

12.1 How Do Cells Replicate?

For life on Earth to exist, cells must replicate. The basic steps in cellular replication are (1) copying the DNA (deoxyribonucleic acid), (2) separating the copies, and (3) dividing the cytoplasm to create two complete cells. This chapter focuses on a process that has been studied for well over a century: how eukaryotic cells replicate. Like much work in biology, the research on eukaryotic cell replication began with simple observations of the process.

What Is a Chromosome?

As studies of cell division in eukaryotes began, biologists found that certain chemical dyes made thread-like structures visible within nuclei. In 1879, Walther Flemming used a dye made from a coal tar to observe these structures and watch them change in the dividing cells of salamander embryos. The threads first appeared in pairs just before cell division and then split to produce single, unpaired threads in the daughter cells. Flemming introduced the term “mitosis,” from the Greek *mitos* (“thread”), to describe this process.

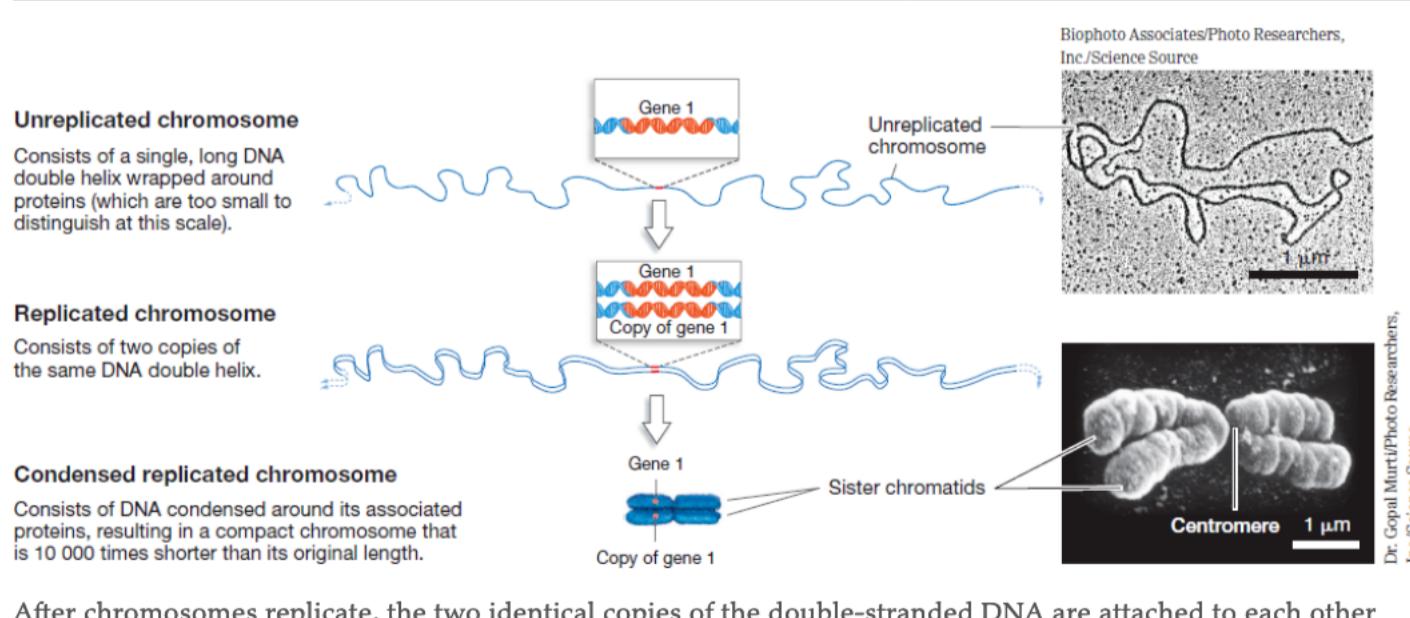
Others studied the roundworm *Ascaris* and noted that the number of threads in a cell was the same before and after mitotic division. All of these cells had the same number of threads.

In 1888, Wilhelm Waldeyer coined the term **chromosome** (‘coloured body’) to refer to these thread-like structures (visible in the chapter-opening photo). Research carried out since then has shown that a chromosome consists of a single long DNA double helix that is wrapped around proteins, called **histones**, in a highly organized manner (see Chapter 19). DNA encodes the cell’s hereditary information, or genetic material. A **gene** is a region of DNA in a chromosome that codes for a particular protein or ribonucleic acid (RNA).

Before mitosis, each chromosome is replicated. As mitosis starts, the chromosomes condense into compact structures that can be moved around the cell efficiently. Then one copy of each chromosome is distributed to each of two daughter cells.

Figure 12.1 illustrates an unreplicated chromosome, the same chromosome after it has been replicated, and the replicated chromosome that has condensed at the start of mitosis. Each of the double-stranded DNA copies in a replicated chromosome is called a **chromatid**. The two chromatids are held together by proteins at a large DNA region called the **centromere** (‘centre part’). Centromeres are often, but not always, found in the middle of chromosomes. Even though a replicated chromosome consists of two chromatids, it is still considered a single chromosome. During mitosis the “sister” chromatids are separated, at which time they become “daughter” chromosomes.

Figure 12.1 Changes in Chromosome Morphology.



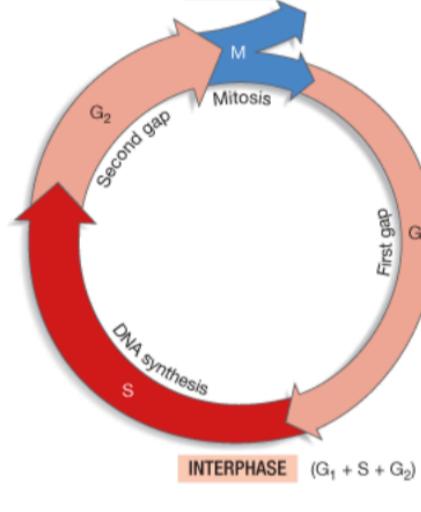
After chromosomes replicate, the two identical copies of the double-stranded DNA are attached to each other along their entire length. Early in mitosis, replicated chromosomes condense and sister chromatids remain attached at a region called the centromere.

If you understand how a chromosome can be made of one or two pieces of DNA, you should be able to draw a model to represent a cell with two different chromosomes before and after the chromosomes are replicated. Use a circle to represent the cell and one of the models above to represent the chromosomes. Label the chromatids.

The Cell Cycle

Figure 12.3 pulls these results together into a comprehensive view of the cell cycle. The cell cycle involves four phases: M phase and an interphase consisting of the G₁, S, and G₂ phases. In the cycle diagrammed here, G₁ phase is about twice as long as G₂ phase, but their actual durations vary depending on the cell type and growth conditions.

Figure 12.3 The Cell Cycle Has Four Phases.



The duration of the G₁ and G₂ phases varies dramatically among cells and organisms.

Why do the gap phases exist? In multicellular organisms, cells perform their functional roles mostly during G₁ phase. G₁ is also the period when the cell “decides” to begin replication and transitions to S phase (as will be explained in [Section 12.3](#)). Before mitosis can take place, a cell uses G₂ phase to prepare for M phase. The time spent in both G₁ and G₂ allows the cell to grow and replicate organelles so it will be able to divide into two cells that can function normally.

Now let's turn to M phase. Once the genetic material has been copied in S phase, how is it divided between daughter cells?

Events in Mitosis

Section 12.2 What happens during M phase?

261

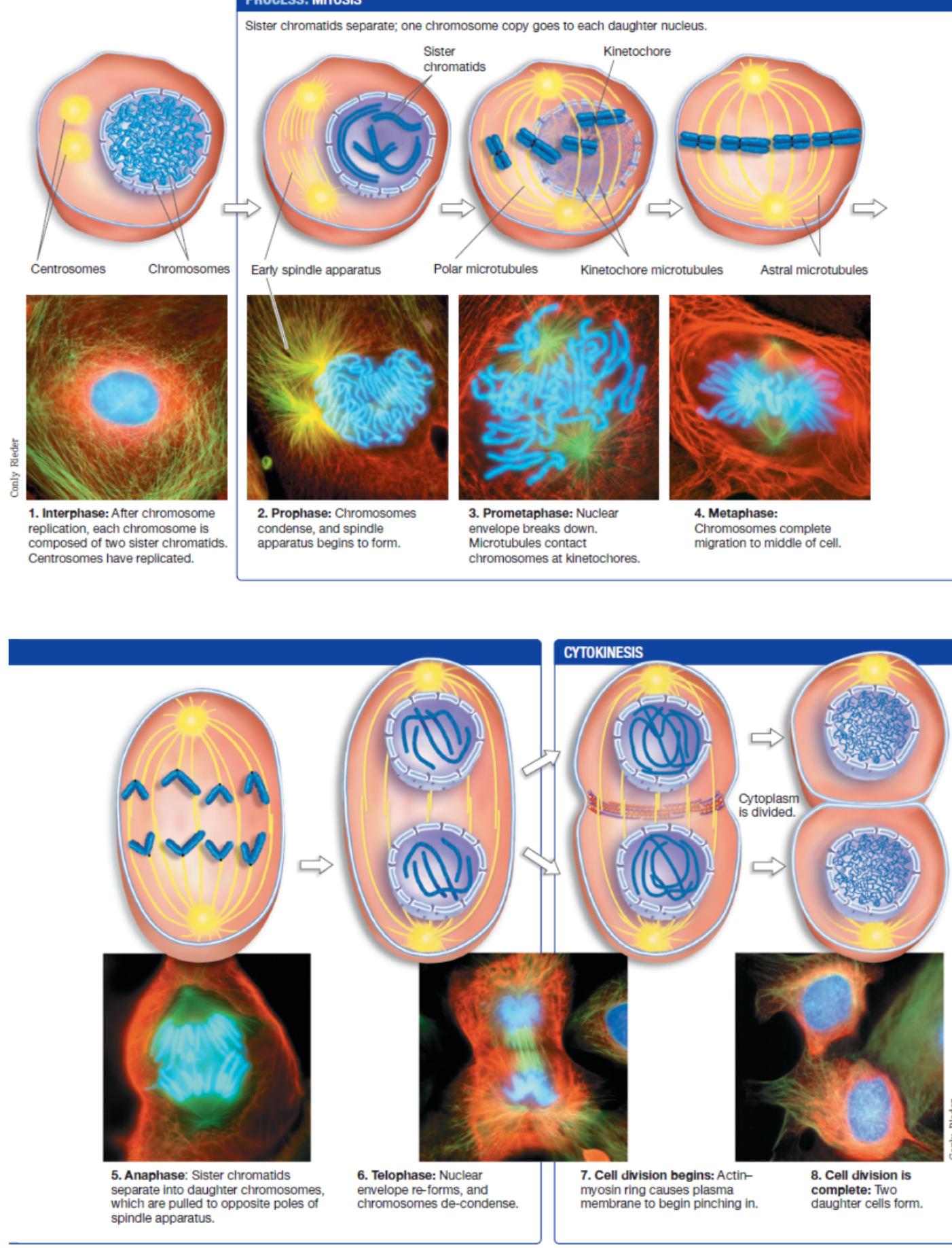
Although mitosis is a continuous process, biologists identify five subphases within M phase on the basis of distinctive events that occur. Some students use the mnemonic device IPPMAT to remember that interphase is followed by the mitotic subphases of prophase, prometaphase, metaphase, anaphase, and telophase.

While mitosis is a complex process, keep in mind its function. At the start of mitosis the cell has a single nucleus and at the end it has two. Mitosis is the replication of a eukaryotic cell's most complex organelle—its nucleus. This is an essential step that must occur before it can divide into two independent cells.

Interphase

To successfully complete mitosis a cell needs to have completed three tasks: (1) DNA replication, (2) cell growth, and (3) microtubule-organizing centre (MTOC) replication (Figure 12.5, step 1). We've discussed the first two events already in this chapter but what of MTOC replication? Recall from Chapter 7 that microtubules are often connected at their bases to MTOCs. Animal and fungal cells have single MTOCs called **centrosomes**. Centrosomes are large protein complexes that contain small bundles of microtubules known as centrioles. Plant cells have hundreds of smaller MTOCs.

Figure 12.5 Mitosis and Cytokinesis.



In the micrographs of newt lung cells under the drawings, chromosomes are stained blue, microtubules are yellow/green, and intermediate filaments are red.

CAUTION If the cell shown in the micrographs has 60 picograms of DNA (6×10^{-11} g) and 22 chromosomes in its G₁ phase, how much DNA and how many chromosomes are in (1) the prophase cell, (2) the anaphase cell, and (3) each daughter cell?

An animal cell that is committed to cell division replicates its centrosome in the cytosol at the same time as it replicates its DNA within its nucleus. The result is the cell shown in [Figure 12.5](#), step 1—the chromosomes are replicated and there are two centrosomes. The centrosomes will play important roles at each step of mitosis and each daughter cell will end up inheriting one.

Plant cells don't have centrosomes but they still need to organize their microtubules during mitosis. Scientists studying the model plant *Arabidopsis* (see [BioSkills 9](#)) recently found out how this occurs. This plant, and presumably others like it, uses a protein called NEDD1. Just before mitosis begins these proteins accumulate on the surface of the nuclear envelope. They form two patches on opposite sides of the nucleus. Microtubules then radiate outwards from these complexes. These structures remain in place for the rest of mitosis, even after the nuclear envelope is temporarily dismantled. NEDD1 protein complexes are not as large as centrosomes but are similar in composition and perform many of the same tasks during mitosis. Other organisms, including animals, use NEDD1 proteins in various MTOCs.

Prophase

Mitosis begins with the events of **prophase** ("before phase"; [Figure 12.5](#), step 2), when chromosomes condense into compact structures. Chromosomes first become visible in the light microscope during prophase.

In the cytoplasm, prophase is marked by the formation of the spindle apparatus. The spindle apparatus is a structure that produces mechanical forces that (1) pull chromosomes to the poles of the cell during mitosis and (2) push the poles of the cell away from each other.

The **spindle apparatus** consists of distinct populations of microtubules anchored at their base to a centrosome (in an animal cell) or NEDD1 complex (in a plant cell). Depending upon the cell type there will be two or three types:

1. **Polar microtubules** extend outwards and overlap with other polar microtubules attached to the opposite MTOC.
2. **Kinetochore microtubules** connect to chromosomes.
3. **Astral microtubules** are found only in animal cells and connect the centrosome to proteins on the inner surface of the plasma membrane.

During prophase in animal cells, the centrosomes move away from each other. In plants and other eukaryotes, the MTOCs are already at opposite sides of the cell.

Prometaphase

As the chromosomes become completely condensed, the nuclear envelope begins to disappear. Microtubules extend into the middle of the cell. Some contact chromosomes and become kinetochore microtubules, while others contact microtubules coming from the opposite side of the cell and become polar microtubules. These events occur during **prometaphase** ("before middle phase"; [Figure 12.5](#), step 3).

Note how the kinetochore regions on each chromosome appear as a constriction. It can't be seen in this figure but there are two kinetochores on each chromosome, one on each chromatid. Microtubules from each pole can only attach to one chromatid or the other.

Early in mitosis, kinesin and dynein motors are recruited to the kinetochore, where they can "walk" the chromosome up and down microtubules. These motors are thought to be very important in the initial attachment of the kinetochore to the plus end of the microtubule. If these ideas are correct, then the process is similar to the way these motors walk along microtubules during vesicle transport (see [Chapter 7](#)).

In all eukaryotes, after the kinetochores have attached to microtubules, chromosomes begin to move to the middle of the cell during prometaphase.

Metaphase

Once the kinetochore microtubules have moved all the chromosomes to the middle of the spindle ([Figure 12.5](#), step 4), the mitotic cells enter **metaphase** ("middle phase"). At this point, the chromosomes are lined up along an imaginary plane between the two spindle poles called the **metaphase plate**.

The formation of the spindle apparatus is now complete. The polar microtubules that extend from each spindle pole overlap in the middle of the cell, thereby forming a pole-to-pole connection.

Each chromosome is held by kinetochore microtubules reaching out from opposite poles and exerting the same amount of tension, or pull. In animal cells, the centrosomes are held in place by the astral microtubules that interact with proteins on the cell membrane.

The alignment of these chromosomes results from the growth and shrinkage of the attached kinetochore microtubules. When chromosomes reach the metaphase plate, the shrinkage of these microtubules at the MTOCs is balanced by slow growth of microtubules at the kinetochores. Since the sister chromatids of each chromosome are connected to opposite poles, a tug of war occurs during metaphase that pulls them in opposite directions.

Anaphase

At the start of **anaphase** (‘‘against phase’’), the cohesins that are holding sister chromatids together at the centromeres split (Figure 12.5, step 5). Because the chromatids are under tension, each replicated chromosome is pulled apart to create two independent daughter chromosomes. By definition, this separation of chromatids instantly doubles the number of chromosomes in the cell.

Two types of movement occur during anaphase. First, the daughter chromosomes move to opposite poles via the attachment of kinetochore proteins to the shrinking kinetochore microtubules. Second, the two poles of the spindle are pushed and pulled farther apart. The motor proteins in overlapping polar microtubules push the poles away from each other. Different motors on the membrane walk along on the astral microtubules to pull the poles to opposite sides of the cell.

During anaphase, then, replicated chromosomes split into two identical sets of daughter chromosomes. Their separation to opposite poles is a critical step in mitosis because it ensures that each daughter cell receives the same complement of chromosomes.

When anaphase is complete, two complete collections of chromosomes are fully separated, each identical with those of the parent cell before chromosome replication.

Telophase

During **telophase** (‘‘end phase’’), the nuclear envelope that dissolved in prometaphase reforms around each set of chromosomes, and the chromosomes begin to de-condense (Figure 12.5, step 6). Once two independent nuclei have formed, mitosis is complete. At this point, most cells will go on to divide their cytoplasm via cytokinesis to form two daughter cells.

Table 12.1 summarizes the key structures involved in mitosis.

Summary Table 12.1 Structures and Proteins Involved in Mitosis

Structure	Definition
Chromosome	A structure composed of DNA and associated proteins.
Chromatin	Chromosomes within an interphase nucleus.
Sister chromatids	The two identical pieces of DNA found in a chromosome after DNA replication.
Centromere	The DNA region that functions as a handle on a chromosome.
Cohesins	Proteins at the centromeres that hold sister chromatids together.
Microtubules	Proteins that have many functions, including moving chromosomes during mitosis.
Kinetochore proteins	Proteins at the centromeres that hold the DNA and microtubules together.
Nuclear lamins	Intermediate filaments on the inner surface of the nuclear envelope that hold the nucleus together during interphase.
Condensins	Proteins along the length of chromosomes that compact them for mitosis.
Centrosomes	The microtubule-organizing centres used in animal cells during mitosis.
NEDD1 complexes	The microtubule-organizing centres used in plant cells during mitosis.

After you’ve studied **Table 12.1** and reviewed **Figure 12.5**, you should be able to make a new table that summarizes what happens to (1) the spindle apparatus, (2) the nuclear envelope, and (3) the chromosomes in each of the five phases of mitosis. You should also be able to explain the purpose of each event in mitosis and what is causing it to occur.