HOW THE CELL DIVIDE?

Cells spend a small part of their life dividing. Cell division is very tightly controlled, ensuring that everything happens correctly (by checkpoints) at the right time and in the right order (by regulation). Cellular division refers to the process by which the living cells divide through complicated process into 2 or more to transmit its genetic material for reproduction, tissue renewal (wound healing), growth and development.

Cell divisions include 2 main events: Cellular and Nuclear divisions. Cellular divisions (Cytokinesis) refer to the process by which cytoplasm and cell components are divided. While, nuclear divisions (Karyokinesis) refer to the process by which a nucleus divides. Two major nuclear divisions are involved in the genetic continuity of the nucleated cells: Mitotic cell division (mitosis) and Meiotic cell division (Meiosis).

Mitosis is the process of cell division in which the daughter cells receive identical copies of DNA of the mother cell. Meiosis is the process of cell division that results in the formation of cells containing half the amount of DNA contained in the parent cell, and having different copies of DNA from one another. The cytoplasm and organelles are usually shared approximately equally between the daughter cells. So, Mitosis creates genetically identical species, while Meiosis increases genetic diversity in a species.

CELL CYCLE

The cell cycle occurs from the completion of one division until the completion of the next division. It involves 3 phases: **Interphase** (G1, S and G2), **Mitosis** (M) followed by **Cytokinesis** (C). The period between M and S is called G_1 stage and that between S and M is G_2 stage (figure below).

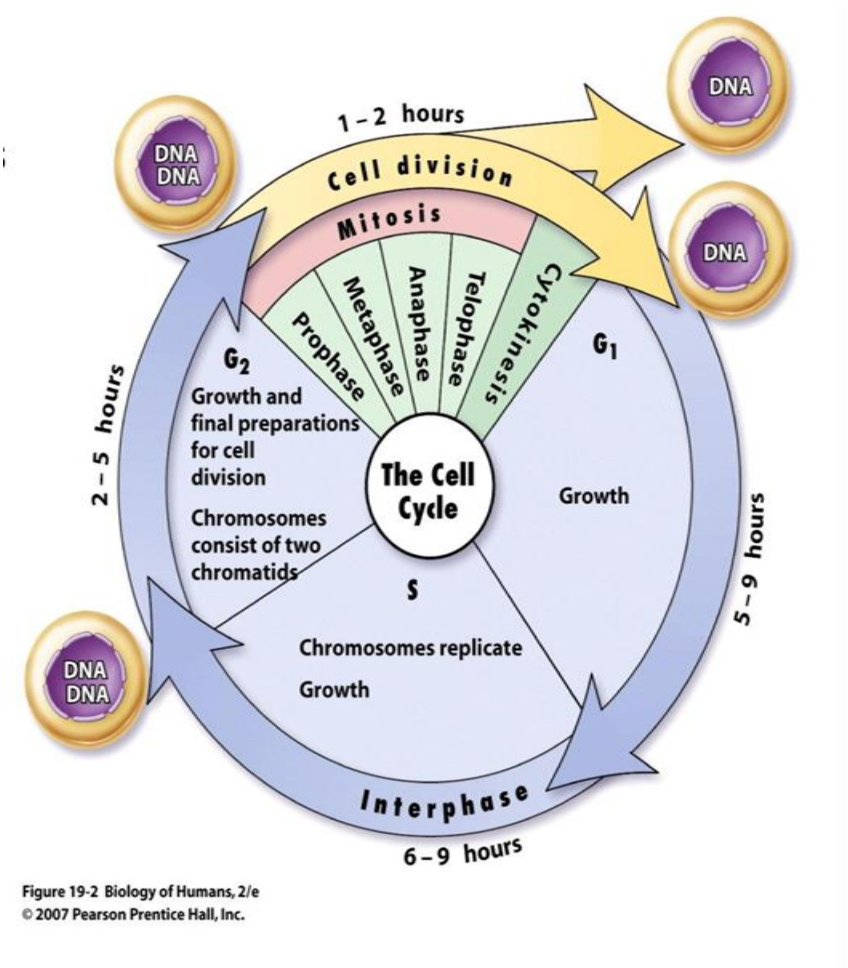


Figure 19-2 Biology of Humans, 2/e

The cell spends 90% of its time in Interphase and only 10% in Mitosis but, the duration of each phase and stage in eukaryotic cells depends on the cell type: For a typical rapidly proliferating normal human somatic cell with a total cycle time of 24 hours (1440 min), the G1 phase might last about 11 hours, S phase about 8 hours, G2 about 4 hours, and M about 1 hour. Other types of cells, however, can divide much more rapidly as budding yeast and embryo cells: Yeast cell has a total cycle time of 2 hours (120 min), the G1 phase might last about 15 mins, S phase about 10 mins, G2 about 90 mins, and M about 5 mins.

Other example:

Cell Type	Total Time
fly embryo	8 minutes
bacteria	20 minutes
human skin	20 - 24 hours
human liver	Once then retain
human nerve	never once mature
Chlamydomonous	14 hours

Note: Some cells divide rapidly as beans, for example take 19 hours for the complete cycle). While other like red blood cells cannot divide at all as they don't contain nucleus. Others, such as nerve cells, lose their capability to divide once they reach maturity. Some cells, such as liver cells, retain but do not normally utilize their capacity for division. Liver cells will divide if part of the liver is removed. The division continues until the liver reaches its former size.

STEPS:

Interphase

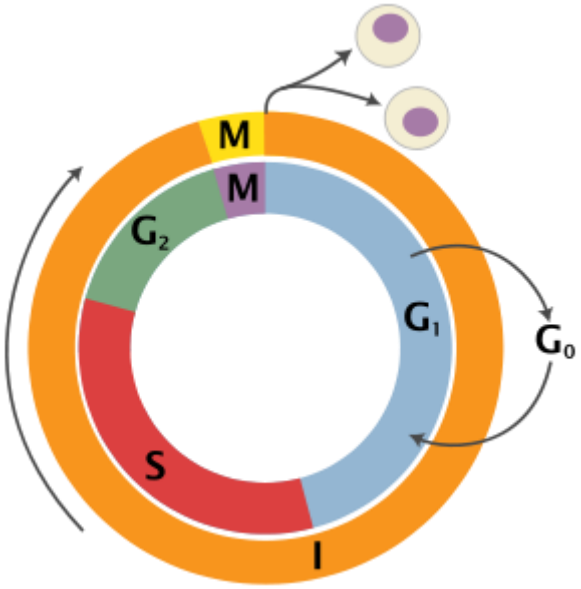
The time between two successive mitotic divisions is known as Interphase (Resting or Growth stage). During interphase, the genetic material in the nucleus is in form of chromatin (uncoiled DNA), which appears only as dark granules within the nucleus. This appearance may be because they are uncoiled, long and thin strands. Both nucleolus and nuclear membranes are present and clearly visible.

In this phase, the cell prepares itself for division through a group of biological processes for cell growth and accumulating nutrients needed for mitosis and duplicating its DNA.

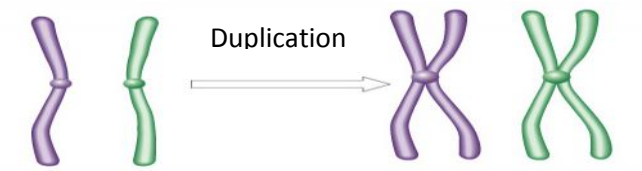
The interphase involves 3 stages called G1, S and G2, respectively.

G1 stage (gap1, Pre-DNA synthesis): It lasting in a range of 4-9 hours depending on the type of eukaryotic cells. The cells become metabolically active (1^{ry} growth) producing RNA and ribosomes for protein synthesis; the cell organelles begin to increase in numbers, and the nucleus and cytoplasm enlarge so, the cell reach their mature size (small in size from previous division). The genetic materials are 2n in number (diploid cell), fully extended and single in structure i.e. a chromatid with a centromere (unduplicated chromosome, Monad).

Cells that have temporarily (reversibly) or permanently stopped dividing are said to have entered a state of quiescence called G_0 phase (Prolonged G1 phase). This phase refers also as non-dividing phase outside of the cell cycle (see figure below) in which the cell will readjusted and stimulated to return to G_1 and thereby reenter the cell cycle.

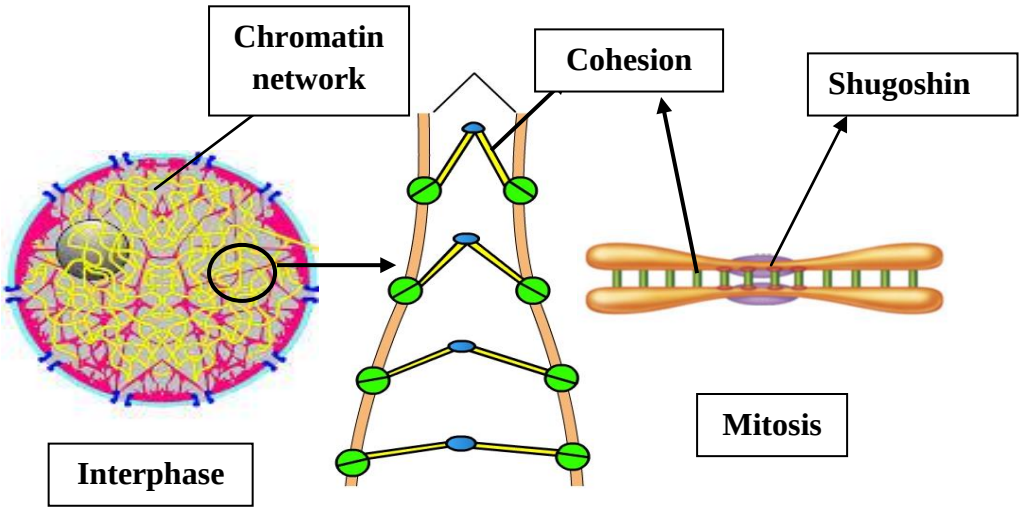


S stage (DNA synthesis): DNA and histone syntheses lasting in a range of 6-9 hours depending on the type of eukaryotic cells. DNA and histone are the main component of chromatids. At the end of this stage, monads have been duplicated and became double in structure i.e. with 2 sister chromatids (duplicated chromosome, dyad) joined by a centromere (figure below) but still diploid (2n).

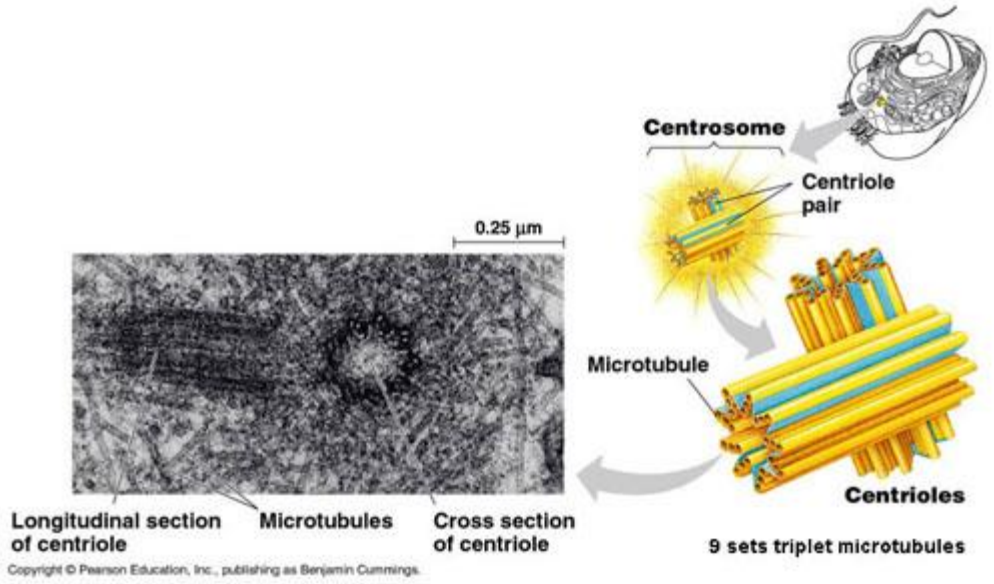


Two unduplicated chromosomes **Two duplicated chromosomes**

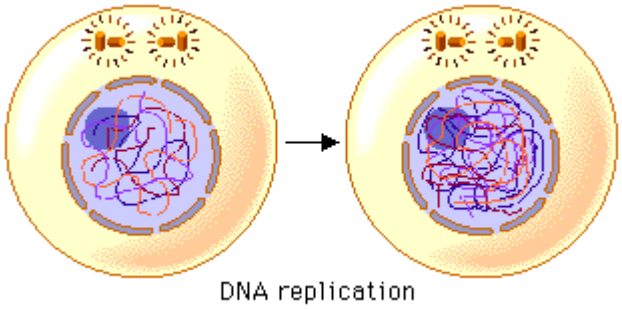
Sister chromatids are held together by multi-subunit protein complexes called **cohesin** and **Shugoshin** in interphase and mitosis (figure below).



G2 stage (gap2, Post-DNA synthesis): This stage lasting from 2-5 hours in some eukaryotic cells. In which the cell synthesis certain component required for mitosis (assemble machinery) as microtubules in plants and microorganisms (centrosomes and centrioles in animal, proteins of spindle fiber, enzymes,... and go to the final preparations of the cell (2^{nd} growth) before divisions. The chromosomes are 2n (diploid) double in structure (dyad) but invisible in this form (uncoil) and the nucleus is filled with chromatin fibers that are formed when the chromosomes are uncoil.



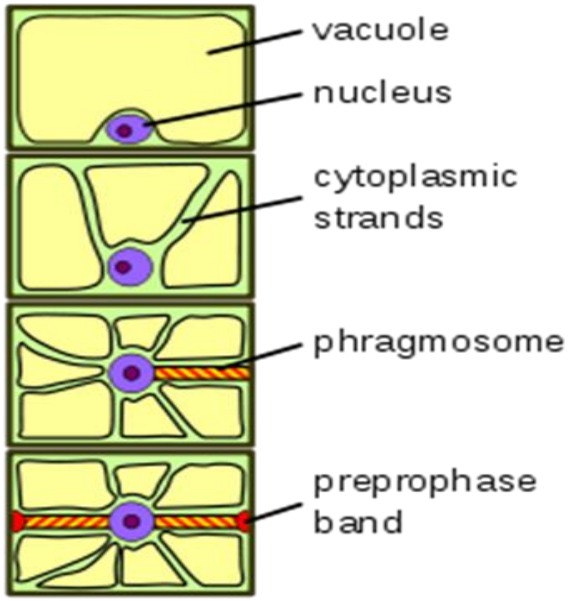
Final Form of Interphase:



Before mitosis, the nucleus (for plant and microorganisms) must to be in central position. To do that the flowing steps happen:

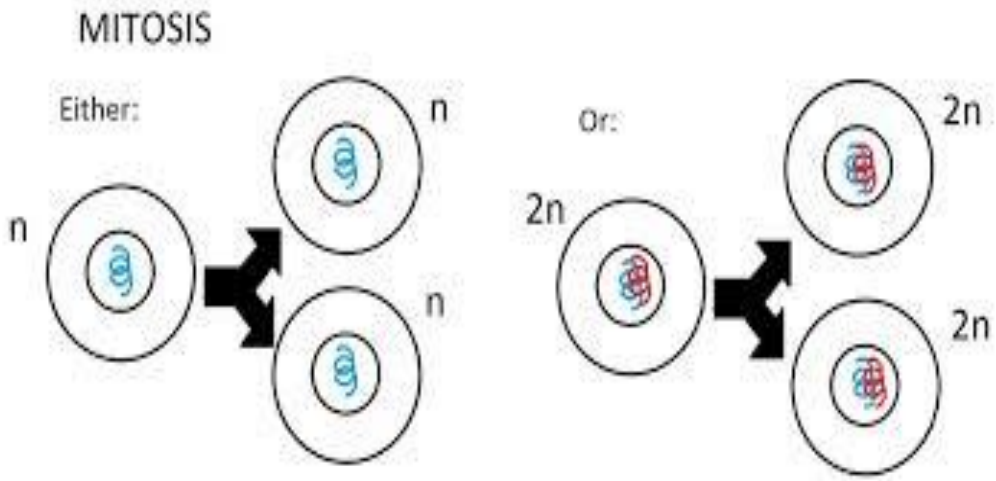
- Initially, cytoplasmic strands forms that penetrate the central vacuole and provide pathways for nuclear migration.
- Actin filaments along these cytoplasmic strands pull the nucleus into the center of the cell.
- These cytoplasmic strands fuse into a transverse sheet of cytoplasm along the plane of future cell division, forming the **phragmosome**.
- Just before mitosis, a dense band of microtubules appears around the phragmosome and the future division plane just below the plasma membrane.
- This preprophase band marks the equatorial plane of the future mitotic spindle as well as the future fusion sites for the

new cell plate with the existing cell wall. It disappears as soon as the nuclear envelope breaks down and the mitotic spindle forms.



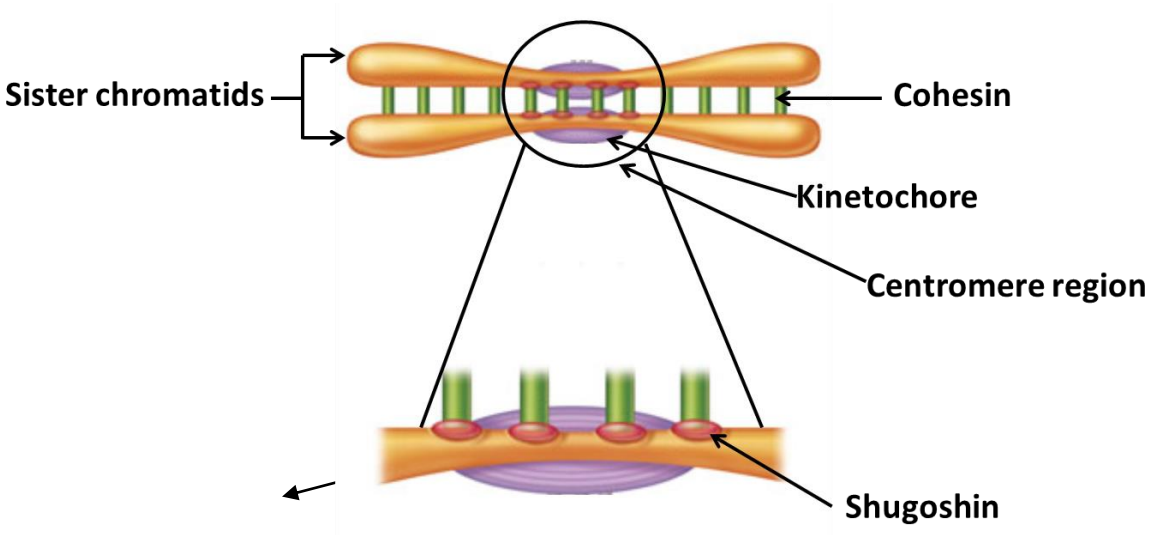
MITOSIS (M-phase)

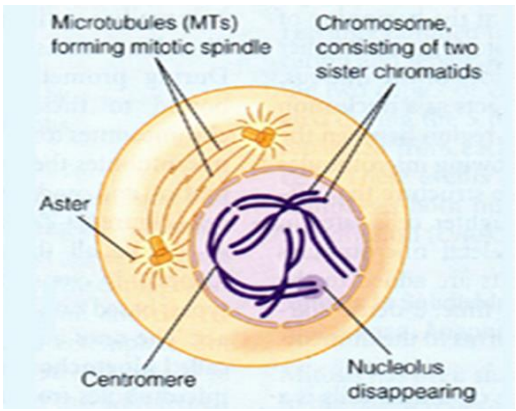
It is the process by which a cell produces two identical daughter cells with complete set of chromosomes. This means that all the chromosomes must be duplicated and separated into two full sets, one at each end of the cell that is splitting in two (figure below). The cell organelles and other material that makes up the cell also split in two.



Mitosis consists of 4 phases known as Prophase, Metaphase, Anaphase and Telophase (figure below).

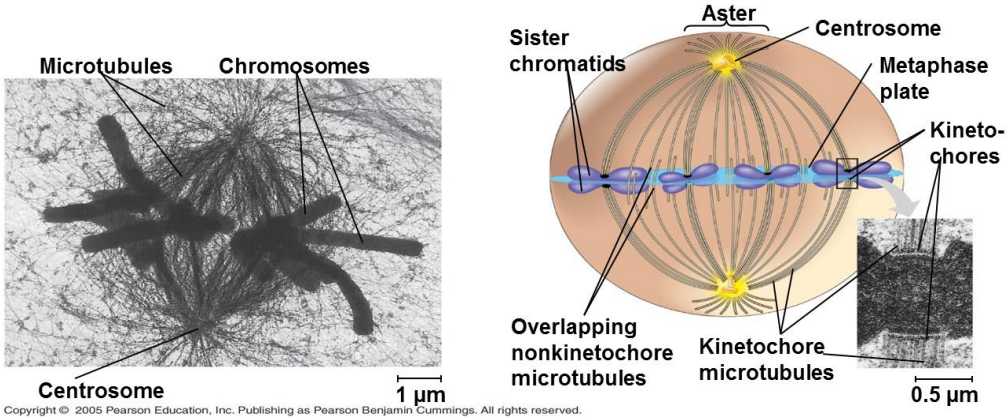
Prophase: In this phase, the sister chromatids condense (coiled) and thickened until they appear as thread-like chromosomes joined by centromere (2n double in structure). Sister chromatids are also held together along their length by **cohesion** but at centromeres region, they are held together by both **cohesin and Shugoshin** proteins (Figure below). Both nuclear envelope and nucleoli start to disappear, while the mitotic spindles begin to form from the centrosomes to control chromosome movement during mitosis (figure below).



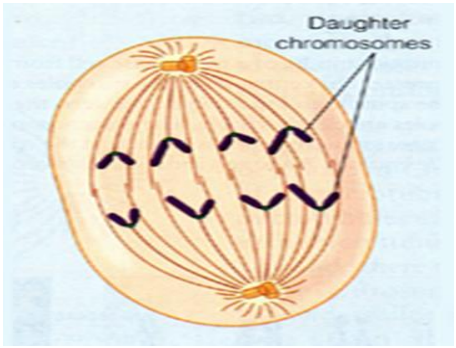


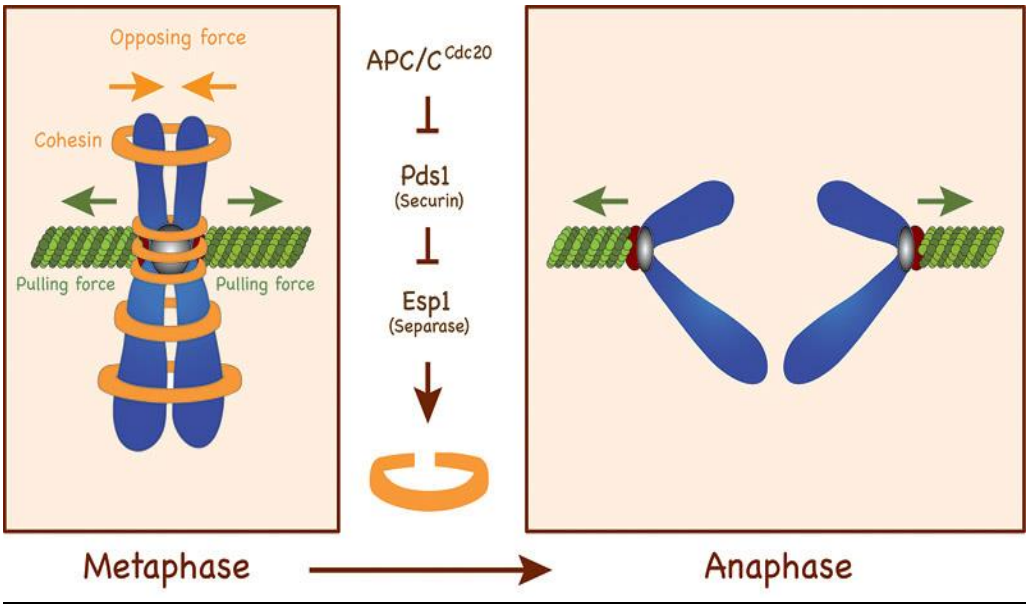
NOTE: The spindle apparatus (figure below) includes the centrosomes in animal cell only.

Metaphase: When the mitotic spindle is fully formed, the chromosomes align themselves along the cell spindle in the middle of the cell (equator, equatorial plates). This movement is due to: Assembly and disassembly of microtubules provide force to move chromosomes with the help of the motor proteins located in kinetochore and poles of cell pull on microtubules to provide force. The metaphase chromosome (2n double in structure) appears as two sister chromatids join together by their centromeres and to the spindles by their kinetochore (figure below). At this stage, separase enzyme (and others) dissolves the cohesion protein along the 2 sister chromatids except at centromere were both cohesion and Shugoshin proteins remains.

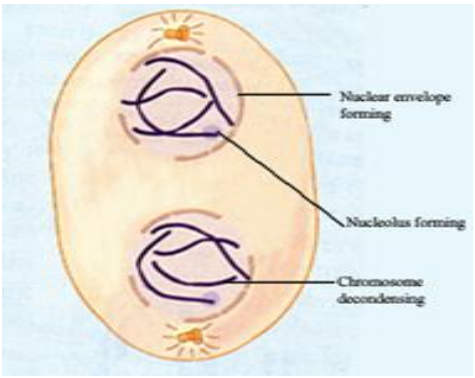


Anaphase: Both cohesion and shugoshin dissolve by proteiolytic enzymes so, the sister chromatids (present in equator) split apart at their centromeres, begin to separate and move to opposite poles of the spindle, segregating one of the two sister chromatids to each of the opposite ends of the cell. In this case, each chromatid became a chromosome. The chromosomes are 2n single in structure (2n monad).

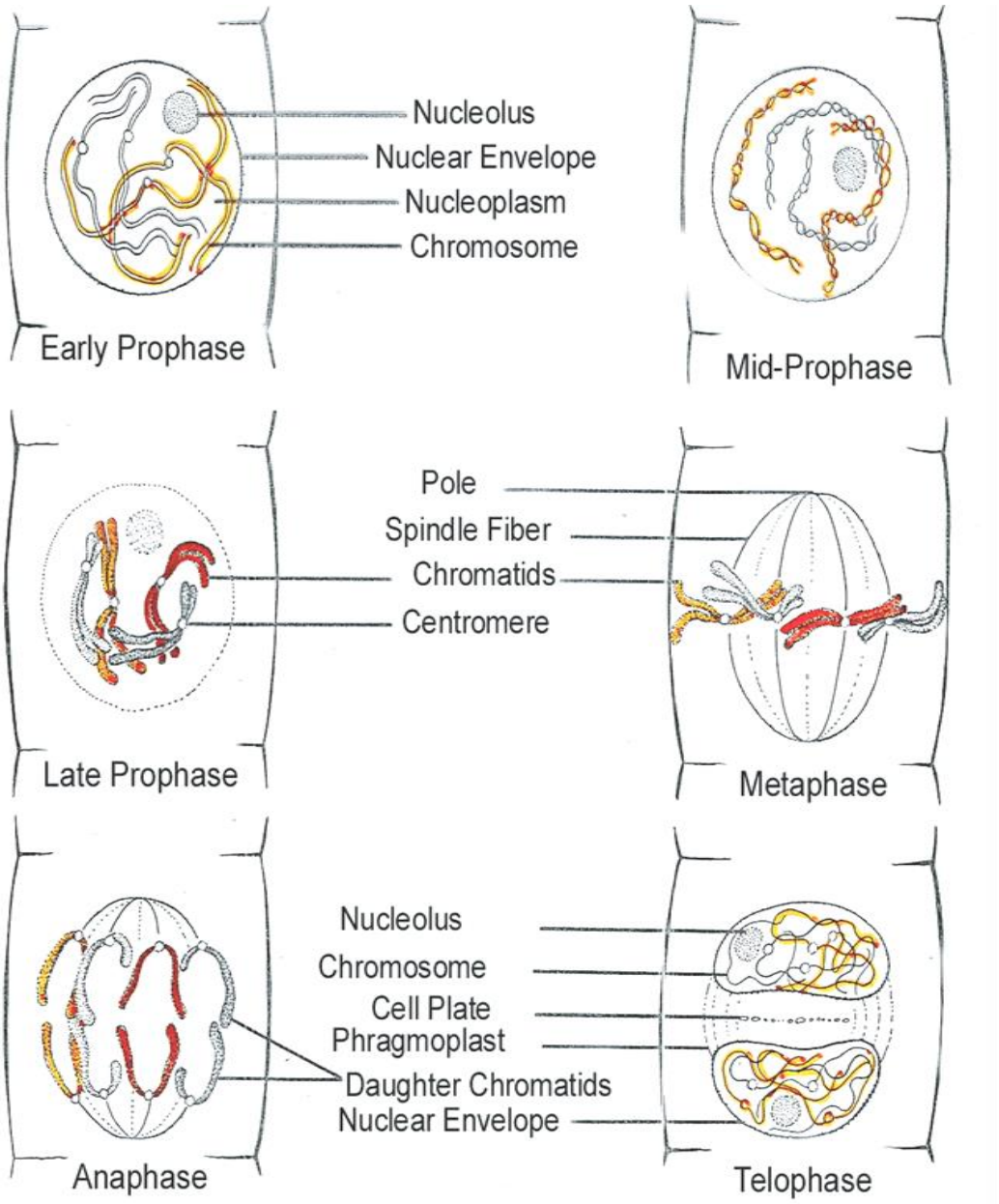




Telophase: A complete set of chromosomes reach each pole of the cell and begin to uncoil (became chromatin). The mitotic spindles, centrosomes and asters begin to disappear (microtubules are broken down into tubulin monomers). The nucleolus and the nuclear envelop reappear around the set of chromosomes. The chromosomes are 2n single in structure. Then the cell prepares to split in two identical daughter cells by a process called cytokinesis.



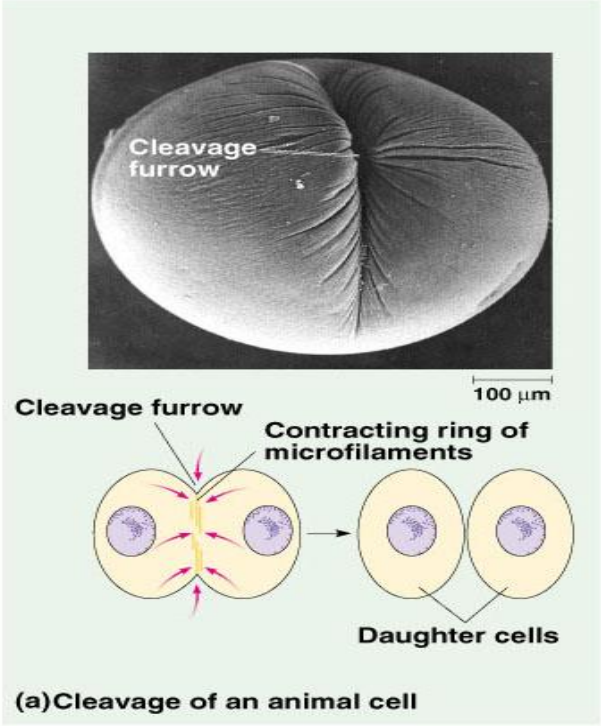
Overall steps in plants:



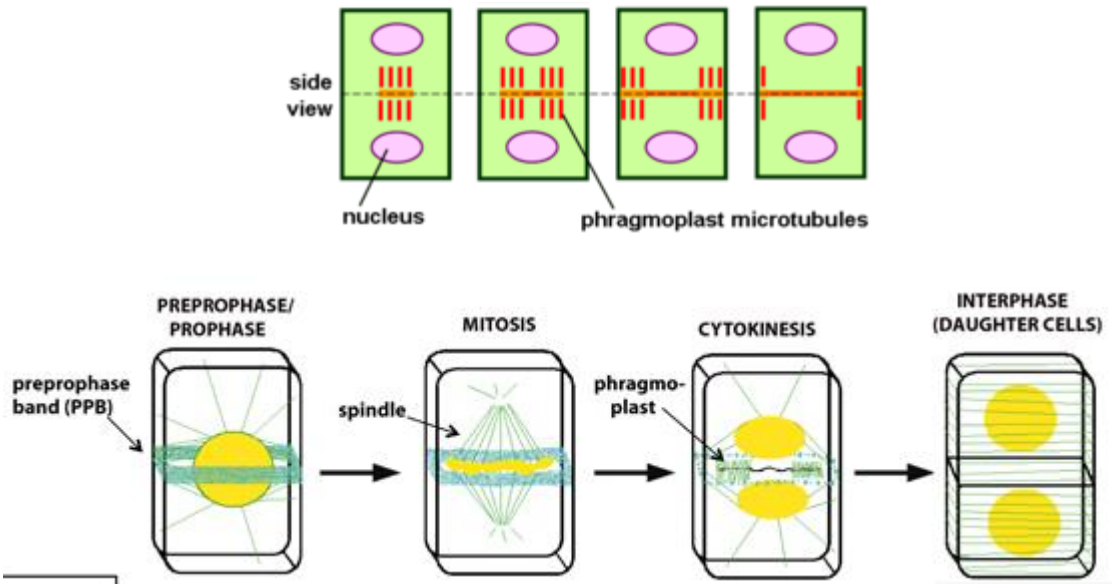
CYTOKINESIS

It usually initiates during the late stages of mitosis (at the end of telophase), and sometimes meiosis, splitting a cell in two, to ensure that chromosome number is maintained from one generation to the next or one cell to another.

In animal, the cell membranes on opposite sides of the cell become pinched-in (constriction) allowing for the cell to divide. The initial structure that forms is called a **cleavage furrow**. The cleavage furrow continues to pinch in, until the two sides are touching. At this point, there will be two new cells.



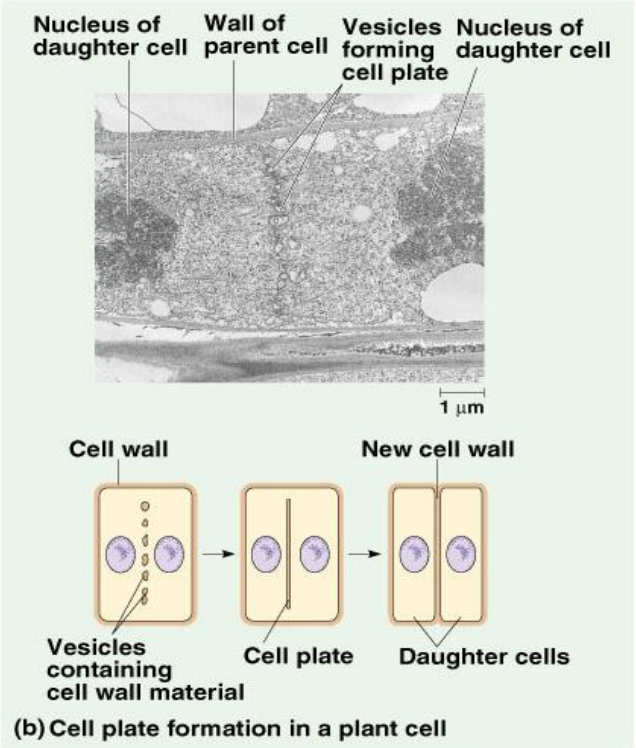
The **phragmoplast** is a plant cell specific structure that forms during late cytokinesis. It serves as a scaffold for cell plate assembly and subsequent formation of a new cell wall separating the two daughter cells.



The phragmoplast is a complex assembly of microtubules (MTs), microfilaments (MFs), and endoplasmic reticulum (ER) elements, that assemble in two opposing sets perpendicular to the plane of the future cell plate during anaphase and telophase.

Once the cell plate has divided the cell into two cells, it forms the **middle lamella.** In the same time the **plasma membrane** of the maim cell split and begin to reform in the both daughter cells. Subsequently, the cell will develop new primary and secondary layers of **cell wall** (figure below).

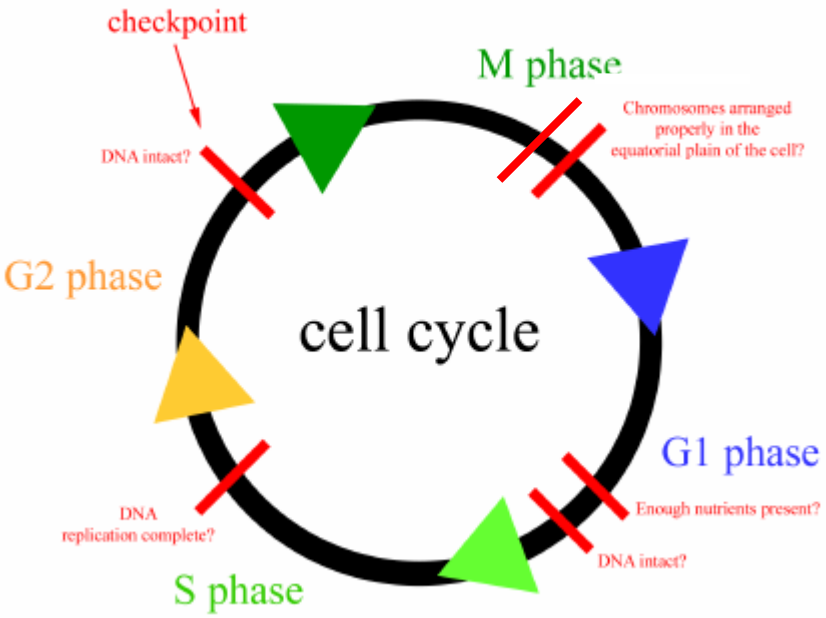
The microtubules and actin filaments within the phragmoplast serve to guide vesicles (from golgi) with cell wall material to the growing cell plate. Actin filaments are also possibly involved in guiding the phragmoplast to the site of the former preprophase band location at the parent cell wall.



(b) Cell plate formation in a plant cell

CELL CYCLE CHECKPOINTS

Maintenance of genomic stability is needed for cells to survive many rounds of division throughout their lifetime without disruption. Key to the proper inheritance of intact genome is the tight temporal and spatial coordination of cell cycle events to monitor the proper execution of cell cycle processes to avoid uncontrolled cell division characterizing malignancy. Those keys are the cell cycle checkpoints.



As we have outlined previously, the cell cycle consists of four primary stages, G1 (GAP 1, 1^{ry} growth), S (Synthesis), G2 (GAP 2, 2^{nry} growth) and M (Mitosis). In order for each of the stages to have good participation in the cycle, DNA must clear all the checkpoints which it encounters along the way.

Multiple checkpoints have been identified as G1 checkpoint, DNA replication checkpoints, G2 checkpoint, antephase checkpoint and Mitotic spindle checkpoint.

G1 checkpoint (restriction point) is located at the end of the G_1 phase, just before entry into S phase (G_1/S) to monitor the size the cell has achieved since its previous mitosis, nutrition, growth factors and also

to evaluate the condition of the DNA. It is a vital checkpoint making the key decision of whether the cell should divide, delay division, or enter a resting stage. If all conditions are "normal", then the cell is allowed to proceed from G1 to the S phase of the cycle. If the cell has not reached an adequate size or if the DNA has been damaged, further progress through the cycle is arrested until these conditions are "corrected."

The **DNA replication checkpoint** is located at the end of the S phase to ensure the good replication of DNA before entering G2 phase.

The **G2 checkpoint** is another checkpoint (after completing S and G2 phases) in which DNA must overcome to complete a successful cycle. In order for this checkpoint to be passed, the cell has to check a number of factors, including DNA, to ensure that the cell is ready for advancing to the M or mitosis phase.

The **mitotic spindle checkpoint** (spindle assembly checkpoint) occurs at metaphase where all the chromosomes should/have aligned at the mitotic plate (equator) and be under bipolar tension (tension of both poles). The tension created by this bipolar attachment is what is sensed, which initiates the anaphase entry i.e. the anaphase will be blocked if the chromatids are not properly assembly on mitotic spindle by their kinetochores. In addition, if this failure to attach correctly to the spindle passes, it causes an unequal segregation of chromosomes (non-disjunction), which can lead to cell death or disease.

The DNA damage and spindle assembly checkpoints are surveillance mechanisms that ensure genomic integrity by delaying cell cycle progression in the presence of DNA or spindle damages, respectively until all chromosomes are correctly attached in a bipolar fashion to the mitotic spindle.

NOTE:

The check for DNA damage in eukaryotic cell division is to successfully pass accurate DNA strands (mutation free) from parental genomes to daughter cells as cells mitotically replicates. The passing of mutation-free DNA will ensure the cycle procedures healthy and functional cells. Mutations (due to either irradiation or chemical modification) will likely lead to cancer.

Signal Mechanisms within the checkpoints can delay (or stall) the cycle until mutations are corrected. If the G1 checkpoint deems the DNA unsuitable for progression it can stop or delay the process sending it into an optional resting phase known as G_0. A special protein referred to as **P53** is essential in the function of the G_1 restriction point as *P53 has the ability to detect mutations in the genes* which pass through the checkpoint.

If mutations are irreversible, they can tag a cell for **self-destruction (cell suicide)** ***via*** **apoptosis** (effector mechanism) and thereby block progression through the cell cycle by eliminating the chance that mutated DNA will be replicated.

Regulation of Eukaryotic Cell Cycle

Not all cells proceed through the stages of the cell cycle at the same rate. Embryonic cells divide very rapidly, while mature cells might divide rarely, or in response to signals such as wounding or growth factors, or not at all.

It should seem obvious that the processes that drive a cell through the cell cycle must be highly regulated and required a number of control mechanisms to ensure that the resultant daughter cells are viable and each contains the complement of DNA found in the original parental cell.

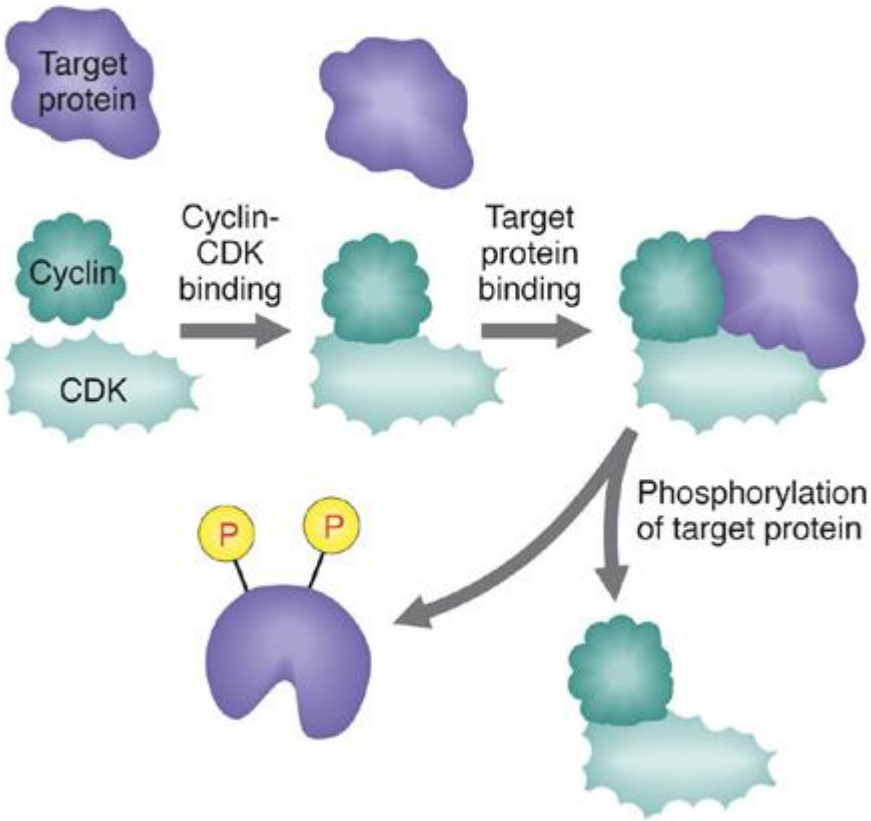
They *control the timing of events* so that each individual process is turned on and off at the appropriate time.

Two key classes of regulatory proteins: cyclins and cyclin-dependent kinases (CDKs) determine a cell's progress through the cell cycle.

What Are Cyclins and Cyclin-Dependent Kinases?

CDKs are enzymatic proteins involved in cell cycle progression. Cdks are defined by their need to bind with cyclin subunits in order for enzymatic activation and modify (transferring phosphate groups from ATP to specific stretches of amino acids in the protein) various protein substrates.

Different types of eukaryotic cells contain different types and numbers of CDKs. For example, yeast has only a single CDK, whereas vertebrates like us have nine, of which four are really critical to the cell cycle (CDK1, CDK2, CDK4, CDK6).



Cyclins are a family of proteins that form the regulatory subunits, while CDKs are the catalytic subunits of the activated complex; cyclins have no catalytic activity and CDKs are inactive in the absence of a partner cyclin. Each cyclin associates with one or two cyclin-dependent kinases to be partially activated.

These proteins are the activators for CDK enzymes. Typically, cyclins are created or destroyed according to whether they are required which directs the cell through the various stages of the cell cycle. When cyclins bind with CDKs, they form a complex where the CDK active site is triggered.

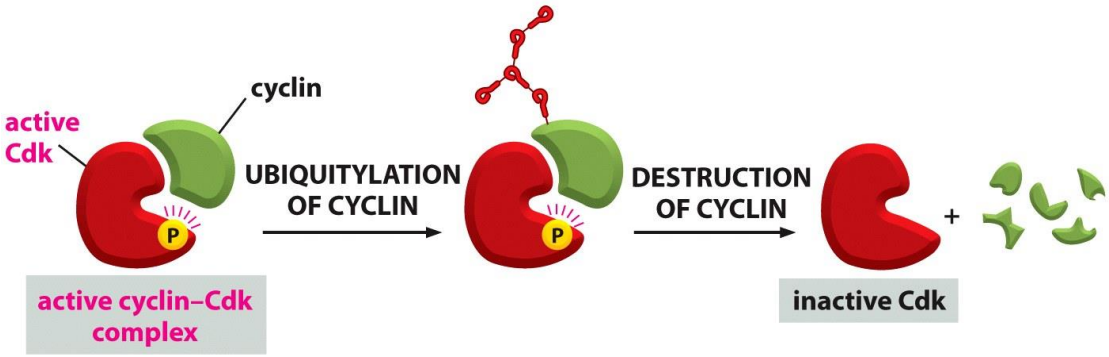


Figure 18-11 Essential Cell Biology 3/e (© Garland Science 2010)

All eukaryotes have multiple cyclins, each of which acts during a specific stage of the cell cycle. All cyclins are named according to the stage at which they assemble with CDKs. Common classes of cyclins include G_1-phase cyclins, G_1/S-phase cyclins, S-phase cyclins, G2-phase cyclins and M-phase cyclins (table below).

Cyclin-CDK Complex	Cyclins	CDK Partners
G_1-CDK	cyclin D (D1, D2, D3)	CDK4, CDK6
G_1/S-CDK	cyclin E	CDK2
S-CDK	cyclin A,B	CDK2
G_2-CDK	cyclin A,B	CDK1
M-CDK		

Specific function of cyclins-CDKs complexes:

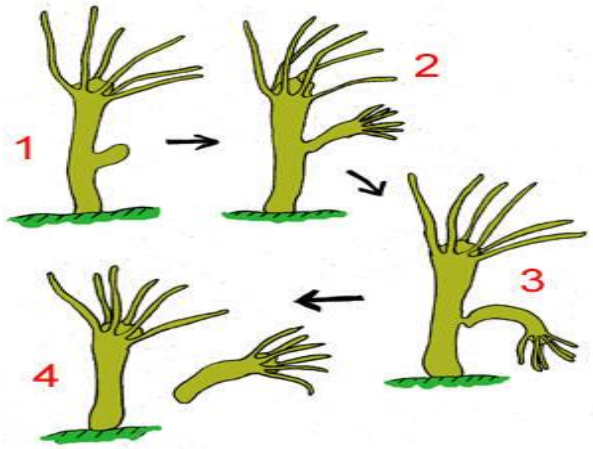
cell cycle stage	cyclins	CDKs	comments
G1	Cyclin D	CDK4&6	Can react to outside signals such as growth factors or mitogens to guide the cell's progress through the G_1 phase to coordinates cell growth with the entry to a new cell cycle.
G1/S	Cyclins E	CDK2	Regulate centrosome and microtubule duplication; important for reaching START required to commit the cell to the process of DNA replication in S-phase.
S	Cyclins B & A	CDK2	Targets are helicases and polymerases required for the initiation and induction of DNA synthesis.
M	Cyclins A & B are synthesized during S	CDK1	- Regulate G2/M checkpoint. - drive the cell's entry to promote the events of mitosis like the assembly of mitotic spindles and alignment of sister-chromatids along the spindles. - Phosphorylate lots of downstream targets as nuclear envelope and initiation of prophase, and subsequently, its deactivation causes the cell to exit mitosis.

IMPORTANCE OF MITOSIS:

Following are the occasions in the lives of organism where mitosis happens:

Asexual Reproduction:

Some organisms produce genetically similar offspring through asexual reproduction. For example; hydra and yeast reproduces asexually by budding. The cells at the surface undergo mitosis and form a mass called bud. Mitosis continues in the cells of bud and it grows into a new individual. The same division happens during asexual reproduction or vegetative propagation in plants and other microbes.

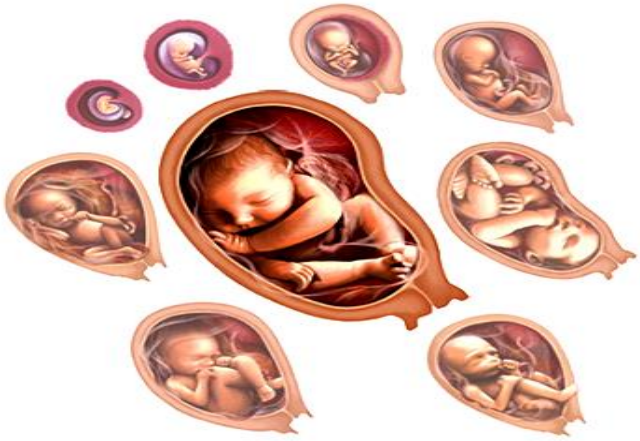


Development and growth:

The number of cells within an organism increase by mitosis. This is the basis of the development of a multicellular body from a single cell i.e., zygote and also the basis of the growth of a multicellular body.

In the fetus, babies and growing children mitosis occurs in most tissues.

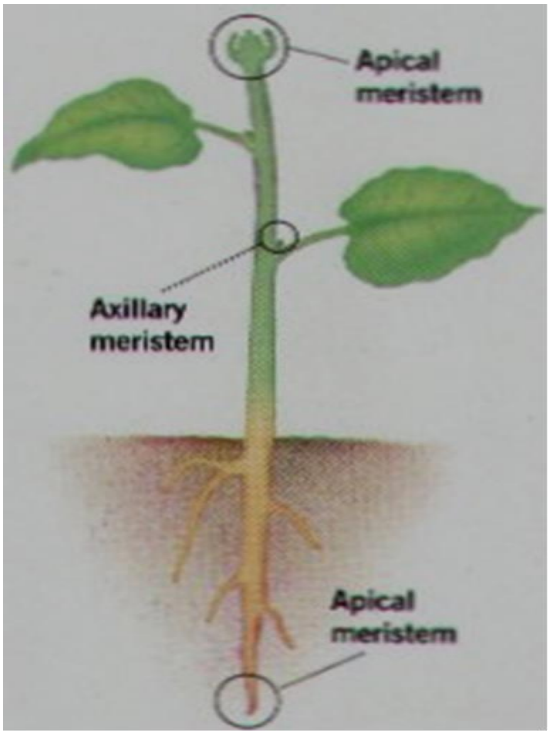
While in adults, however, most tissues do not proliferate but mitosis occurs regularly at the following sites:



1. Red bone marrow – for production of blood cells (erythropoiesis)
2. Lymphoid tissue - formation of lymphocytes (lymphooiesis)
3. Testes – for spermatogenesis (production of spermatozoa)
4. Epidermis - replacement of superficial skin cells
5. Hair follicles - hair growth
6. Gastro-intestinal tract - renewal of epithelium

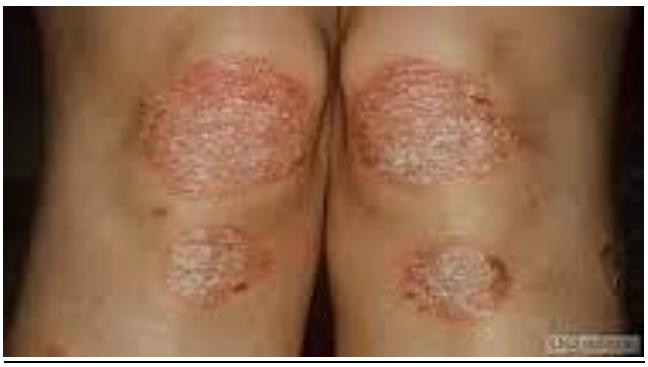
Note that most of the neural cells do not perform mitosis so; any damage in them cannot be repair.

In plants, mitotic cell division mainly takes place in special regions called meristems. They are either present in Shoot apex or axillary buds or root tips of the plants for development and growth.



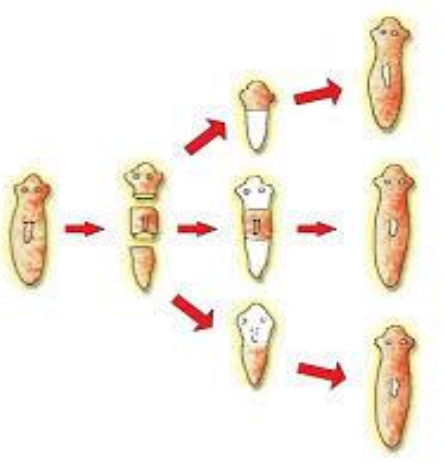
Cell Replacement:

In some parts of body, e.g. skin and digestive tract, cells are constantly sloughed off and replaced by new ones. New cells are formed by mitosis and so are exact copies of the cells being replaced.

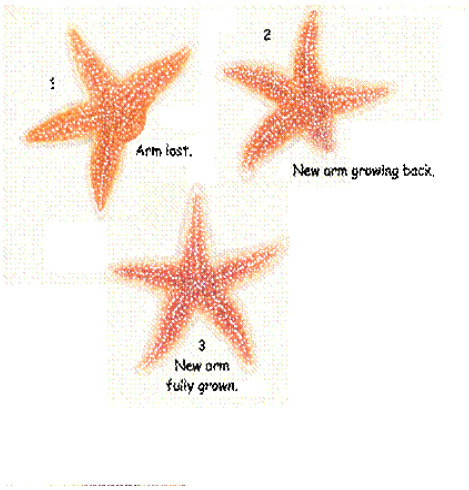


Regeneration:

Some organisms can regenerate (form *de novo*) their parts of bodies. The production of new cells is achieved by mitosis. For example; hydra, sea star and flat worms regenerate their lost part through mitosis.



Flat worm



Sea Star