Medical Genetics Course

DNA packaging, chromosomes and cell division

Lecture 2

maalshboul@just.edu.jo

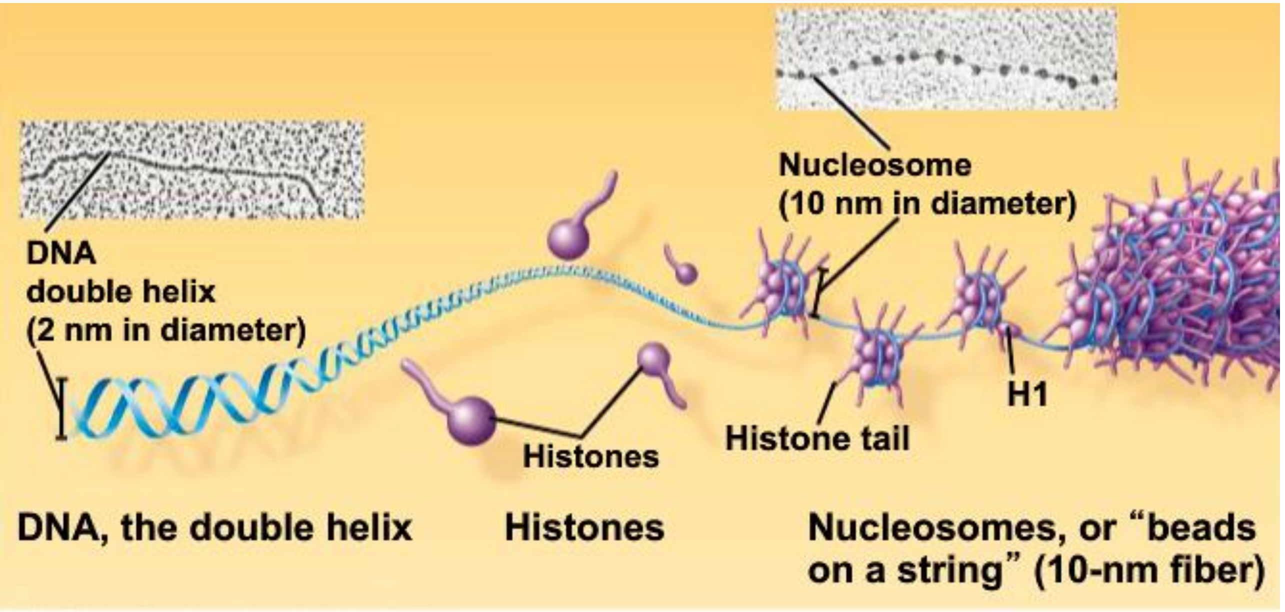
Learning points for this lecture

- Describe how chromosomes are packed during cell division
- Describe the results of mitosis and meiosis, and how these are achieved

Chromosome Structure

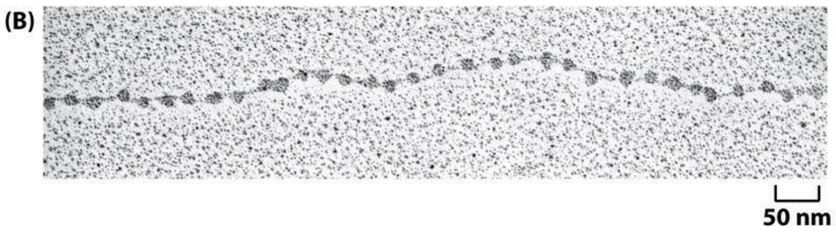
Chromatin: nuclear DNA plus all the proteins bound to it

- 2 main groups of proteins involved in folding/packaging eukaryotic chromosomes
  - **Histones** = positively charged proteins filled with amino acids lysine and arginine that bond
  - Histone protein sequence is highly conserved among eukaryotes
  - DNA is wound around histone proteins to produce nucleosomes
- **Nonhistones**
- less positive (Many are acidic and negatively charged
- Highly variable in cell types, organisms, and at different times in the same cell type
- have role in compaction



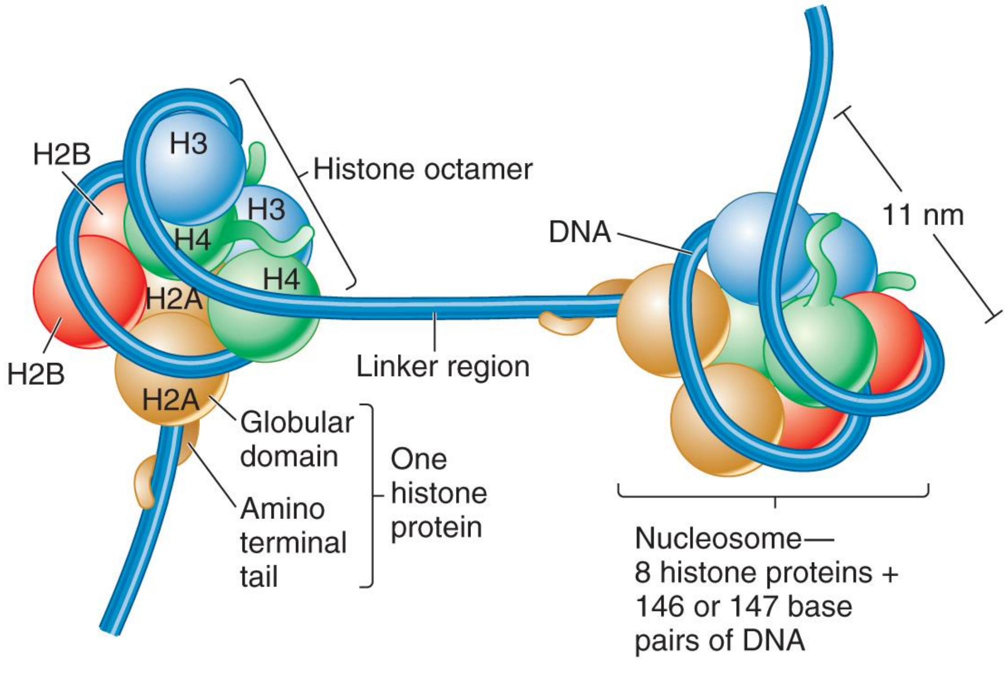
Model for Chromatin Structure

- Chromatin is linked together every 200 bps
- Chromatin arranged like “beads on a string” (electron microscope)

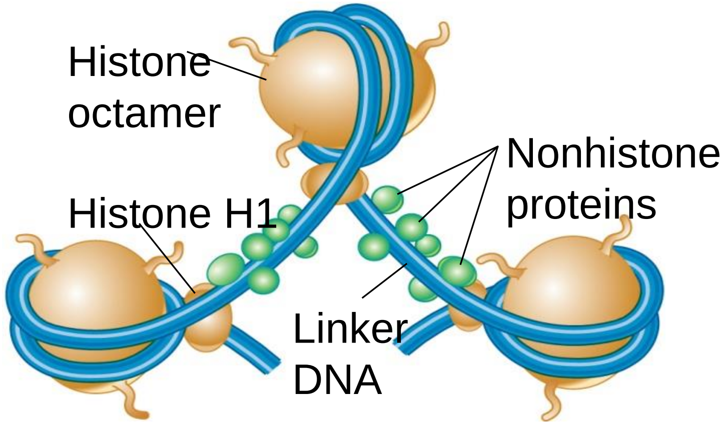


Nucleosome

- 8 histones in each nucleosome
- 146 bps per nucleosome core particle with 53 bps for linker DNA (H1)



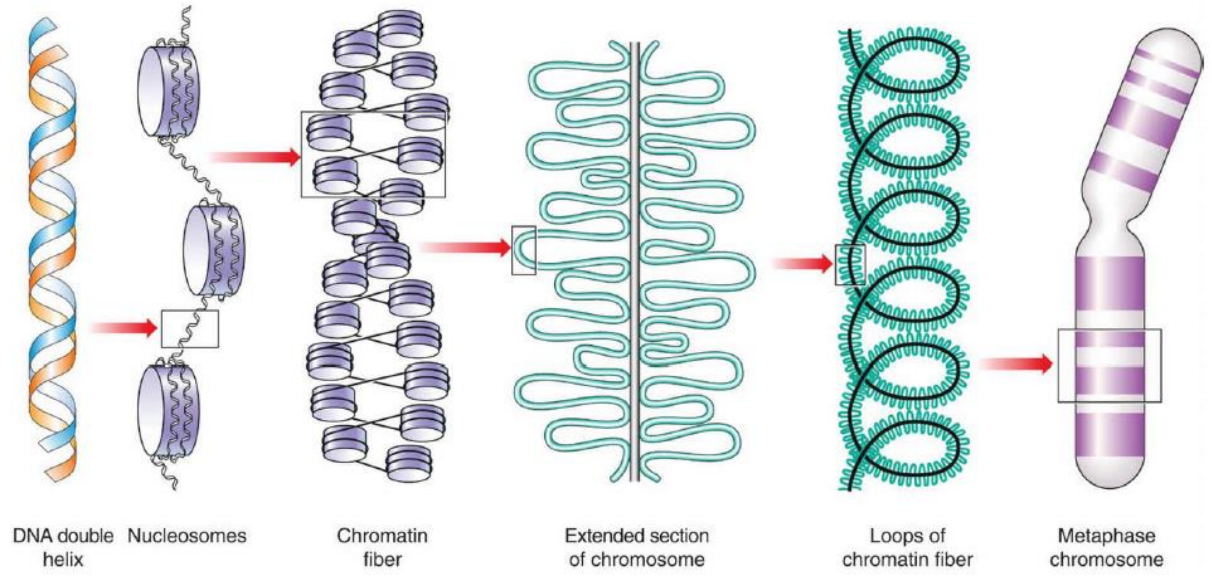
(a) Nucleosomes showing core histone proteins

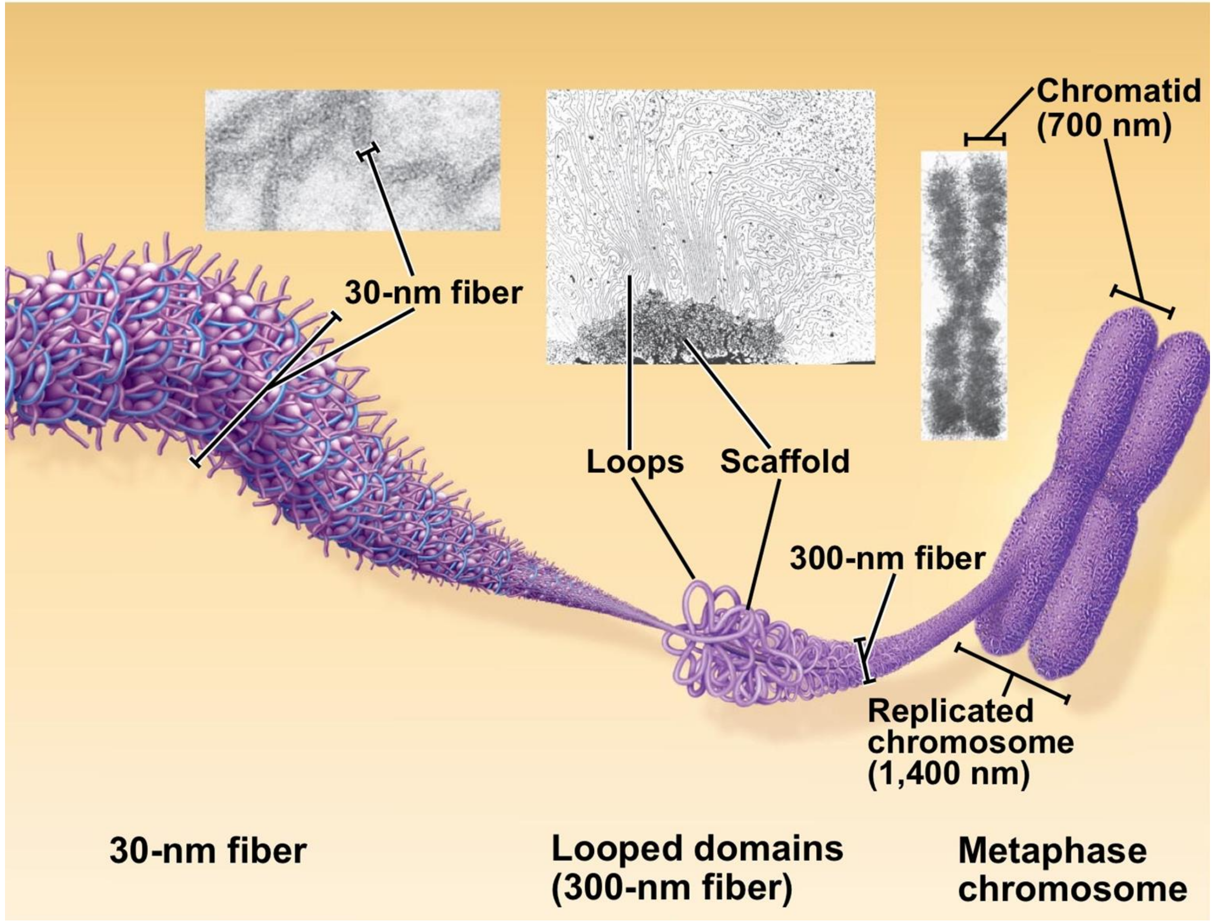


(c) Nucleosomes showing linker histones and nonhistone proteins

histone octomer
Chromatosome!

The packaging of DNA into chromosomes involves several orders of DNA coiling and folding

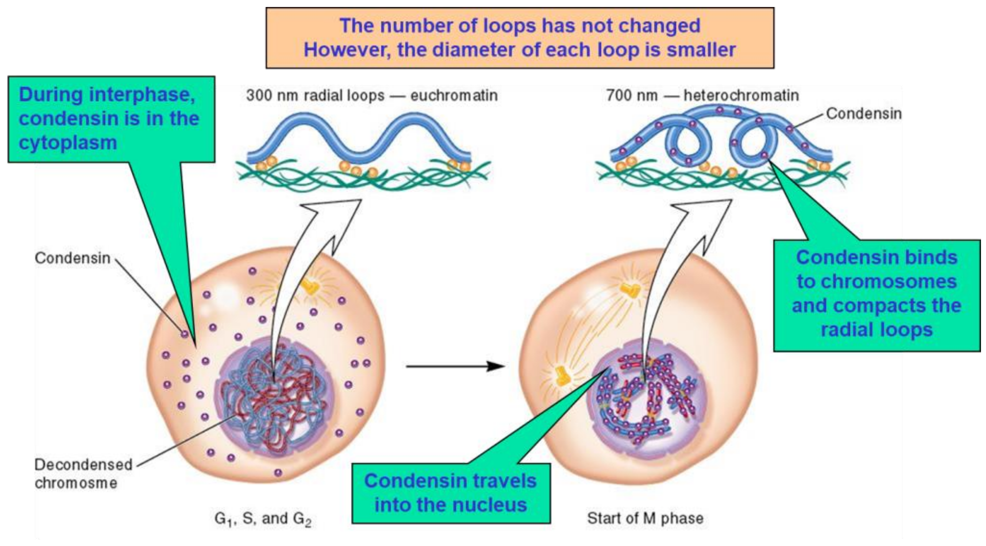


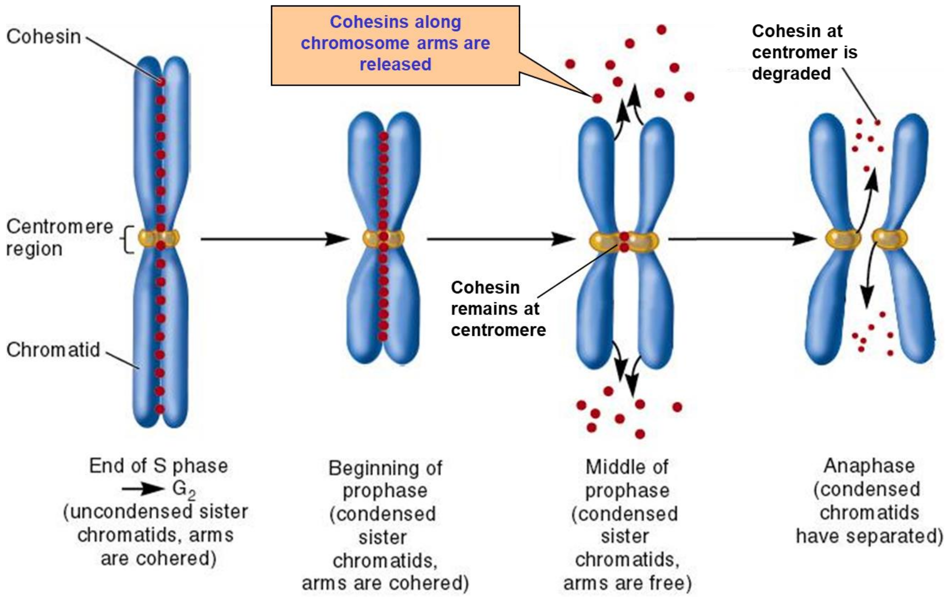


- DNA is further compacted when the DNA nucleosomes associate with one another to produce 30 nm chromatin (Solenoid model)-6/turn
- Is probably the major type of chromatin in the nucleus during interphase
- **DNA supercoiling** is very important for DNA packaging by reducing the space required by the DNA.

The formation of metaphase chromosomes is also promoted by two multi protein complexes
(SMC proteins -> structural maintenance of chromosomes):

- Condensin: are responsible for condensing chromosomes at mitosis.
  - group of proteins that bind to chromosomes as a cell enters prophase, causing the chromosomes to become more compact and visible under a light microscope.
- Cohesin:
  - Promote binding of the sister chromatids after S phase and until the middle of the prophase.
  - At anaphase the cohesins bound to the centromere are degraded by a specific protease.





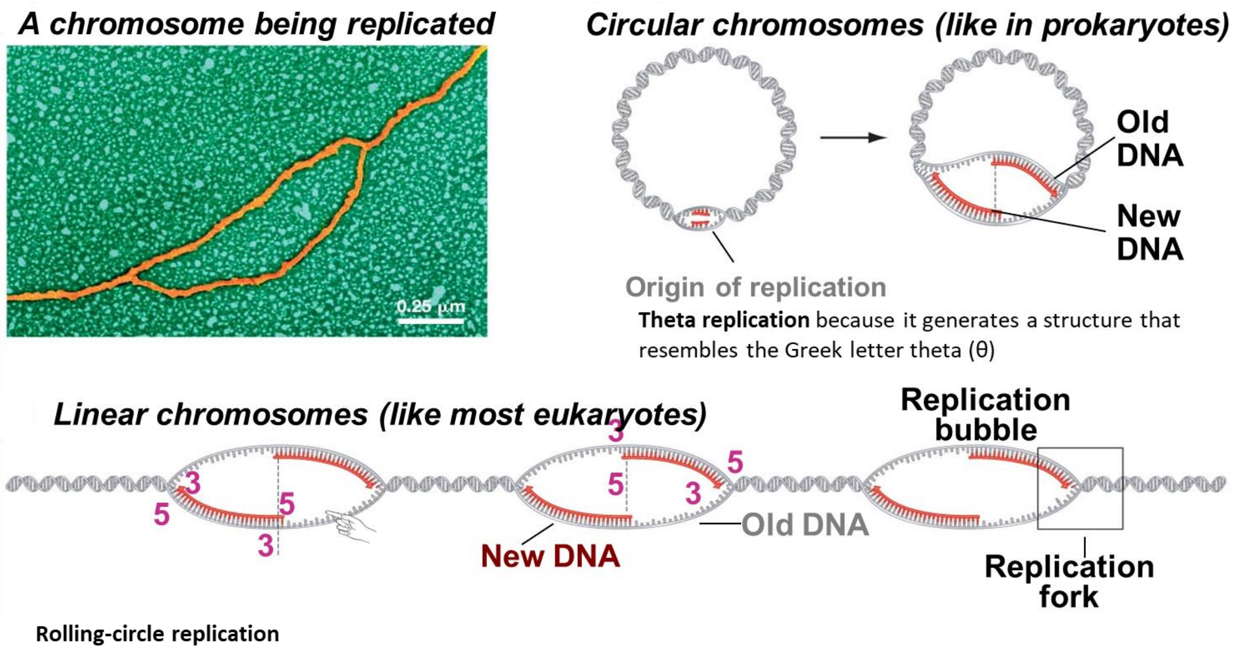
Chromatin Is Coiled into Higher-Order Structures

- Interestingly, histone H1 is very important in stabilizing chromatin higher-order structures, and 30-nanometer fibers form most readily when H1 is present.
- Processes such as **transcription and replication require the two strands of DNA to come apart temporarily**, thus allowing polymerases access to the DNA template. However, the presence of nucleosomes and the folding of chromatin into 30-nanometer fibers pose barriers to the enzymes that unwind and copy DNA.
- It is therefore important for cells to have means of opening up chromatin fibers and/or removing histones transiently to permit transcription and replication to proceed.

- Chromosomes contain three types of regions that are required for replication and segregation:

1. **Origins of replication!**

- Replication origins—certain DNA sequences along each chromosome at which DNA replication can be initiated;



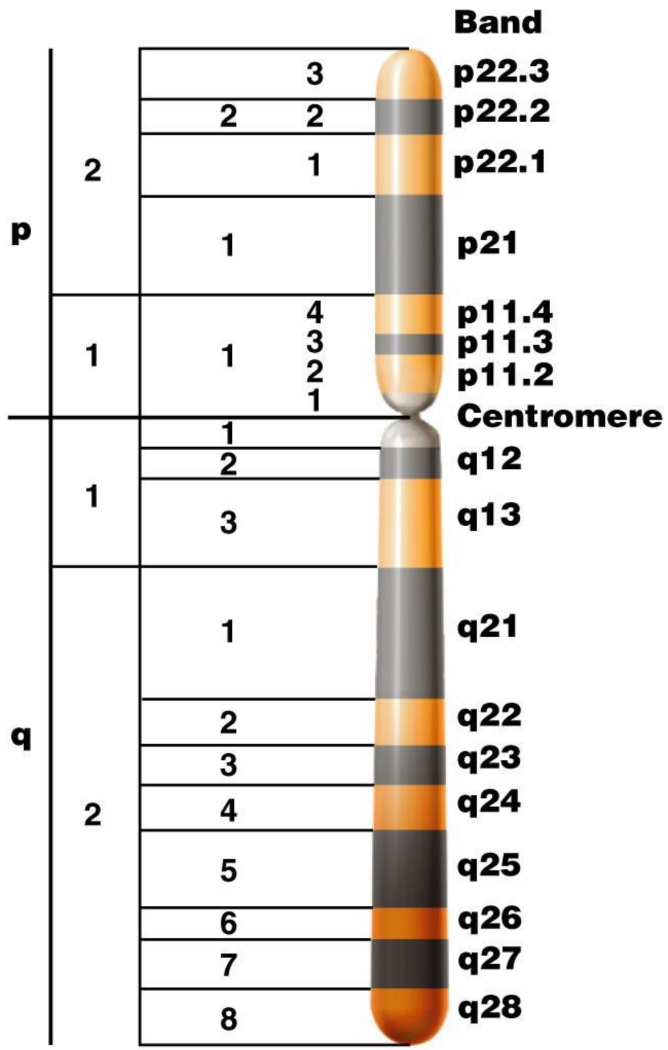
Why do eukaryotic chromosomes have multiple origins?

Why is replication able to go in both directions?

- Chromosomes contain three types of regions that are required for replication and segregation:

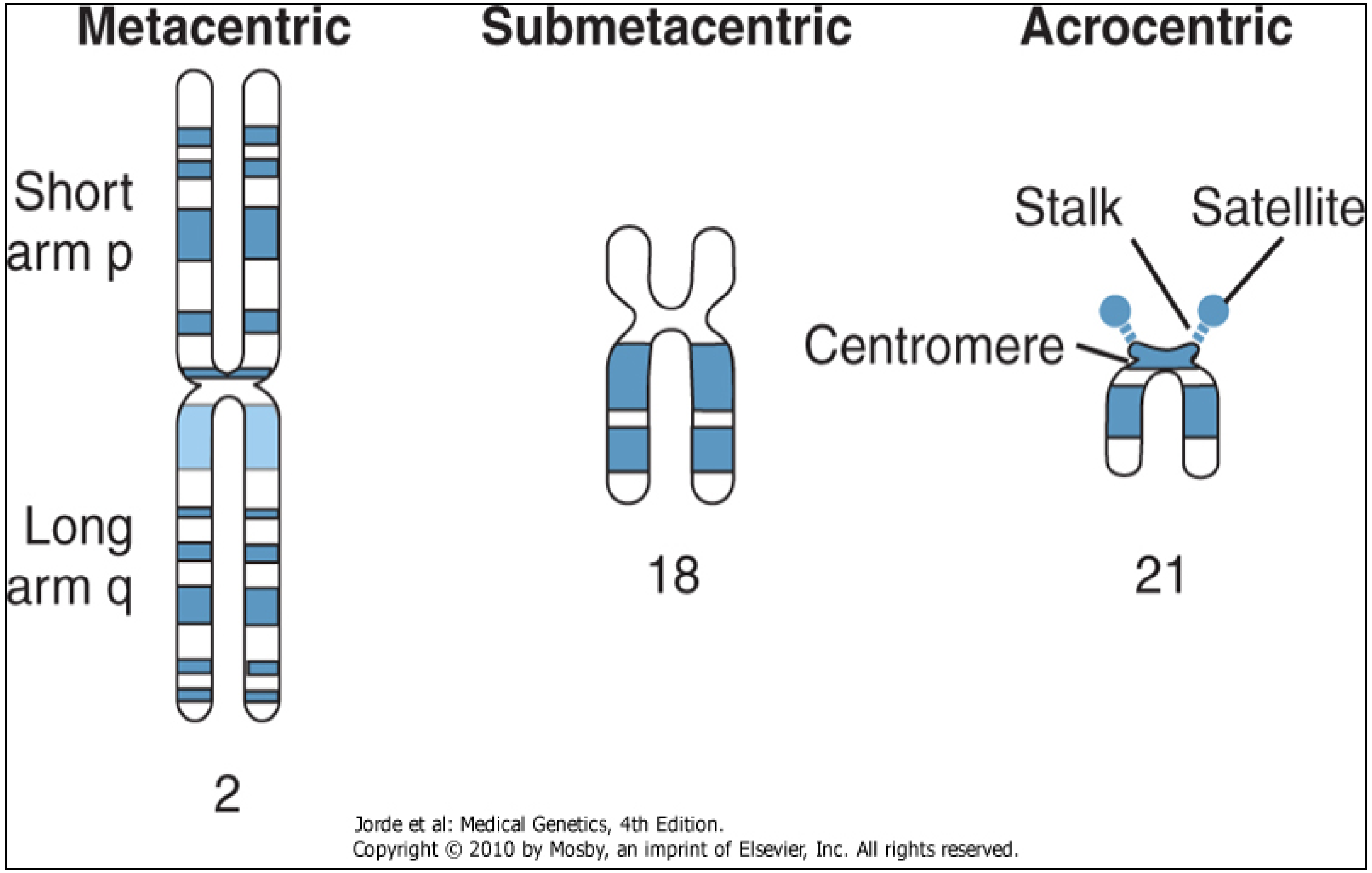
2. Each chromosome has a constriction point called the centromere, which divides the chromosome into two sections, or “arms.”
    - The short arm of the chromosome is labeled the “p arm.”
    - The long arm of the chromosome is labeled the “q arm.”

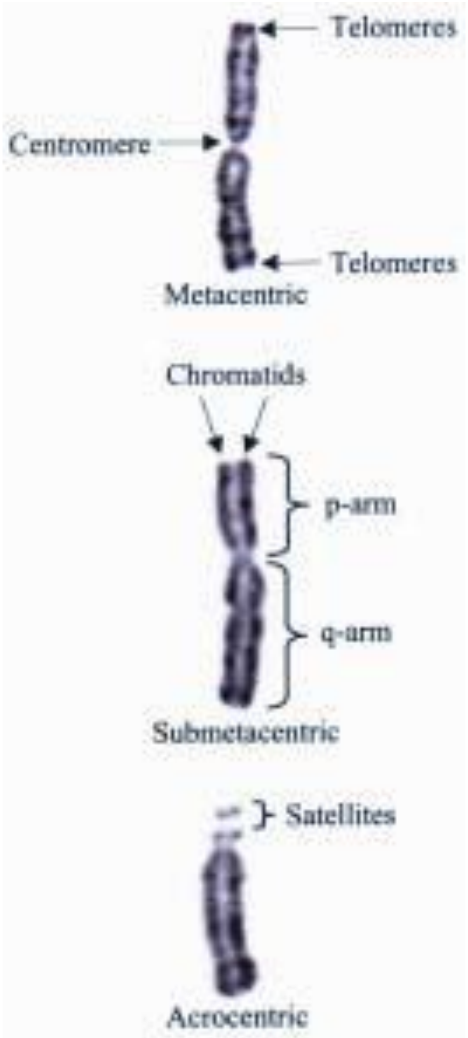
- The location of the centromere on each chromosome gives the chromosome its characteristic shape, and can be used to help describe the location of specific genes
- The centromere is essential for chromosome segregation. And is used for the formation of the kinetochore: A group of proteins that link the centromere to the spindle apparatus. (ensuring proper segregation)!



Chromosome Shape

Metacentric- centromere is located in the middle of chromosome
Submetacentric- centromere is displaced from the center
Acrocentric – centromere is placed near the end





3. The **telomere** is essential for the stability of the chromosome tips.

- Each end of a linear chromosome is composed of a special DNA–protein structure called a telomere
- Telomeres -> prevent chromosomal rearrangements so brings stability. It prevents chromosomes shortening: They protects the chromosome from digestion via enzymes and the telomeres do replicate so the chromosomes do not become shortened

Packaging of DNA into Chromosomes

- The compaction of interphase chromosomes is not completely uniform

Heterochromatin

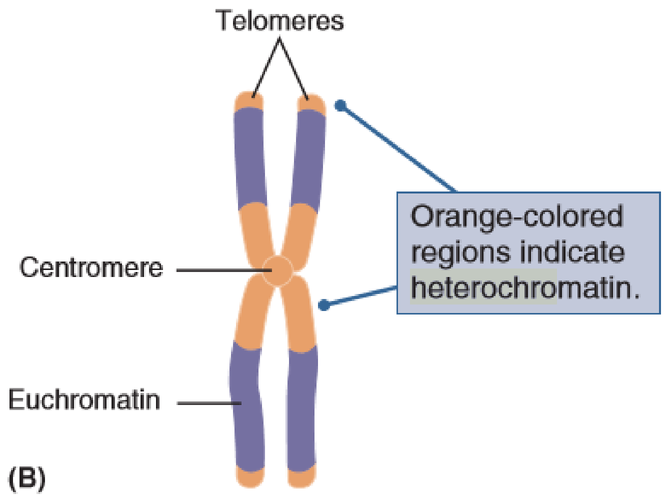
- diff types of condensed chromatin
- Darkly stainable by many standard dyes used to make chromosomes
- Represents 10% of chromosome
- Tightly compacted regions of chromosomes
- Transcriptionally inactive (in general, do not always encode proteins.)
- Responsible for function of telomeres, centromeres
- Replicate late S phase
- Tight binding of histone H1.

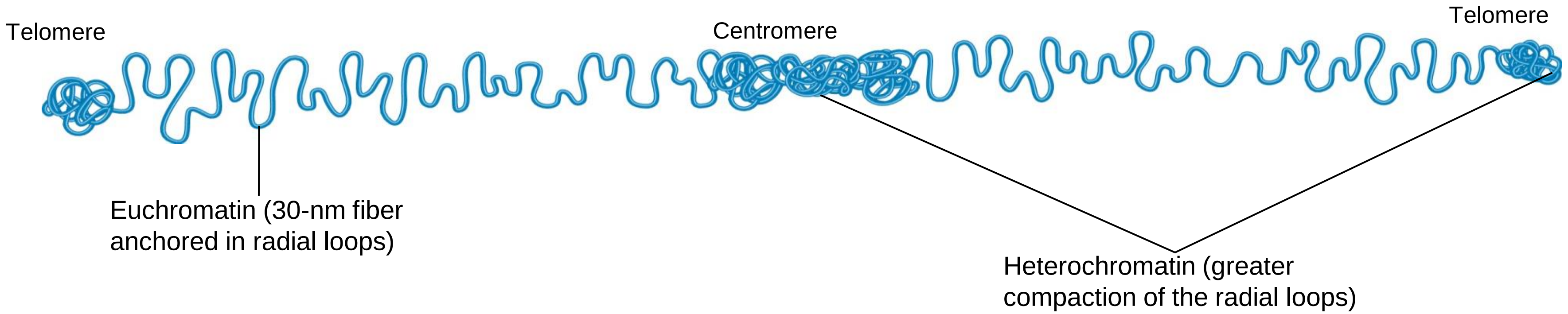
-> Constitutive heterochromatin: the chromosomal regions that are always heterochromatic and permanently inactive.
-> Facultative heterochromatin: chromatin that can occasionally interconvert between hero chromatin and euchromatin (X chromosome converting to a heterochromatic Barr body).

Packaging of DNA into Chromosomes

<u>Euchromatin</u>

- Less condensed regions of chromosomes
- Usually areas where gene expression is occurring
- Composed of all types of chromatin structures- 30 nm fibers, loops, etc
- 90% of chromatin
- Replicate throughout S phase
- Weak binding of histone H1 molecules





Metaphase Chromosomes

- As cells enter M phase, the level of compaction changes dramatically
  - By the end of prophase, sister chromatids are entirely heterochromatic
  - Two parallel chromatids have an overall diameter of 1,400 nm

- These **highly condensed metaphase chromosomes** undergo little gene transcription

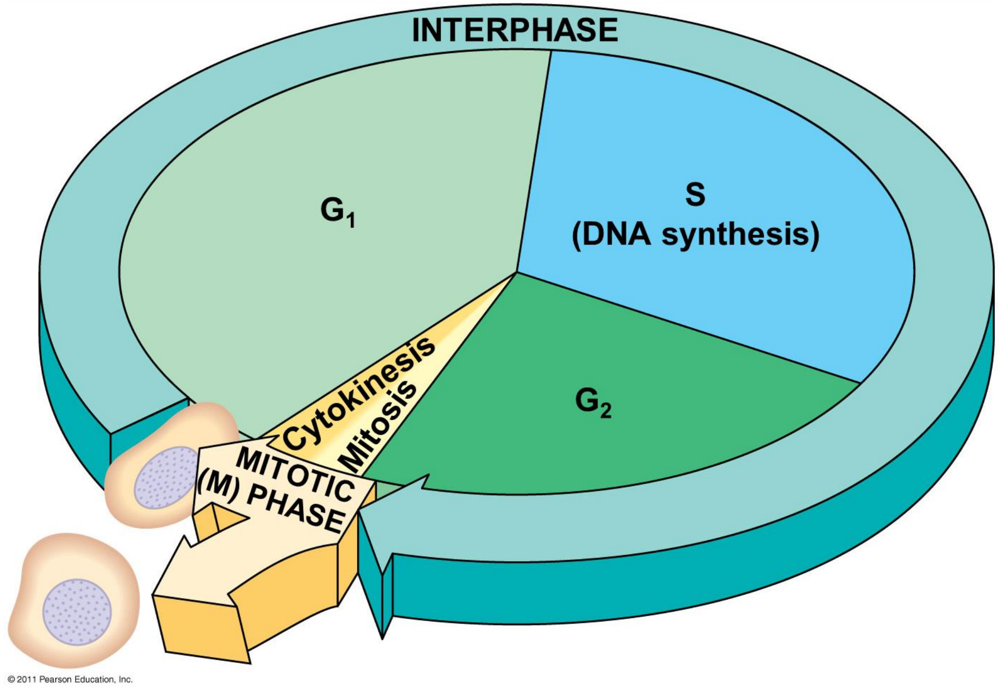


Figure 12.6

Human chromosomes

- DNA and associated proteins are organized into chromosomes
- Human somatic cells are **diploid** and have 22 pairs of **autosomes** AND 1 set of **sex** chromosomes (XX or XY)= total of 46
  - Females XX
  - Males XY
- Germ cells are haploid and contain 22 chromosomes plus 1 sex chromosome (X or Y)

Cell Division

- The aims of cell division:

  1- Replace died cells

  2- Repair of the damaged cells such as wound cells

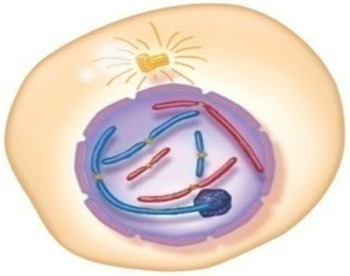
  3- Reproduction of living organisms
- An understanding of cell division is basic to an understanding of cytogenetics. Dividing cells are needed in order to study chromosomes using traditional cytogenetic techniques, and many cytogenetic abnormalities result from errors in cell division.
- **There are two types of cell division: mitosis and meiosis.**
- **Mitosis** is the division of somatic cells, whereas **meiosis** is a special type of division that occurs only in gametic cells.

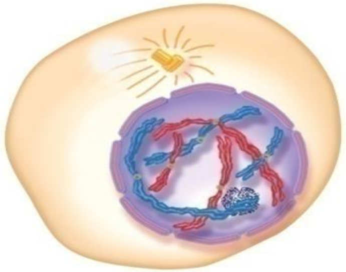
cell cycle

- it is the series of growth and development steps, a cell undergoes between its “birth”—formation by the division of a mother cell—and reproduction—division to make two new daughter cells
- **Stages of the cell cycle**
- To divide, a cell must complete several important tasks: it must grow, copy its genetic material (DNA), and physically split into two daughter cells.
- Cells perform these tasks in an organized, predictable series of steps that make up the cell cycle.
- The cell cycle is a cycle, rather than a linear pathway, because at the end of each go-round, the two daughter cells can start the exact same process over again from the beginning.

- The cell cycle consists of the alternation of cell division (mitosis and cytokinesis) and interphase.
- DNA replication and protein synthesis take place during interphase.

- **Gap 1** (G1)– many cytoplasmic organelles are constructed; RNA, protein and other molecules are synthesized; cell almost doubles in size
- **Synthesis** (S)– DNA is replicated and chromosomes duplicate, forming 2 sister chromatids attached at the centromere
- **Gap 2** (G2)– more cell growth; mitochondria divide; spindle precursors form


G_1 phase


S phase

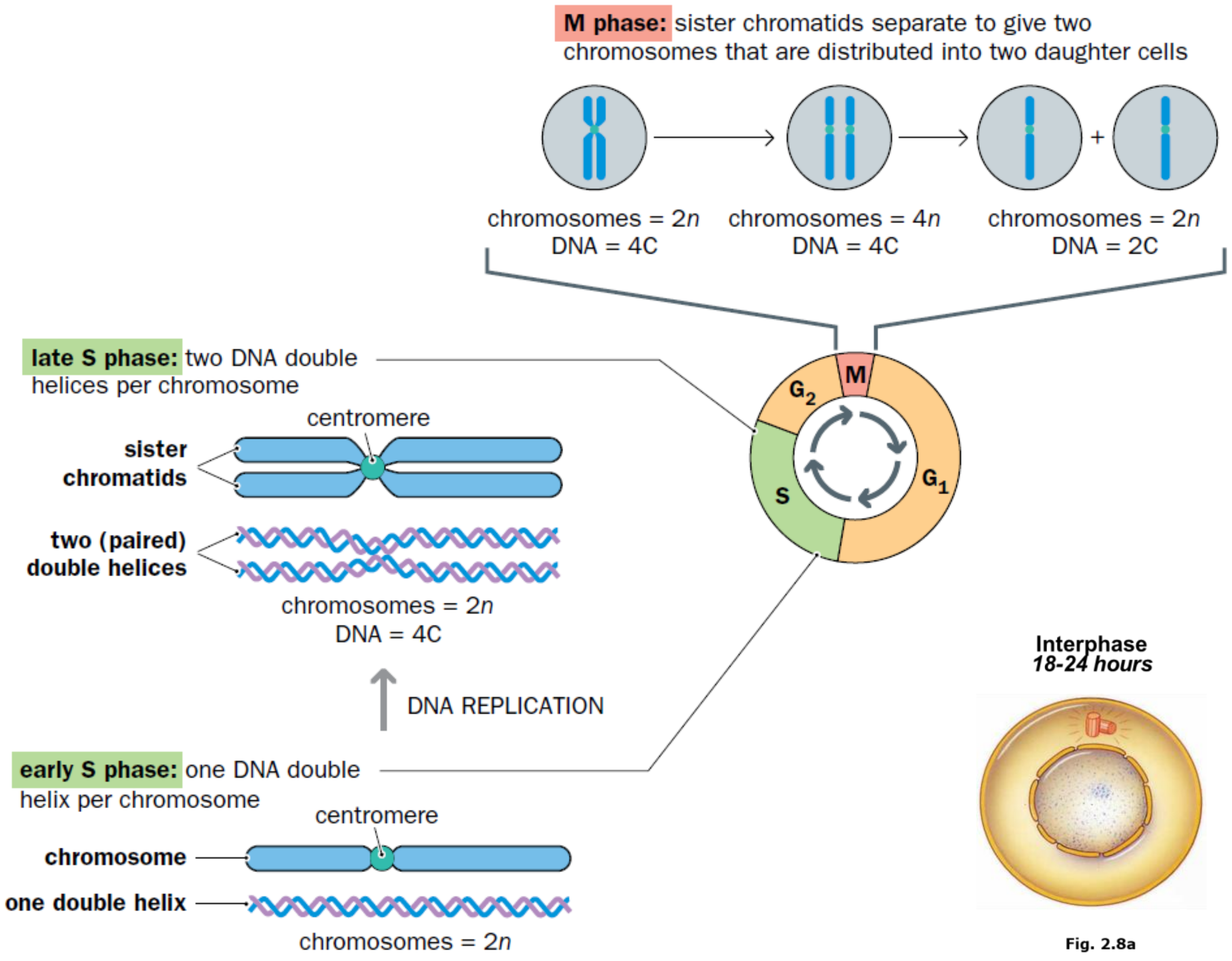


Fig. 2.8a

Mitosis

- It then progresses into the M phase of the cycle where mitosis occurs
- The primary purpose of mitosis is to distribute the replicated chromosomes to the two daughter cells
- This stage known as equational division (Cells have the same equal No. of chromosomes).
  - In humans for example,
    - The 46 pairs of sister chromatids are separated and sorted
    - Each daughter cell thus receives 46 chromosomes

Mitosis has five stages that have distinct cytological characteristics

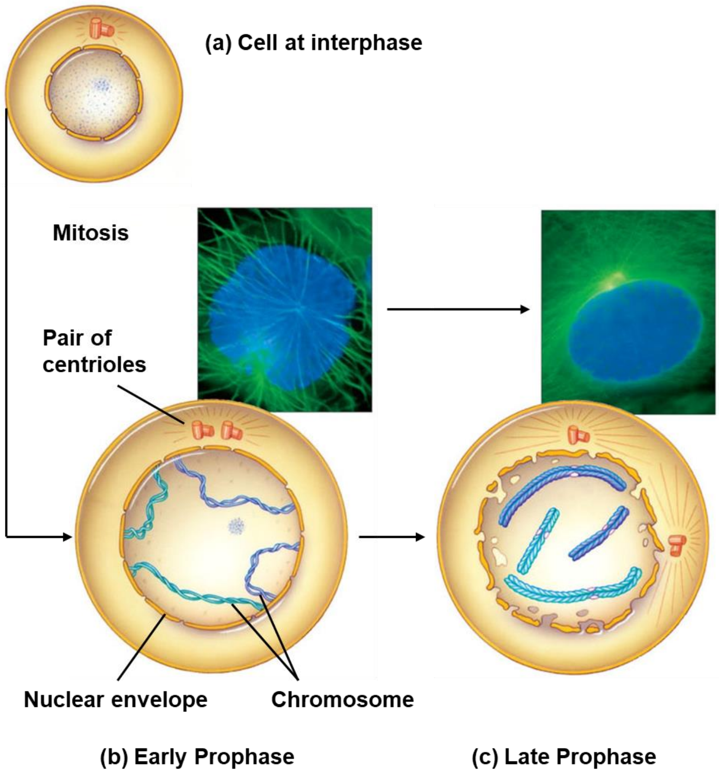
The five stages of mitosis and their major events

- Prophase - chromosomes condense and become visible
- Prometaphase – spindle forms and sister chromatids attach to microtubules from opposite centrosomes
- Metaphase – chromosome align at the cell's equator
- Anaphase – sister chromatids separate and move to opposite poles
- Telophase – chromosomes decondense and are enclosed in two nuclei
- Cytokinesis overlaps the latter stages of mitosis

During Prophase

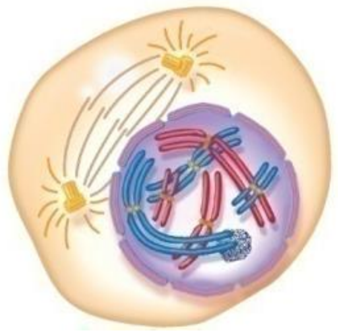
- Chromosomes condense and become visible
- Nuclear envelope dissociates into smaller vesicles
- Centrosomes separate and move apart toward opposite poles
- The mitotic spindle apparatus is formed
  - Composed of microtubules (MTs)

PROPHASE



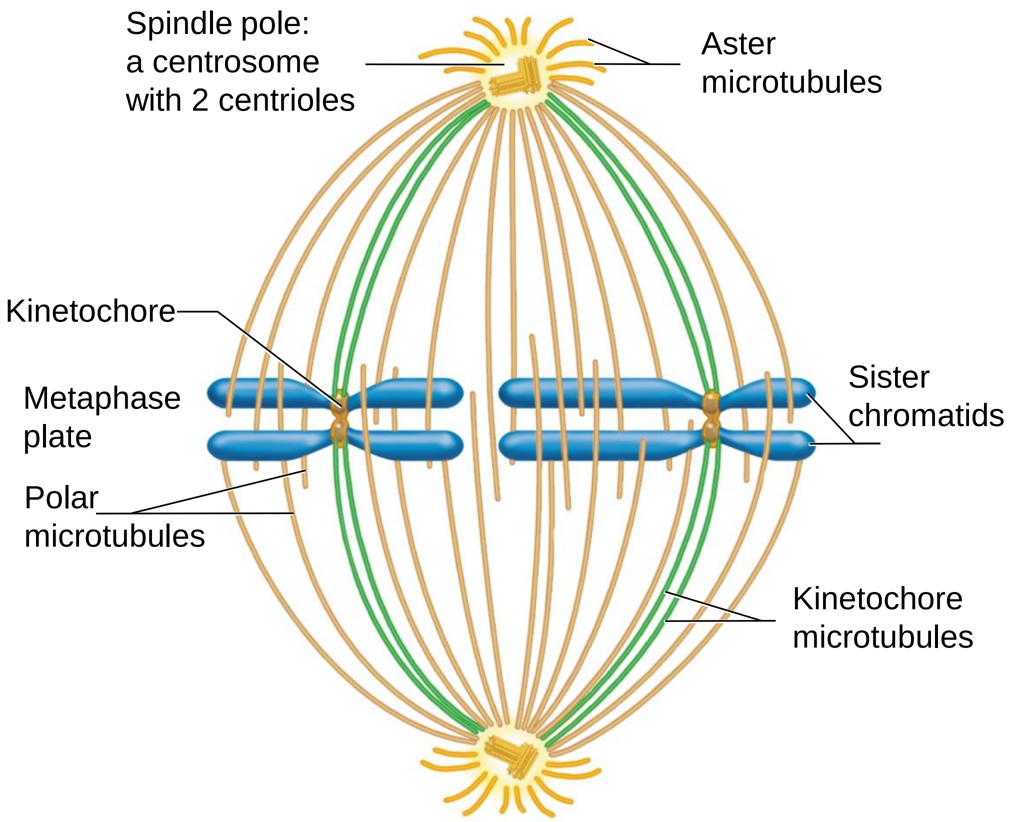
The Mitotic Spindle Apparatus

- Spindle apparatus is formed from microtubule-organizing centers (MTOCs: called centrosomes)
- Microtubules are formed by rapid polymerization of tubulin proteins
  - Centrosomes lie at each spindle pole
  - A pair of centrioles is within each centrosome
- Some plants do not have centrosomes
  - The nuclear envelope functions as an MTOC



The Mitotic Spindle Apparatus

- There are three types of spindle microtubules
  1. Aster microtubules
     - Help position the spindle
  2. Polar microtubules
     - Help to “push” the poles away from each other
  3. Kinetochore microtubules
     - Attach to the **kinetochore**, which is bound to the centromere of each individual chromosome



During Prometaphase

- Nuclear envelope breaks down
- Centrosomes move to opposite ends of the cell, forming the spindle poles
- Spindle fibers interact with the sister chromatids
- Kinetochore microtubules grow from the two poles
  - The two kinetochores on a pair of sister chromatids are attached to kinetochore MTs on opposite poles

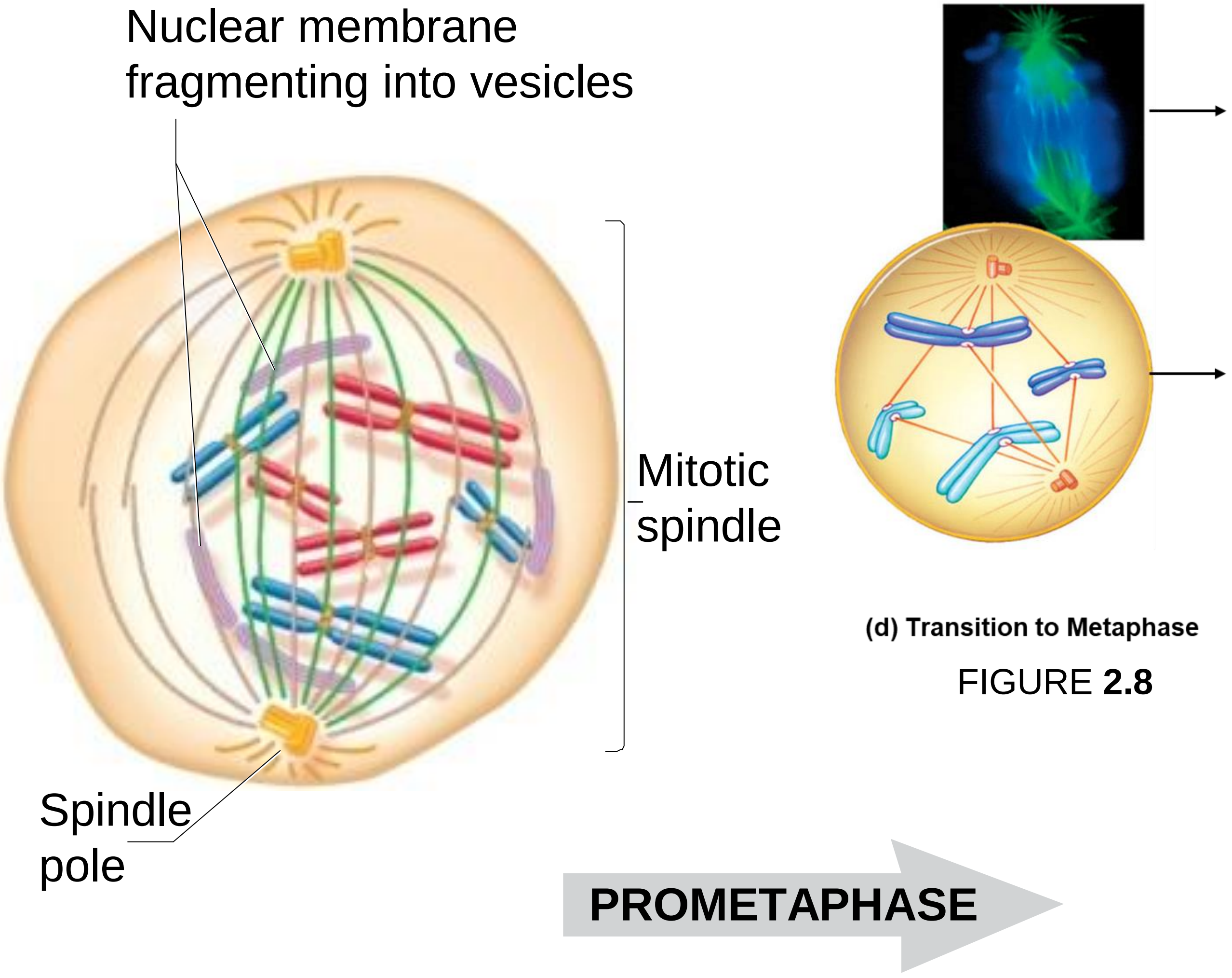
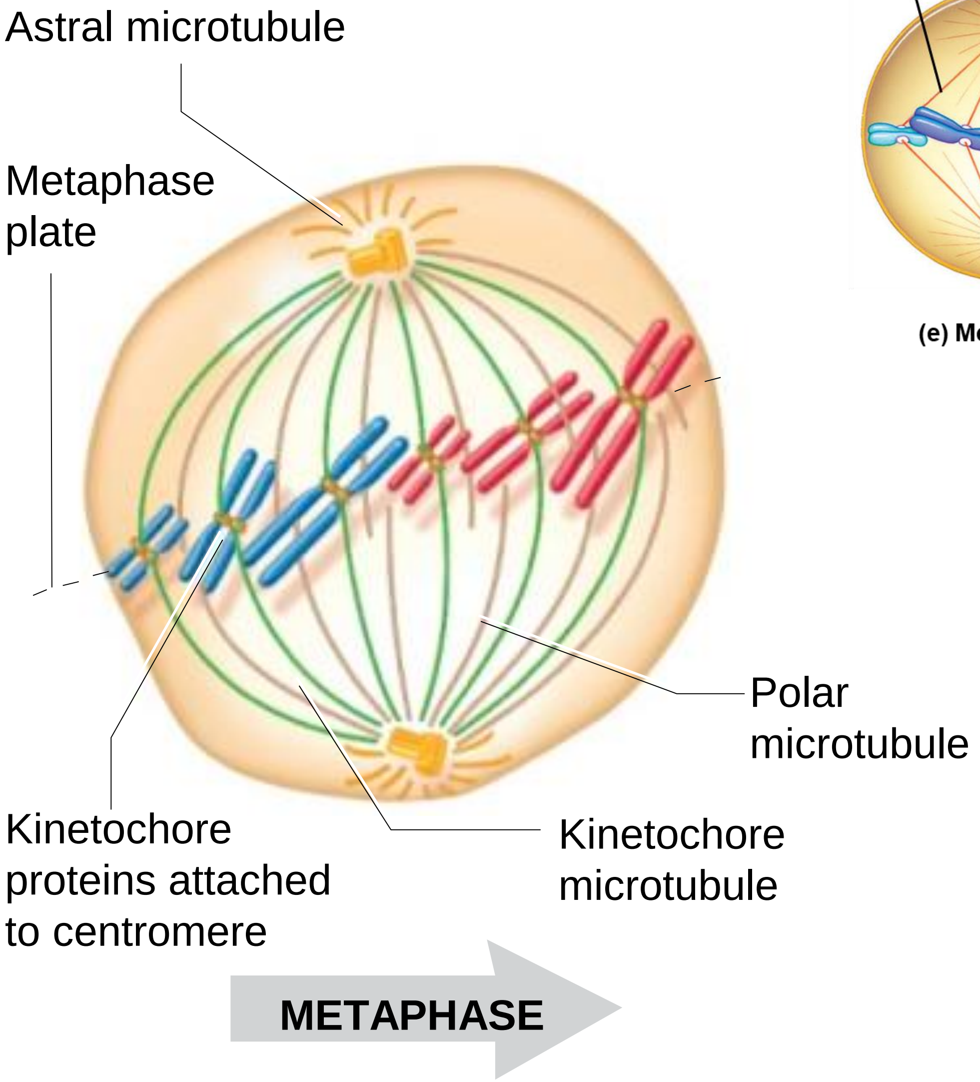
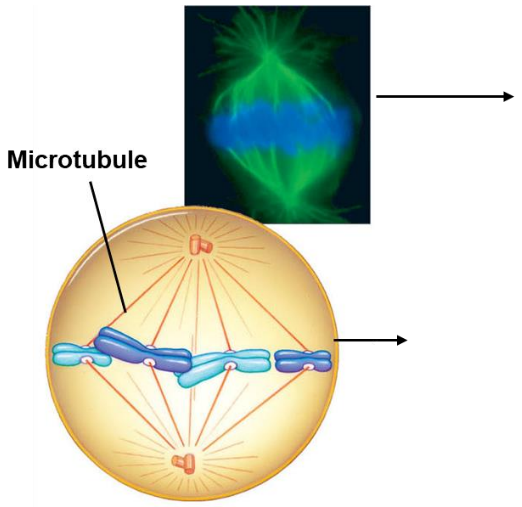


FIGURE **2.8** Stages of mitosis. Only two pairs of chromosomes from a diploid (2*n*) cell are shown here. The photographs show mitosis in a mouse cell; the DNA is stained blue and the microtubules (spindle fibers) are stained green.

During Metaphase

- Chromosomes become arranged a single plane, the metaphase pla between the two centrosomes.
- Each pair of chromatids is attache to both poles by kinetochore microtubules
- Forces pushing and pulling chromosomes to or from each pol are in balanced equilibrium

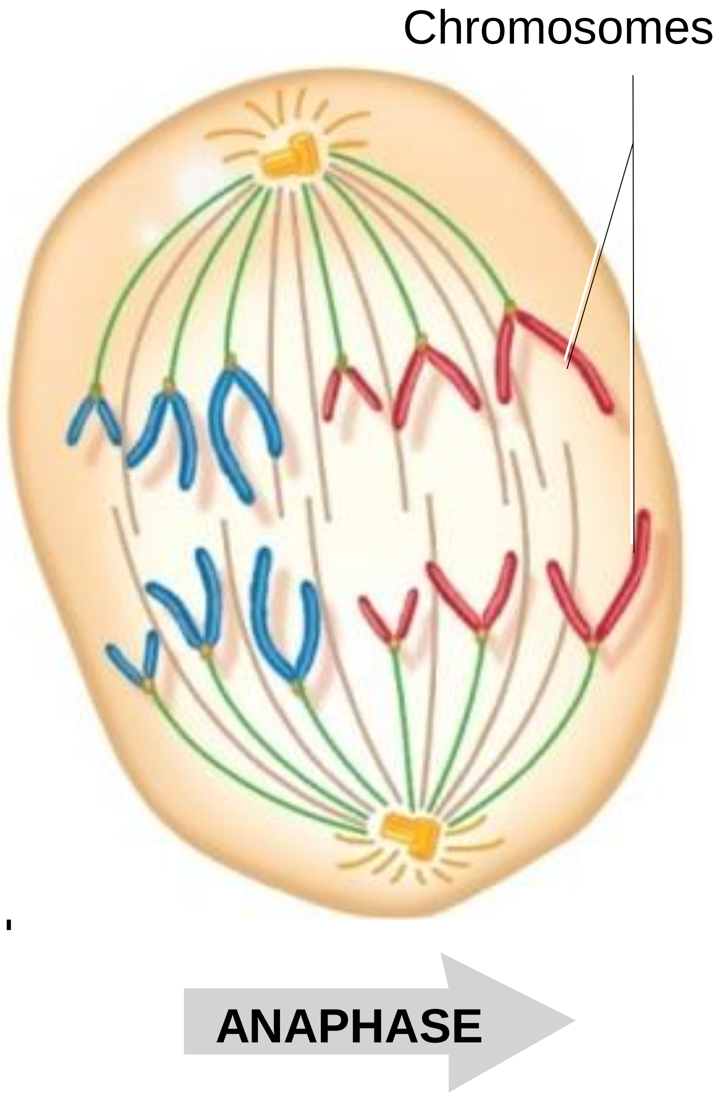


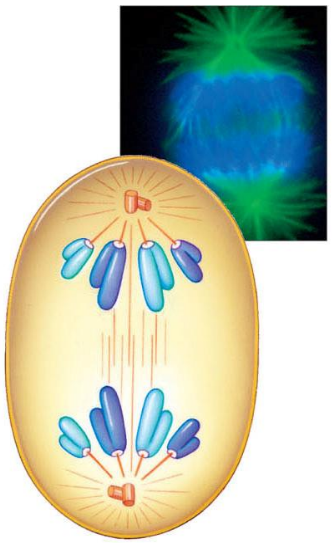


(e) Metaphase

During Anaphase

- Anaphase begins when the sister chromatids separate and move toward opposite spindle poles.
- The connection holding the sister chromatids together is broken
- Each chromatid, now an individual chromosome, is linked to only one pole
- As anaphase proceeds
  - Kinetochore MTs shorten
    - Chromosomes move to opposite poles
  - Polar MTs lengthen
    - Poles themselves move further away from each other





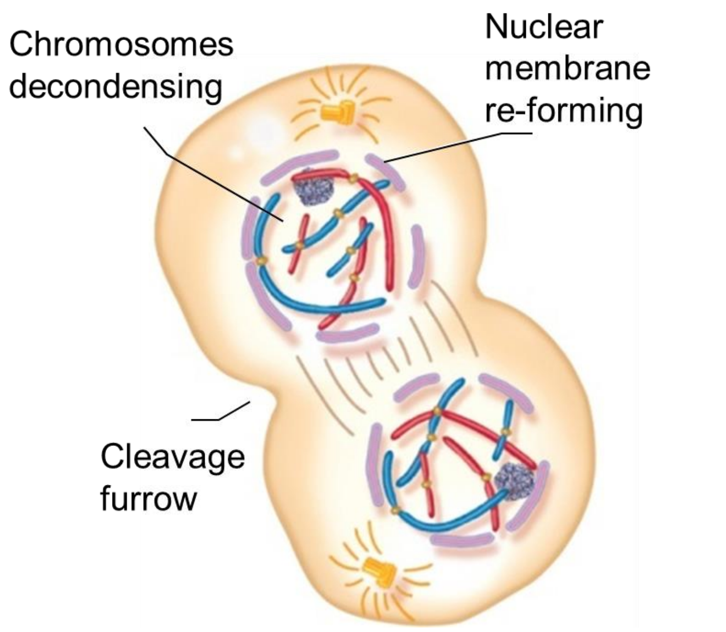
(f) Anaphase

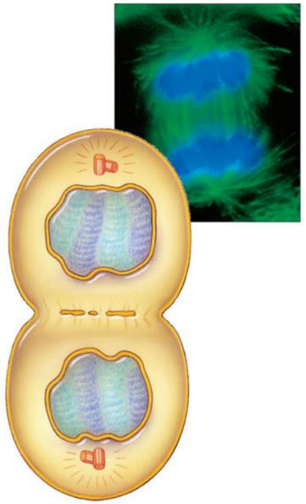
During Telophase & Cytokinesis

The mitotic phase is divided into (2) stages:

- **Mitosis**: the nucleus (and all its contents, including the duplicated chromosomes) divide and are evenly distributed to the ‘daughter’ cells
- **Cytokinesis:** the cytoplasm is divided into 2

  - Chromosomes reach their respective poles
  - Nuclear membrane reforms to form two separate nuclei
  - Spindle fibres disperse
  - The chromosomes relax and lengthen, once again disappearing from view (decondensed).
  - In most cases, mitosis is quickly followed by cytokinesis





(g) Telophase

TELOPHASE AND CYTOKINESIS