

Lecture 8:

Part A: Cell Signaling

Aims:

- Examples of how cells communicate with each other to get the job done!

Alberts *et al*, Chap 16

Background

Cells respond to their environment.

Organelles within a cell respond to their environment within the cell.

Each organelle has a defined function for regular cell metabolism

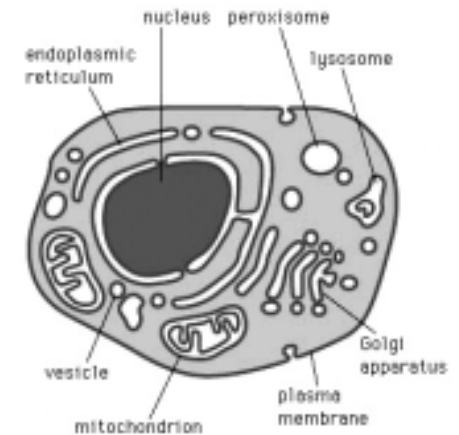
-> cell survival

Responses to

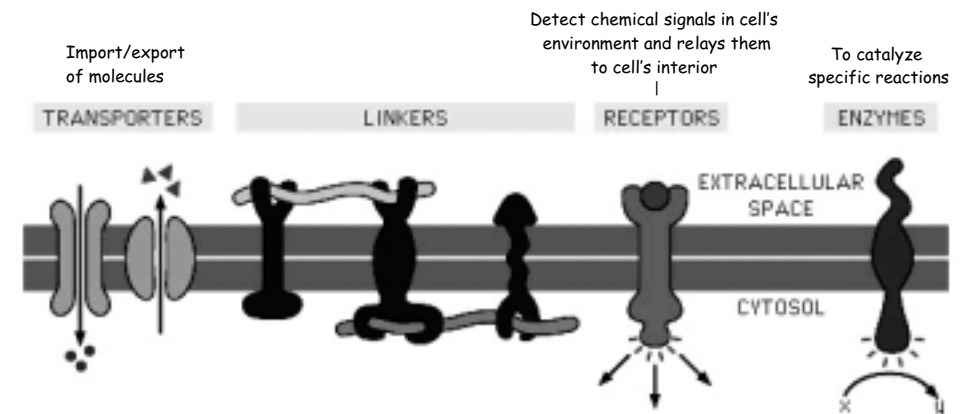
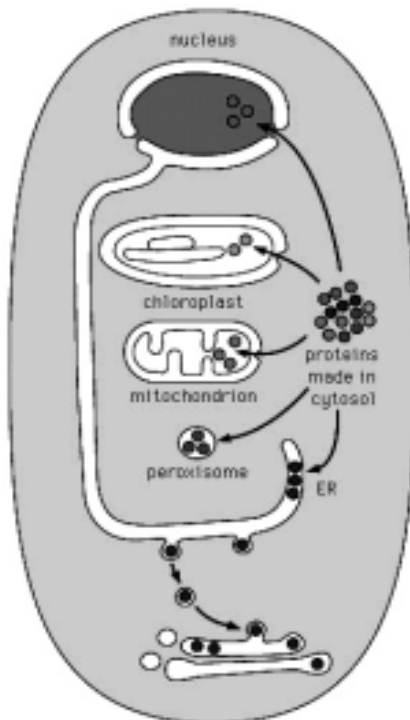
- electrochemical gradients
- cell surface proteins
- growth factors, hormones
- light
- touch
- neurotransmitters
-and more!

Regulation of cell metabolism:

- transcriptional level
- proteins



Proteins



General Principles of Cell Signaling

Signal Transduction:

Conversion of a signal from one physical or chemical form to another.

The process by which a cell produces a response to an extracellular signal.

Successive steps along this pathway.

Cell X (Signaling cell) produces a particular molecule
which is detected by

Cell Y by means of a receptor (protein Cell Y that receives signal)

Then the incoming extracellular signal is converted
to intracellular signals that direct cell behavior.

So a signal is received and transduced -> referred to as cell signaling

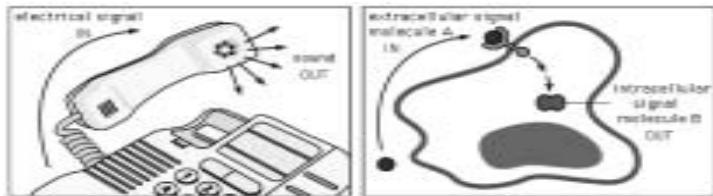


Fig 16.2

Forms of Cell Signaling:

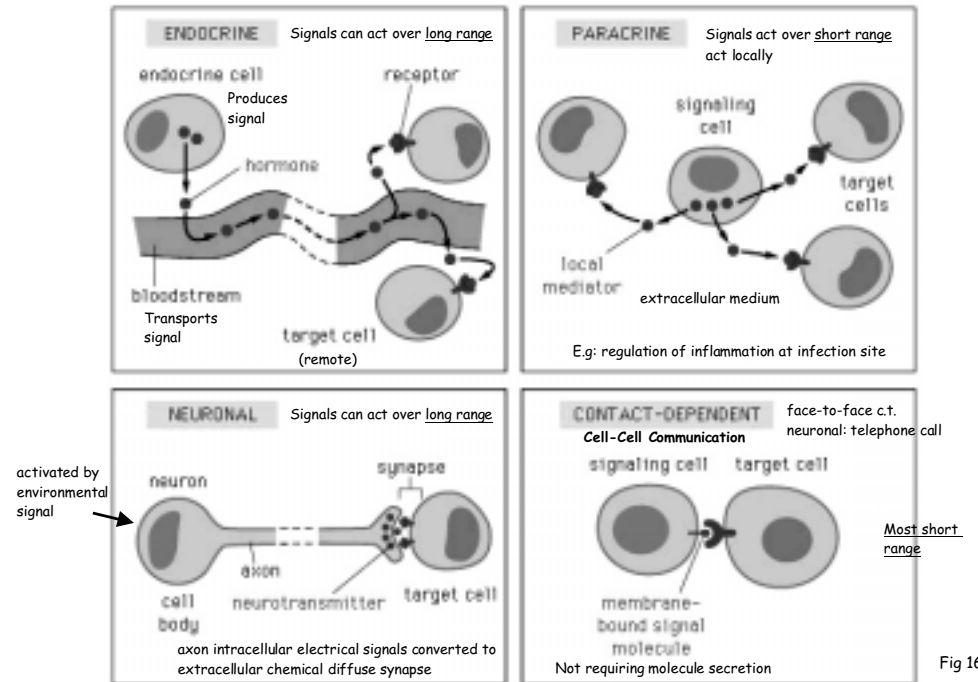


Fig 16.3

A Neuron

Arrow indicates direction in which signals are conveyed,
away from the cell body.

Dendrites receive signals from axons of other neurons.

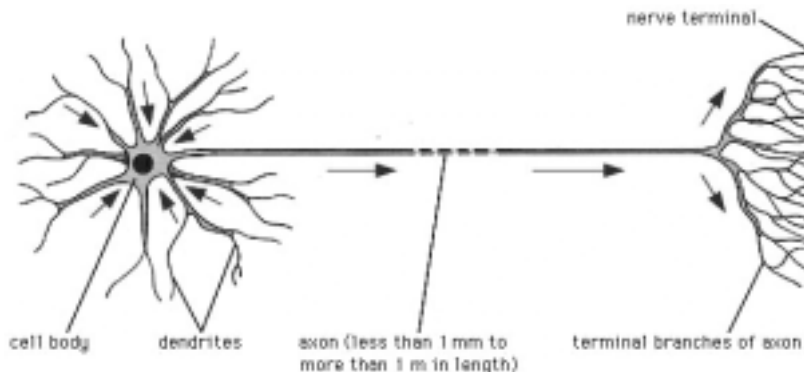


Table 15-1 Some Examples of Signal Molecules

Signal Molecule	Site of Origin	Chemical Nature	Some Actions
Hormones			
<i>Adrenaline</i>	adrenal gland	derivative of the amino acid tyrosine	increase blood pressure, heart rate, and metabolism
<i>Cortisol</i>	adrenal gland	steroid (derivative of cholesterol)	affect metabolism of proteins, carbohydrates, and lipids in most tissues
<i>Estrogen</i>	ovary	steroid (derivative of cholesterol)	influence and maintain secondary female sexual characteristics
<i>Glucagon</i>	alpha cells of pancreas	peptide	stimulates glucose synthesis, glycogen breakdown, and lipid breakdown in, e.g., liver and fat cells
<i>Insulin</i>	beta cells of pancreas	protein	stimulates glucose uptake, protein synthesis, and lipid synthesis in, e.g., liver cells
<i>Testosterone</i>	testis	steroid (derivative of cholesterol)	influence and maintain secondary male sexual characteristics
<i>Thyroid hormone (thyroxine)</i>	thyroid gland	derivative of the amino acid tyrosine	stimulates metabolism of many cell types
Local Mediators			
<i>Epidermal growth factor (EGF)</i>	various cells	protein	stimulates epidermal and many other cell types to proliferate
<i>Fibroblast growth factor (FGF)</i>	various cells, including blood platelets	protein	stimulates many cell types to proliferate
<i>Nerve growth factor (NGF)</i>	various neuronal tissues	protein	promotes survival of certain classes of neurons, promotes growth of their axons
<i>Histamine</i>	mast cells	derivative of the amino acid histidine	cause blood vessels to dilate and become leaky, helping to cause inflammation
<i>Nitric oxide (NO)</i>	nerve cells; endothelial cells lining blood vessels	dissolved gas	cause smooth muscle cells to relax; regulate nerve cell activity
Neurotransmitters			
<i>Acetylcholine</i>	nerve terminals	derivative of choline	excitatory neurotransmitter at many nerve-muscle synapses and in central nervous system
<i>gamma-Amino-butyric acid (GABA)</i>	nerve terminals	derivative of the amino acid glutamic acid	inhibitory neurotransmitter to control nervous system
Contact-dependent Signaling Molecules			
<i>Delta</i>	prospective neurons; various other embryonic cell types	transmembrane protein	inhibits neighboring cells from becoming specialized in some way as signaling cell

Each cell responds to a limited set of signals

- Each cell has a specific function -> specialized cells.
- The same signal molecule can induce different responses in different target cells.
- Different receptors generate different intracellular signals.

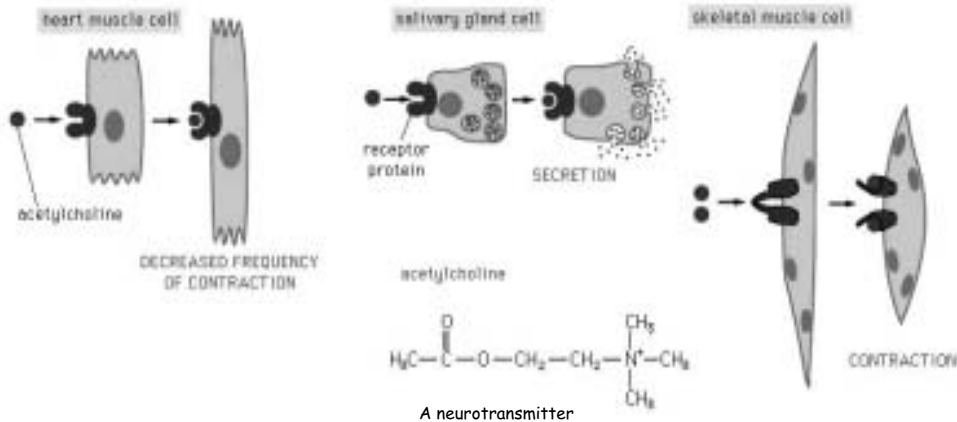
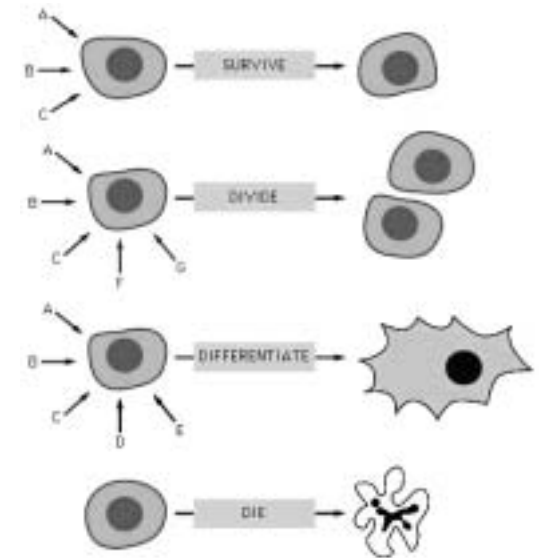


Fig 16.5

The presence of one signal modifies the responses to another -> an intercellular relay system for different signals interacting

One combination of signals may enable a cell to survive,
another to differentiate,
another to divide,
in absence of signal -> death.



Receptors relay signals via intracellular signaling pathways

Signaling cascades -> Information flow

Function to:

1. **Transfer signal** from point at which it's received on cell to cell machinery that will make the response
2. **Transform the signal** into a molecular form able to stimulate that response
3. **Amplify the signal** received -> stronger signal
so few extracellular signal molecules are enough to initiate a large response
4. **Distribute signal** to influence several processes in parallel.
The signal can diverge at any step in pathway and relay to a different intracellular targets -> complex response
5. **Signal transmission can be modulated** (influenced): each stop of signaling cascade is open to interference by other factors so transmission of signal can be modulated dependent on conditions inside or outside of cell

An intracellular signaling cascade:

A receptor protein on cell surface transduces an extracellular signal into an intracellular signal, initiating a signaling cascade of many steps

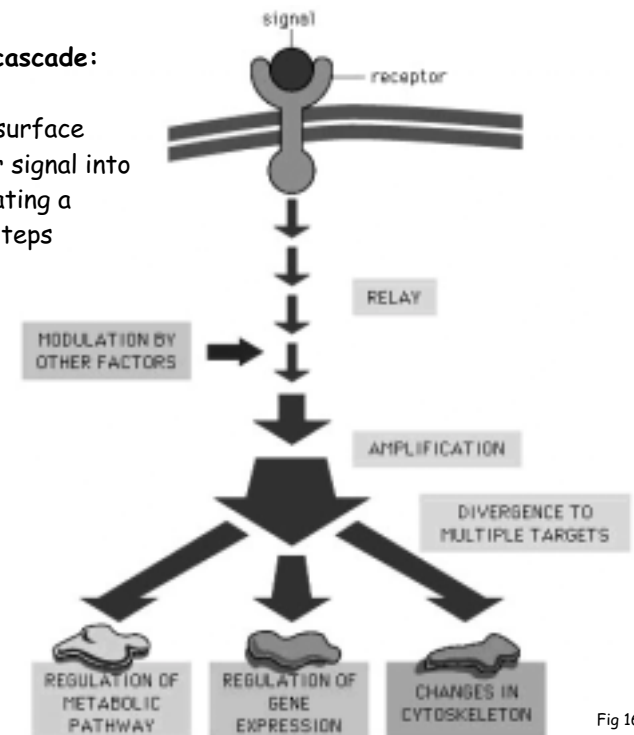


Fig 16.8

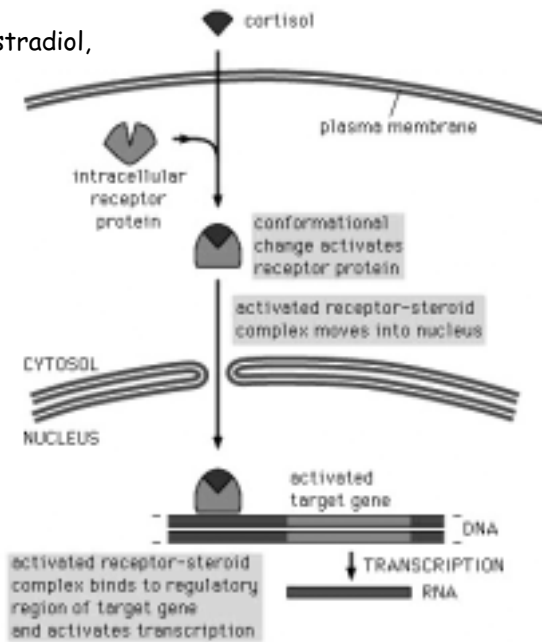
Some signal molecules can cross the plasma membrane

Small hydrophobic molecules

(E.g steroid hormones: cortisol, estradiol, testosterone, thyroid hormone)

diffuse across membrane and bind intracellular receptors

(e.g in cytosol or nucleus)



Cortisol activates a gene regulatory protein

Fig 16.12

Cell-surface receptor binding of signal molecule causes change in receptor, which relays message without the need for the signal molecule itself to enter the cell

Example: Nitric oxide enters cells and activates enzymes directly

triggers smooth muscle relaxation in blood vessel wall, reducing workload on heart & consequently the O_2 requirement of heart muscle

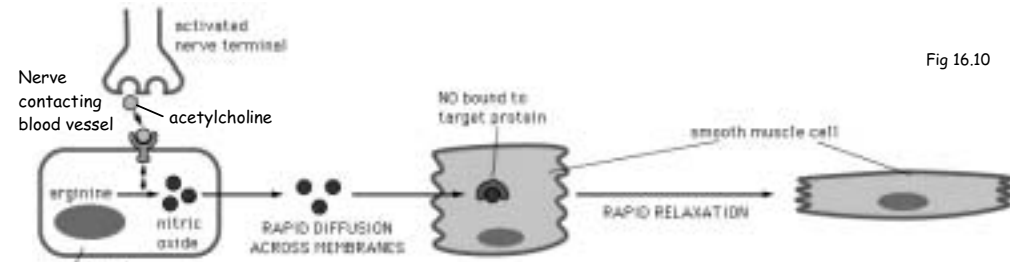
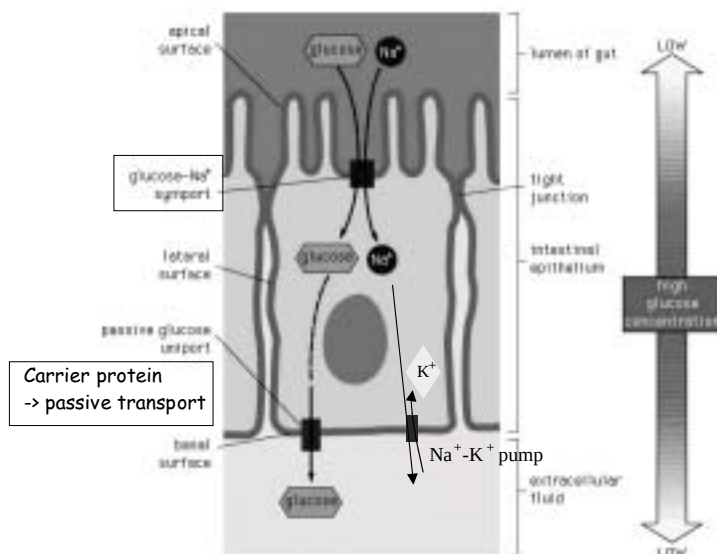


Fig 16.10

Acetylcholine released by nerve terminals in blood vessel wall activates endothelial cells lining blood vessel to make and release NO which diffuses out of cell and into adjacent smooth muscle cell.

Example: Two types of glucose carriers enable gut epithelial cells to transfer glucose across the gut lining



Intracellular signaling cascades act as a series of molecular switches

E.g Enzyme-linked and G-linked receptors

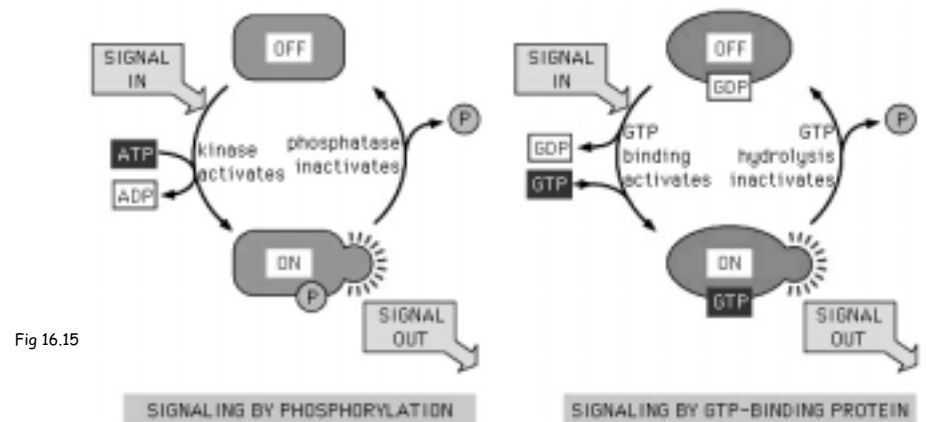


Fig 16.15

Intracellular signaling prot is activated by the addition of a phosphate group & inactivated by the removal of P.

Signaling prot is induced to exchange its bound GDP to GTP. Hydrolysis of bound GTP to GDP switches prot off.

Response to extracellular signal may be fast or slow

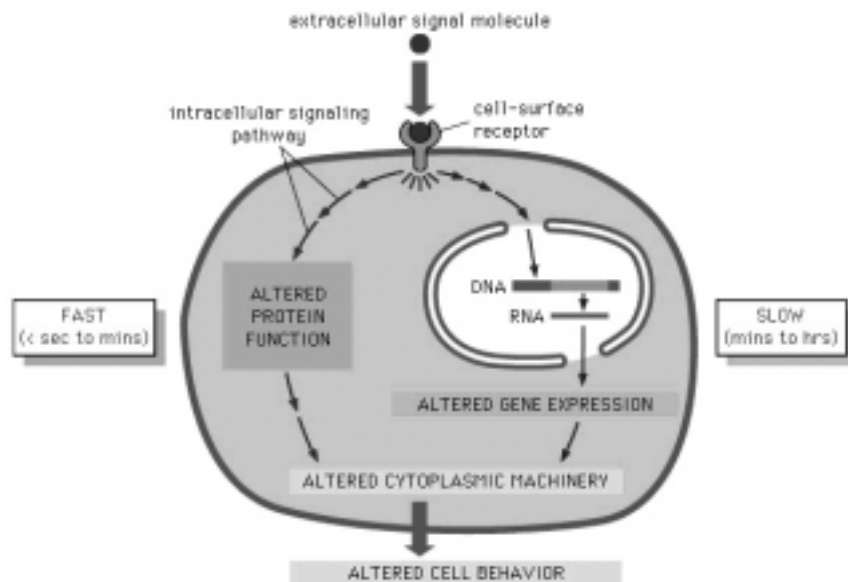


Fig 16.23

Protein kinase networks integrate information to control complex cell behavior:

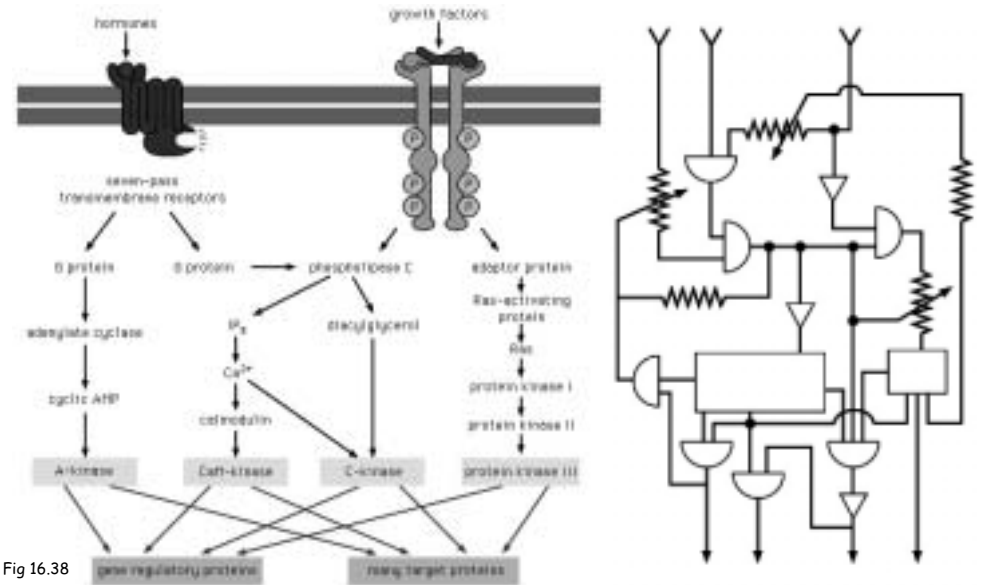


Fig 16.38

Four Parallel intracellular signaling pathways & the connections between them

Network of integrating devices

General Principles of Cell Signaling

- Signals can act over long or short range
- Each cell responds to a limited set of signals
- Receptors relay signals via intracellular signaling pathways
- Intracellular signaling cascades act as a series of molecular switches
- Some signal molecules can cross the plasma membrane
- There are three main classes of cell-surface receptors -
 - Enzyme-linked receptors
 - G Protein Receptors
 - Ion Channel Receptor
- Networks integrate information to control complex cell behaviors

Points for drug targeting

Part B:

Cell Division, Cell Cycle, Cell Death & Cell Differentiation

Aims:

- **Cell Division:** Overview
- **Cell Cycle:** $G_1/S/G_2/M$
 - Cell cycle control
 - Cancer
- **Cell Death:** Apoptosis, Necrosis, Opsonization
- **Cell Differentiation:**
 - Cell Types; Tissues
 - Stem cells

Cell Division

Cell Cycle: cycle of duplication of cell contents and then division into two
Involves orderly sequence of events:

1. chromosome replication, cell growth;
2. chromosome separation;
3. cell division.

Cell division
in bacteria

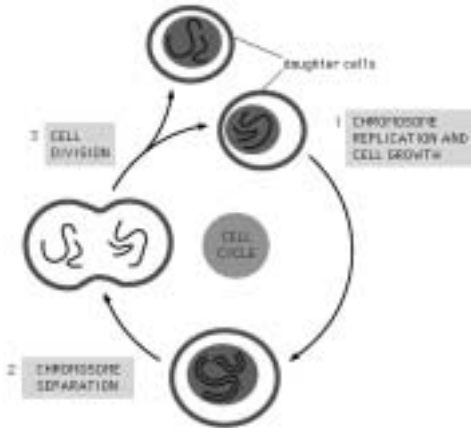
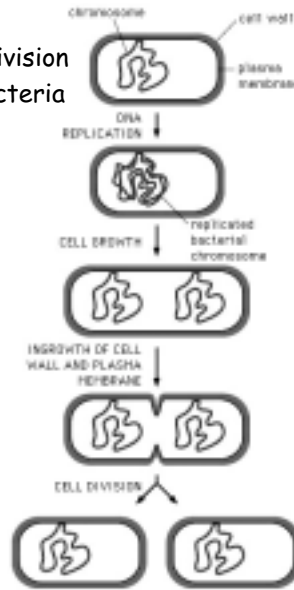


Fig 18.1

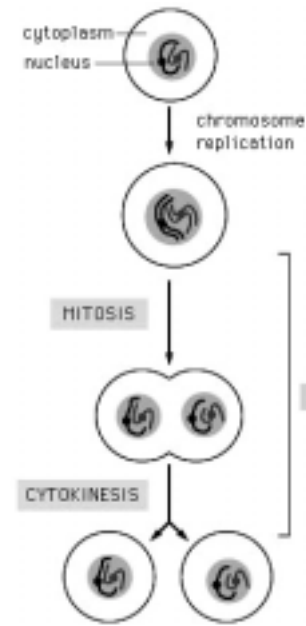
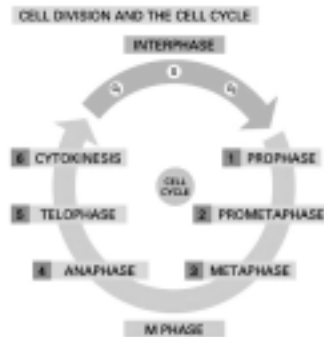
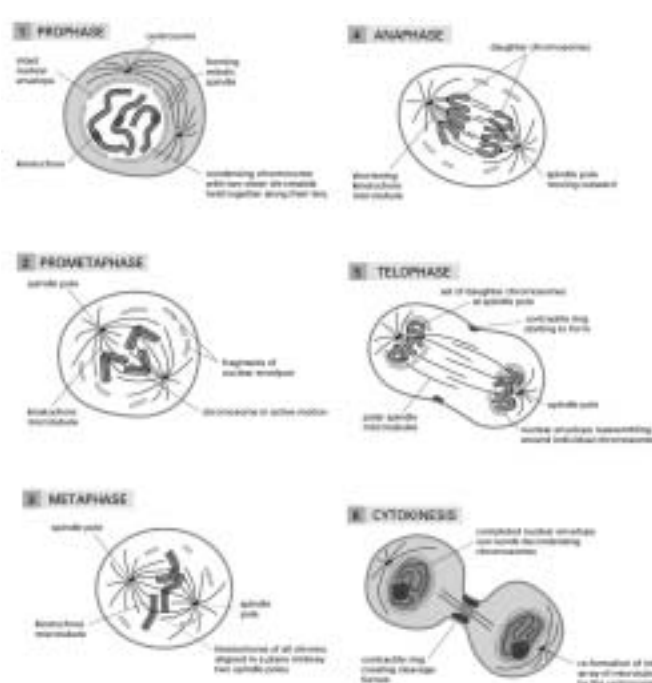


Fig 19.1

Before mitosis begins each chromosome has been replicated and consists of two identical chromatids (sister chromatids) joined together by proteins (kinetochores)

During mitosis: these proteins are cleaved, sister chromatids split to become independent sister chromosomes, which are pulled to opposite poles of the cell by the mitotic spindle



Before mitosis begins each chromosome is replicated & consists of two identical chromatids joined together by prots

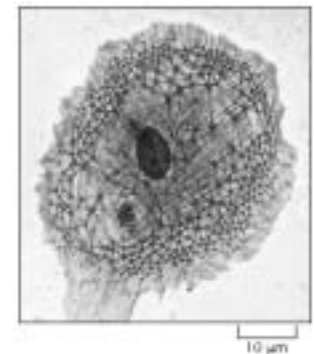
During mitosis: these prots are cleaved, sister chromatids split, now independent sister chromosomes are pulled to opposite poles of cell by mitotic spindle

Cytoskeleton

- Intricate network of protein filaments extending throughout cytoplasm
- shape
- directed movement
- "bones" and "muscle" of the cell

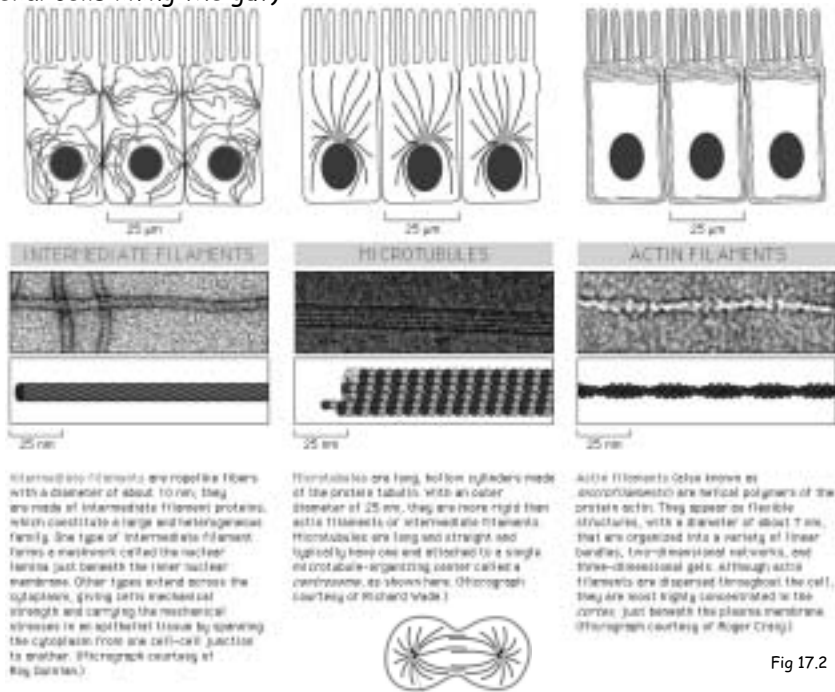
Important in:

- Crawling on surface
- Contraction of muscle cells
- Changes in shape during embryo development
- Cell division: segregation of chromosomes into the two daughter cells in mitosis



Fibroblast stained
with coomassie blue

The three types of protein filaments that form the cytoskeleton
(Cells: epithelial cells lining the gut)



Cell cycle duration varies from one cell type to another

For example:	<u>Cell type</u>	<u>Cell-cycle times</u>
	Early frog embryo cells	30 mins
	Yeast cells	1.5 - 3 hrs
	Intestinal epithelial cells	~12 hrs
	Mammalian Fibroblasts in culture	~20 hrs
	Human liver cells	~1 year

The eucaryotic cell cycle is divided into four phases:

M, G₁, S, G₂

Four phases of eucaryotic cell cycle:

- During **interphase** the cell grows
Transcription, translation, grow in mass
- During **M phase** growth stops,
nucleus divides (mitosis) then
cell divides (cytokinesis)

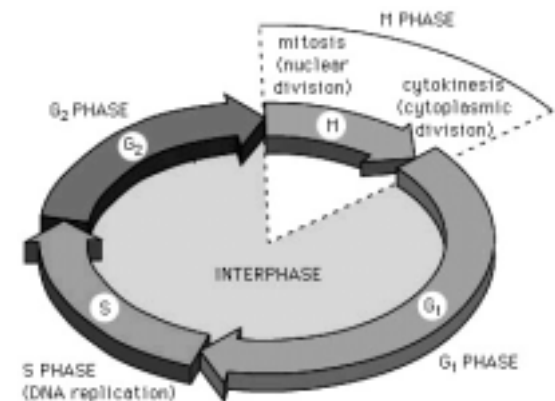
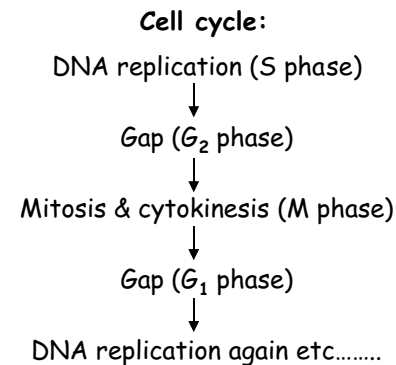
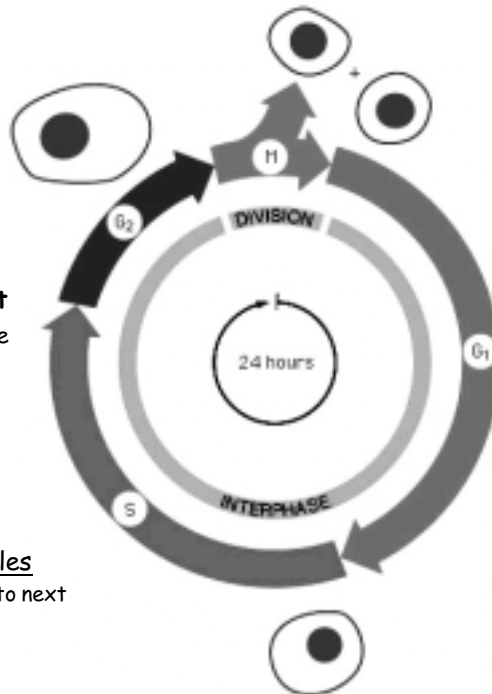
Interphase:

period b/w one M phase and the next
- made up of 3 remaining phases of cell cycle

- **S phase:** replication of DNA
- **G₁** is gap between M and S phases
- **G₂** is gap between S and M phases

Together **G₁** and **G₂** give time for cell
to grow and duplicate cytoplasmic organelles
Cell decides during G phases whether to proceed to next
phase or allow time to prepare

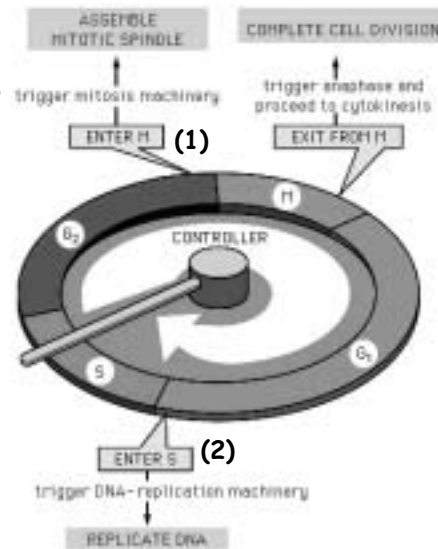
Chromosome condensation marks end of
G₂ phase; Chromosomes visible in light
microscope as long threads



Cell-control system: a control system that ensures correct progression
through the cell cycle by regulating the cell cycle machinery.
Factory: central control system that switches machinery on/off at correct
times, coordinating all activities that go to make the final product.

The Cell-Cycle Control System:

- Activates enzs & other proteins responsible for carrying out each process at the appropriate time, then it deactivates them once process is completed
- Ensures each stage of the cycle is completed before the next is begun
- Takes into account extracellular conditions
- Central role in regulation of cell numbers in tissues of the bodymalfunction -> cancer
- Controlling cell survival or death



A central control system triggers the major processes of the cell cycle

-> Checkpoints

1. G_2 checkpoint: Is all DNA replicated?
Is cell big enough?
2. G_1 Checkpoint: Is cell big enough?
Is env. favorable?
Is DNA damaged?

The Cell-cycle control system is based on cyclically activated protein kinases

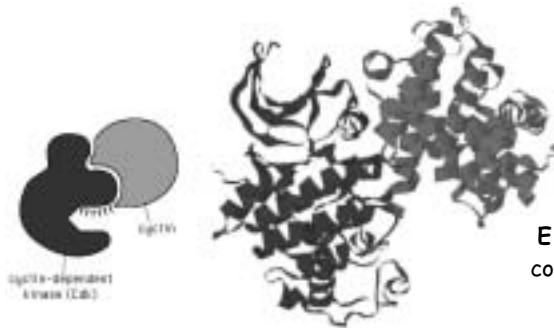
Protein kinases are activated & deactivated as needed

Cyclins: proteins that bind to cell-cycle kinases before the kinases can become enzymatically active a regulatory protein

Thus kinases of the cell-cycle control system are known as

Cyclin-dependent protein kinases (Cdks)

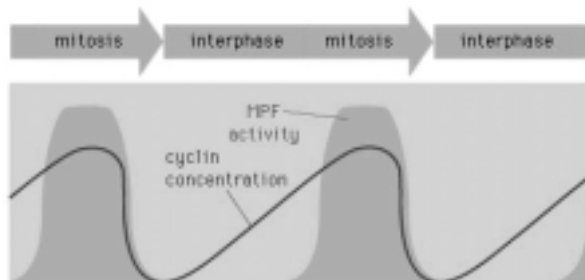
"Cyclins" because their concentrations vary in a cyclic fashion during cell cycle



Cyclin-Cdk complex

E.g M-phase promoting factor kinase complexed to specific cyclin
-> M phase

Cyclin-dependent protein kinases are regulated by the accumulation and destruction of cyclin



The rise and fall in MPF activity & cyclin:
[MPF] doesn't change during cell cycle, only its activity changes
[Cyclin] increases/falls

Fig 18.8

The activity of Cdks is further regulated by their phosphorylation & dephosphorylation

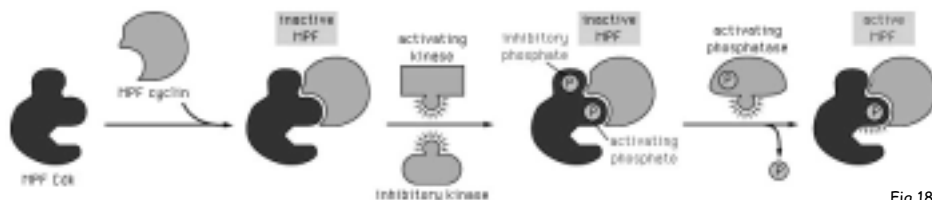
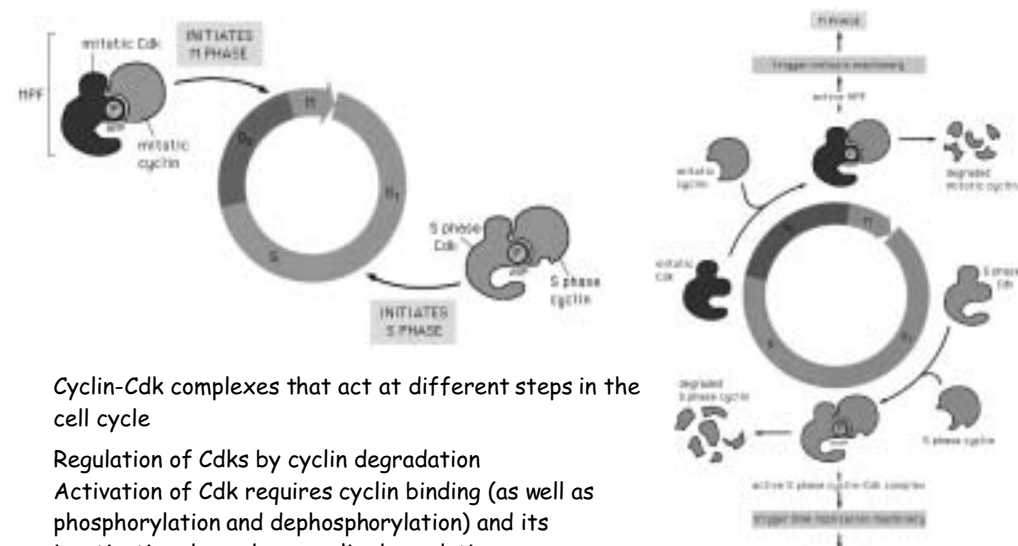


Fig 18.11

Different cycling-Cdk complexes trigger different steps in the cell cycle

Positive feedback e.g activated MPF indirectly activates more MPF



Cyclin-Cdk complexes that act at different steps in the cell cycle

Regulation of Cdks by cyclin degradation
Activation of Cdk requires cyclin binding (as well as phosphorylation and dephosphorylation) and its inactivation depends on cyclin degradation

Fig 18.13

The cell cycle can be halted in G_1 by Cdk inhibitor proteins

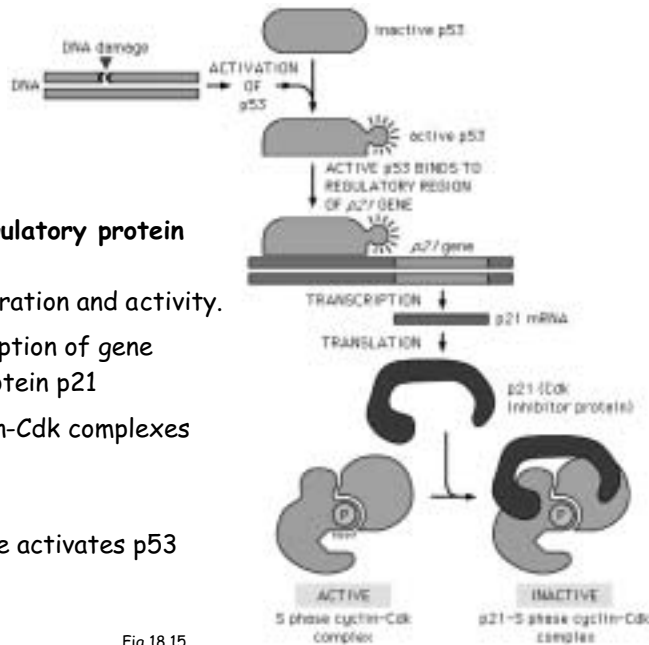


Fig 18.15

Example p53, a gene regulatory protein

DNA damaged

→ increase in p53 concentration and activity.

→ P53 stimulates transcription of gene

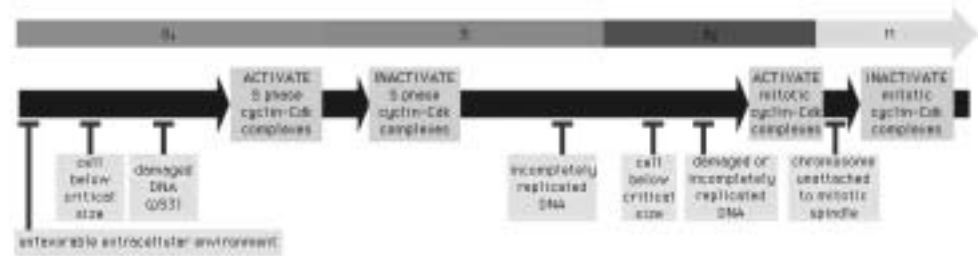
encoding Cdk inhibitor protein p21

→ P21 binds S phase cyclin-Cdk complexes and inactivates them

→ cell cycle arrests in G_1

Unknown how DNA damage activates p53

Cells can dismantle their control system and withdraw from the cell cycle



Some cell cycle checkpoints:

- Incomplete or deranged intracellular processes

- Unfavorable extracellular environment

OR....

- Cell may enter a modified G_1 state: G_0 (G zero)

- Cell-cycle control system is partly dismantled: many of Cdks & cyclins disappear

E.g Liver cells divide only once a year or two

Certain epithelial gut cells divide more than twice a day to continuously renew gut lining

Cells divide if need arises

IL2 pathway

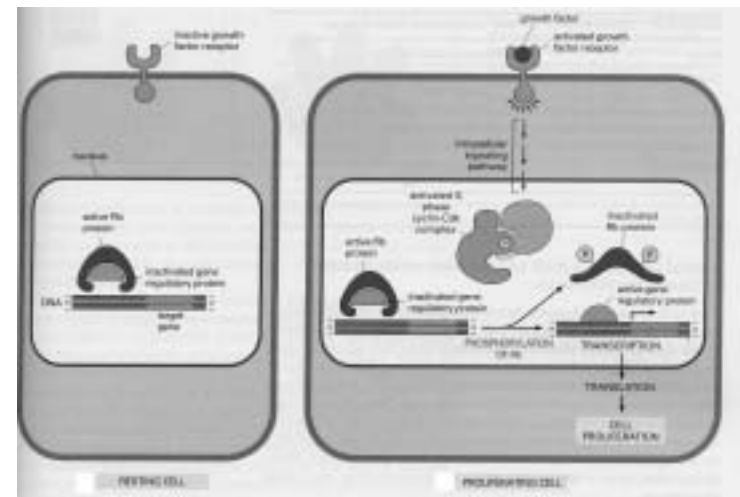
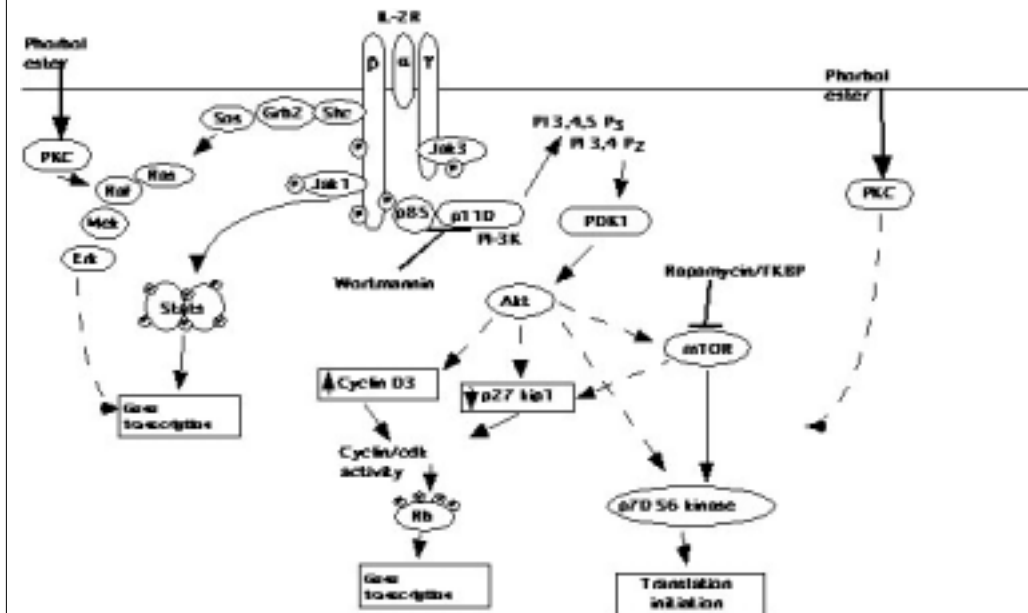


Fig 18.23

Rb protein is missing or defective → rare childhood eye tumor: retinoblastoma

Rb is abundant in nucleus of all vertebrate cells; binds to particular gene regulatory proteins preventing them from stimulating transcription of genes required for cell proliferation

Extracellular signals (e.g growth factors) stimulate cell proliferation leading to activation of G_1 cyclin-Cdk complexes that phosphorylate Rb, altering its conformation so it releases its bound gene regulatory proteins which then activate genes required for cell proliferation.

Control of cell numbers in multicellular organisms - Cell Proliferation

Cell proliferation (multiplication) depends on signals from other cells

E.g growth factors: epidermal growth factor:

- stimulates proliferation of skin cells

Cell senescence: Animal cells have a built-in limitation on the number of times they will divide

Survival factors: Signals from other cells to avoid programmed cell death

Cell Death.....

Cell death

Necrosis: death as a result of injury or toxicity

Cells swell and burst -> inflammatory response

Apoptosis: programmed cell death -> cell suicide

- Cells shrink and condense, with the involvement of specific cellular proteins
 - Collapse of the cytoskeleton
 - Distinct fragmentation of nuclear DNA
 - Altered cell surface -> initiates phagocytosis of cell
 - Phagocytosis: process by which particulate material is engulfed by a cell
- E.g macrophage (a type of white blood cell) phagocytosing bacterium (Greek, makros, big, phagein, to eat)
- Does not affect neighboring cells
 - Traditionally mediated by intracellular proteolytic (proteases: caspases) cascade
 - Proteases are activated by cleavage in response to signals inducing apoptosis
 - Suicide machinery is regulated by signals
 - killer -> activating suicide machinery
 - survival -> suppressing suicide machinery

Opsonization: death by cell budding

Vesicles form from cytosol and cell membrane resulting in cell disintegration

Necrosis.....apoptosis in tissue culture

Apoptosis in developing tissue and engulfed by neighboring cell

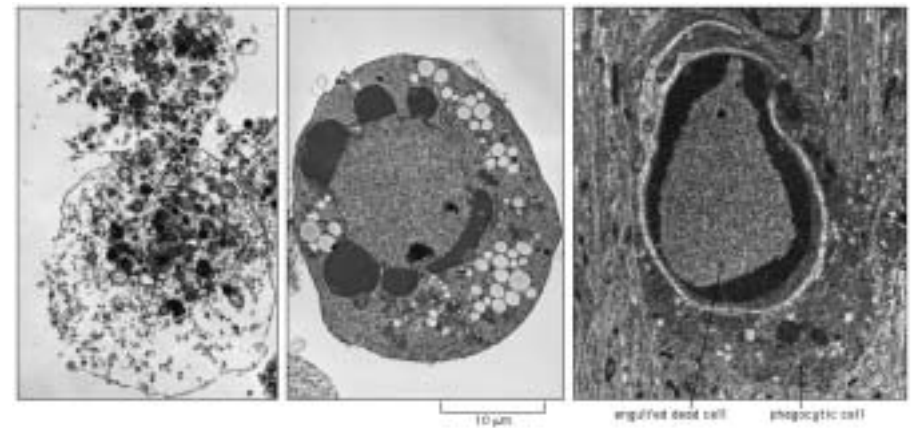


Fig 18.20

Apoptosis in developing mouse paw

Cells undergoing apoptosis (red)
→ elimination of tissue between the developing digits



Apoptosis occurs during metamorphosis of a tadpole into a frog

Cells in tadpole tail are induced to undergo apoptosis → tail is lost
Stimulated by increase in thyroid hormone in blood

Fig 18.19



Cell death: Over production of nerve cells for target cells:

cells receive insufficient amounts of survival factor to suppress suicide program
→ apoptosis

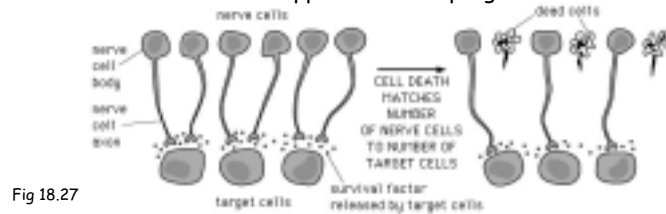
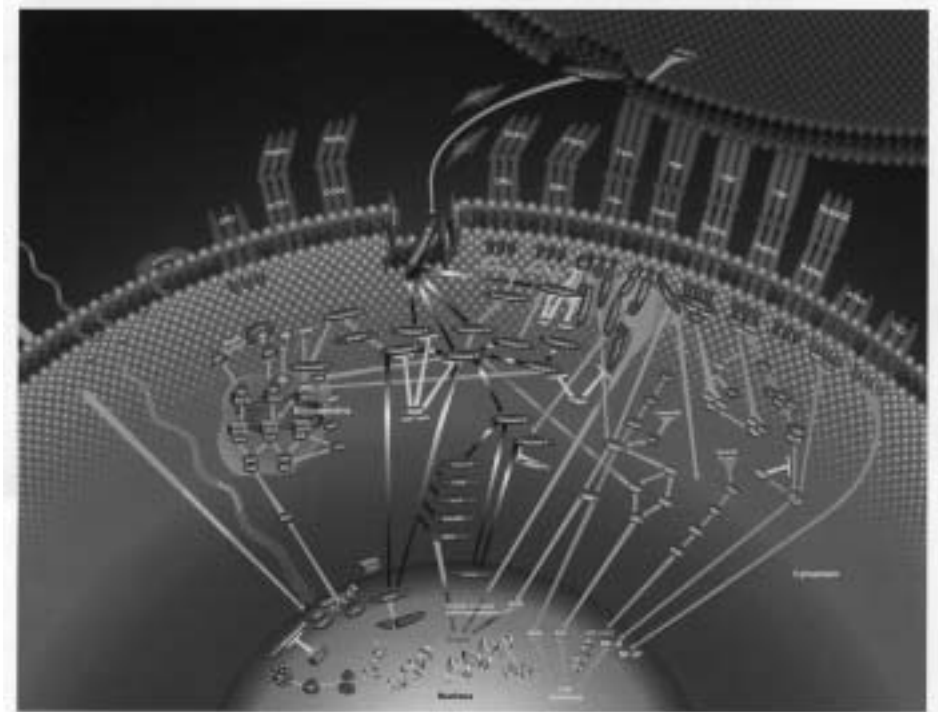


Fig 18.27



Cancer cells

Cancer: disease arising from mutant cells that escape normal controls on cell division - proliferation & survival; cells invade and colonize the tissues of the body

Faulty proliferation

Mutations at:

Proliferation genes: encode proteins that help promote cell division

→ overexpression of gene → excessive cell proliferation

mutant gene: oncogene - cancer promoting gene

normal gene: proto-oncogene (damage to one → cancer)

Antiproliferation genes: encode prots that help stop the cell cycle at checkpoints

→ excessive cell proliferation

normal gene: tumor-suppressor genes

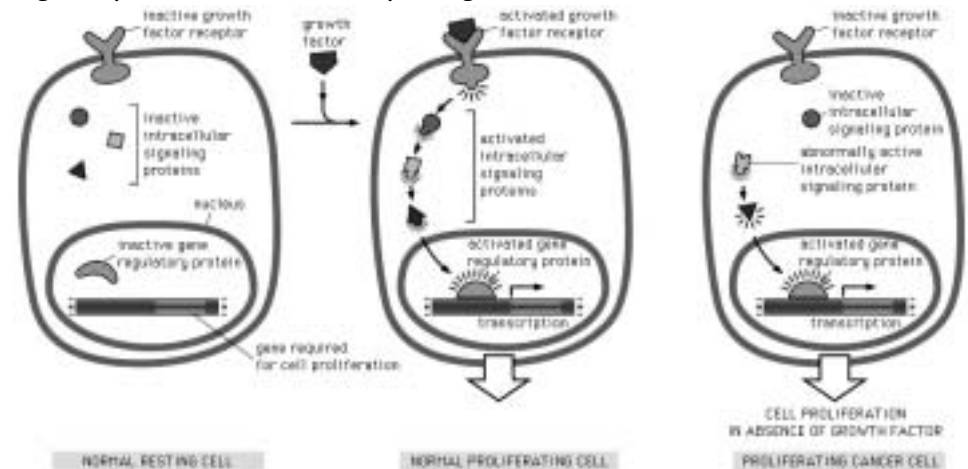
inactivation/loss of both copies of gene (in diploid cell) → cancer

Oncogenes can be created from any gene encoding prots of signaling pathways in response to cell growth factors → abnormally active protein

I.e. expressed by cells not normally expressing it; in excessive amounts; in form whose activity is uncontrolled

→ proliferation even in absence of appropriate extracellular signals

E.g Cell proliferation caused by oncogene

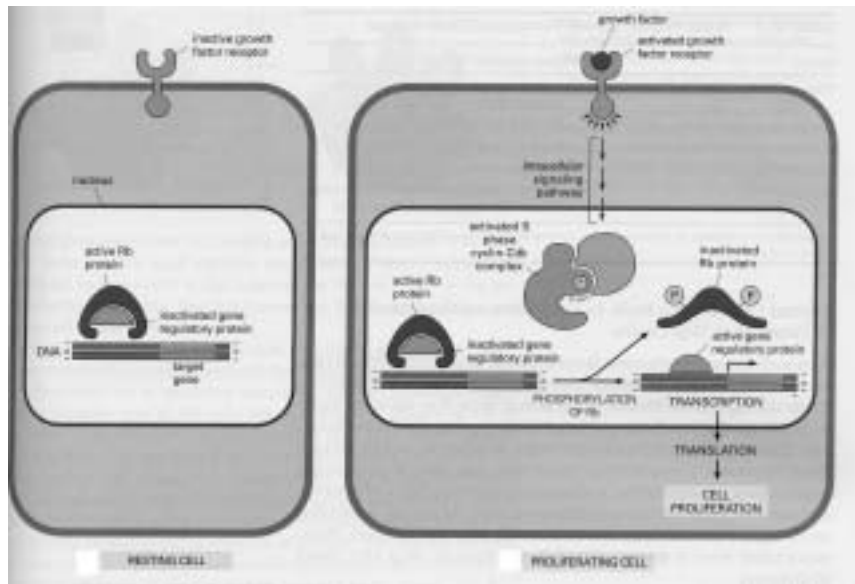


Intracellular signaling prots & genes normally activated by extracellular growth factors are inactive

Stimulation by extracellular signal (growth factor), signaling prots & genes become active
→ cell proliferates

Mutation in proto-oncogene encodes intracellular signaling prot (normally activated by extracellular signal) created oncogene - that encodes altered form of signaling prot that's active even in absence of growth factor binding

Example of tumor-suppressor gene: retinoblastoma (Rb) gene
 Inactivation of both copies of Rb gene → removes brake in cell cycle
 → cancer (especially in retina)



Cell Differentiation

Differentiated Cells & Tissues

Tissues: organized assembly of specialized cells that forms a distinct part of a plant or an animal

Differentiated cell types:
 Over 200 types of cells in the human body
 Assembled into a variety of types of tissue
 e.g. epithelial, nervous, connective, muscle

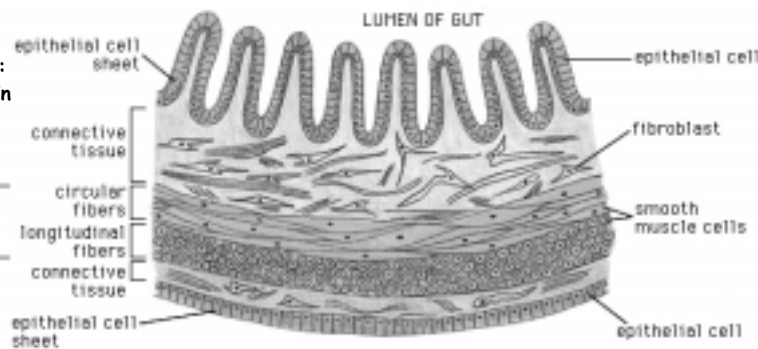
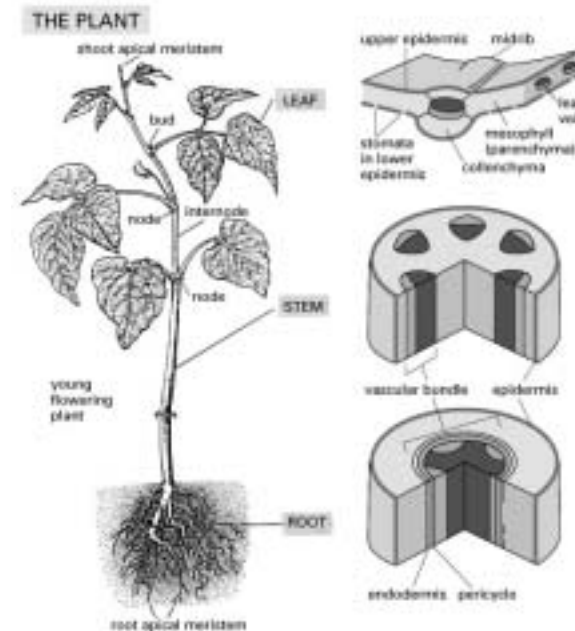


Fig 21.2

Example of the organization of cells into tissues:
 Cross-section through part of the intestinal wall of a mammal
 It's constructed from epithelial tissue, connective tissue, and muscle tissue.
 Each tissue is an organized assembly of cells held together by cell-cell adhesions, extracellular matrix, or both

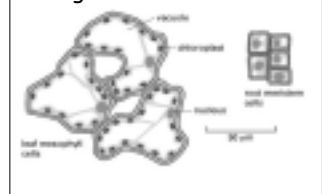
The cell types and tissues from which higher plants are constructed



3 main types of organs:
 Leaves, stems, roots

Each from 3 tissue systems:
 Ground
 Dermal
 Vascular

E.g Ground:



Different ways of packing cells together to form an epithelial sheet

The polarized organization of an epithelial sheet

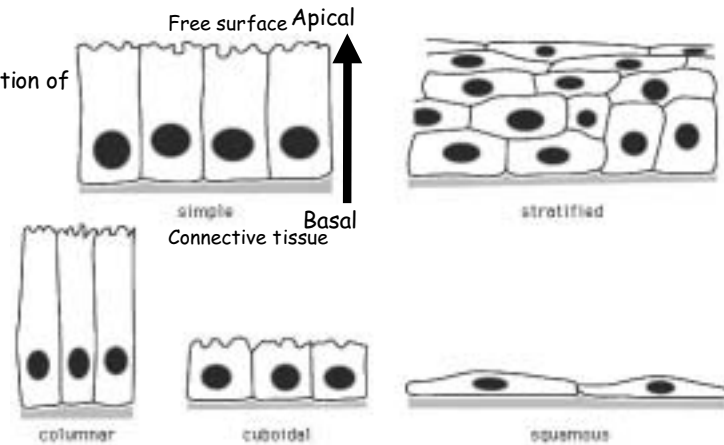


Fig 21.17

Summary of main types of cell-cell junctions found in animal epithelia

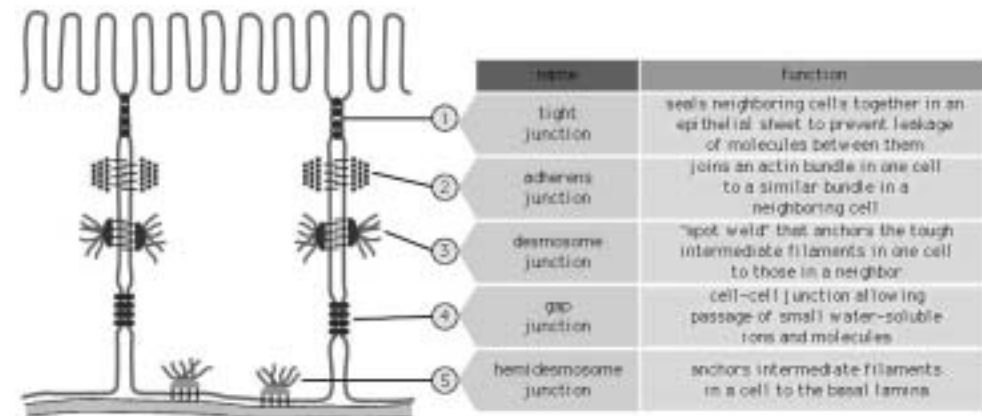
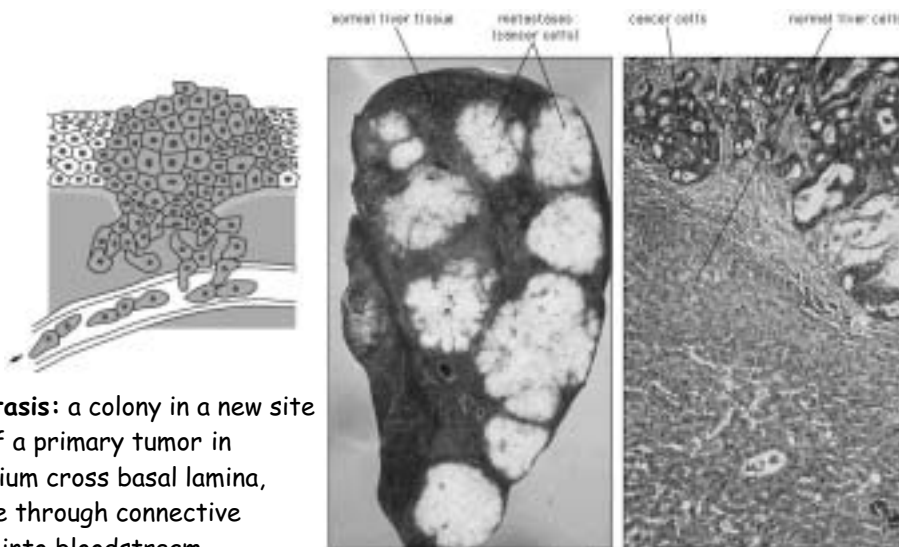


Fig 21.21

Mutations in a single dividing cell can cause it and its progeny to violate the normal controls → cancer



Metastasis: a colony in a new site
Cells of a primary tumor in epithelium cross basal lamina, migrate through connective tissue, into bloodstream

Metastases in human liver originating from primary colon tumor

Higher magnification
Stained cells

Fig 21.42

Stem Cells

Stem Cells

- Precursor cells
- Generate a continuous supply of terminally differentiated cells
 - cells unable to divide further
 - e.g red blood cells, surface epidermal cells, cells lining gut
- Retained in the corresponding tissues along with the differentiated cells
- Not terminally differentiated & can divide without limit (lifetime of animal)
- Each daughter cell has a choice:
 - it can remain a stem cell or terminally differentiate

THUS job of stem cell:
to produce cells that will carry out specialized function

Stem cells are not terminally differentiated but are determined:
- they express gene regulatory proteins to ensure their progeny will be of the appropriate types

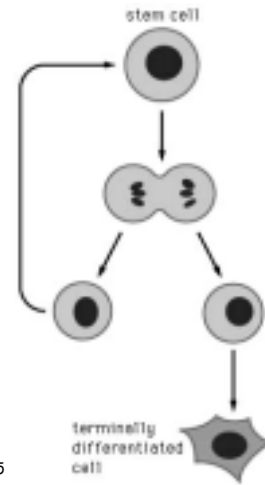


Fig 21.35

A single type of stem cell may give rise to several types of differentiated progeny

E.g Stem cells of the intestine produce:
absorptive cells, goblet cells
(mucus-secreting cells)

Hemopoiesis:
process of blood formation

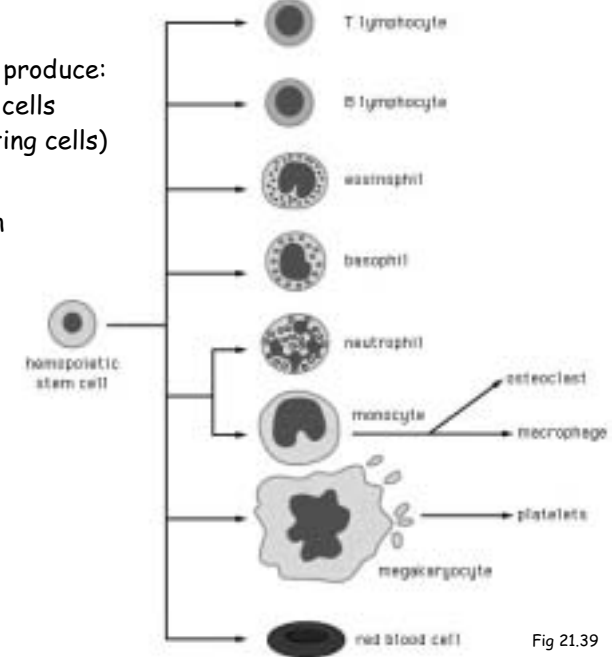
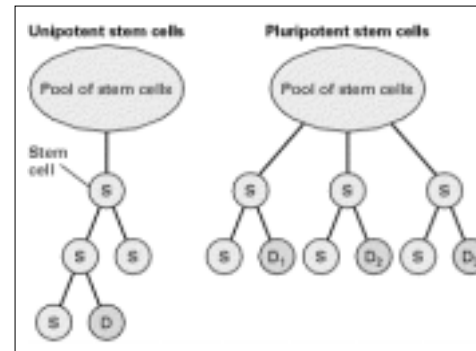
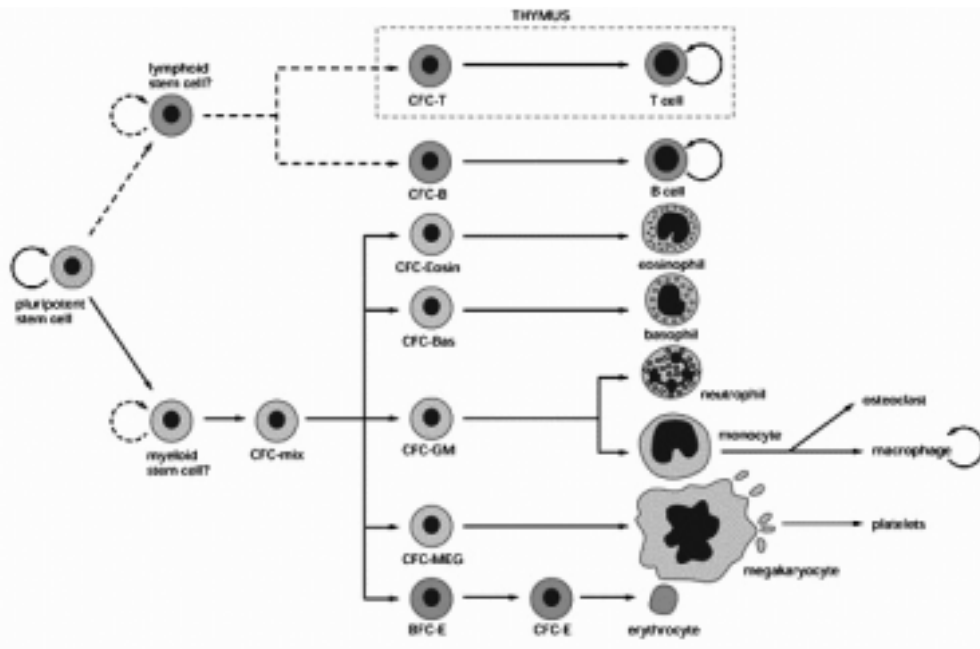
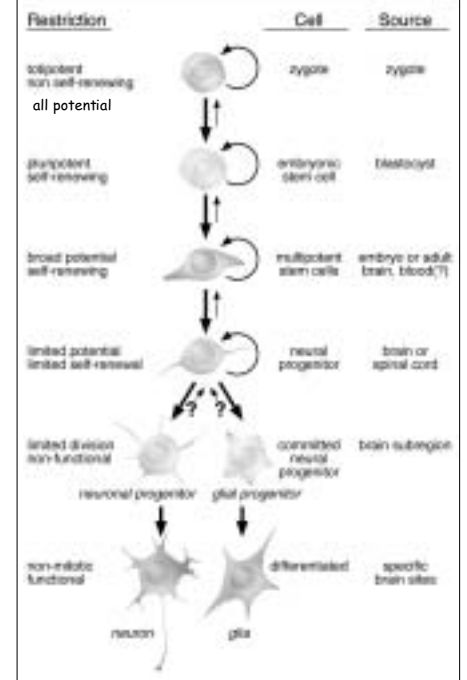


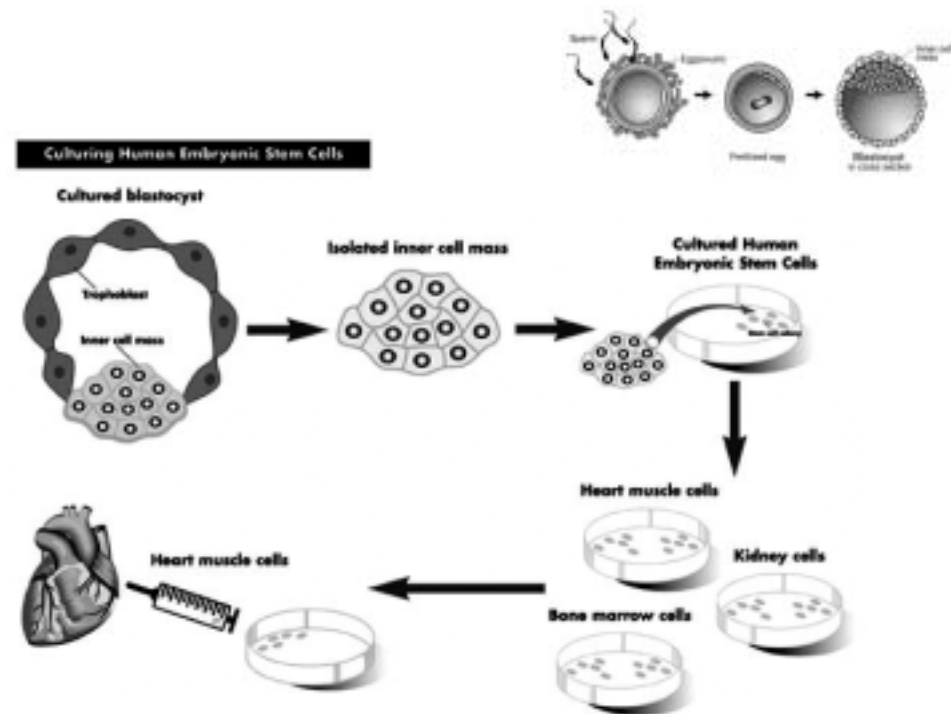
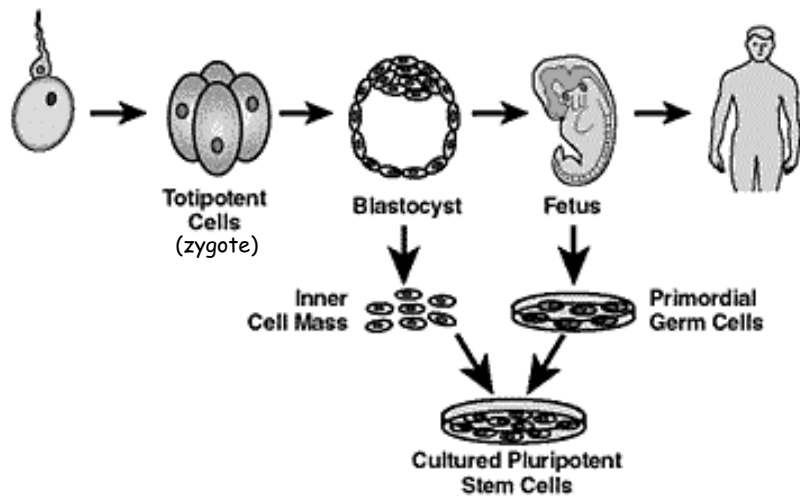
Fig 21.39

Hematopoiesis:



Potential Stem Cells with Neural Capability





Stem cells in Therapy

Stem cells can also proliferate:

Important for growth and tissue repair, normal maintenance

E.g Transfusion of a few hemopoietic stem cells into a mouse whose blood stem cells were destroyed -> repopulate animal with new blood cells and prevent anemia

In humans using bone marrow transplants as a treatment of leukemia

Many diseases are associated with death or damage to cells/tissue
e.g Alzheimers (brain cells)

Using transplant organs to replace damaged tissue:

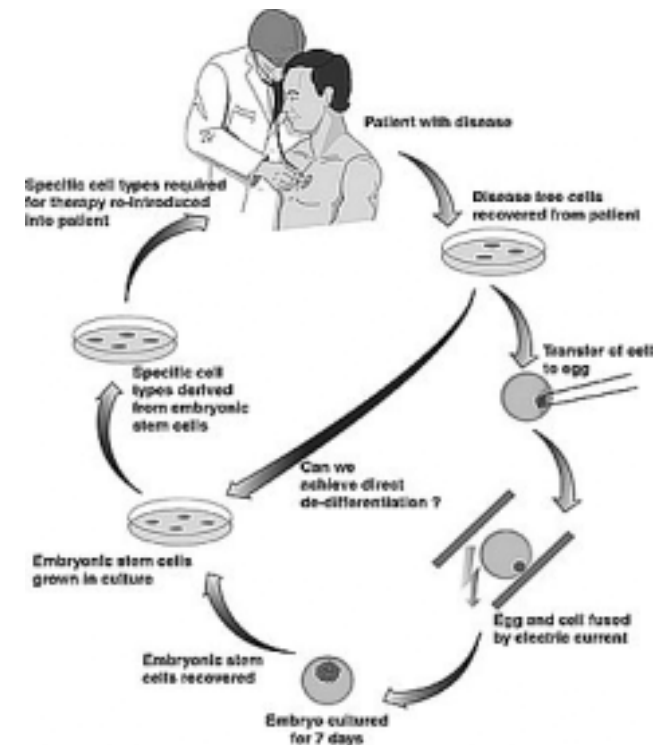
- Good - known source; surgical methods; architecture is there
- Bad - difficult to find genetically matched donor; longevity & safety
- Ugly - donor is not always known e.g Parkinson's disease
more transplants needed than there are donors

Stem cells can help:

An unspecialized cell with the **potential** to become what you like

E.g of naturally occurring stem cell -> embryonic stem cell

Stem Cell Therapy



Some examples of use of embryonic stem cells:

- partial repair of spinal cord damage in rat
- partial repair of multiple sclerosis in rat
- partial repair of parkinsons in rat
- in culture: differentiation to muscle, neurons, pancreatic cells
- studies of disease mechanisms
- repeated demonstration that embryonic stem cells can contribute to any adult cell

Ethical and Legal Issues:

- ethical status of early human embryo - frozen embryos after IVF
- human embryo construction and destruction

Use of Adult stem cells:

- bone marrow transplants
- many recent in vitro differentiation
 - induced differentiation of brain stem cells to many different cells

Technical limits:

- source, supply, rejection

Scientific issues about possible use of adult stem cells to treat disease:

Pro - know source (bone marrow)

- genetic match ok if autologous
- genetic match for bone marrow transplant

Con - unknown potency - pluripotent, multipotent, unipotent?

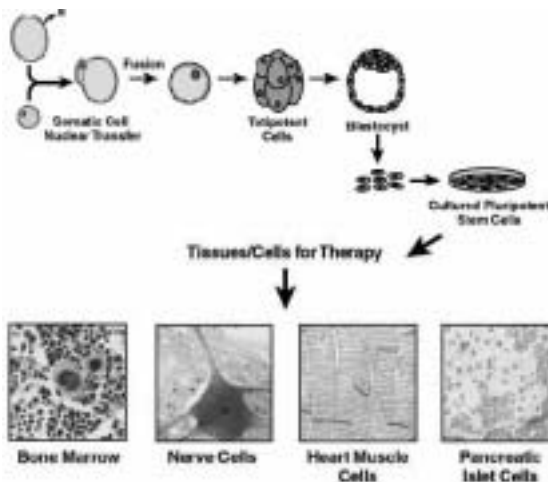
- rare - no source identified for many diseases
- substantial growth and identification issues
- longevity and safety
- new replacement methods
- genetic match/longevity

Nuclear transplant therapy:

Egg, remove nucleus, transfer nucleus from somatic cell -> differentiation

Pros - source: adult cells and oocyte; theoretical pluripotency

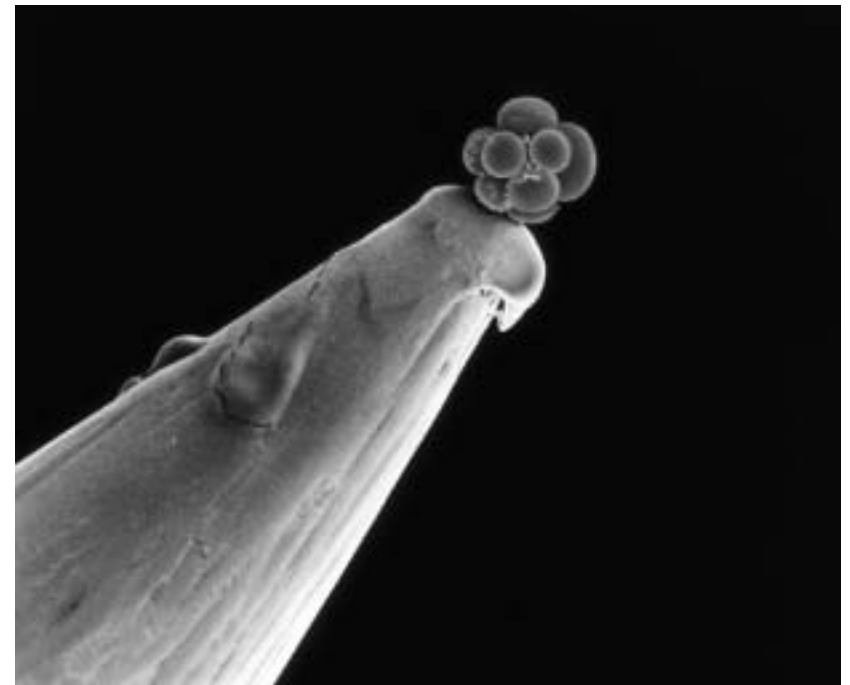
Con - surgery, longevity, architecture



Examples of NT to generate adult animal: sheep, goat, cow, pig, mouse

Efficiency poor,
Frequent abnormal development,
Success/danger in humans ??
Ethical: early embryo
Legal

Stem cell cluster



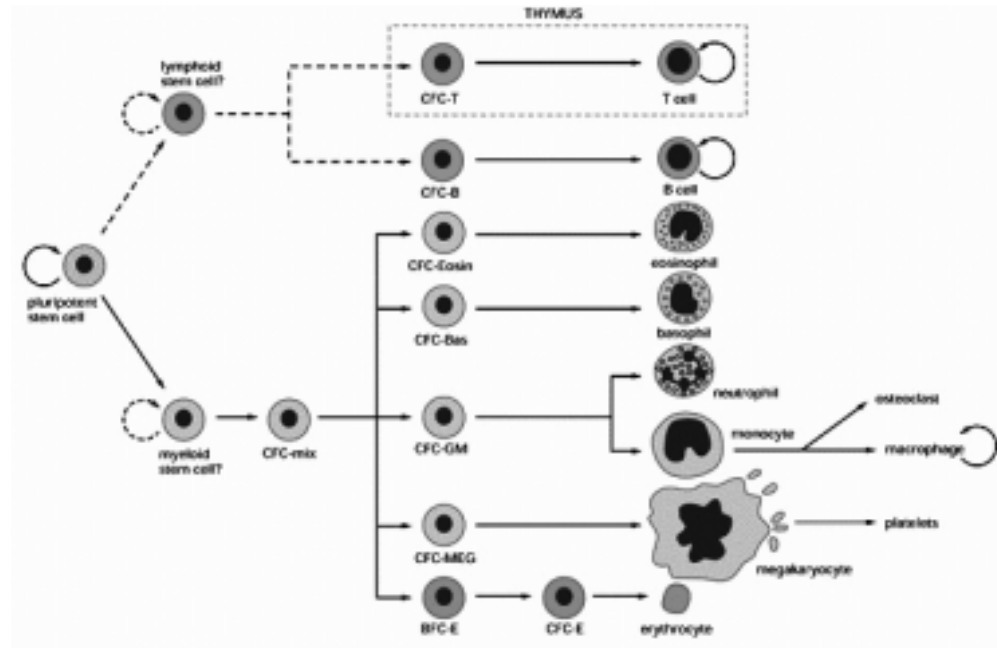
Part C: Immunology

Aims:

- **Antibodies**
 - Definition, Production, Purification
- **Immune response**
 - Lymphocytes: B & T
 - MHC
- **Vaccination**
 - Regular
 - Gene therapy

Section off
Alberts *et al*, Chap 5

Hematopoiesis/Hemopoiesis: process of blood formation



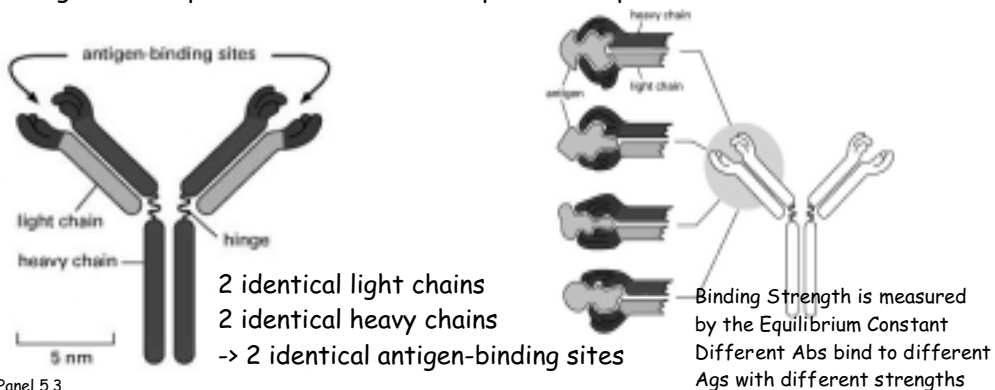
Antibodies

Antibody/Immunoglobulin:

Protein produced by **B lymphocytes** in response to a foreign molecule or an invading organism
Binds tightly to the foreign molecule or cell, thereby inactivating it or marking it for destruction

Antigen:

Molecule that is recognized by and binds specifically to an antibody molecule
It generates/provokes the immune response that produces the antibodies

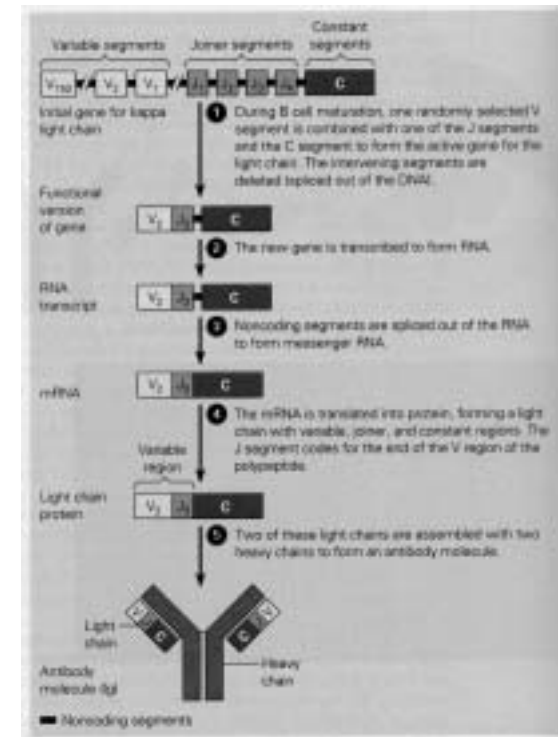


Panel 5.3

The genetic basis of antibody diversity

Building blocks

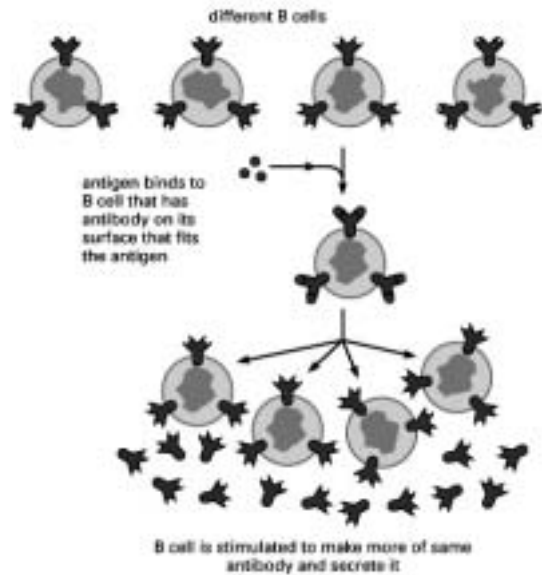
Ability to combine any several hundred unique variable (V) segments with any four joiner (J) segments to form unique light chains



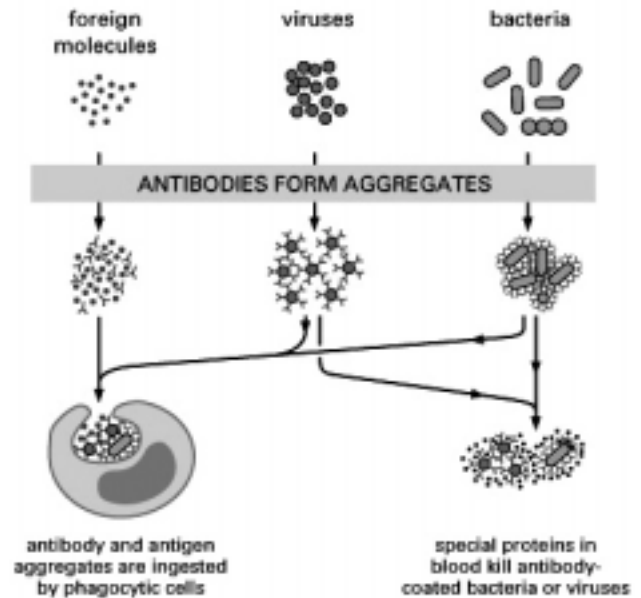
B cells

A class of white blood cells that make Abs

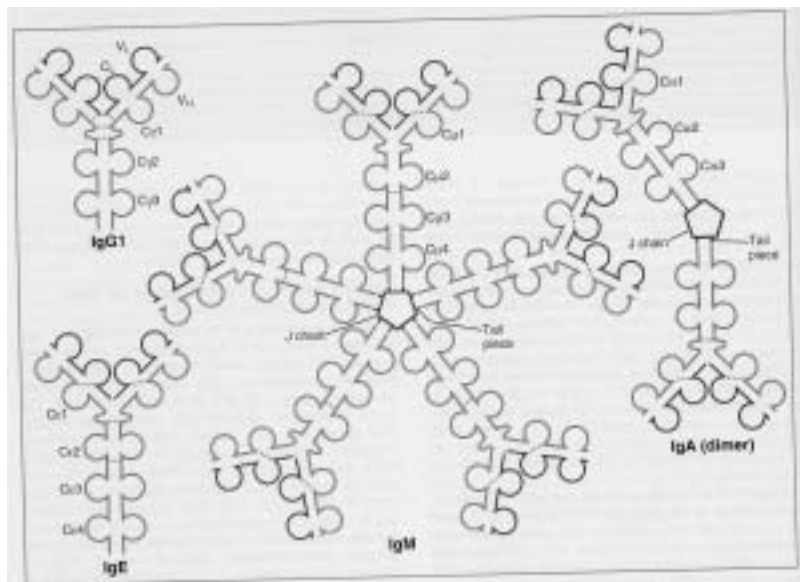
Resting B cell carries on its surface different membrane-bound antibody molecule
- serves as a receptor to recognize specific antigen



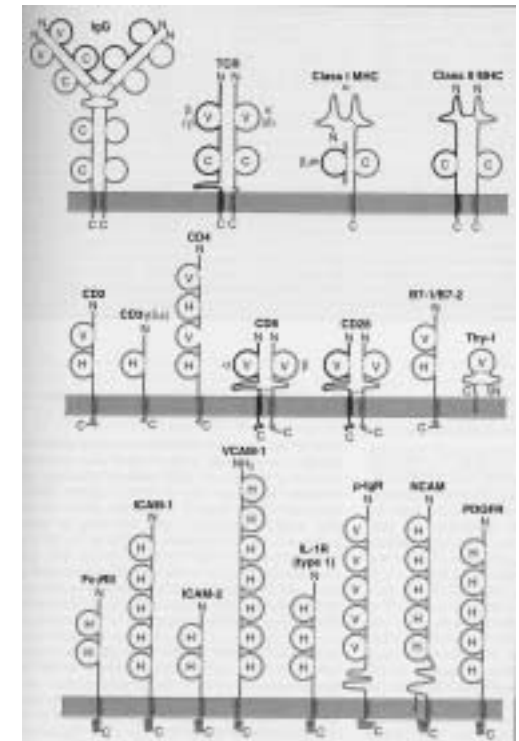
Antibodies: a defense against infection



Immunoglobulin Isotypes



The Immunoglobulin Superfamily



The Immune Response

A set of reactions carried out by the immune system following the detection of a foreign invader in a healthy human.

First line of defense: the skin

- Impenetrable barrier
- Contains sweat and oil glands the secretion of which gives skin acidic (pH 3-5) surface, enough to inhibit growth of microorganisms
- Sweat contains lysozyme -> destroys bacterial cell walls

Other routes of entry:

- **Digestive tract:** saliva (lysozyme), acidic stomach
- **Respiratory tract:** lined with cilia inhibits passage to lungs

Second Line of Defense:

- **Leucocytes:** White blood cells
 - ingest invading microorganisms: phagocytes, macrophages, neutrophils
 - kill own body's infected cells
- **Antimicrobial proteins:** complement system
- **Inflammatory response**

Third Line of Defense: The immune system

Types of Blood cells

Blood is composed of a fluid called plasma and several different types of cells circulating within that fluid

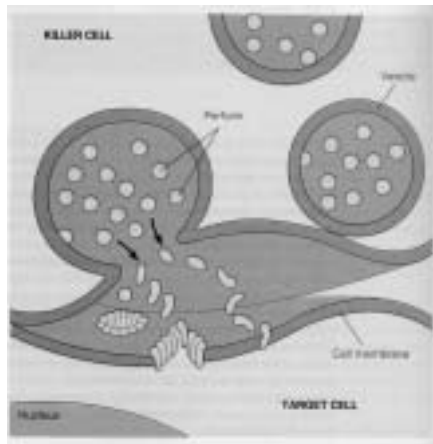
CELLULAR ELEMENTS 45%		
CELL TYPE	NUMBER (per mm ³ of blood)	FUNCTIONS
Erythrocytes (red blood cells)	5-8 million	Transport oxygen and help transport carbon dioxide
Leukocytes (white blood cells)	5,000-10,000	Defense and immunity
Basophil		
Lymphocyte		
Natural killer cell		
Neutrophil		
Macrophage		
Platelets	250,000-400,000	Blood clotting

Second Line of Defense....

- Leucocytes: White blood cells
 - ingest invading microorganisms: phagocytes, macrophages, neutrophils
 - kill own body's infected cells
 - Natural killer cells kill body's own cells that are infected (e.g by virus)



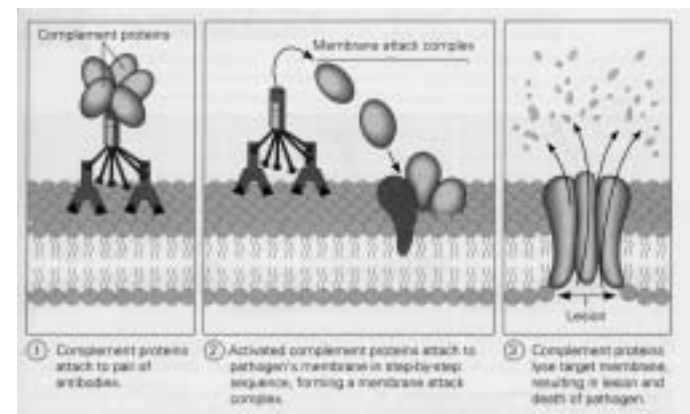
Phagocytosis by a macrophage
Probes surroundings with cytoplasmic extensions
Bacteria are engulfed
Electron micrograph



NK cells bind abnormal cells
Perforin form pores -> cells ruptures

Second Line of Defense....

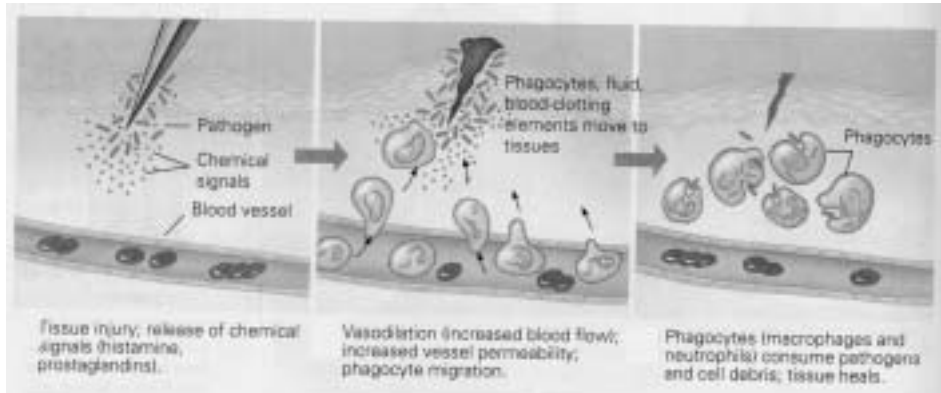
- Antimicrobial proteins: complement system



Complement proteins kill infected cells by forming a membrane attack complex
-> cell lysis

Second Line of Defense....

• Inflammatory response



Triggered when cells of injured tissue release chemical signals

- That induce increased permeability of capillaries and increased blood flow to affected area
- Chemoattractants are released -> attract phagocytes (macrophages & neutrophils)
- Phagocytes engulf pathogens and cell debris -> tissue heals

Immune System-

It's qualities include:

Diversity: to respond to a wide array of antigens

Specificity: can respond to the one antigen present rather than all antigens

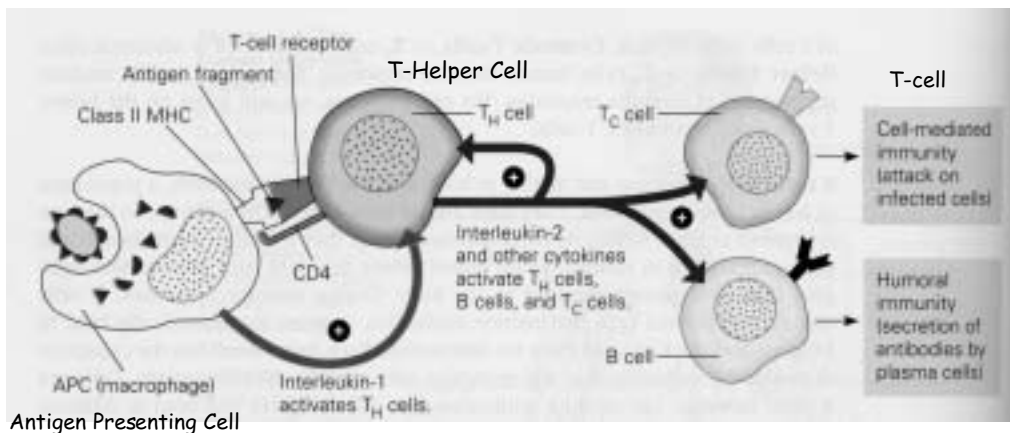
Memory: for previously encountered antigens

-> a faster response on subsequent encounters

Divisions of the Immune system:

- **Humoral immune response** - B-Cells stimulated to produce antibodies
- **Cell-mediated immunity** - T-Cell Mediated Response (Natural Killer Cells)
- **Complement system** - cascade of proteins to lyse cells

Some of the players.....

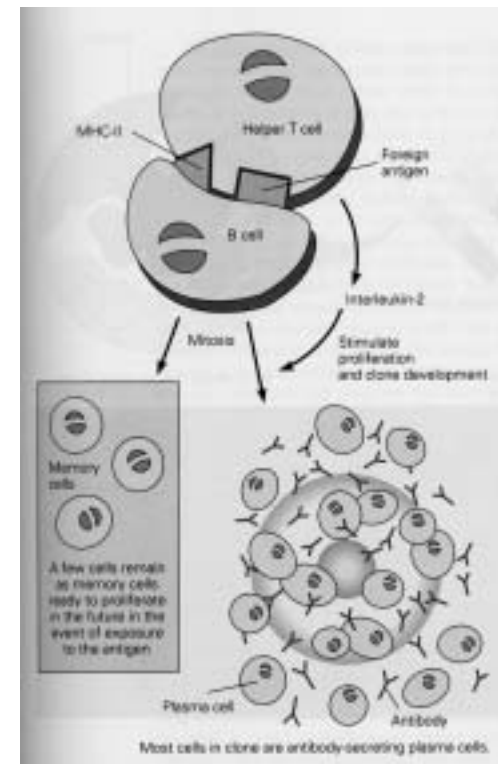


Humoral Immune Response

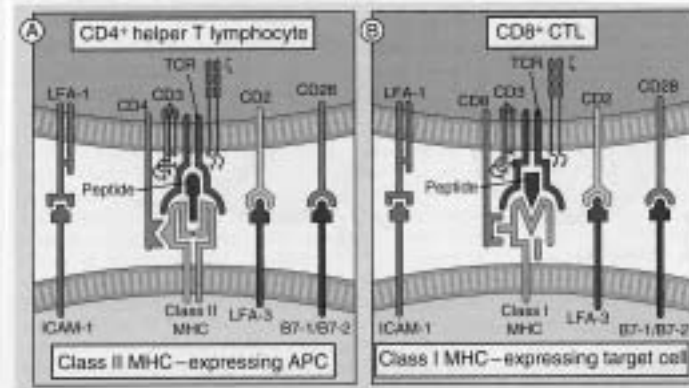
Activated T helper cell interacts with B cell that's engulfed antigen and displays a fragment of the antigen along with class II MHC proteins

T_H then secretes IL2 which stimulates B cell to divide repeatedly and differentiate into plasma (antibody secreting) cells and memory cells

Secreted antibodies specifically bind and inactivate the antigen that stimulated the immune response



The diagram illustrates the molecular components of a T cell receptor complex. The TCR is shown at the top, with a label 'Specific recognition of antigen' pointing to its extracellular domain. Below the TCR are the CD3 and CD28 molecules, which are part of the 'Signal transduction' pathway. Further down are the CD4/CD8 and CD2 molecules, which are involved in 'Signal transduction and adhesion'. At the bottom is the Integrin molecule, which is responsible for 'Adhesion'. The entire complex is embedded in a cell membrane, represented by a wavy line.

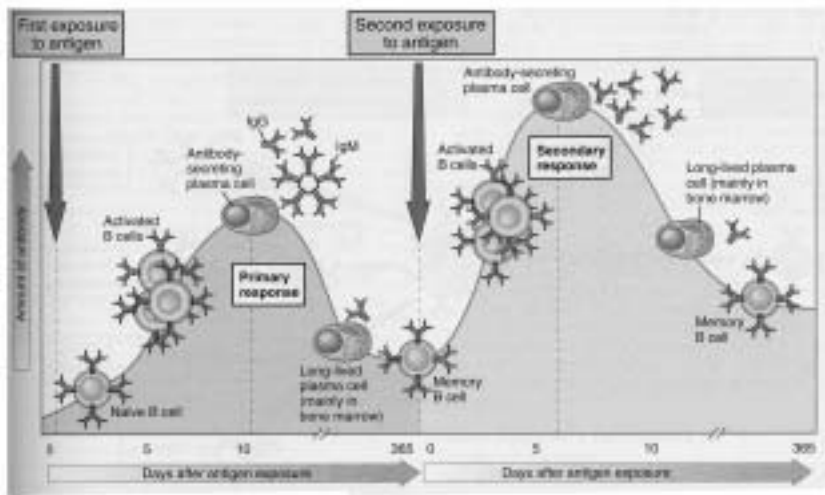


The diagram illustrates the TCR-mediated signaling pathway. It begins with the interaction between the T cell's TCR (CD3/CD8) and the APC's antigen presentation. This triggers a signaling cascade involving Lck, ZAP-70, and various phosphorylation events. Key biochemical intermediates include PLCγ1 activation, increased cytosolic Ca²⁺, Diacylglycerol (DAG), and the exchange of GDP for GTP on Ras. These lead to the activation of Calcineurin, PKC, and ERK/JNK. Finally, these active enzymes lead to the activation of transcription factors NFAT, NF-κB, and AP-1.

	Antigen recognition	Effector functions
B lymphocyte		Antibody secretion
Helper T lymphocyte		Activation (proliferation and differentiation) of T and B lymphocytes Activation of macrophages Inflammation
Cytolytic T lymphocyte (CTL)		Target cell lysis
Natural killer (NK) cell		Target cell lysis

The graph illustrates the kinetics of antibody production. The y-axis represents 'Serum antibody titer' and the x-axis represents 'Weeks' from 0 to 12. Two antigen exposures are shown: 'Antigen A' at week 2 and 'Antigen A + Antigen B' at week 7. The legend indicates that dark grey shading represents 'Anti-A' and light grey shading represents 'Anti-B'. The first exposure to Antigen A results in a 'Primary anti-A response' peaking at week 4. The second exposure, which includes both Antigen A and Antigen B, results in a 'Secondary anti-A response' (peaking at week 9) and a 'Primary anti-B response' (peaking at week 9). The secondary response to Antigen A is significantly higher than the primary response.

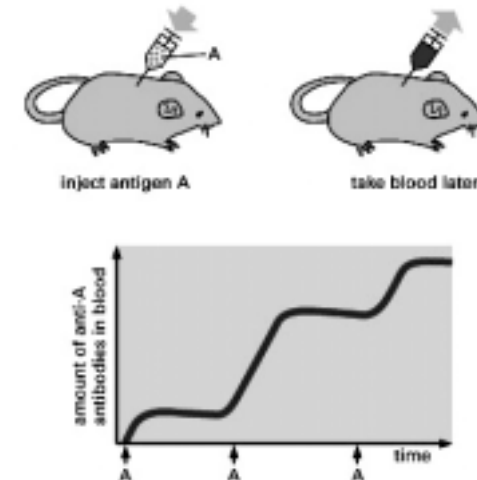
Kinetics of primary & secondary humoral immune response:



Raising antibodies in animals

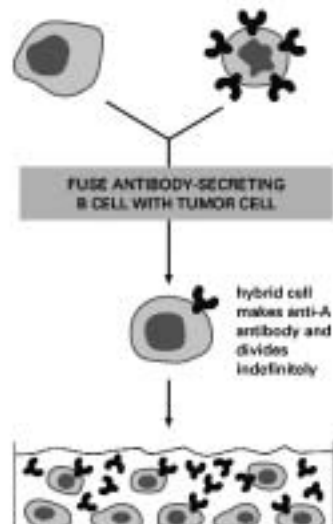
Common animals used: mouse, rabbit, sheep, goat

Repeated injections of the same antigen at intervals of several weeks stimulates specific B cells to secrete large amounts of antibodies into bloodstream



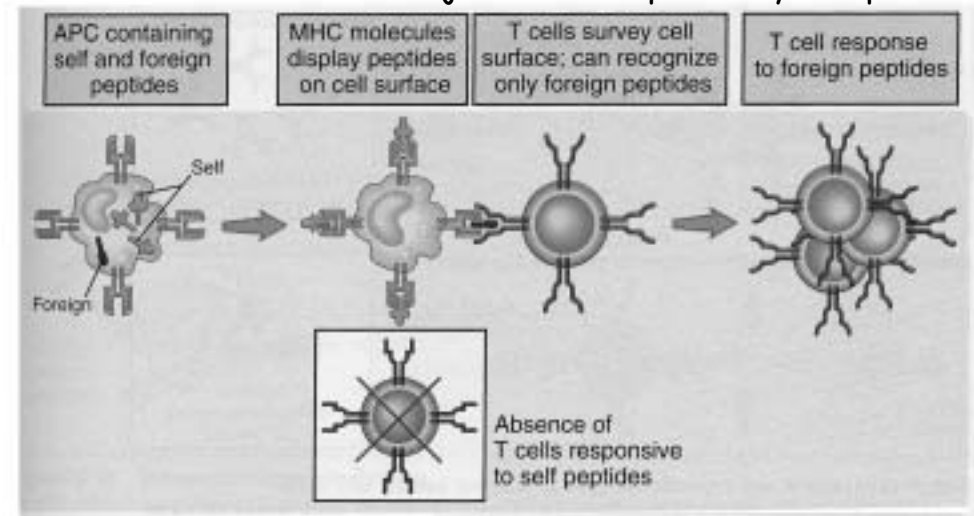
Monoclonal antibodies

To obtain large amounts of a particular antibody molecule
Fusion of B cell taken from an animal injected with antigen A
With a cell of a B cell tumor
→ Hybrid cell that secretes anti-A antibodies

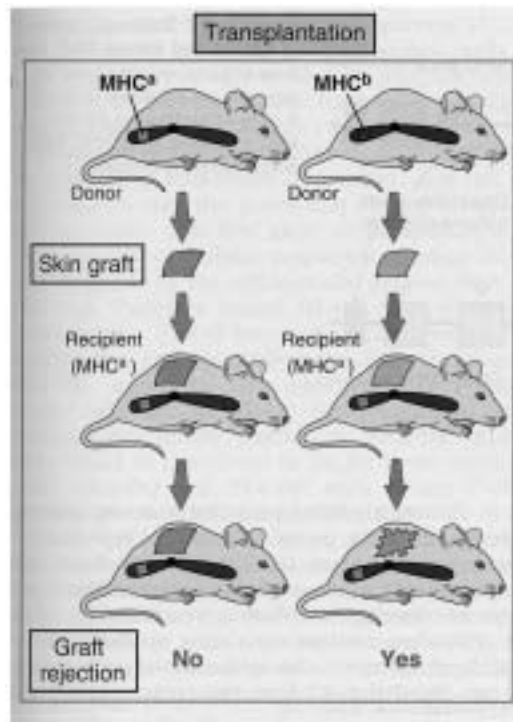


T cells survey antigen presenting cells for foreign peptides:

MHC - Major Histocompatibility Complex

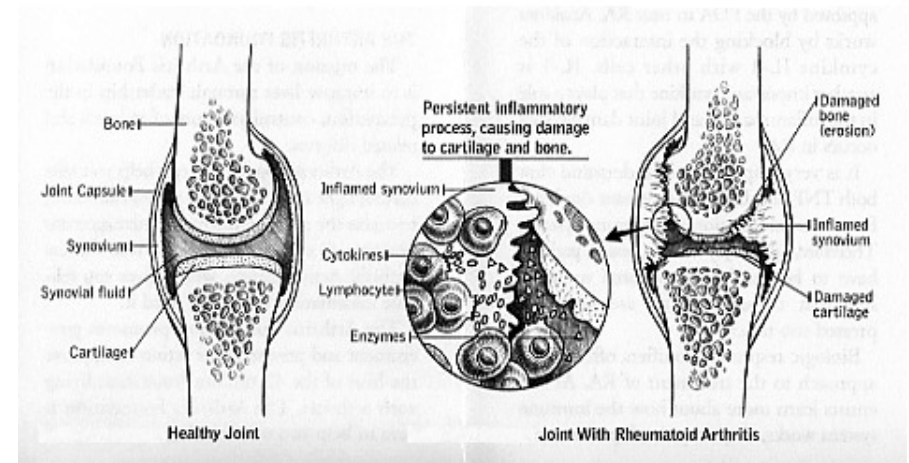


No distinction of self → autoimmune disease



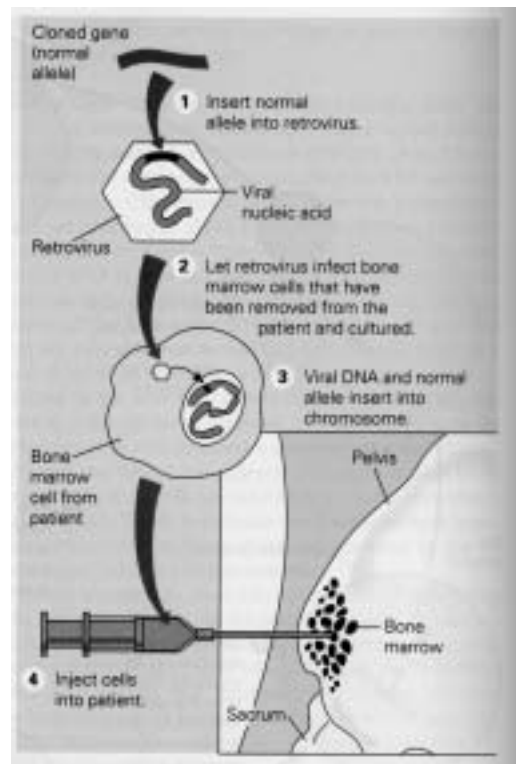
**MHC -
Major
Histocompatibility
Complex**

Rheumatoid Arthritis

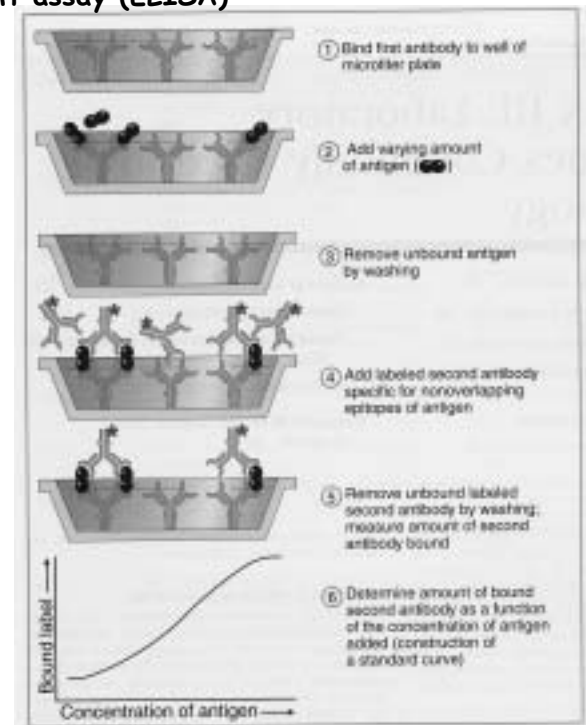


Gene Therapy

- Usually bone marrow cells are cultured from patient
- Retroviral vector containing desired gene is added to these cells
- Some take up retrovirus and incorporate DNA into their genome
- These cells are returned to patient through blood or into bone marrow

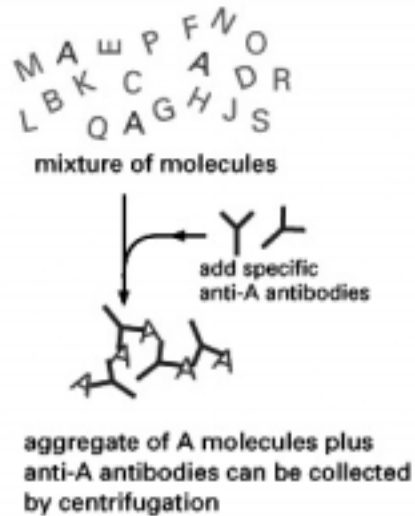


Enzyme linked immunosorbent assay (ELISA) Or radioimmunoassay



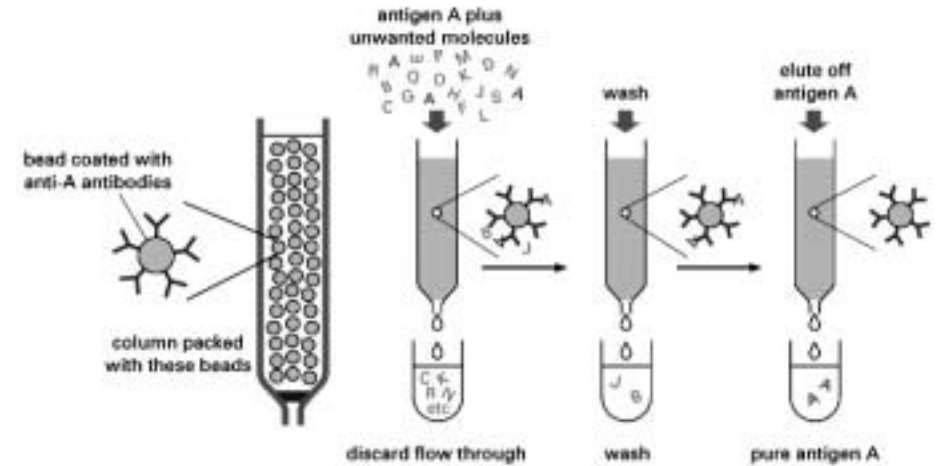
Antibodies used to purify molecules.....

Immunoprecipitation:



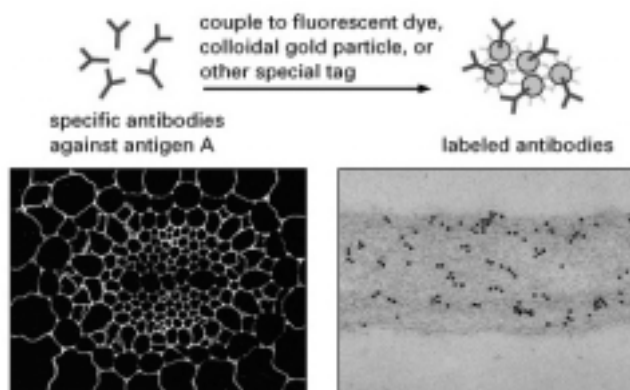
Antibodies used to purify molecules.....

Immunoaffinity column chromatography:



Antibodies used as molecular tags:

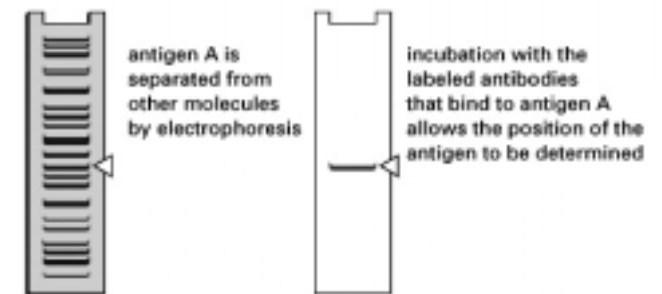
Microscopic detection



Fluorescent Ab binds to Ag in tissue
Gold-labeled Ab
Detected by electron microscope
Ag: pectin in plant cell walls

Antibodies used as molecular tags:

Biochemical detection



Sensitivity is increased by using multiple layers of Abs
-> sandwich
Detection of smaller numbers of Ag molecules

