Mitosis Lab

Wednesday, February 8, 2017 12:25 PM

LABORATORY 3. MITOSIS AND MEIOSIS

OVERVIEW

In this laboratory, you will investigate the process of mitosis and meiosis. The first part is a study of mitosis. You will use prepared slides of onion root tips to study plant mitosis and to calculate the relative duration of the phases of mitosis in the meristem of root tissue. Prepared slides of the whitefish blastula may be used to study mitosis in animal cells and to compare animal mitosis with plant mitosis.

The second part is a study of meiosis. You will simulate the stages of meiosis by using chromosome models. You will study the crossing over and recombination that occurs during meiosis. You will observe the arrangements of ascospores in the asci from a cross between wild type *Sordaria fimicola* and mutants for tan spore coat color in this fungus. These arrangements will be used to estimate the percentage of crossing over that occurs between the centromere and the gene that controls the tan spore color.

OBJECTIVES

Before doing this laboratory you should understand:

- the events of mitosis in animal and plant cells;
- the events of meiosis (gametogenesis) in animal and plant cells; and
- the key mechanical and genetic differences between meiosis and mitosis.

After doing this laboratory you should be able to:

- recognize the stages of mitosis in a plant or animal cell;
- calculate the relative duration of the cell cycle stages;
- describe how independent assortment and crossing over can generate genetic variation among the products of meiosis;
- use chromosome models to demonstrate the activity of chromosomes during meiosis I and meiosis II;
- relate chromosome activity to Mendelian segregation and independent assortment;
- demonstrate the role of meiosis in the formation of gametes in a controlled experiment using an organism of your choice;
- calculate the map distance of a particular gene from a chromosome's center or between two genes using an organism of your choice;
- compare and contrast the results of meiosis and mitosis in plant cells; and
- compare and contrast the results of meiosis and mitosis in animal cells.

INTRODUCTION

All new cells come from previously existing cells. New cells are formed by the process of cell division which involves both replication of the cell's nucleus (**karyokinesis**) and division of the cytoplasm (**cytokinesis**).

There are two types of nuclear division: mitosis and meiosis. **Mitosis** typically results in new somatic (body) cells. Formation of an adult organism from a fertilized egg, asexual reproduction, regeneration, and maintenance or repair of body parts are accomplished through mitotic cell division. **Meiosis** results in the formation of either gametes (in animals) or spores (in plants). These cells have half the chromosome number of the parent cell. You will study meiosis in Exercise 3B.

Where does one find cells undergoing mitosis? Plants and animals differ in this respect. In higher plants the process of forming new cells is restricted to special growth regions called **meristems**. These regions usually occur at the tips of stems or roots. In animals, cell division occurs anywhere new cells are formed or as new cells replace old ones. However, some tissues in both plants and animals rarely divide once the organism is mature.

To study the stages of mitosis, you need to look for tissues 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.

EXERCISE 3A.1: Observing Mitosis in Plant and Animal Cells Using Prepared Slides of the Onion Root Tip and Whitefish Blastula

Roots consist of 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 elongation** is the area in which growth occurs. The **region of maturation** is where root hairs develop and where cells differentiate to become xylem, phloem, and other tissues (Figure 3.1).

Figure 3.1: Median Longitudinal Section

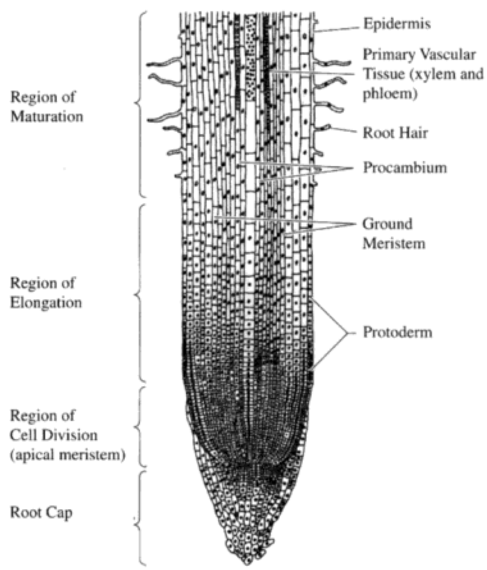


Figure 3.1b: Apical Meristem Tip Close Up

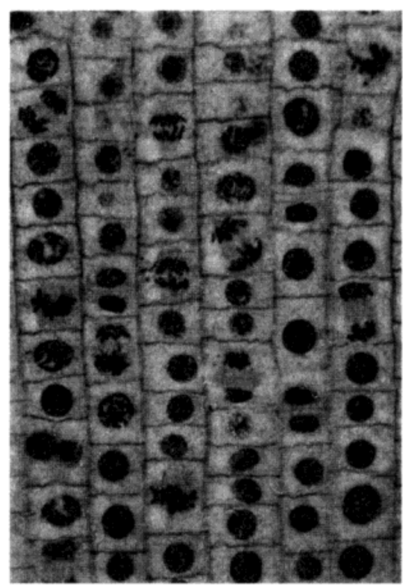
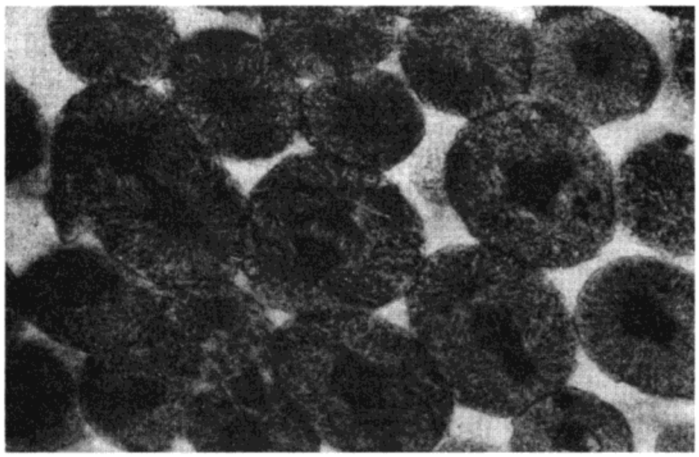


Figure 3.2: Whitefish Blastula

The whitefish blastula is excellent for the study of cell division. As soon as the egg is fertilized it begins to divide, and nuclear division after nuclear division follows. You will be provided with slides of whitefish blastula which have been sectioned in various planes in relation to the mitotic spindle. You will be able to see side and polar views of the spindle apparatus.



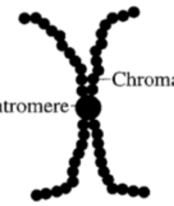
Procedure

Examine prepared slides of either onion root tips or whitefish blastula. Locate the meristematic region of the onion, or locate the blastula with the 10X objective, and then use the 40X objective to study individual cells. For convenience in discussion, biologists have described certain stages, or phases, of the continuous mitotic cell cycle, as outlined below. Identify one cell which clearly represents each phase. Sketch and label the cell in the box provided.

1. The nondividing cell is in a stage called **interphase**. The **nucleus** may have one or more dark-stained **nucleoli** and is filled with a fine network of threads, the **chromatin**. During **interphase**, DNA replication occurs.

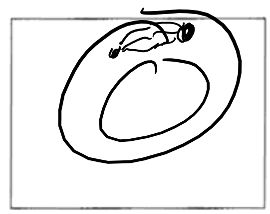
Interphase

Figure 3.3



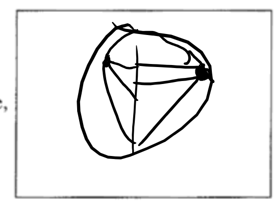
Chromosome
(replicated)

2. The first sign of division occurs in **prophase**. There is a thickening of the chromatin threads, which continues until it is evident that the chromatin has condensed into **chromosomes**. With somewhat higher magnification you may be able to see that each chromosome is composed of two **chromatids** joined at a **centromere**. As prophase continues, the chromatids continue to shorten and thicken. In late prophase the nuclear envelope and nucleoli are no longer visible, and the chromosomes are free in the cytoplasm. Just before this time the first sign of a spindle appears in the cytoplasm; the spindle apparatus is made up of **microtubules**, and it is thought that these microtubules may pull the chromosomes toward the poles of the cell where the two daughter nuclei will eventually form.



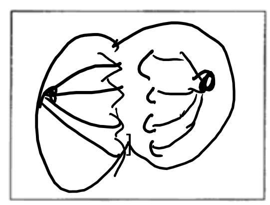
Prophase

3. At **metaphase**, the chromosomes have moved to the center of the spindle. One particular portion of each chromosome, the centromere, attaches to the spindle. The centromeres of all the chromosomes lie at about the same level of the spindle, on a plane called the metaphase plate. At metaphase you should be able to observe the two chromatids of some of the chromosomes.



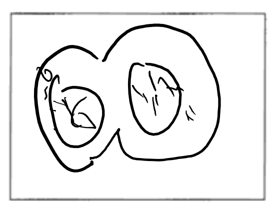
Metaphase

4. At the beginning of **anaphase**, the centromere regions of each pair of chromatids separate and are moved by the spindle fibers toward opposite poles of the spindle, dragging the rest of the chromatid behind them. Once the two chromatids separate, each is called a chromosome. These daughter chromosomes continue poleward movement until they form two compact clumps, one at each spindle pole.



Anaphase

5. **Telophase**, the last stage of division, is marked by a pronounced condensation of the chromosomes, followed by the formation of a new nuclear envelope around each group of chromosomes. The chromosomes gradually uncoil to form the fine chromatin network seen in interphase, and the nucleoli and nuclear envelope reappear. **Cytokinesis** may occur. This is the division of the cytoplasm into two cells. In plants, a new cell wall is laid down between the daughter cells. In animal cells, the old cell will pinch off in the middle along a **cleavage furrow** to form two new daughter cells.



Telophase

Analysis Questions

1. Why is it more accurate to call mitosis "nuclear replication" rather than "cellular division"?

When cell division occurs, the nucleus is replicated rather than divided.

2. Explain why the whitefish blastula and onion root tip are selected for a study of mitosis.

In these specific areas, rapid cell division occurs

EXERCISE 3A.2: Time for Cell Replication

Overview

To estimate the relative length of time that a cell spends in the various stages of cell replication, you will examine the meristematic region of a prepared slide of the onion root tip. The length of the cell cycle is approximately 24 hours for cells in actively dividing onion root tips.

Procedure

It is hard to imagine that you can estimate how much time a cell spends in each phase of cell replication from a slide of dead cells. Yet this is precisely what you will do in this part of the lab. Since you are working with a prepared slide, you cannot get any information about how long it takes a cell to divide. What you can determine is how many cells are in each phase. From this, you can infer the percent of time each cell spends in each phase.

1. Observe every cell in one high power field of view and determine which phase of the cell cycle it is in. This is best done in pairs. The partner observing the slide calls out the phase of each cell while the other partner records. Then switch so the recorder becomes the observer and vice versa. Count at least two full fields of view. If you have not counted at least 200 cells, then count a third field of view.

2. Record your data in Table 3.1.

Table 3.1

	Number of Cells				Percent of Total Cells Counted	Time in Each Stage
	Field 1	Field 2	Field 3	Total		
Interphase	360	299	463	372	91.8	1321.9
Prophase	28	30	25	23	6.4	92.1
Metaphase	4	4	5	4	0.8	11.5
Anaphase	2	1	5	1	6.4	5.8
Telophase	2	3	6	4	0.6	8.6
			Total Cells Counted	1120		

3. Calculate the percentage of cells in each phase.

Consider that it takes, on average, 24 hours (or 1,440 minutes) for onion root-tip cells to complete the cell cycle. You can calculate the amount of time spent in each phase of the cell cycle from the percent of cells in that stage.

Percent of cells in stage X 1,440 minutes = _________ minutes of cell cycle spent in stage

Questions

1. If your observations had not been restricted to the area of the root tip that is actively dividing, how would your results have been different?

The cells would not be actively dividing

2. Based on the data in Table 3.1, what can you infer about the relative length of time an onion root-tip cell spends in each stage of cell division?

Most cells spend time in Interphase in the root tip of onion cell