Name: _____
Bio AP

Lab: Cell Division: Mitosis
(Modified from Carolina Cell Respiration & AP Biology Investigative Labs)

BACKGROUND:

One of the characteristics of living things is the ability to replicate and pass on genetic information to the next generation. Cell division in individual bacterial and archaea usually occurs by binary fission. Mitochondria and chloroplasts also replicate by binary fission, which is evidence of the evolutionary relationship between these organelles and prokaryotes.

Cell division in eukaryotes is more complex. It requires the cell to manage a complicated process of duplicating the nucleus, other organelles and multiple chromosomes. This process, called the cell cycle, is divided into here parts: interphase, mitosis, and cytokinesis (figure 1). In the first growth phase (G_1), the cell grows and prepares to duplicate its DNA. In the synthesis phase (S), the chromosomes are replicated. In the second growth (G_2), the cell prepares to divide. In mitosis, the duplicated chromosomes are separated into two nuclei. In most cases, mitosis is followed by cytokinesis, when the cytoplasm divides and organelles separate into daughter cells. This type of cell division is asexual and is important for growth, renewal, and repair of multicellular organisms.

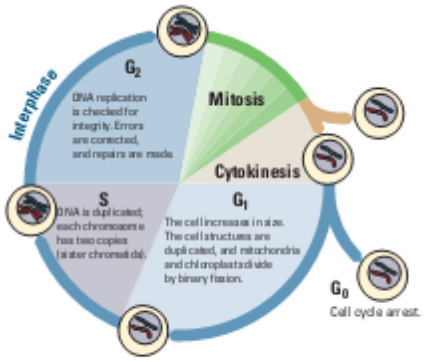


Figure 1. The Cell Cycle Showing Interphase, Mitosis, and Cytokinesis

Cell division is tightly controlled by complexes made of several specific proteins. These complexes contain enzymes called cyclin-dependent kinases (CDKs), which turn on or off the various processes that take place in cell division. CDK partners with a family of proteins called cyclins. One such complex is mitosis-promoting factor (MPF), sometimes called maturation-promoting factor, which contains cyclin A or B and cyclin-dependent kinase (CDK). (See fig. 2a). CDK is activated when it is bound to cyclin, interacting with various other proteins that, in case allow the cell to proceed from G_1 into mitosis. The level of cyclin changes during the cell cycle (fig. 2b). In most cases, cytokinesis follows mitosis.

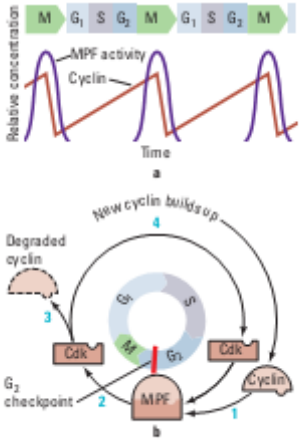


Figure 2a-b. MPF Production During the Cell Cycle

As shown in figure 3, different CDKs are produced during the phases. The **cyclins** determine which processes in cell division are turned on or off and in what order by CDK. As each cyclin is turned on or off, CDK causes the cell to move through the stages in the cell cycle.

Cyclin and CDKs do not allow the cell to progress through its cell cycle automatically. There are three check points a cell must pass through: the G_1 checkpoint, G_2 checkpoint, and the M-spindle checkpoint. (Fig 4). At each of the three checkpoints, the cell checks that it has completed all of the tasks needed and is ready to proceed to the next step its cycle. Cells pass the G_1 checkpoint when they are stimulated by appropriate external growth factors; for example, platelet-derived growth factor (PDGF) stimulates cells near a wound to divide so that they can repair an injury. The G_2 checkpoint checks for damage after DNA is replicated and therefore if there is damage it prevents the cell from going into mitosis. The M-

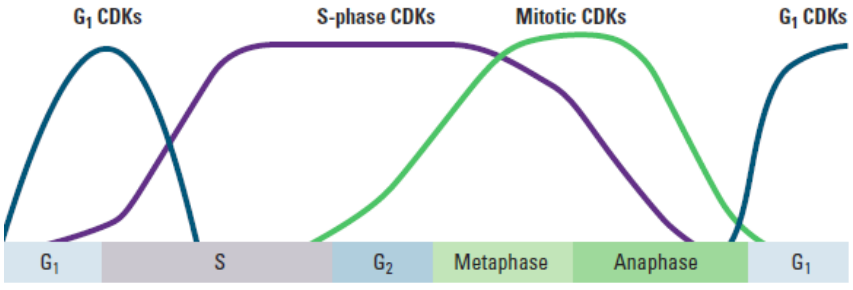


Figure 3. Levels of CDKs During the Cell Cycle

spindle checkpoint (metaphase) assures that the mitotic spindles of microtubules are properly attached to the kinetochores (anchor sites on the chromosomes). If the spindles are anchored correctly properly, the cell does not continue on through mitosis. The cell cycle is regulated very precisely. Mutations in cell cycle genes that interfere with proper cell cycle control are found very often in cancer cells.

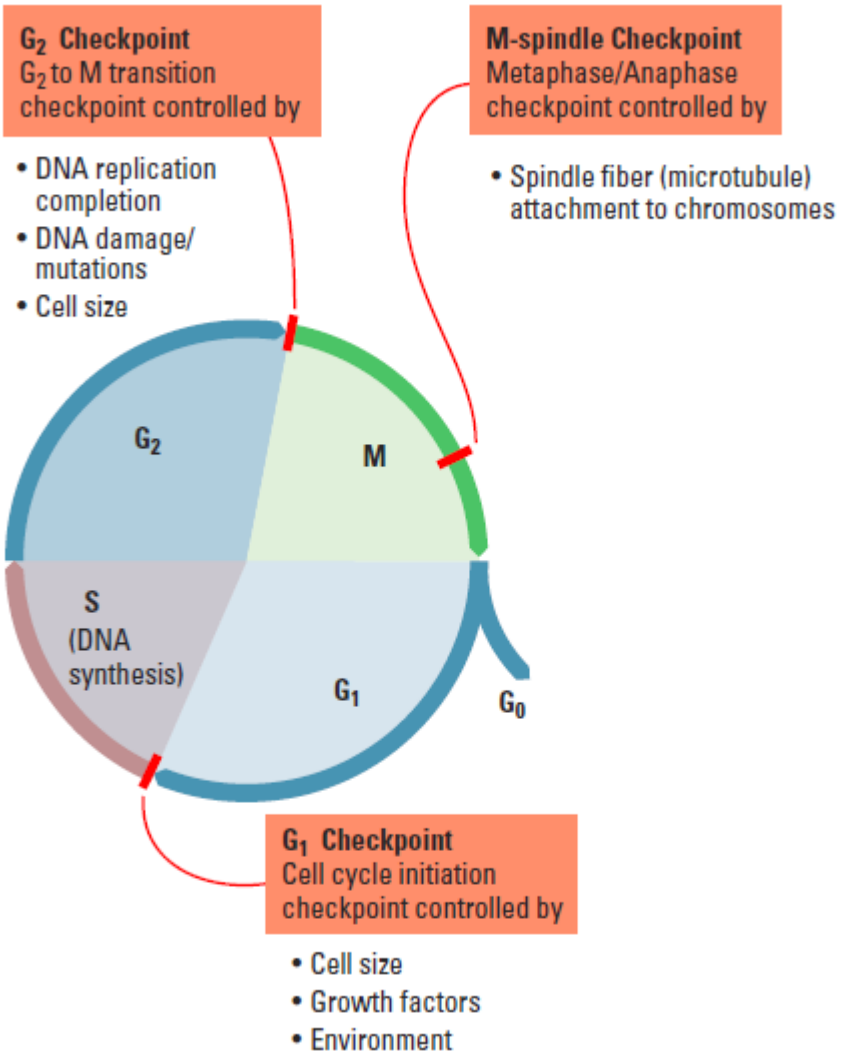


Figure 4. Diagram of the Cell Cycle Indicating the Checkpoints

LEARNING OBJECTIVES:

- To describe the events in the cell cycle and how these events are controlled
- To explain how DNA is transmitted to the next generation via mitosis

GENERAL SAFETY:
Follow your teacher's directions. Do not work in the laboratory without your teacher's supervision.

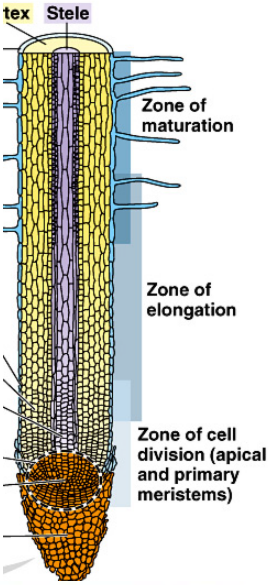
THE INVESTIGATION:
You will examine the apical meristem of an onion root, diagramming a cell in each of the major phases of mitosis. You will then determine the percentage of time a cell spends in each of the stages.

INTRODUCTION:
In order to see how well you understand and can explain the key concepts of mitosis and meiosis, answer the following:

1. How did you develop from a single-celled zygote to an organism with trillions of cells? What is true about the genetic information in one of the cells found in your toe and one of the cells found in your kidney?
2. Why is it important that DNA be replicated prior to cell division?
3. What would happen to the chromosome number of a species if gametes were formed by mitosis and not meiosis? Explain.
4. Are sister chromatids exact copies of each other? Defend your answer.

MATERIALS:
Prepared slide: onion root tip
Microscope

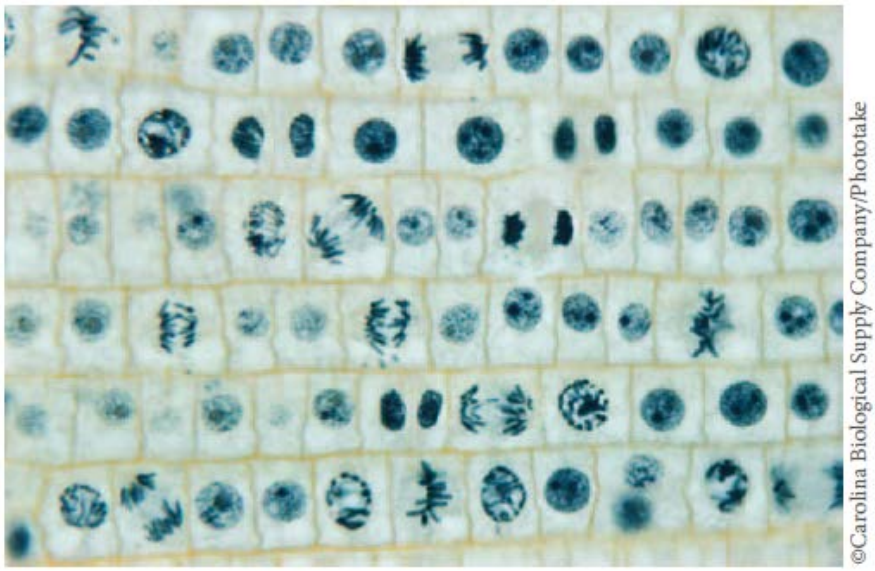
Part 1: MITOSIS;
You will use prepared slides of onion root tips to study plant cell mitosis and to calculate the relative duration of the phases of mitosis in the meristem of root tissue. To study the stages of mitosis you need to look for tissue where there are many cells in the process of mitosis. This restricts your search to the tips of growing plants such as the onion root tip, or in the case of animals, to developing embryos such as the whitefish blastula.
Roots have different regions. The root cap functions in protection. The apical meristem is the region that contains the highest percentage of cells undergoing mitosis. The region of maturation is where root hairs develop and where cells differentiate.



Procedure:

1. Examine the apical meristem region of the onion root tip. Draw a *labeled* diagram of a cell in interphase and in each of the stages of mitosis (prophase, metaphase, anaphase and telophase). Be sure to include the power of magnification and your field of view.

2. To estimate the relative length of time that a cell spends in the various stages of mitosis, you will examine the meristematic region of a prepared slide of the onion root tip. Using the high power objective (40x) locate the meristematic region. Count all the cells in the field and tabulate the number of cells that are in interphase and in each stage of mitosis (prophase, metaphase, anaphase, and telophase)



3. Complete the table below:

	Number of Cells			Total number of cells in field 1,2,and 3	Percent of total cells counted
	Field 1	Field 2	Field 3		
Interphase					
Prophase					
Metaphase					
Anaphase					
Telophase					
Total number of cells counted					

Questions:

1. Prepare a Bar graph of your results showing the percent of time spent in each stage of the cell cycle.
2. Based upon your results, what can you infer about the relative length of time an onion root tip cell spends in each stage of the cell cycle?
3. Complete the table below:

Stage	KEY CHANGES occurring in the cell (including identification characteristics)
Prophase	
Metaphase	
Anaphase	
Telophase	