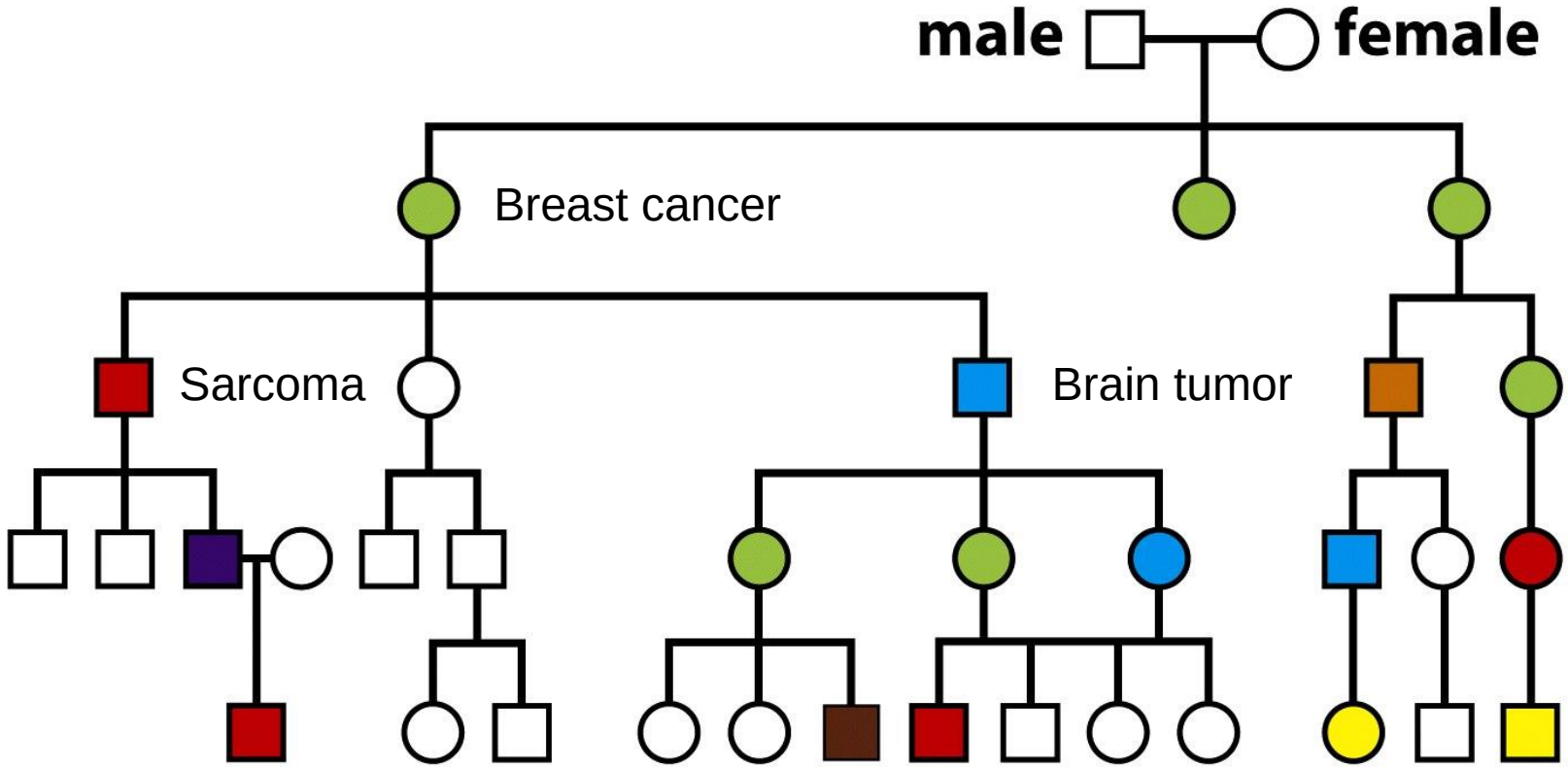
- Causative gene: *APC*
- Normally causes 100 or more colorectal adenomas
- If left untreated, almost 100% of cases will develop colorectal cancer
- Prophylactic colectomy while the person is in their 20s is recommended (total colectomy or total colectomy/ileal pouch-anal anastomosis).

(2) Lynch syndrome)

- Main causative genes: *MLH1*, *MSH2*, *MSH6*, *PMS2*
- High incidence of colorectal cancer, endometrial cancer.
- Right-sided colorectal cancer, onset at a young age
- Onset of related cancers such as ovarian cancer, stomach cancer, biliary tract cancer, renal pelvis/ureter cancer, brain tumor, and sebaceous adenoma.
- Microsatellite instability-high (MSI-H)

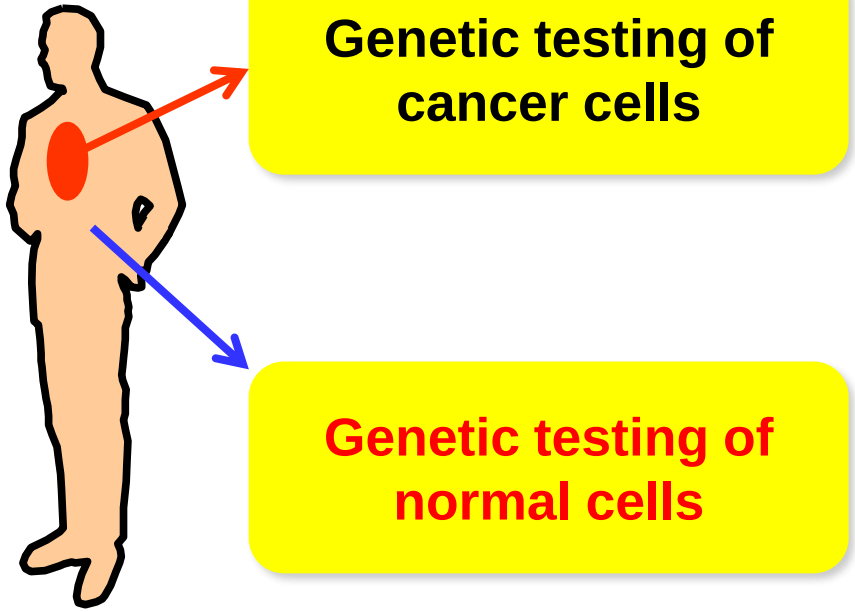
Li-fraumeni Syndrome

This is a rare inherited disorder in which various cancers, including breast cancer, sarcoma, and brain tumors occur frequently in a family.
The disease is caused by a mutation in the p53 gene.
A major feature of families with Li-Fraumeni syndrome is that family members develop different cancers even if they have a mutation in the same gene.



(From *The Biology of Cancer*)

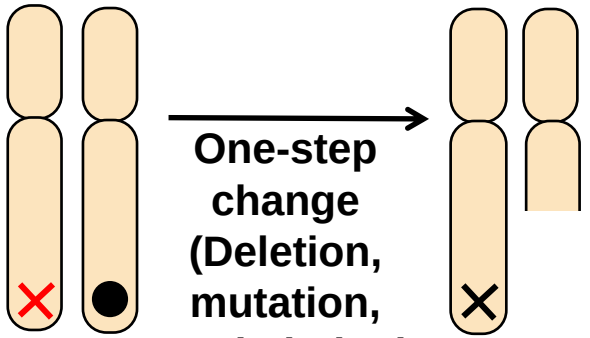
Tumor-suppressor Genes: Include Genes That Cause Familial (Hereditary) Tumors



Genetic testing of cancer cells

Genetic testing of normal cells

Genetic predisposition: Individual genetic differences (germline mutation)



BRCA1, BRCA2 (hereditary breast cancer)
APC (familial adenomatous polyposis)
TP53 (Li-Fraumeni syndrome)

It is important to provide individualized treatment based on the genetic testing of normal tissue (blood, etc.).

Genetic Testing and Individualized Treatment