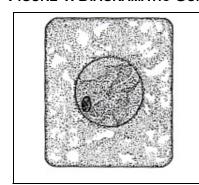
LAB ____. MITOSIS AND CANCER

One of the basic tenets of biology is that all new cells come from living cells. New cells are formed by the process of cell division which includes both the division of the cell's nucleus (<u>mitosis</u>) and the division of the cells cytoplasm (<u>cytokinesis</u>). Mitotic nuclear division typically rests in new <u>somatic</u> (body) cells. All of the following processes are accomplished through mitotic cell divisions:

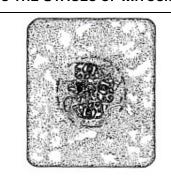
- Formation of an adult organism from a fertilized egg (zygote)
- Asexual reproduction (as in the reproduction of an Amoeba or Paramecium)
- Regeneration (as in the re-growth of a starfish arm)
- Repair or maintenance of body parts (as in the healing of a wound or the replacement of epithelial tissue)
- Growth (as in growth in height of a tree or growth in size of a maturing animal)

Although, once it has started, mitosis is a continuous process, scientists have divided it into distinct stages for convenience and ease of study and understanding of the cellular events. These are illustrated below (for plant cells) in Figure 1.

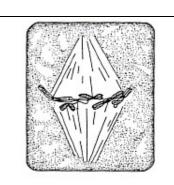
FIGURE 1. DIAGRAMATIC GUIDE TO THE STAGES OF MITOSIS



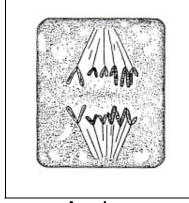
Interphase



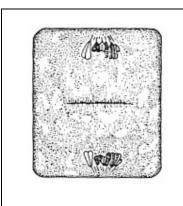
Prophase



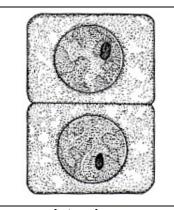
Metaphase



Anaphase



Telophase



Interphase of new daughter cells

In this lab, we will be viewing sections of tissue containing mitotic cells and determining what stages each of the cells is in and estimating how long each stage lasts. We will also be comparing normal cells and cancer cells to see how mitosis is altered in cancer cells.

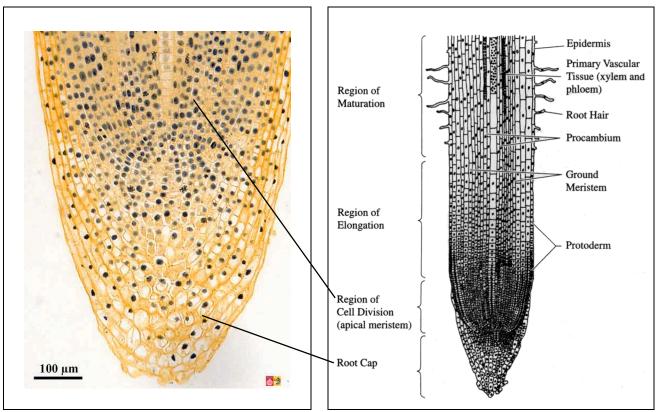
Where does one find cells undergoing mitosis? It depends if you are studying plants or animals. Plants and animals differ in this respect. In higher plants (gymnosperm and angiosperm), the process of forming new cells is restricted to special growth regions called **meristems**. These regions usually occur at the tips of the stems and roots of the plant. In animals, cell division occurs in many tissues and in many organs throughout the body — whenever cells receive the appropriate signals to form new cells, possibly for growth or to repair an injury or to replace old cells. However, some tissues in both plants and animals rarely divide once the organism is mature, like nerve cells or muscle cells in animals or mature xylem or phloem in plants.

PART 1. NORMAL MITOSIS IN PLANTS

- 1. Obtain a compound microscope and a slide of a longitudinal section through an onion root tip. Clean both before proceeding.
- 2. Scan the side under low power first and locate the <u>apical meristem</u>, the region of rapidly dividing cells directly behind the protective <u>root cap</u>. Refer to Figures 2 and 3 for guidance.

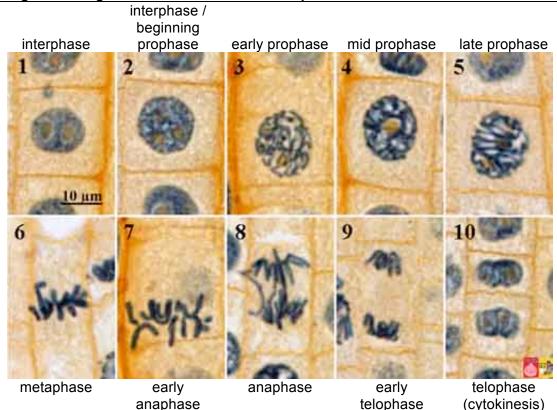
Figure 2. Onion root tip photograph

Figure 3. Onion root tip diagram



3. Switch to high power and center your slide in the apical meristem region, so that you have a field of view in which there is a wide selection of cells in various stages of mitosis (including interphase). Be sure to adjust your light for optimum viewing. Refer to Figure 4 for guidance.

Figure 4. Stages of mitosis in onion root tip cells



- 4. You are now going to identify the stage of each cell in your field of view. Starting at the top right corner of the field, record the stage of each cell in Data Table 1 as Area 1. Count your cells in a systematic manner.
- 5. After completing the count in this first area (Area 1), move your slide to a new area of the apical meristem region and perform the stage identification and count a second time. Again record the stage of each cell in Data Table 1, but list this count in Area 2.
- 6. Repeat the count a third time in a third area of the apical meristem region and record the stage of each cell in Data Table 1, but list this count in Area 3.
- 7. Return to viewing the slide of the onion root tip. Find a representative cell for each stage of mitosis and draw a clear diagram in the accompanying space. Be sure to draw only what you see, but include all details that are visible. Your drawings will not necessarily look exactly like the ones in Figure 1.
- 8. Now in Data Table 1, sum across the count areas to find the total number of cells in each phase.

9. Calculating the time for each stage of mitosis: There is a direct relationship between the number cells counted in a given stage of mitosis and the time that that stage takes to complete. This may be calculated if the total time for mitosis in onion root tip cells is known. (That total time is measured from interphase to interphase.) It is generally accepted that the total time for mitosis in onion root tip cells is 720 minutes (12 hours). Therefore, to calculate the time for each stage of mitosis in these onion cell, set up a ratio of the number of cells in each phase, compared to the total number of cells counted. Then multiply this fraction by the total time (720 minutes) needed to complete one mitotic division. In other words, the time for a specific phase is equal to:

Number of cells in a specific phase

Total number of cells counted

X 720 minutes = time for specific phase

- 10. Using your data, calculate the time required for the completion of each stage of mitosis in onion root tip cells. Be sure to use the totals for all three count areas. Record these calculated results in the appropriate column of Data Table 1.
- 11. Now, calculate the *percentage* of the cell cycle spent in each phase and record it in Data Table 1.
- 12. Prepare a bar graph to illustrate your results. Your independent variable is the five stages of mitosis and your dependent variable is the "time (minutes) to complete stage".

DATA TABLE 1. COUNT AND TIMING OF CELLS IN VARIOUS STAGES OF MITOSIS

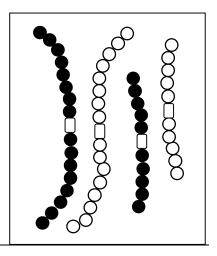
Stage of Mitosis	# Cells in Area 1	# Cells in Area 2	# Cells in Area 3	Total # Cells	Time in Minutes	% of Cell Cycle
INTERPHASE						
PROPHASE						
METAPHASE						
ANAPHASE						
TELOPHASE						
			Totals		720	100%

FIGURE 5. OBSERVED STAGES OF MITOSIS

Interphase	Prophase	Metaphase
	110011111111111111111111111111111111111	
Anaphase	Telophase	Cytokinesis

PART 2. SIMULATING MITOSIS

1. Using the pop-it beads provided by your teacher to model chromosomes, demonstrate your understanding of the process of mitosis. Begin with a cell with 4 chromosomes (2 pairs of homologous chromosomes, therefore the diploid number = 4). Distinguish the pairs of chromosomes from each other by size (one pair long and one pair short). Distinguish the members of each homologous pair by color. The magnet will represent the centromere. Use construction paper to represent the rest of the cell. Demonstrate to your teacher the process of mitosis in this cell.



Teacher's initials _____

PART 3. MITOTIC DIVISION IN CANCER CELLS

A key characteristic of cancer cells is that they are no longer constrained by the standard cell cycle controls that normally coordinate cell division activity. Consequently the timing of mitosis in cancer cells is altered. You may have heard of cancer cells being "runaway" which have no controls on their rate of reproduction. It is this characteristic that allows some cancer cells to grow and spread quite rapidly. In this section of the lab, you will analyze data to determine the differences in timing of mitosis between normal stomach cells and cancerous stomach cells of the chicken.

DATA TABLE 2. Mitosis in Normal Chicken Stomach Cells

DATA TABLE 3. Mitosis in Cancerous Chicken Stomach Cells

Otomach Cens	Officken Glomach Gens						
Stage of Mitosis	Total # Cells	Time in Minutes	% of Cell Cycle	Stage of Mitosis	Total # Cells	Time in Minutes	% of Cell Cycle
INTERPHASE	440			INTERPHASE	424		
PROPHASE	40			PROPHASE	50		
METAPHASE	8			METAPHASE	12		
ANAPHASE	2			ANAPHASE	3		
TELOPHASE	10			TELOPHASE	11		
Totals	500	625	100%	Totals	500	448	100%

- 1. Study the data in Table 2 (Mitosis in Normal Chicken Stomach Cells). Assume that the total time needed for one normal mitotic division of these cells is 625 minutes. Calculate, in the same manner as before, the total time needed for each normal phase of mitosis. Also, calculate the *percentage* of the cell cycle spent in each phase. Enter these data in the appropriate column of Data Table 2.
- 2. Repeat the same analysis for the data in Table 3 (Mitosis in Cancerous Chicken Stomach Cells). In the case of cancer cells, however, the total time needed for one mitotic division is only 448 minutes. Also, calculate the *percentage* of the cell cycle spent in each phase. Enter these data in the appropriate column of Data Table 3.
- 3. Prepare a bar graph to illustrate these results. Your independent variable is the five stages of mitosis and your dependent variable is the "time (minutes) to complete stage". Plot both the data for normal cells and for cancerous cells on the same graph.

Name		
Name		

SUMMARY QUESTIONS

Referring to your data and graphs for <u>onion root tip cells</u> in Part 1, answer the following questions.

1.	Which stage in the mitotic cycle takes the most time? What <i>percentage</i> of the total time is this?
2.	Why do you think that this stage (in Question 1) takes so much longer? What activities, in relation to mitosis are occurring during this phase?
3.	Which stage is the <u>second</u> longest? What <u>percentage</u> of the total time is this?
4.	Again, what activities, in relation to mitosis are occurring during this phase?
5.	List the remaining stages, in order, from longest to shortest duration.

Na	me AP Biology
	Referring to your data and graphs for <u>normal and cancer chicken stomach cells</u> in Part 3, answer the following questions.
6.	How do the data for each phase in the <u>normal</u> chicken cell compare with that of the onion root tip cell? Are the <u>percentages</u> of time for the two longest phases similar? Are you able to make any generalized conclusions based on the information.
7.	How does the timing of the <u>complete</u> cell cycle differ in <u>normal</u> chicken cell vs. <u>cancerous</u> chicken cells?
8.	Which stage exhibits the most dramatic difference in timing between normal and cancerous chicken cells?
9.	What nuclear and cytoplasmic changes would you expect to find in cancer cells, as compared to their normal counterparts? (HINT: What events would be most affected by the alteration in timing sequence of mitosis?)