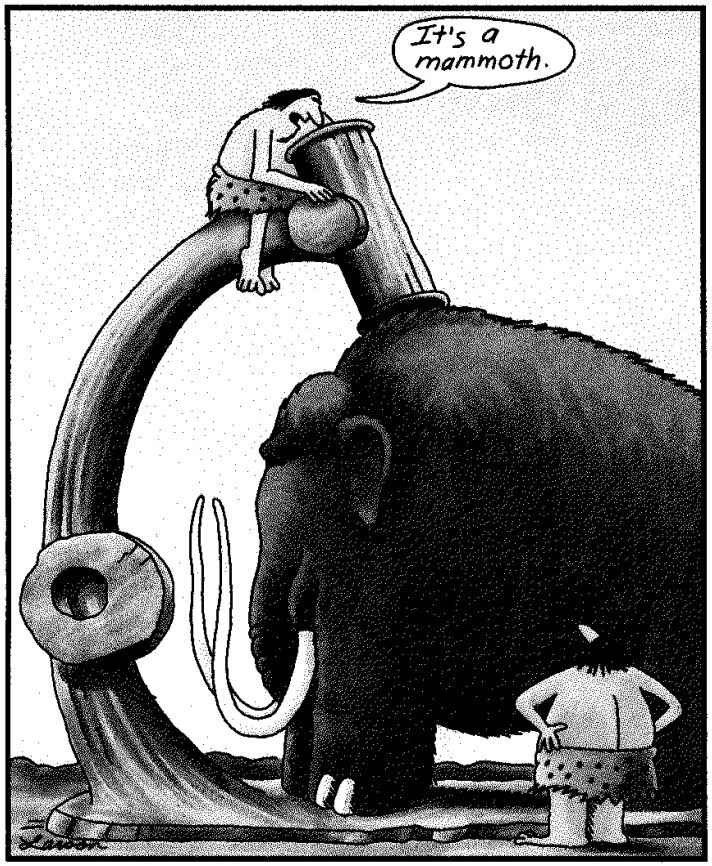
Tour of the cell: cell, cell cycle and origin of cancer

Marina Marjanovic, Ph.D.

Associate Director, Imaging at Illinois
Beckman Institute for Advanced Science and Technology
Adjunct Associate Professor, Bioengineering
University of Illinois at Urbana-Champaign

Biophotonics Summer School 2012

Biophotonics = general term for all techniques that deal with the interaction between biological structures and photons



Early microscopes

Properties of life

1. Living things are highly organized.
2. Living organisms are homeostatic.
3. Living organisms reproduce themselves.
4. Living organisms grow and develop.
5. Living organisms respond to stimuli.
6. Living organisms are adapted.
7. Living organisms can take energy from the environment and change its form.

Organizational levels of life

Simple molecules

↓

Macromolecules

↓

Organelles

↓

Cells

↓

Tissues

↓

Organs

↓

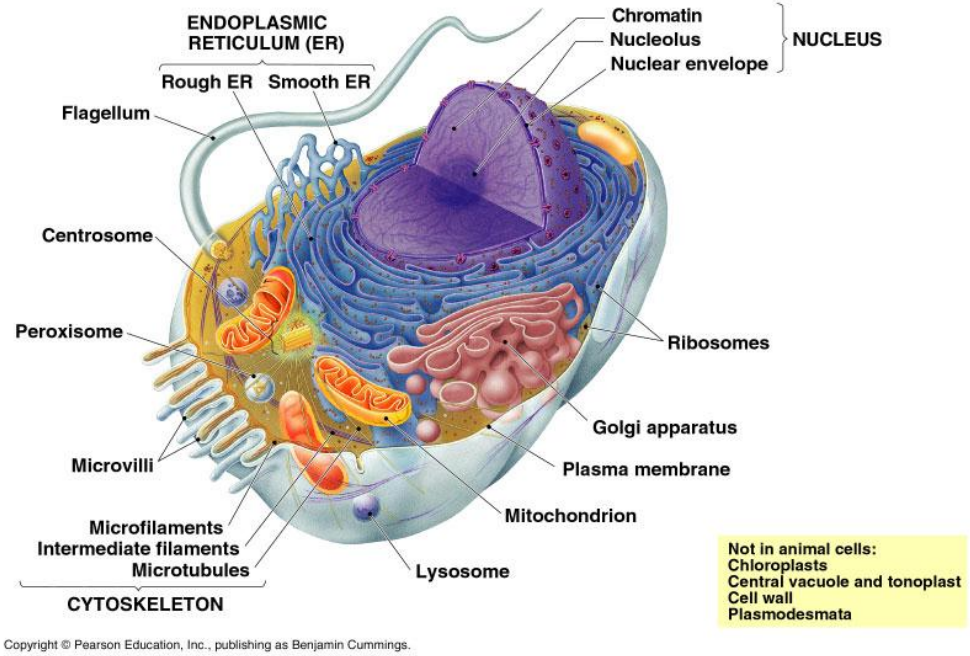
Organ systems

↓

Organism

Cell = smallest unit of life

Overview of a (animal) cell

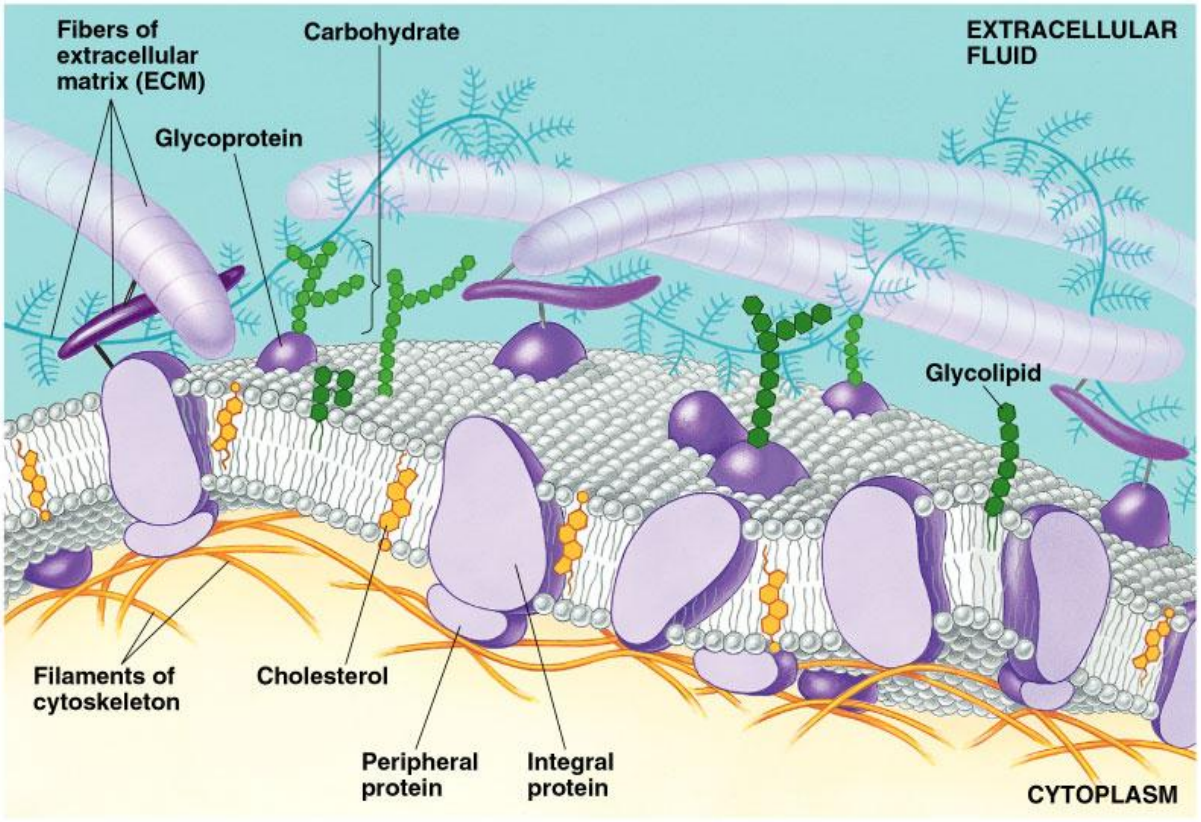


- All cells are enclosed by a **membrane** that regulates the passage of materials between the cell and its surroundings.
- DNA (genetic material) is found in the **nucleus**.
- Surrounding the nucleus is the **cytoplasm** which contains various **organelles.**

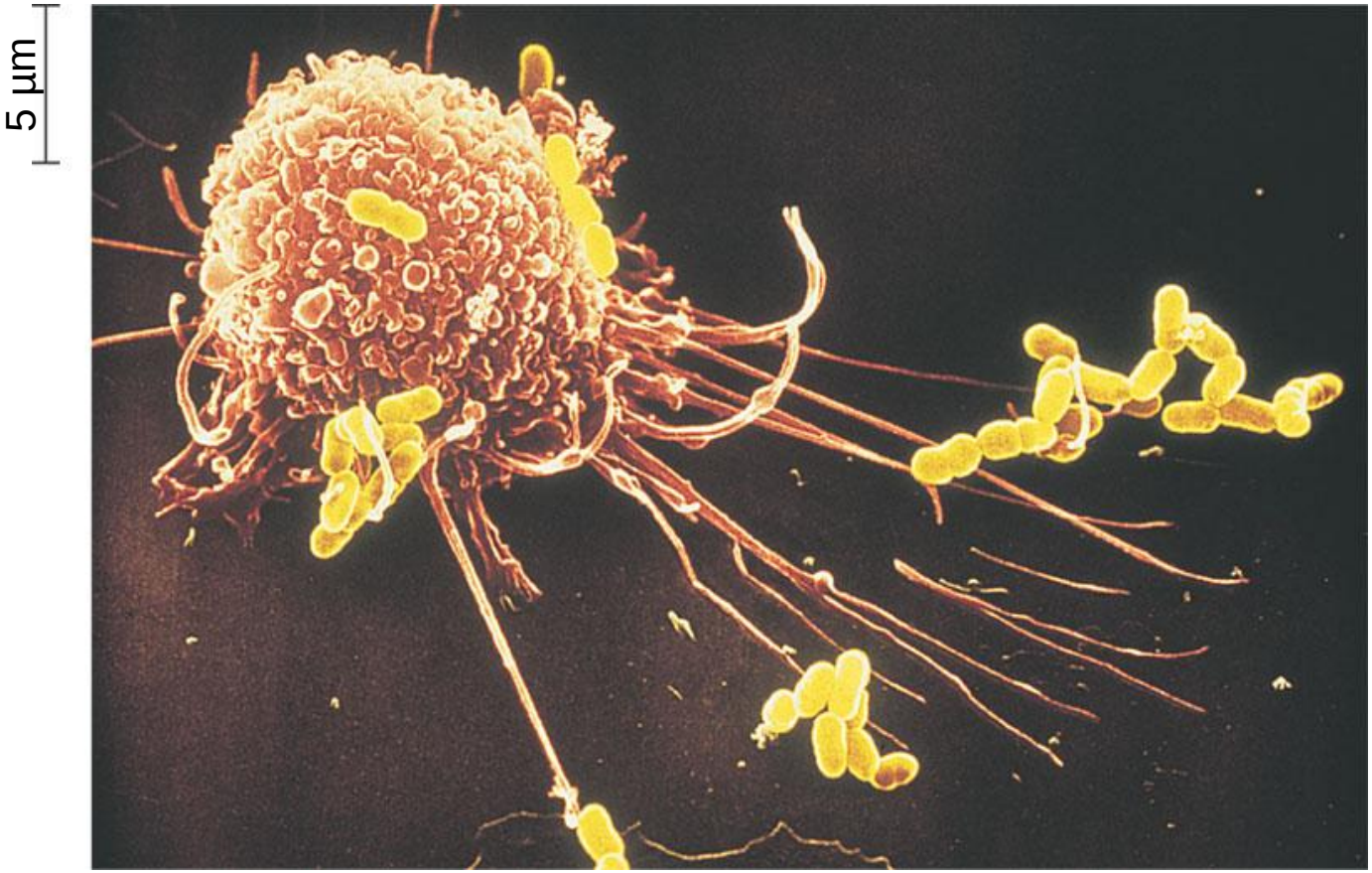
Complex structure of the extracellular matrix (ECM)

Functions of the ECM include:

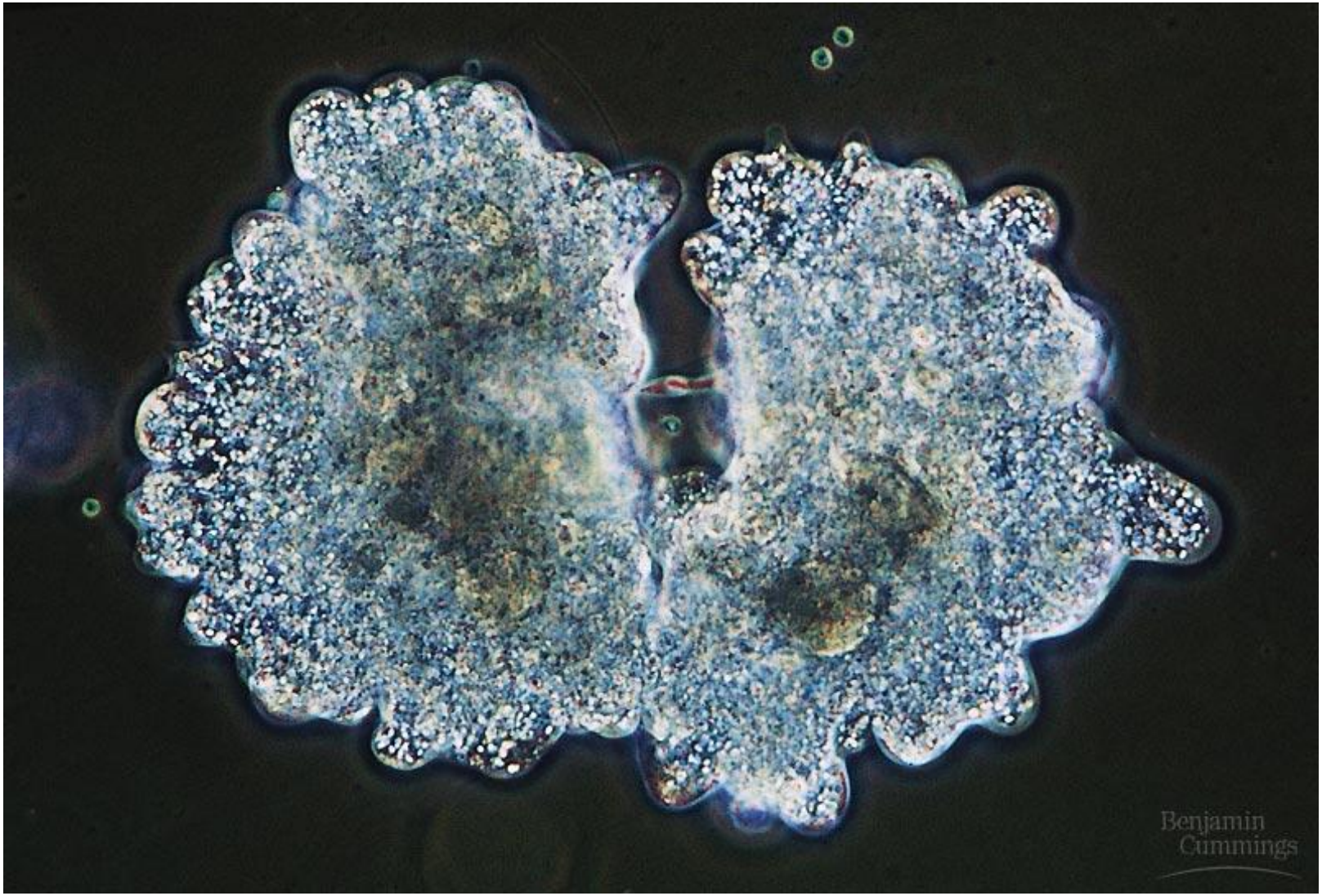
- support
- adhesion
- movement
- regulation



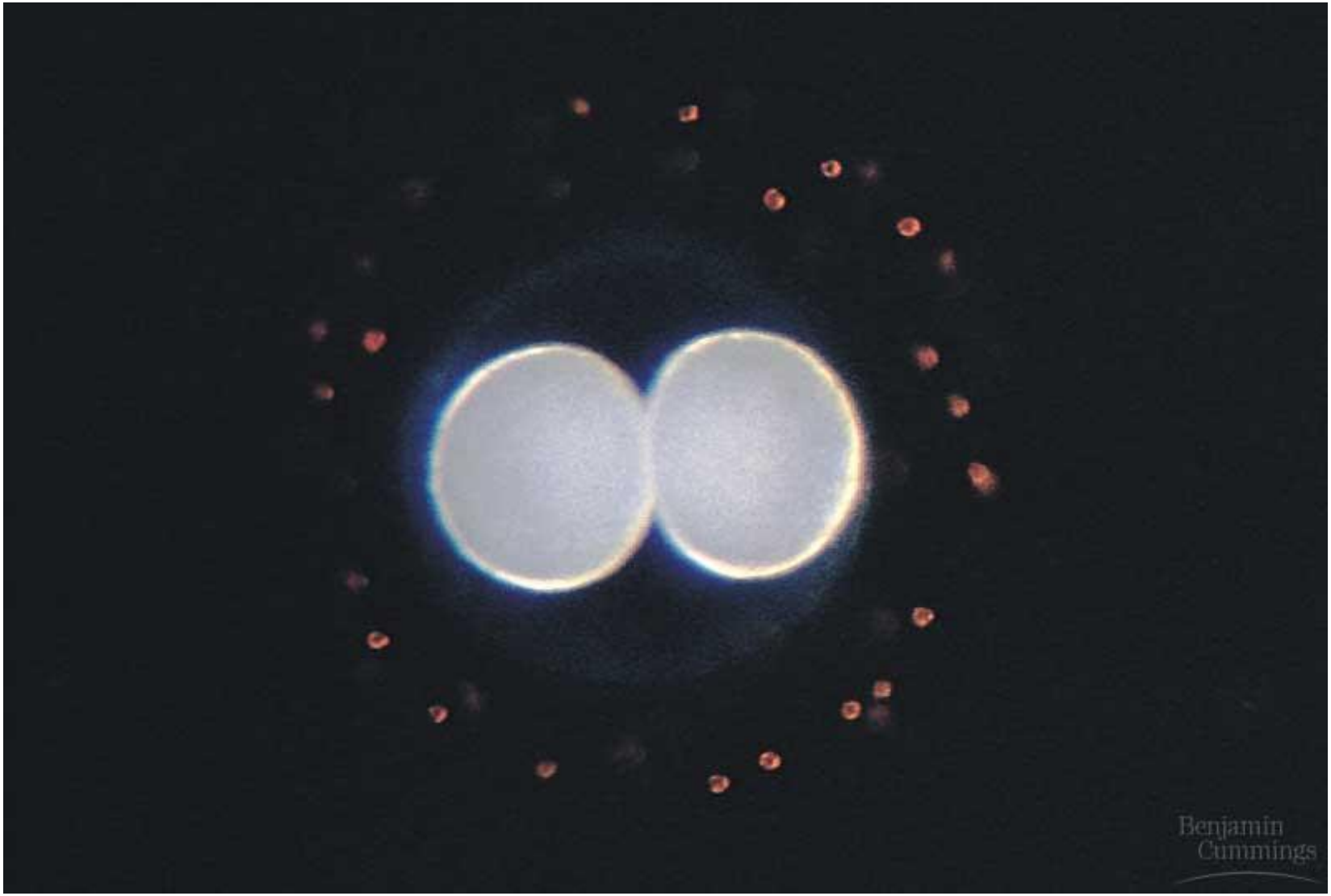
- Cells rely on the integration of structures and organelles in order to function.
- Cell is a living unit greater than the sum of its parts.



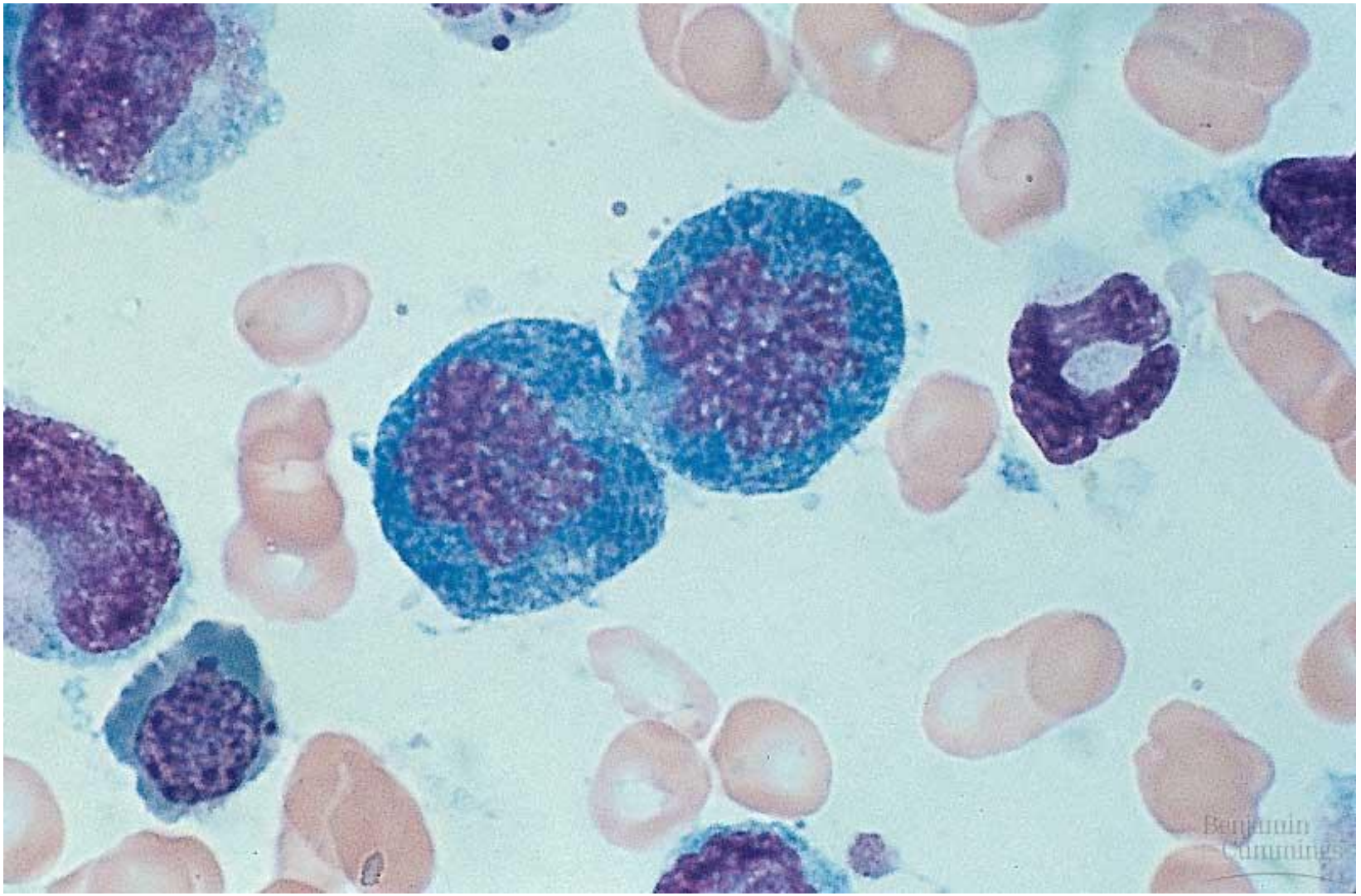
The functions of cell division: reproduction



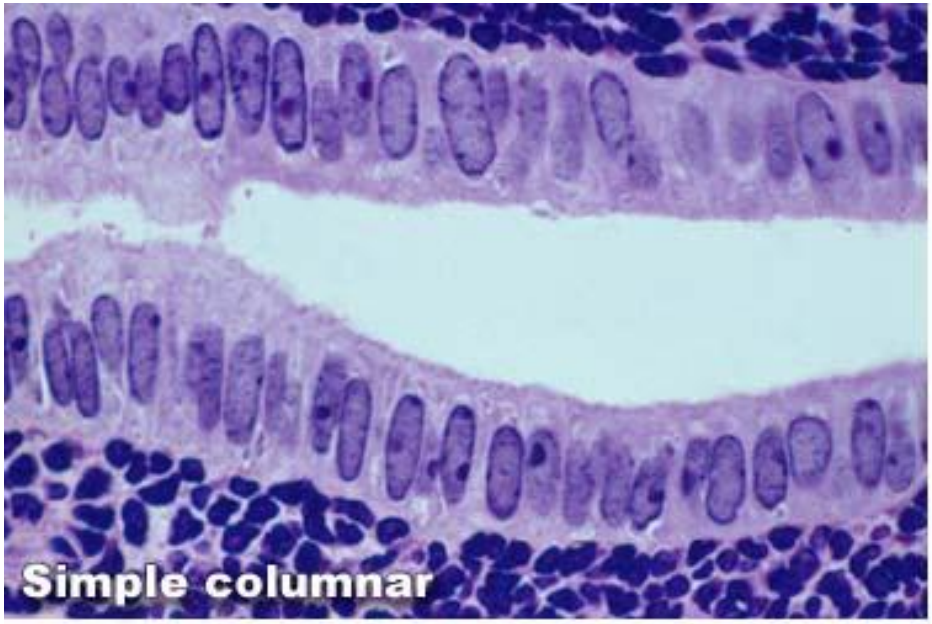
The functions of cell division: growth and development

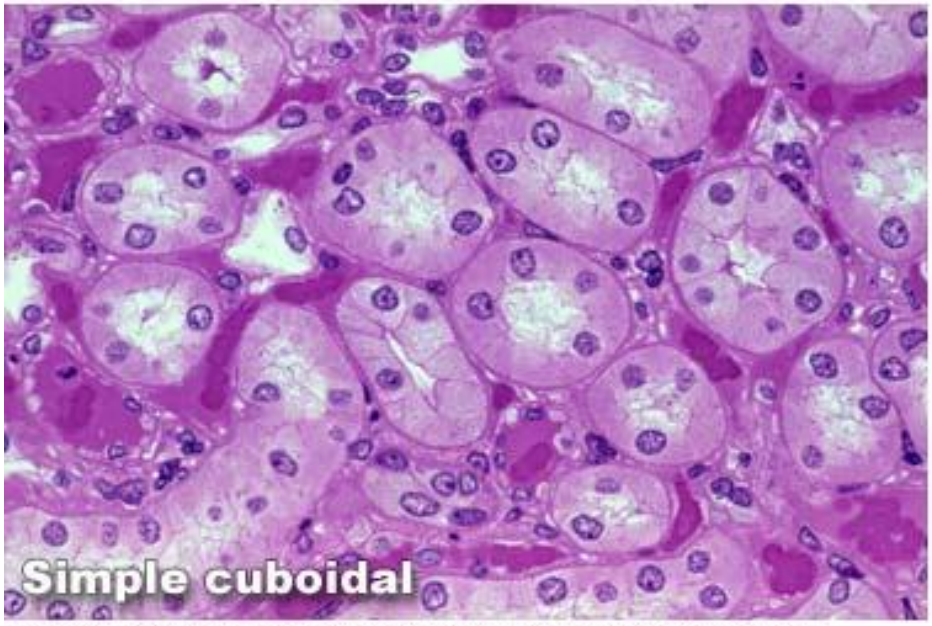


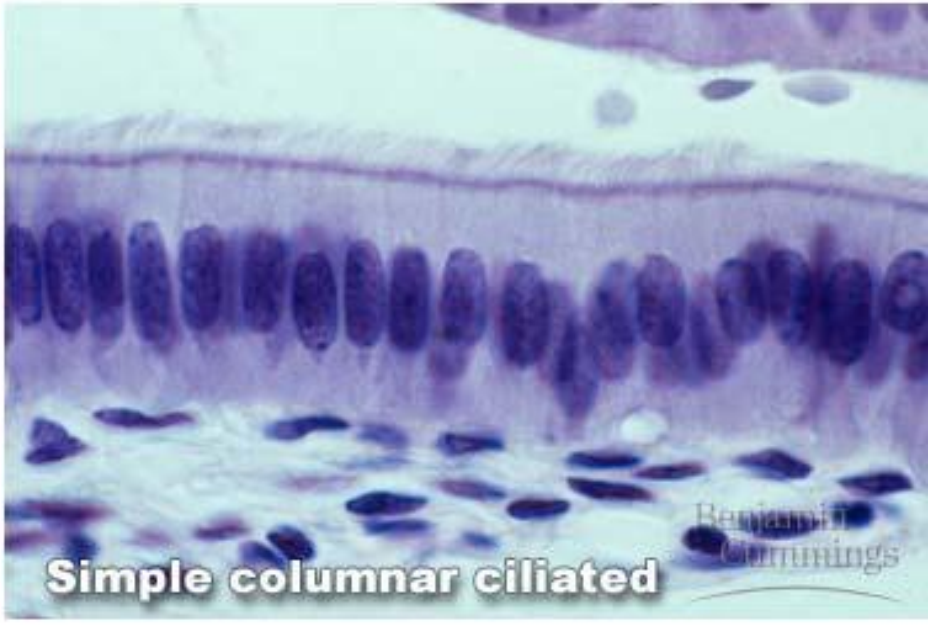
The functions of cell division: tissue renewal



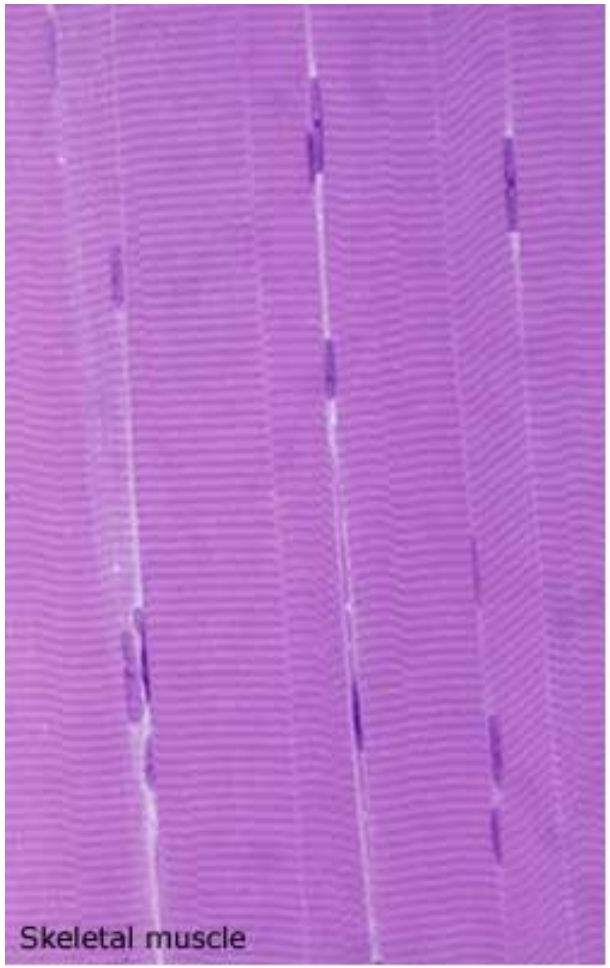
Epithelial tissues

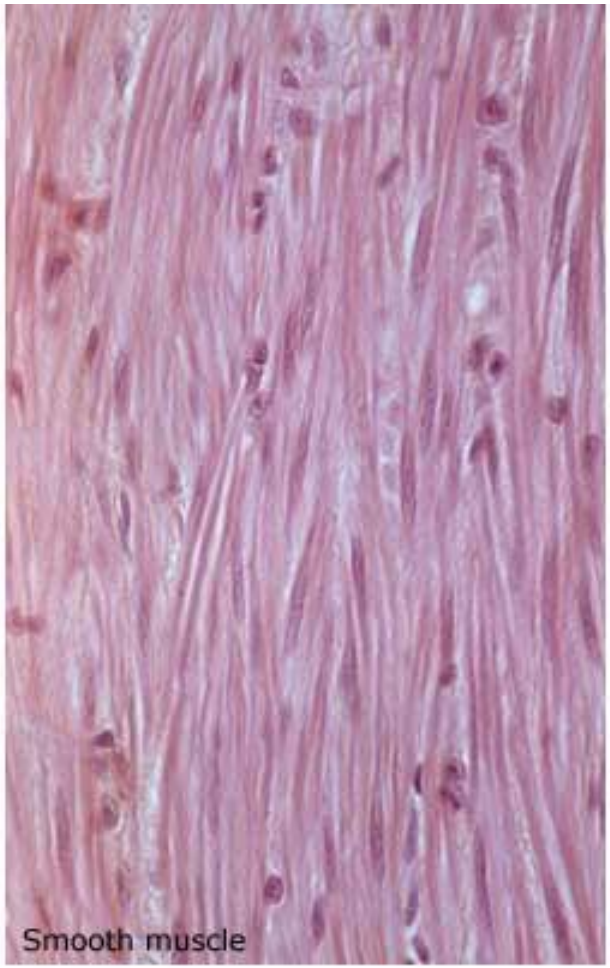


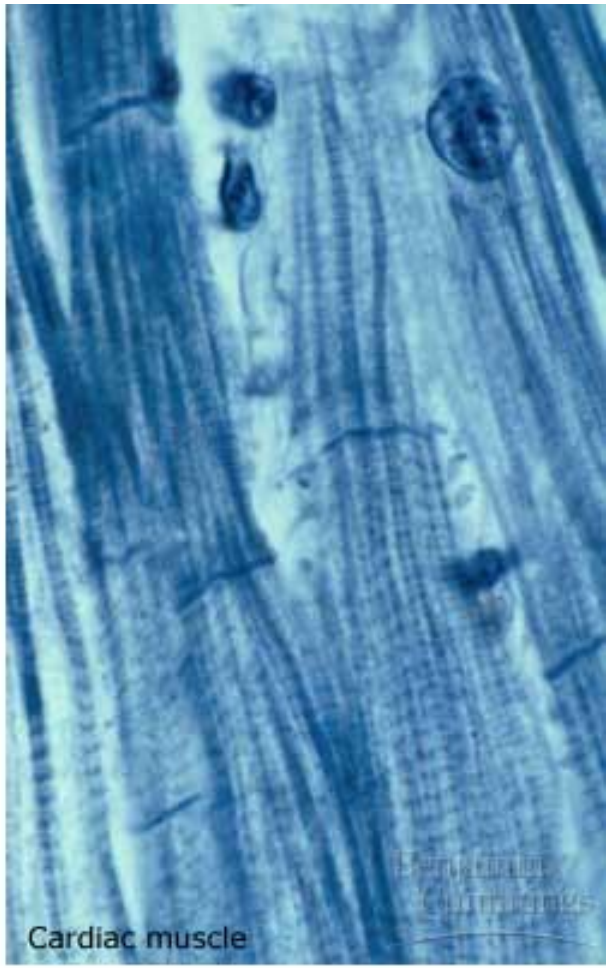




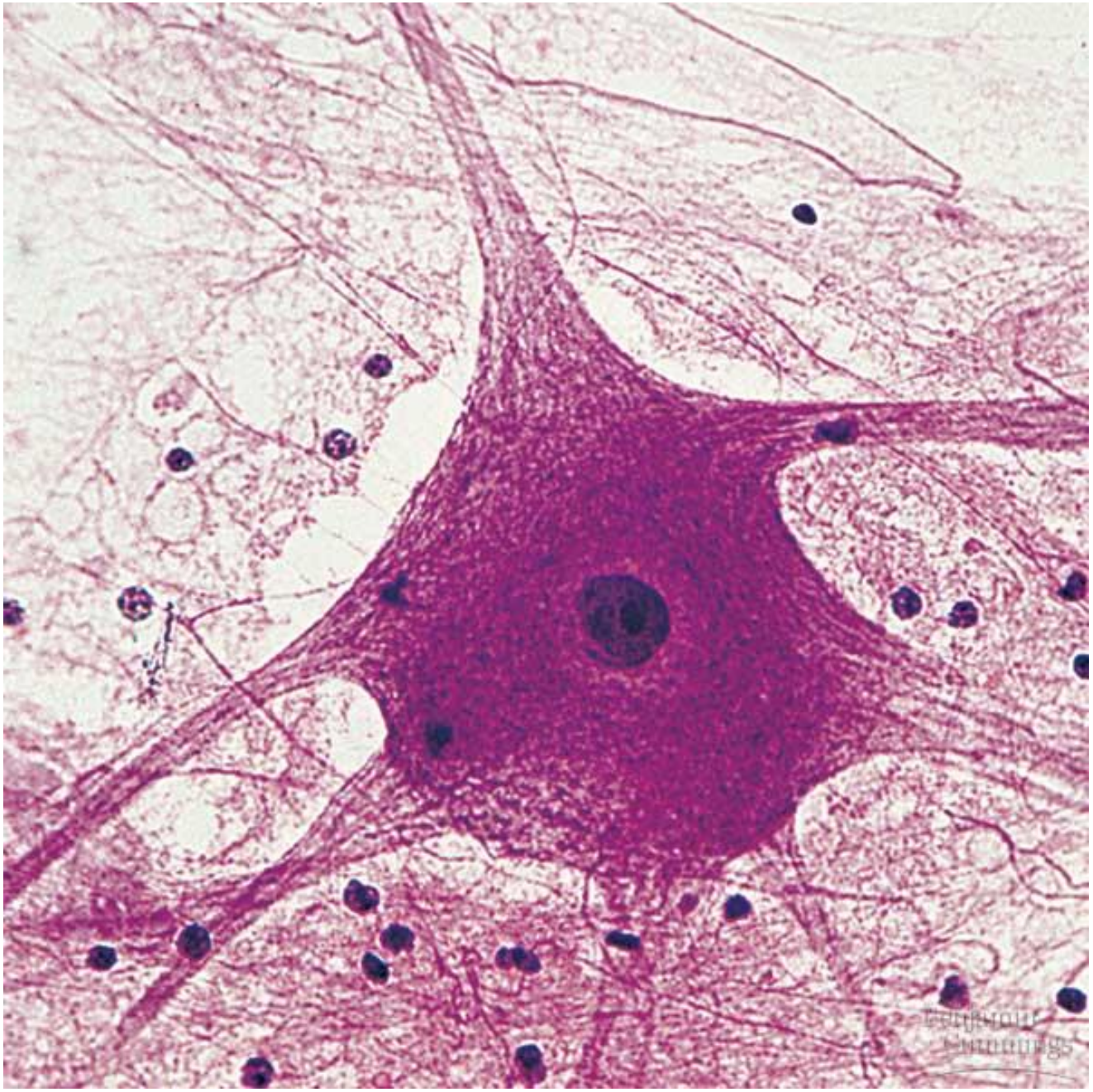
Muscle tissues



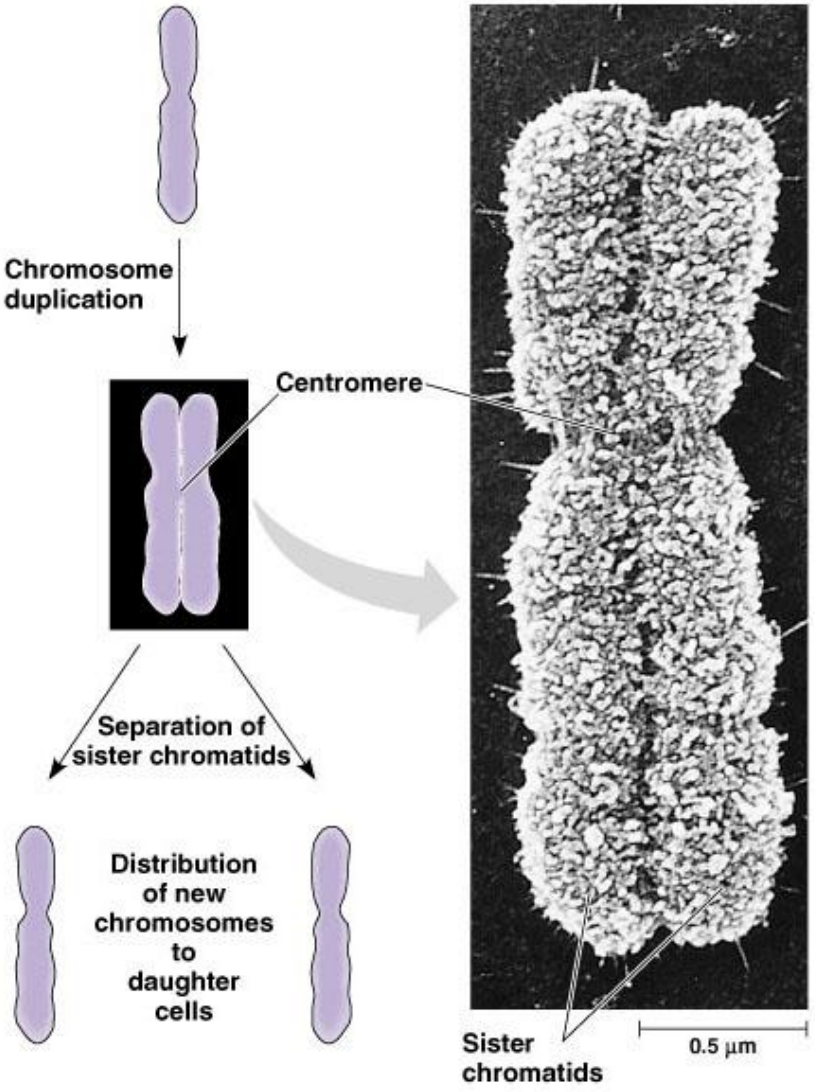




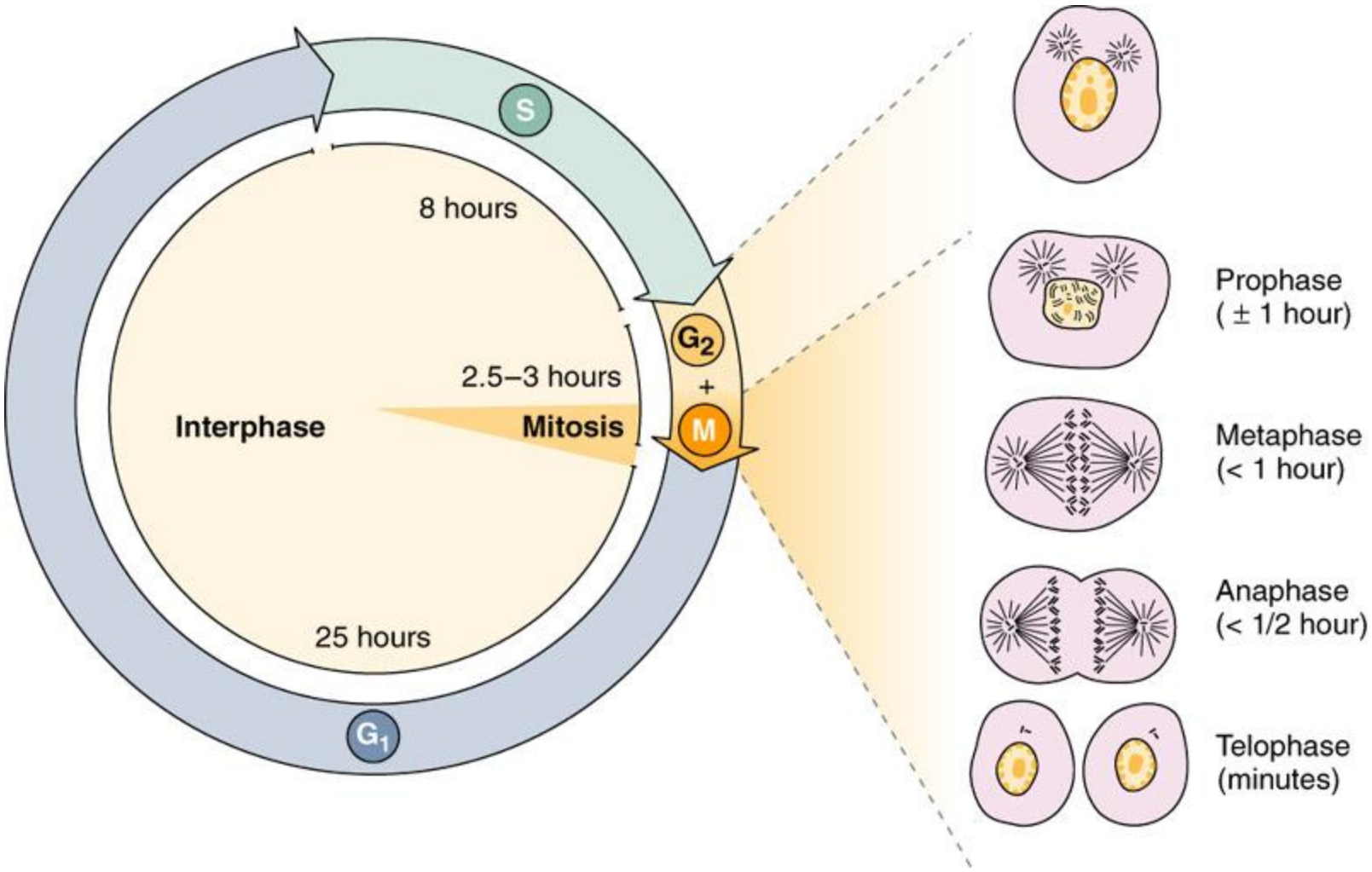
Neuron: a basic cell of the nervous tissue



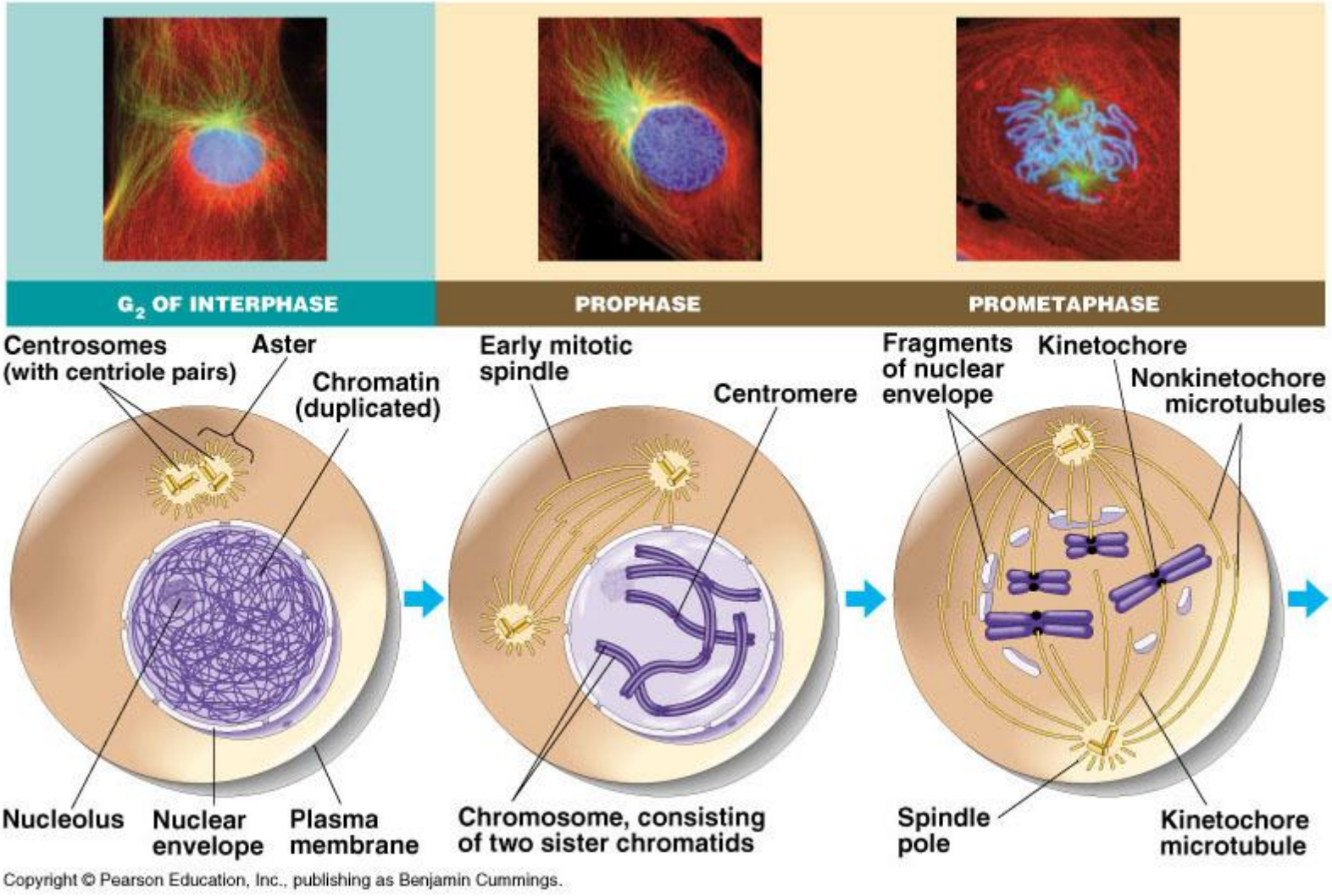
Chromosome formation and replication



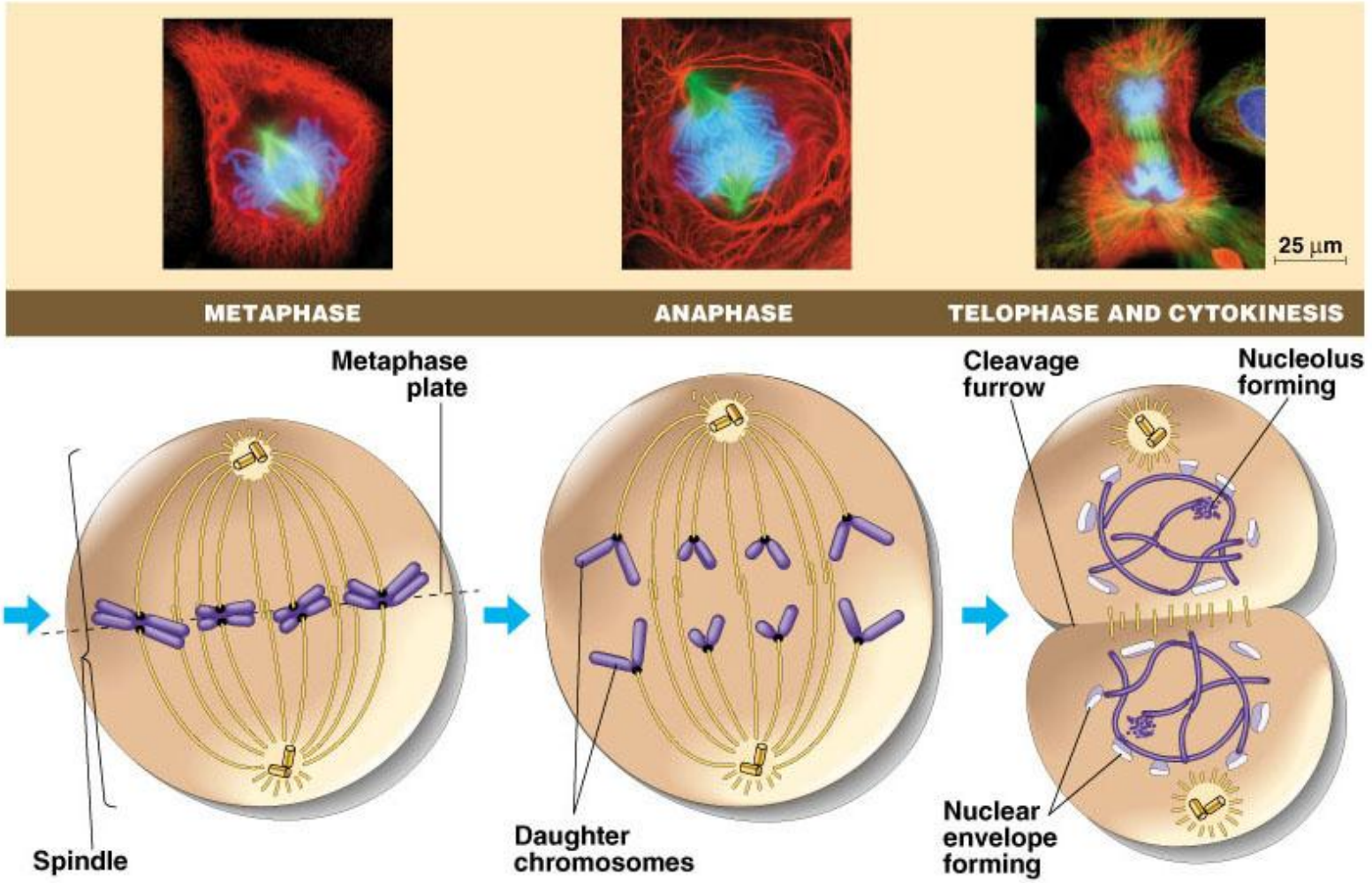
Cell cycle


S
8 hours
G_2
+
M
2.5–3 hours
Interphase
Mitosis
25 hours
G_1
Prophase
(± 1 hour)
Metaphase
(< 1 hour)
Anaphase
(< 1/2 hour)
Telophase
(minutes)

Stages of mitotic cell division in an animal cell

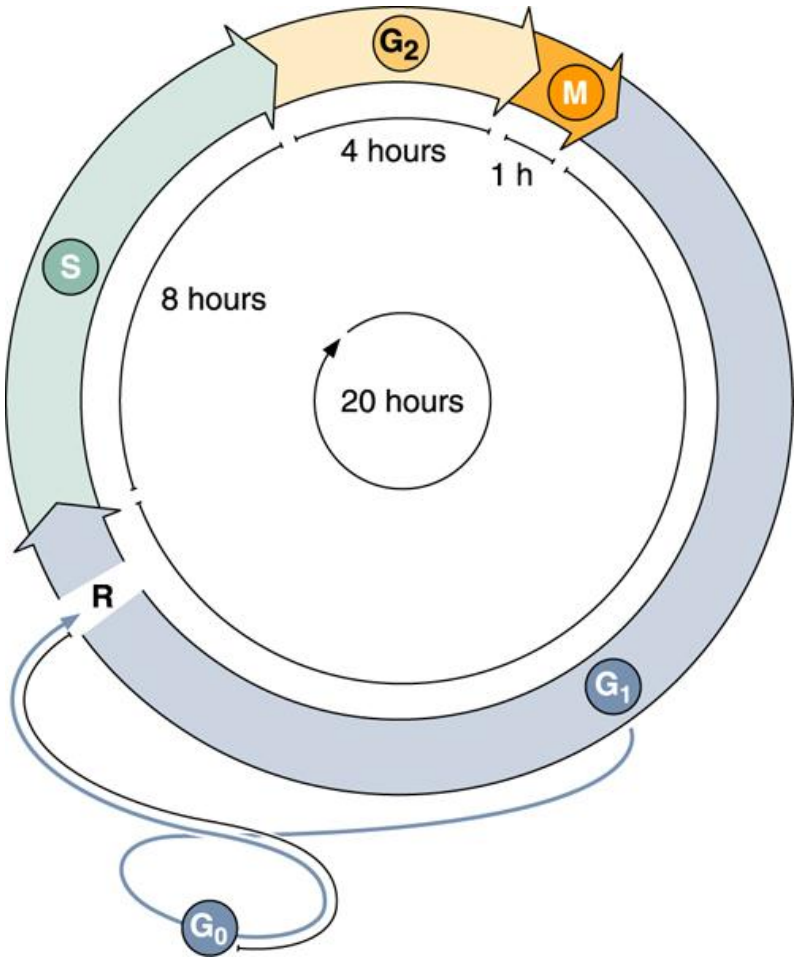


Stages of mitotic cell division in an animal cell



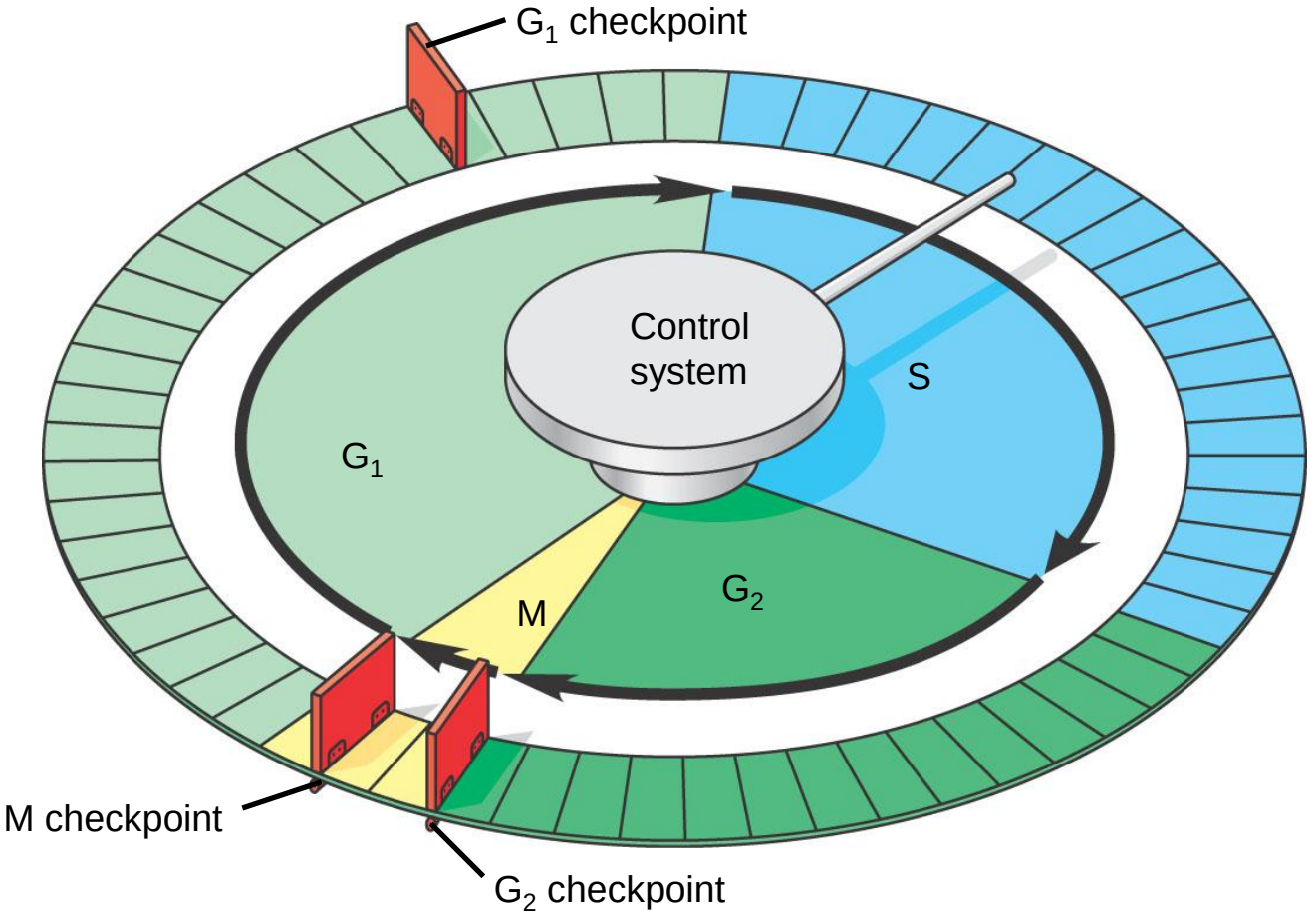
Cell cycle

- The frequency of cell division varies with the type of cell
- These cell cycle differences result from regulation at the molecular level

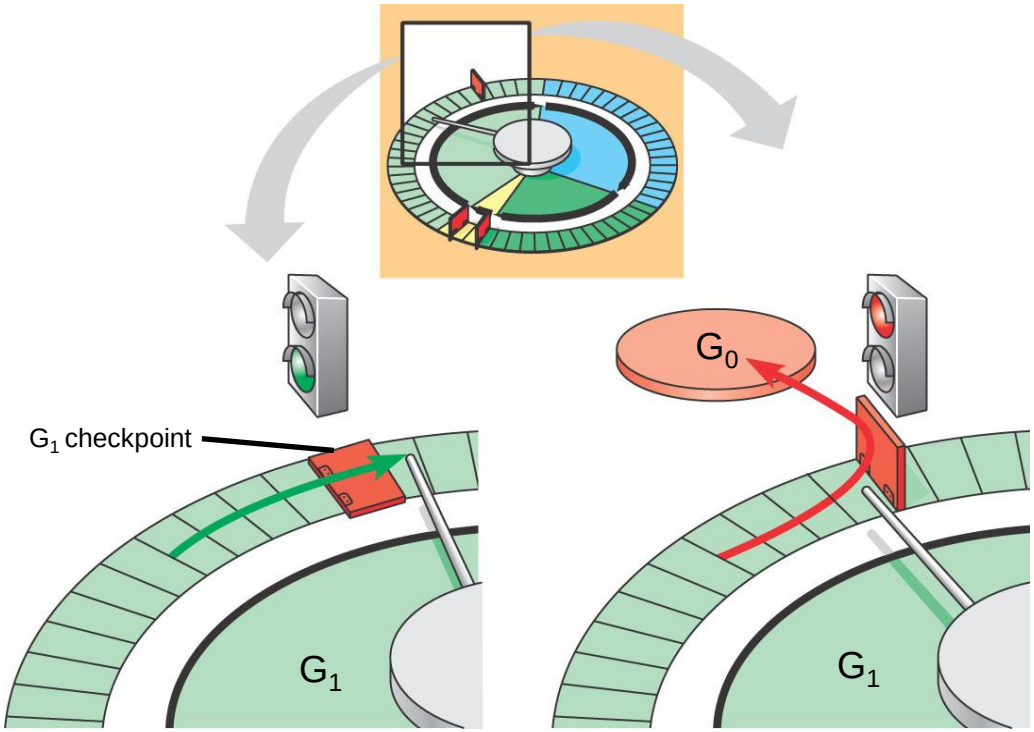


The cell cycle control system

- The sequential events of the cell cycle are directed by a distinct cell cycle control system, which is similar to a clock



- The clock has specific checkpoints where the cell cycle stops until a go-ahead signal is received.
- Both internal and external signals control the cell cycle checkpoints.
- Cancer cells do not respond normally to the body's control mechanisms and form tumors.

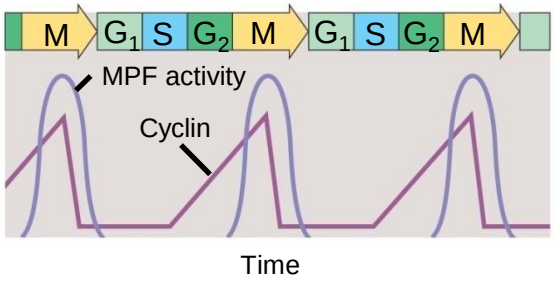


(a) If a cell receives a go-ahead signal at the G_1 checkpoint, the cell continues on in the cell cycle.

(b) If a cell does not receive a go-ahead signal at the G_1checkpoint, the cell exits the cell cycle and goes into G_0, a nondividing state.

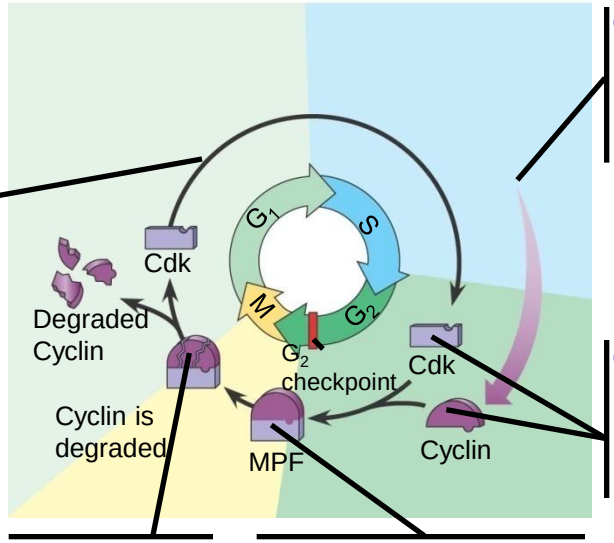
- Two types of regulatory proteins are involved in cell cycle control: cyclins and cyclin-dependent kinases (Cdks)
- The activity of cyclins and Cdks fluctuates during the cell cycle

(a) Fluctuation of MPF activity and cyclin concentration during the cell cycle



MPF = cyclin + cyclin-dependent kinase
MPF = 'M-phase-promoting factor'

(b) Molecular mechanisms that help regulate the cell cycle



1 Synthesis of cyclin begins in late S phase and continues through G_2. Because cyclin is protected from degradation during this stage, it accumulates.

2 Accumulated cyclin molecules combine with recycled Cdk molecules, producing enough molecules of MPF to pass the G_2 checkpoint and initiate the events of mitosis.

3 MPF promotes mitosis by phosphorylating various proteins. MPF's activity peaks during metaphase.

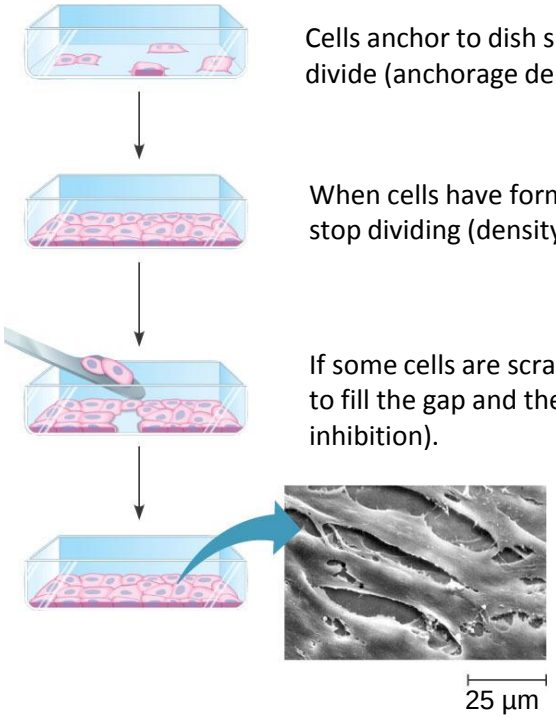
4 During anaphase, the cyclin component of MPF is degraded, terminating the M phase. The cell enters the G_1 phase.

5 During G_1, conditions in the cell favor degradation of cyclin, and the Cdk component of MPF is recycled.

Effect of external factors on cell division

- In density-dependent inhibition crowded cells stop dividing.
- Most animal cells exhibit anchorage dependence in which they must be attached to a substratum to divide.

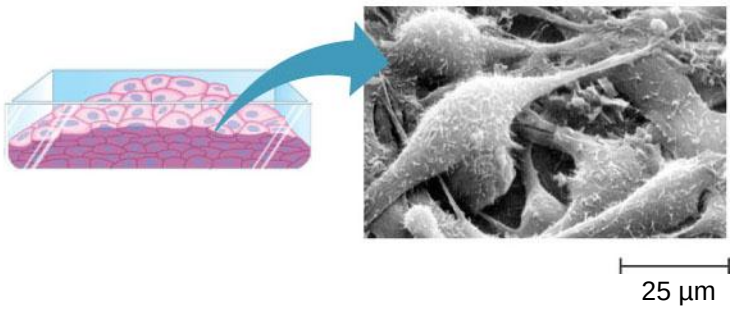
(a) Normal mammalian cells. The availability of nutrients, growth factors, and a surface for attachment limits cell density to a single layer. Normally, cells divide 20-50 times.



- Cancer cells exhibit neither density-dependent inhibition nor anchorage dependence.

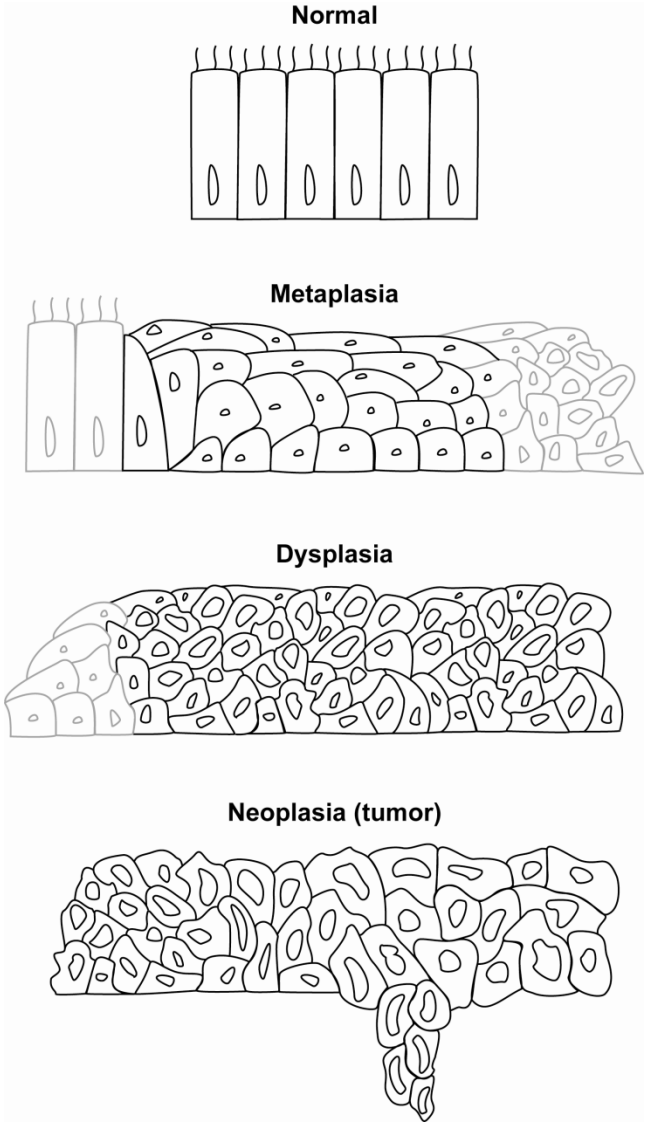
Cancer cells do not exhibit anchorage dependence or density-dependent inhibition.

(b) Cancer cells. Cancer cells usually continue to divide well beyond a single layer, forming a clump of overlapping cells. They can divide indefinitely ("immortal" cells).



Properties of a transformed cell:

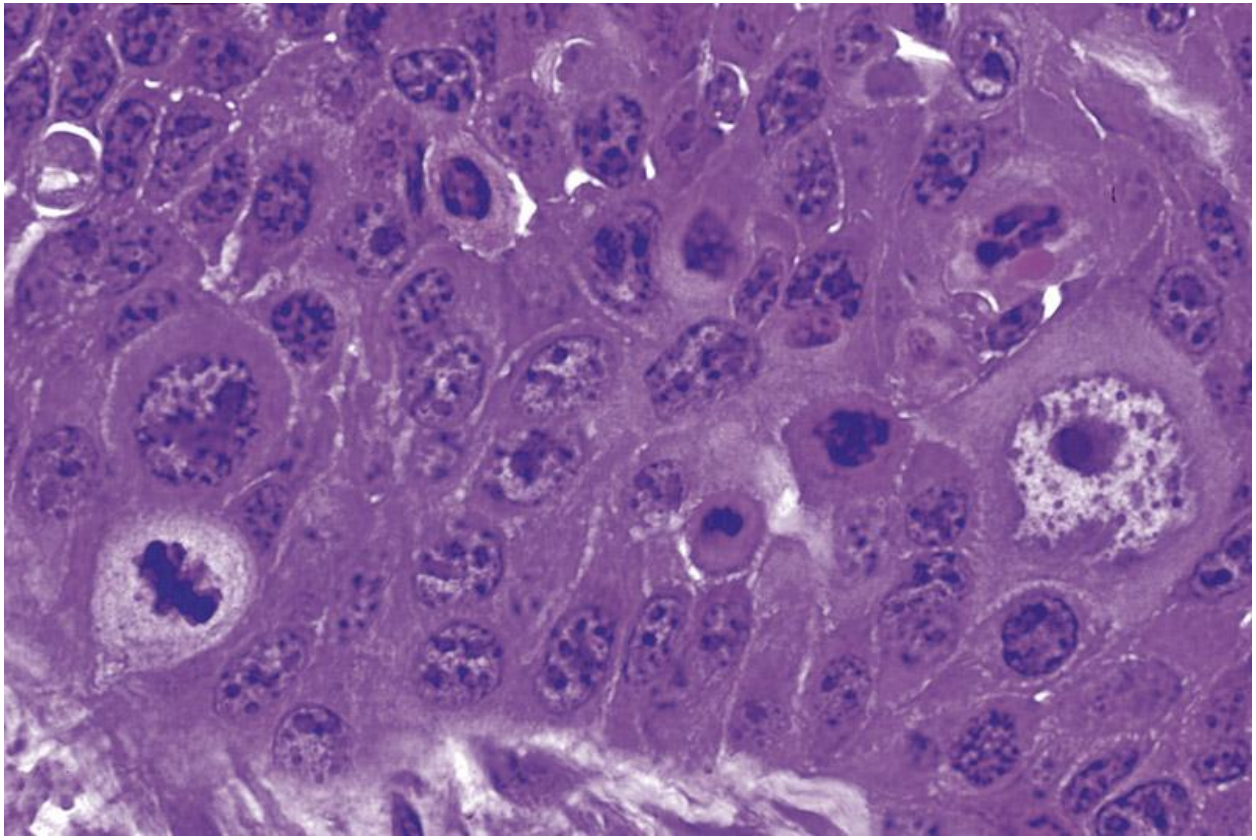
- Unusual number of chromosomes
- Abnormal metabolism
- Loss of normal cellular functions
- Loss of density-dependent inhibition
- Loss of anchorage dependence
- Release of signal molecules that cause growth of blood vessels toward the tumor



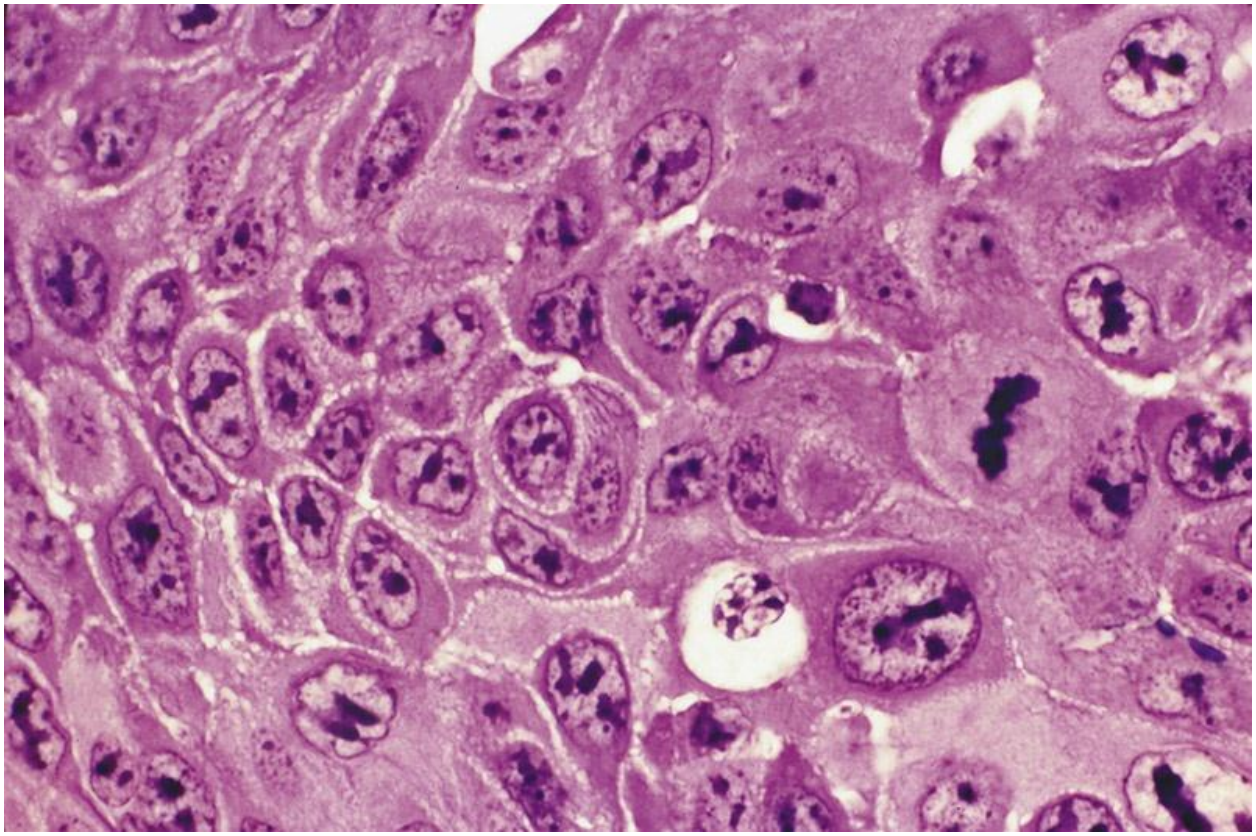
- change in the cell/tissue appearance

- loss of cell uniformity
- diversity in nuclear size and shape
- increased number of cells in mitosis
- pre-malignant change (carcinoma in situ)

neoplasia = new growth

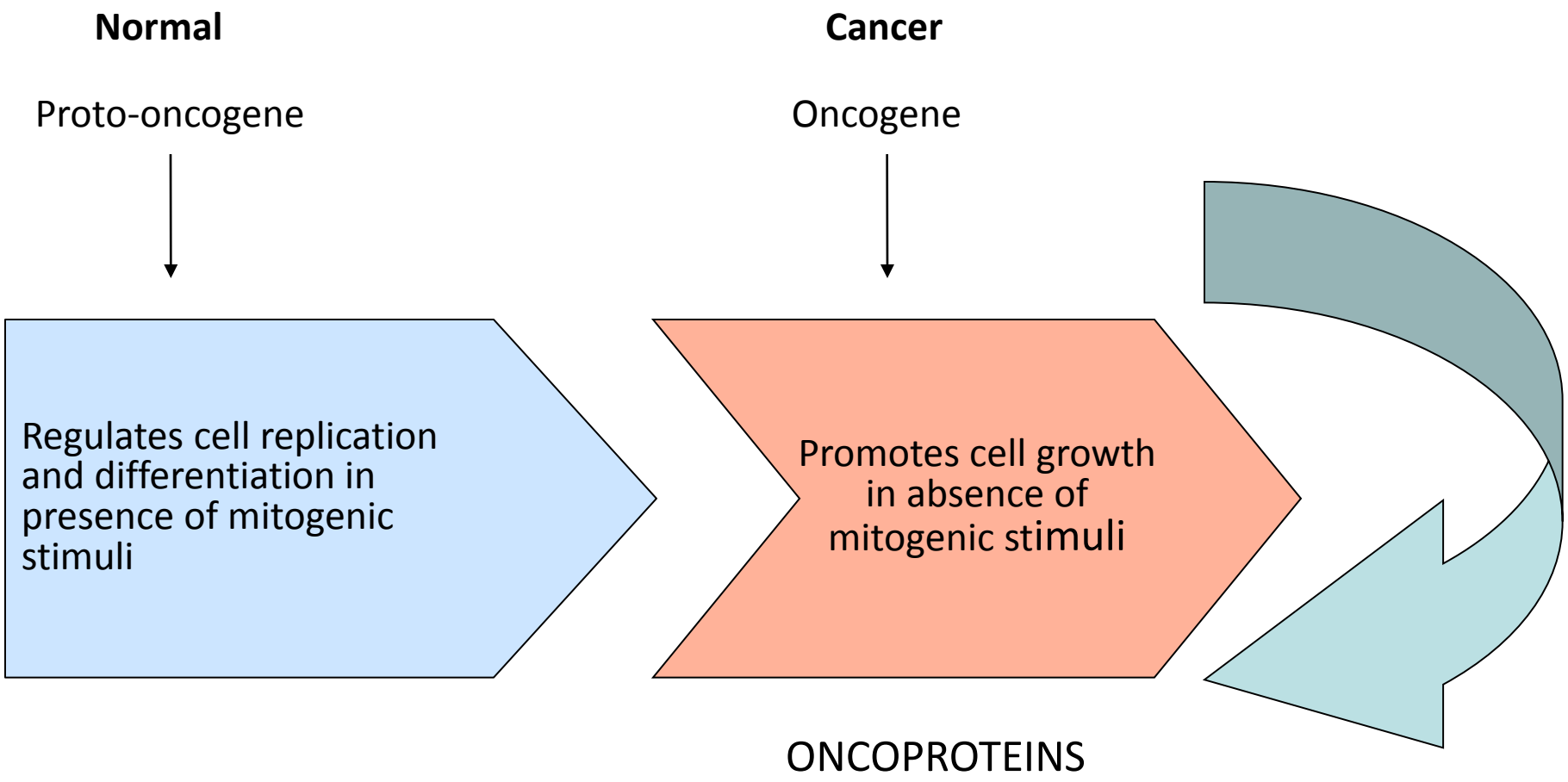


Section of a malignant epithelial skin tumor (squamous cell carcinoma). An increase in the number of cells in mitosis and diversity of nuclear morphology are signs of malignancy.

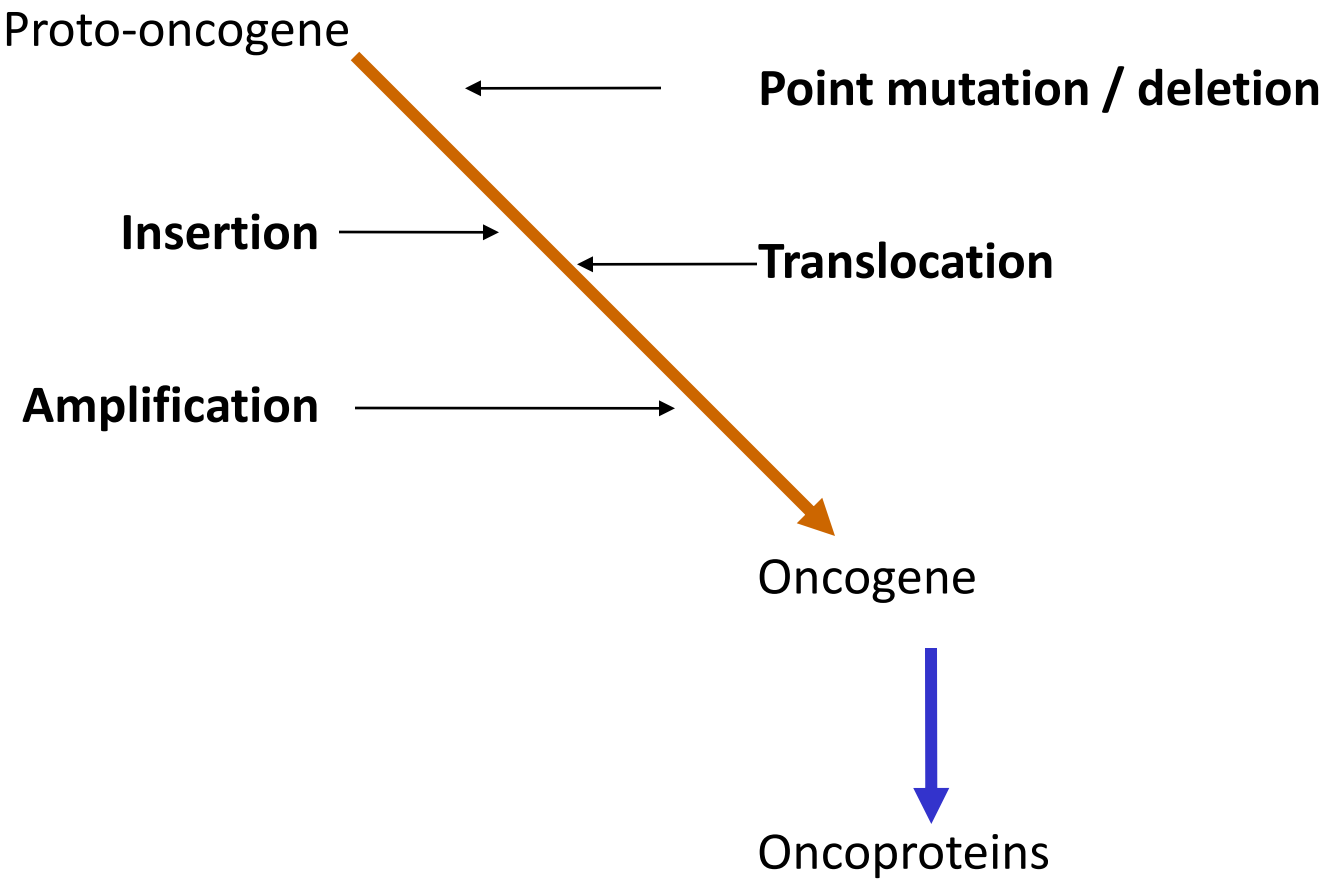


Section of a fast-growing malignant epithelial skin tumor showing an increased number of cells in mitosis and great diversity of nuclear morphology.

Oncogenes: genes that promote independent growth

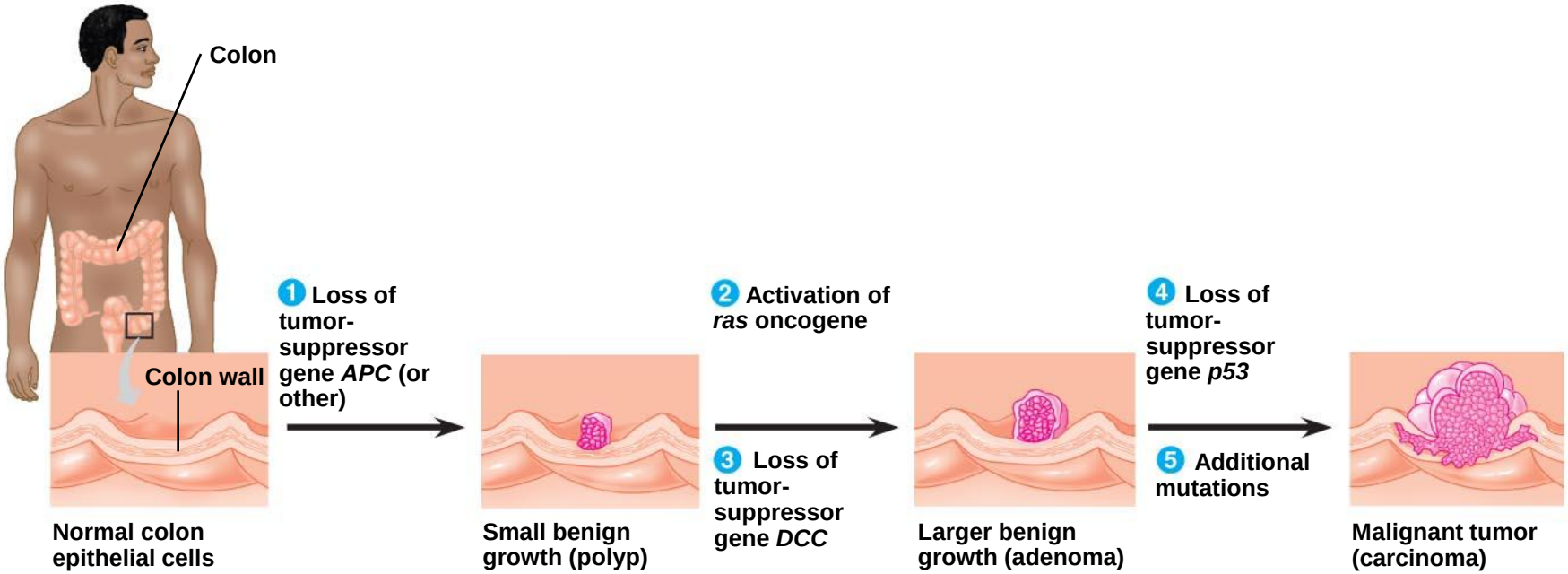


Oncogenes - How are they formed?



Multi-step model for the development of colorectal cancer

- Progressive, non lethal DNA damage leading to uncontrolled cell division



- Purposeless, pathologic proliferation of cells characterized by loss of control over cell division.

- DNA damage at growth control genes is central to development of neoplasm: Non-lethal genetic damage that causes clonal proliferation of affected cells.

- Types:

Benign
- slow growing
- capsulated
- non-invasive
- do not metastasize
- well differentiated

Malignant
- fast growing
- non capsulated
- invasive
- metastasize
- poorly differentiated

- Hanahan and Weinberg (2000). The Hallmarks of Cancer. Cell 100, 57-70.

  - proposing six common acquired capabilities that allow cancer cells to survive, proliferate and disseminate

  - Cell's most cited article (referenced over 10,000)

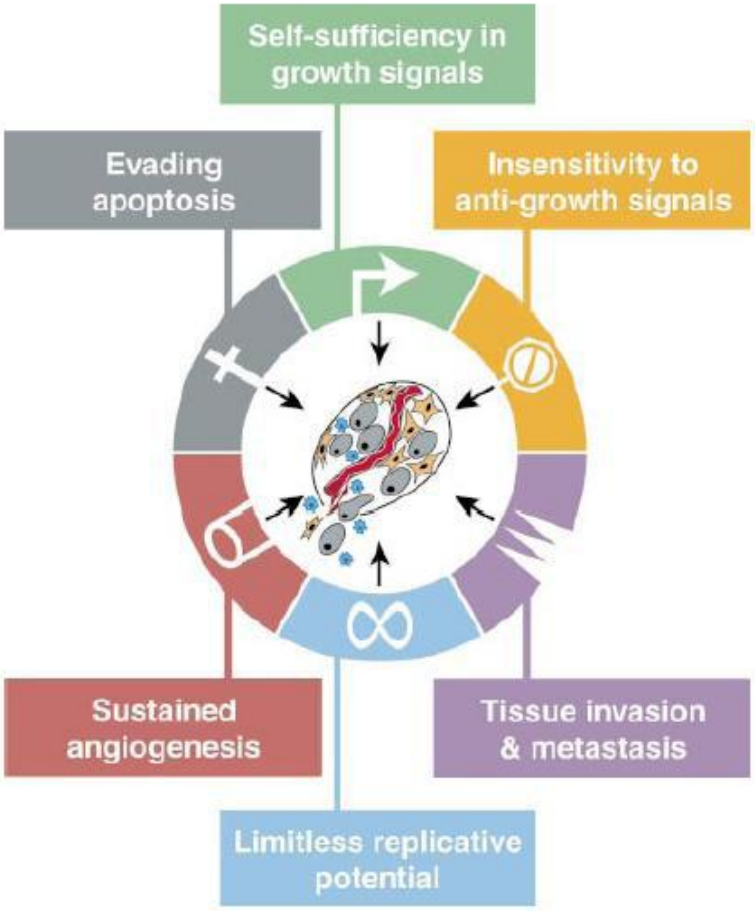
- Hanahan and Weinberg (2011). Hallmarks of Cancer: The Next generation. Cell 144, 646-674.

  - adding two new capabilities and two enabling characteristics based on research data

Six essential traits

Six acquired capabilities shared by most human tumors:

1. Self-sufficiency in growth signals
2. Insensitivity to anti-growth signals
3. Evasion of programmed cell death (apoptosis)
4. Limitless replicative potential
5. Sustained angiogenesis
6. Tissue invasion and metastasis



Self-sufficiency in growth signals

Normal cells:

- Require growth signals before moving into an active proliferative state
- Normal cells cannot proliferate without such signals
- The growth signals are extracellular and they are transmitted into the cell by binding of signal molecules (growth factors or matrix components) to transmembrane receptors

Tumor cells:

- Do not need stimulation from external signals to divide
- Tumors can mimic normal growth signals or generate their own growth signals

Insensitivity to antigrowth signals

Antigrowth signals:

- Include soluble growth inhibitors and immobilized inhibitors in the extracellular matrix

- Block proliferation
  - Cells may be forced out of active cycle into quiescent state (G_0)
  - Cells may be induced to remove their proliferation potential by being induced to postmitotic states (e.g. neurons)

- Response to antigrowth signals is associated with cell cycle clock
  - Cells monitor environment
  - Decide to proliferate, be quiescent or enter postmitotic state

Insensitivity to antigrowth signals

Normal cells:

- Tissues constrain cell multiplication by instructing cells to postmitotic or differentiated states

Tumor cells:

- Avoid antigrowth signals
- Avoid terminal differentiation (overexpression of some proteins will impair cell differentiation and promote growth)

Evading apoptosis

– Tumor cell population expansion depends on:
  - Rate of cell proliferation
  - Rate of cell attrition

– Apoptosis (programmed cell death or cell suicide) is a major barrier to cancer

– Steps of apoptosis (30-120 min)
  - Cellular membranes are disrupted
  - Nuclear skeleton is broken down
  - Chromosomes are degraded
  - Nucleus and other organelles fragmented

Apoptosis = programmed cell death (removal of transformed cells, removing of damaged cells, shaping of the embryo-morphogenesis)

- cell and nucleus become compact (pyknotic nucleus)
- DNA is fragmented
- DNA and cytoplasm fragments are forming vesicles that are detaching
- vesicles are engulfed by macrophages, but no inflammatory reaction
- prevents formation of tumors

Necrosis = accidental cell death (pathological process)

- cells swell and burst
- macrophages engulf the debris by phagocytosis
- secrete molecules that activate other immunodefensive cells and promote inflammation

Evading apoptosis

Normal cells:

- Programmed to die after a certain number of divisions or if they become damaged
- Apoptotic pathways respond to DNA damage, activity of oncogenes, lack of oxygen, etc.
- Primary way of removing transformed cells

Tumor cells:

- Able to sometimes bypass apoptosis
- Over 50% of human cancers show loss of p53 protein function, main DNA damage sensor that can induce apoptosis

Limitless replicative potential

Normal cells:

- Finite replicative potential that does not depend on external signals
- During every cycle 50-100 bp loss of telomeric DNA from the ends of every chromosome
- 60-70 divisions

Tumor cells:

- Upregulate expression of telomerase enzyme, which adds some repeating base pairs to the end of telomeric DNA
- Telomere length is above a threshold and permits unlimited multiplication (immortalization)

Sustained angiogenesis

Normal cells:

- Blood vessels supply oxygen and nutrients
- All cells within 100 μm of a capillary blood vessel
- Angiogenesis (blood vessel growth) depends on counterbalance of positive and negative signals to encourage or block blood vessel growth

Tumor cells:

- Develop their own angiogenic ability
- Activate angiogenesis (‘angiogenic switch’) by changing the balance of angiogenesis inducers and inhibitors
- Offers a therapeutic target (e.g. anti-VEGF antibodies impair neovascularization and tumor growth)

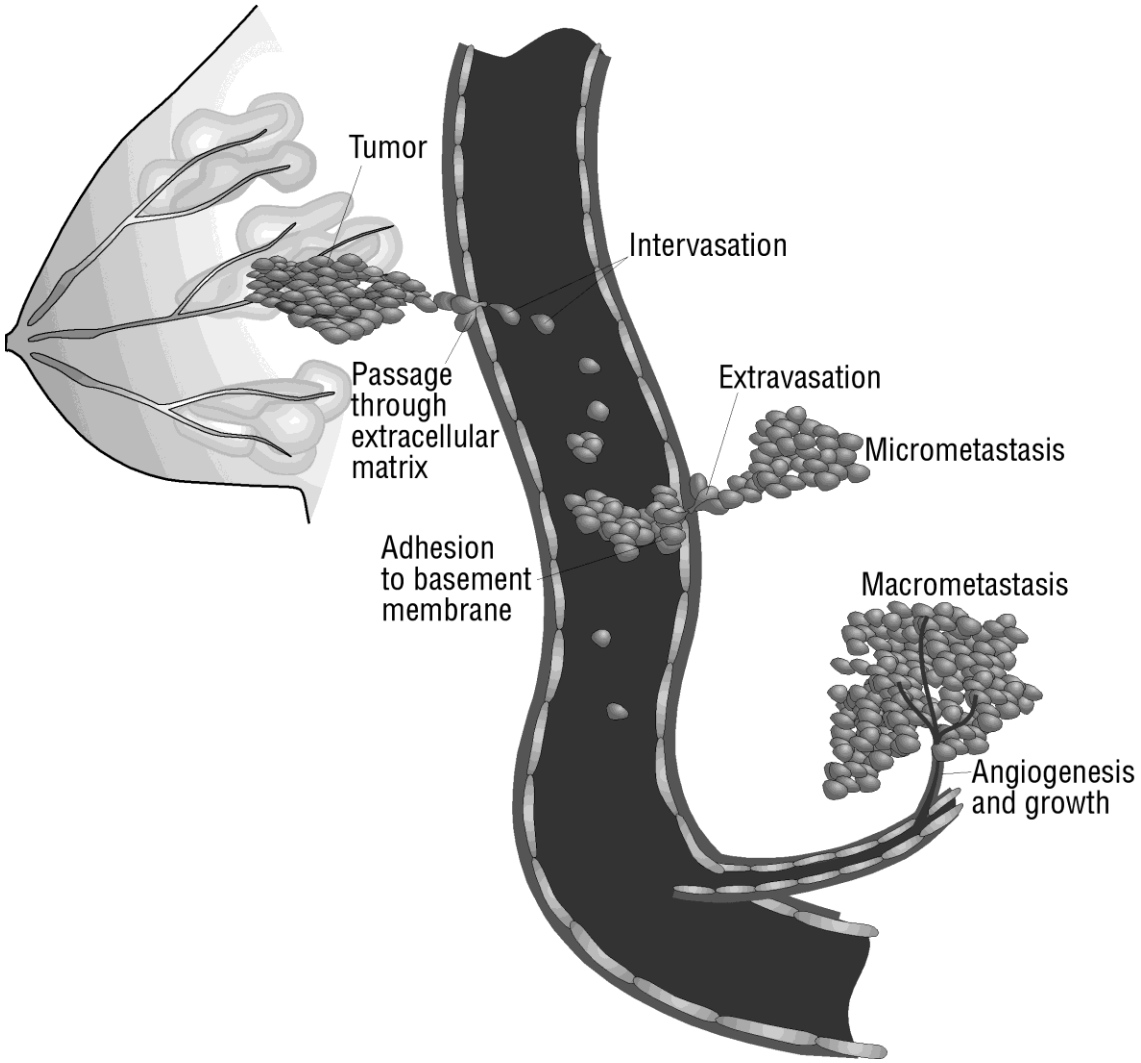
Tissue invasion and metastasis

– Overview
  - Enable cancer cells to leave primary tumor and colonize other part of body where space and nutrients are not limiting
  - Cause 90% of human cancer deaths
  - Invasion and metastasis depend on all of the other five acquired hallmark capabilities

– Characteristic changes in specific binding proteins
  - Cell-cell adhesion molecules
  - Integrins (link cells to extracellular matrix)

– Increased activity of extracellular proteases
  - Docking of active proteases on cell surface can facilitate cancer cell invasion across blood vessel walls and through normal epithelial cell layers

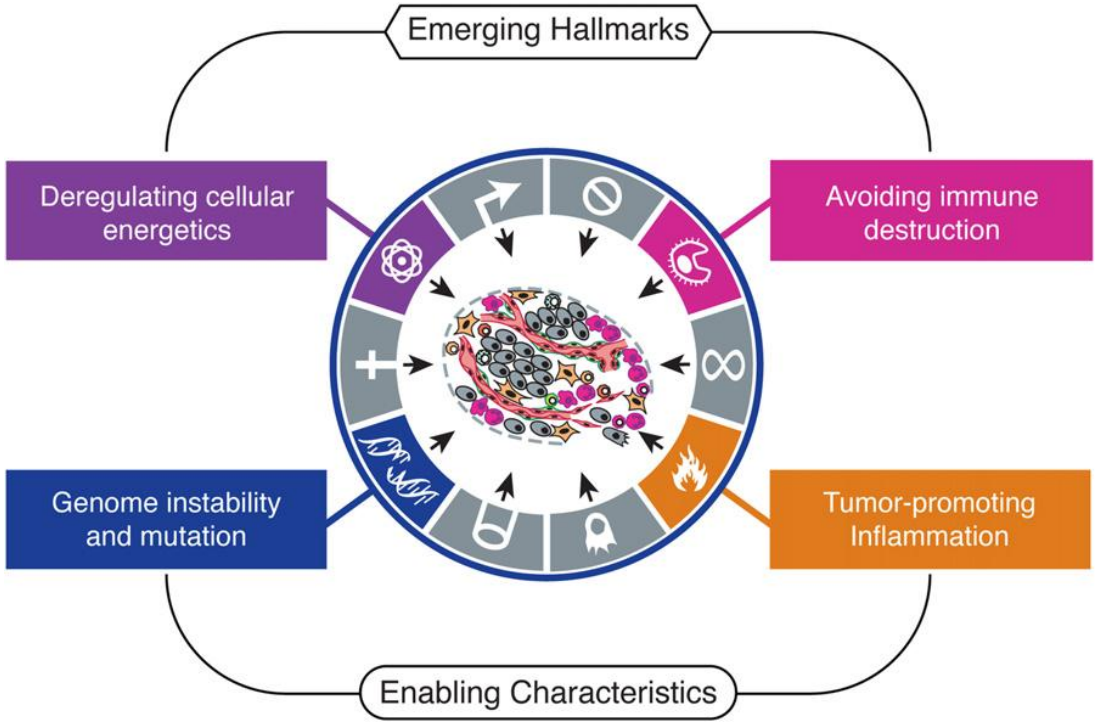
- Main feature of malignancy
- Major cause of cancer related morbidity and mortality



What is new?

In addition to six core hallmarks research in the past 10 years suggests:

- Two emerging hallmark capabilities
- Two enabling characteristics
- Significance of tumor microenvironment



Genome instability and mutation

- Most capabilities are acquired through changes in the genome of the cancer cells or regulation of gene expression
- In normal cells DNA monitoring and repair systems ensure that mutations are rare events; spontaneous mutations leading to development of tumor would be very unlikely during the human life span
- Loss of function of maintenance and repair systems (e.g. p53 protein) leads to genome instability and accumulation of mutations during cancerogenesis

Tumor-promoting inflammation

- Inflammation by innate immune cells has been regarded as a defense mechanism against cancer cells
- Tumor-associated inflammatory responses can actually stimulate tumor growth by supplying: growth factors to the tumor microenvironment, survival factors that limit apoptosis, enzymes that promote angiogenesis, etc.
- Inflammation is in some cases present at the early stages of cancerogenesis; inflammatory cells can release mutagens and thus accelerate the further development of malignancy

Reprogramming energy metabolism
(‘metabolic switch’)

- Increased cell proliferation requires increased energy production

- Normal cells produce most of their energy through aerobic degradation of glucose (glucose $\longrightarrow$ pyruvate $\longrightarrow$ CO_2)

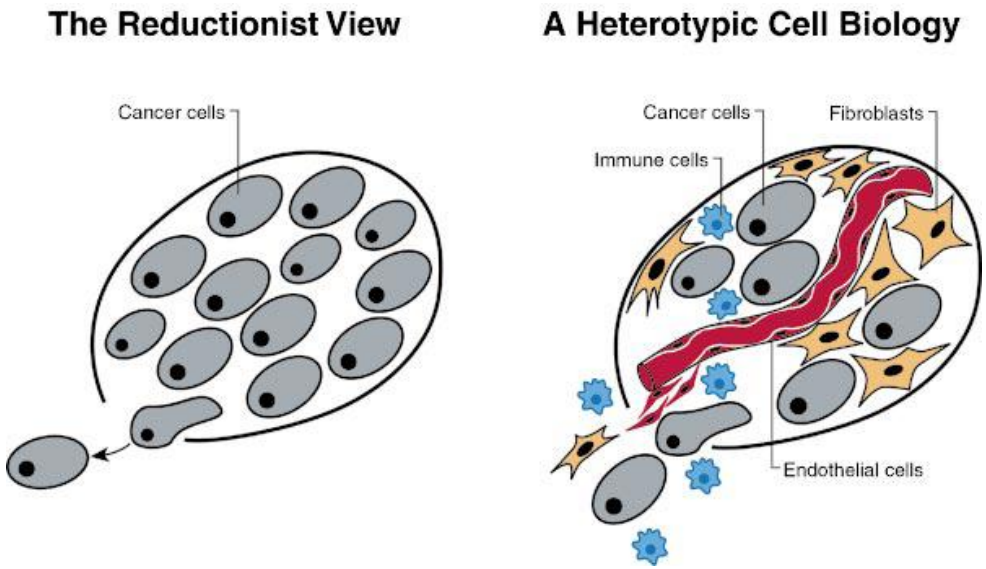
- Cancer cells limit metabolism to less efficient glycolysis (glucose $\longrightarrow$ pyruvate $\longrightarrow$ lactate) even in the presence of oxygen:
  - Supporting possible hypoxic conditions in the tumor
  - Using glycolytic intermediates in biosynthetic pathways necessary for production of macromolecules in the fast proliferating cells

- Mixed populations of cells in some tumors; symbiosis of lactate-producing and lactate-utilizing cells

Evading immune destruction

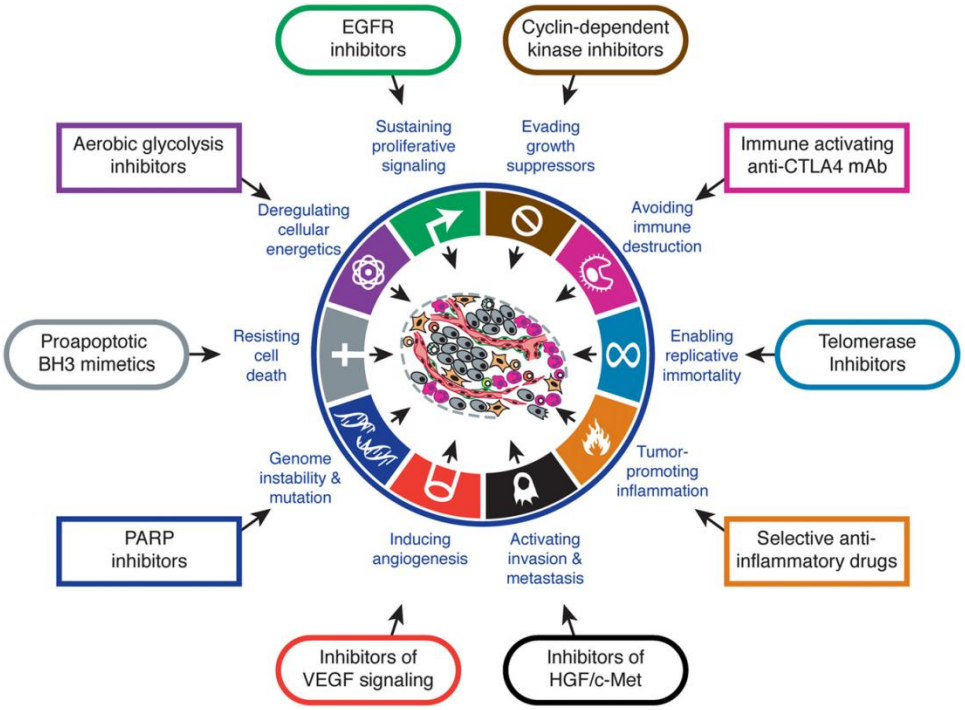
- Immune system is capable of recognizing and eliminating cancer cells
- Tumors develop more often in immunodeficient mice and immunocompromised humans
- Cancer cells possibly evade immunological destruction by disabling some cells of the immune system

Tumor microenvironment



- Tumors are complex organs, not a collection of cancer cells
- Cancer cells can recruit normal cells in their surroundings as active collaborators of their neoplastic growth; e.g. inflammatory cells attracted to sites of tumor growth may promote, rather than eliminate cancer cells
- Tumor = cancer cells + stromal cells + extracellular matrix (ECM)
- Dynamic interactions between cancer and stromal cells are due to their progressive changes during the multistep transformation of normal cells to malignant cells

Therapeutic targeting



- Cancer drugs targeting single hallmark or enabling characteristics are often not efficient
- More effective therapies require selective co-targeting of multiple hallmark capabilities or enabling characteristics

Future vision

Recognition of the widespread applicability of these concepts will increasingly affect the development of new means to treat human cancer.