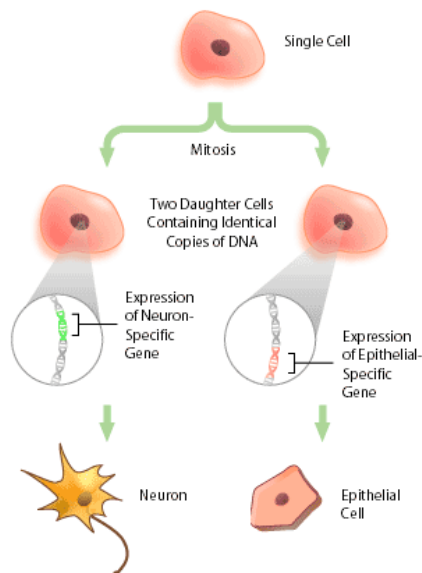


Cell Division, Cancer, and Chemotherapy

HASPI Medical Biology Lab 05

Background/Introduction



Cell Differentiation & Cell Division

The human body starts as a single cell in the womb, and grows and develops until it reaches more than 100 trillion cells as an adult. From that single cell develops over 200 different types of cells—such as skin cells, muscle cells, and neurons—that come together to create complex tissues and organs within the human body. The process by which basic cells become more specialized to perform a specific function is known as **differentiation**. It is important to understand that ALL of the cells in the body have the exact same DNA, but depending on their function only USE the DNA instructions that are needed. For example, cells in the pancreas specifically use the gene to create the protein insulin, and they are the ONLY cells that use that part of the DNA.

<http://blogs.scientificamerican.com/a-blog-around-the-clock/files/2011/09/a2-cell-differentiation.gif>

How does a basic cell know what type of cell to become? This is a very complex question and a lot of molecular research focuses on getting a more exact understanding of how this works. Most simply, the basic cells, such as **stem cells**, are activated to start dividing and producing specific proteins of the cell they will become. These cells then begin dividing themselves to produce a tissue that can perform a specific function. For example, a stem cell is activated to divide and become a red blood cell by producing proteins like hemoglobin that are needed to carry oxygen throughout the body. This red blood cell will continue to divide and produce many red blood cells that all perform the function of carrying oxygen. As you can see, cell differentiation and cell division go hand in hand.

How does the body actually grow? It adds more cells through cell division, or **mitosis**.

Mitosis most commonly will occur in one of these three circumstances:

- **Growth and Development** – Multicellular organisms grow and develop by adding more cells. In general, cells do not get bigger, and instead just add more cells.
- **Regeneration** – In some multicellular organisms, such as starfish (sea stars), limbs or body parts can be regenerated through mitosis.
- **Replacement** – Different cell types have different life spans. When a cell dies, it must be replaced. A list of the average life spans for some common cell types can be found below.

Table 1. Cell Types & Life Span

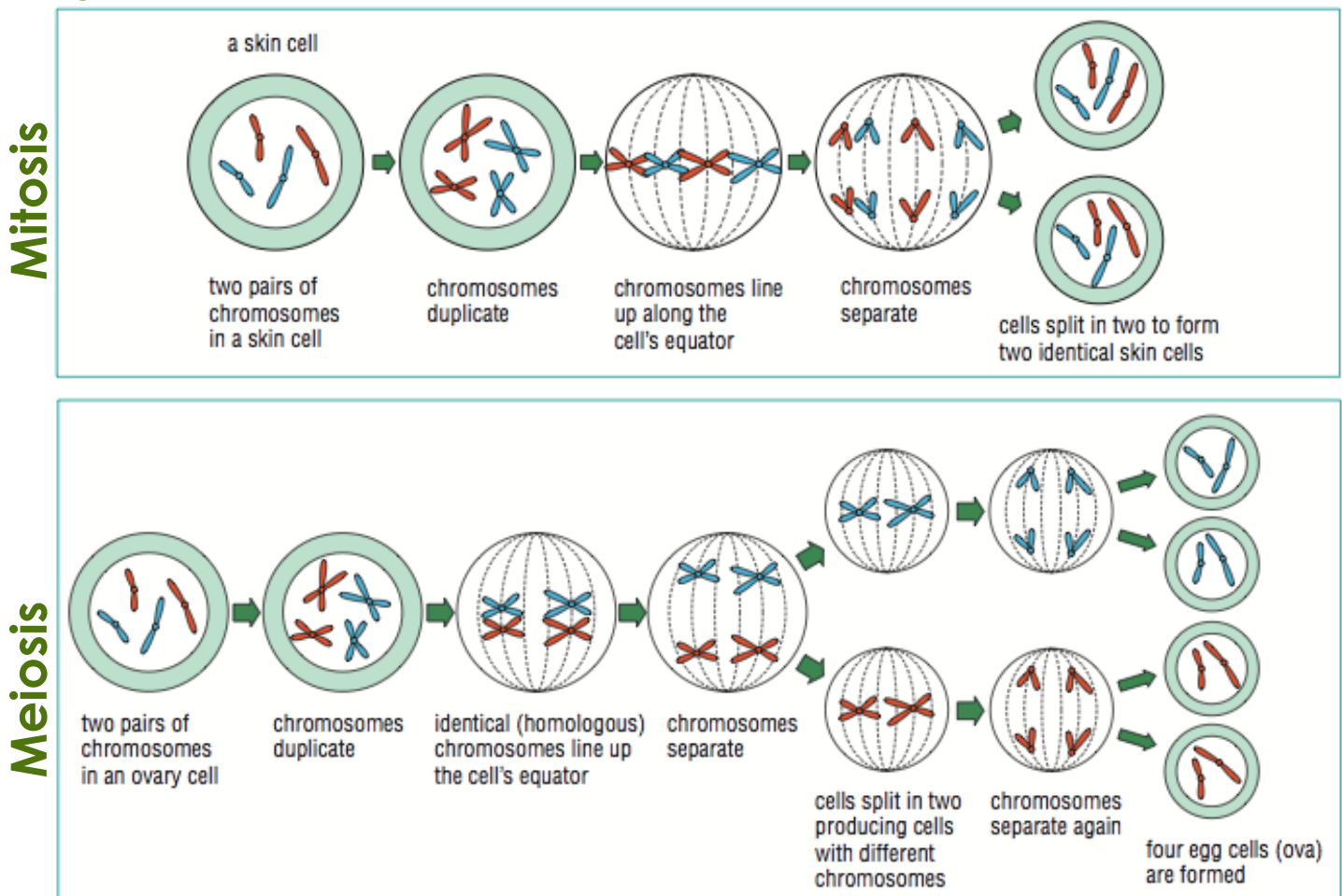
Cell Type	Life Span
Stomach cells	2 days
Sperm cells	2-3 days
Colon cells	3-4 days
Platelets	10 days
Skin cells	2-4 weeks
White blood cells	2 months – 1 year
Red blood cells	4 months
Pancreas cells	1 year or more
Bone cells	25-30 years
Brain cells	70+ years

Mitosis NOT Meiosis!

Mitosis and meiosis are very similar processes, with very different outcomes. In mitosis, a cell splits in two and both cells that are created are identical, meaning they have the exact same DNA. All of the cells that make up the body, called **somatic cells**, undergo mitosis. On the other hand in meiosis, a cell splits into four and all of the cells created are different, meaning they have different DNA. **Germ cells**, specifically sperm and egg, are the only cells in the body that undergo meiosis. This is an important difference because if germ cells divided by the process of mitosis, we would all be clones of our parents!

There are three important goals of mitosis: Replication of DNA, segregation of DNA, and cytokinesis through which the cells actually separate. A cell spends more than 95% of its life preparing for mitosis and less than 5% actually undergoing mitosis. During this preparation time, called interphase, the cell performs its normal functions and replicates its DNA. Once all of the cell's DNA have been replicated, it organizes the DNA into chromosomes with two identical halves, called sister chromatids. Through mitosis the sister chromatids are split apart into two separate sides of the cell, and then the cell separates through cytokinesis. In this way, each new cell contains the exact same copy of DNA, and the two are identical to each other. Mitosis is broken up into stages that you will learn more about in Part A of this lab. Diagram A below summarizes the steps of mitosis and shows a comparison between mitosis and meiosis.

Diagram A. Mitosis vs. Meiosis



<https://year11biology2012.wikispaces.com/file/view/Cell%20Division%20by%20mitosis%20and%20meiosis.png/361089422/Cell%20Division%20by%20mitosis%20and%20meiosis.png>

Cell Division Without Regulation = Cancer

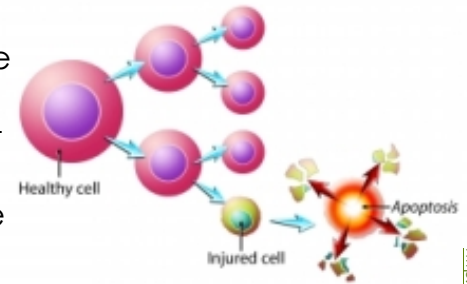
Mitosis is a strictly regulated process. Cells do not divide unless they are needed. For example, neurons in the brain can live up to 70 + years so these cells never undergo mitosis. Each cell type has genes that regulate when the cell should divide and when the cell should die. When mutations occur within the genes that regulate mitosis and cell death, tumors and cancer can result. Essentially, you can think of cancer as a homeostatic imbalance between cells being produced and cells dying.

For example, look at a patch of skin cells on the back of your hand. Let's say there are approximately 1,000 cells in an area of skin. These skin cells live for approximately 10 days so every day, 100 of those cells die. This means that every day 100 cells undergo mitosis to replace those that have died in order to maintain the 1,000 cells.

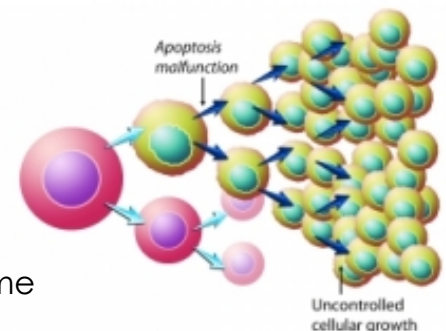
What happens if a single one of these cells has its **regulatory gene(s)** mutated and is no longer being controlled? It starts dividing twice a day and every cell it produces will have the same mutation and start dividing twice a day as well. To continue the example above, after 5 days of this growth there would be 2,048 mutated cells. Now add in the fact that these cells also don't die after the 10 days. These groups of mutated cells will form a tumor that pushes on the tissues around them and can even impact how the surrounding tissues and organs function. Eventually, a few of these cells could enter the bloodstream and travel to other locations in the body where they will continue to divide and create new tumors. It is important to note that most cancers do not divide this quickly, and our bodies have systems in place that try to catch mutated cells before they can cause cancer. Unfortunately, some of these cells are still able to get past the protective measures.

Cancer can be passed on through genes or caused by exposure to toxins in the environment. Substances that can cause cancer are known as **carcinogens**. When substances cause cancer, they are actually causing mutations in the genes that regulate cell division and cell death.

Normal Cell Division

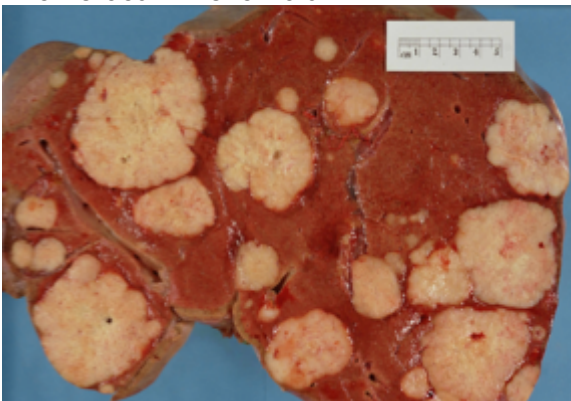


Cancer Cell Division



http://uvrhealth.com/Plone/ebsco_images/7250.jpg

Diagram B. A liver section with numerous white tumors



http://upload.wikimedia.org/wikipedia/commons/5/53/Secondary_tumour_deposits_in_the_liver_from_a_primary_cancer_of_the_pancreas.jpg

Mutations in regulatory genes can be caused by:

- **Heredity** – passed from parents to offspring; examples include breast cancer, skin cancer, colon cancer
- **Spontaneous mutations** – random errors that occur in the DNA replication process
- **Chemical or toxic substance exposure** – examples include asbestos, cigarette smoke, benzene
- **Radiation** – examples include radon, UV radiation, x-rays
- **Pathogens** – examples include hepatitis, human papillomavirus (HPV), Epstein-Barr Virus (EBV)

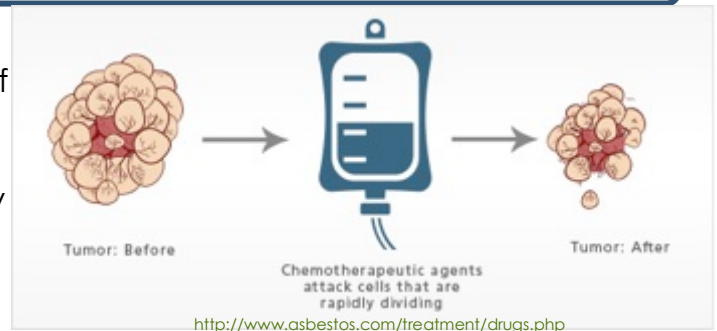
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Chemotherapy

Chemotherapy (also called “chemo”) is a type of cancer treatment that uses drugs to destroy cancer cells. Chemotherapy works by stopping or slowing the growth of cancer cells, which grow and divide quickly, but it can also harm healthy cells that divide quickly, such as those that line your mouth and intestines or cause your hair to grow. Damage to healthy cells may cause side effects. The most common side effects are hair loss (alopecia), decreased production of red and white blood cells (myelo- and immunosuppression), and inflammation of the digestive tract lining (mucositis).



The majority of chemotherapy drugs target cells that divide quickly. They are designed to impair mitosis by destroying damaging cells, specifically tumor cells. For this reason, aggressive tumors and cancers that grow quickly are extremely sensitive to chemotherapy. Slower growing or solid tumors do not respond as well, due to their slower growth rate, as well as the fact that some cells at the center of solid tumors are no longer dividing, making chemotherapy completely ineffective. For these types of cancers, chemotherapy is often used in combination with surgical removal and/or radiation therapy.

Administering the correct dosage of chemotherapy can be tricky. Chemotherapy drugs are cytotoxic (toxic to cells), and administering an excessive dose may run the risk of killing the patient. On the other hand, administering a dose that is too low may be ineffective on the targeted tumor.

Review Questions – answer questions on a separate sheet of paper

1. How many different types of cells are in the human body? Give 5 examples.
2. Explain differentiation. How can cells perform different functions when they all have the exact same DNA?
3. How do cell differentiation and cell division work together?
4. What are the three circumstances where mitosis, or cell division, will occur?
5. Do you think that cell types with shorter or longer life spans go through mitosis more often? Explain your answer.
6. What is the difference between somatic and germ cells? Give an example of each.
7. What is the major difference between the outcomes of mitosis and meiosis?
8. What are the three goals of mitosis?
9. Why are regulatory genes so important?
10. Compare and contrast normal cell division and cancer cell division.
11. Explain how an uncontrolled mutation in a single cell can lead to the development of a tumor and cancer.
12. List and give an example of a possible cause of cancer.
13. How are chemotherapy drugs capable of killing off cancerous cells while not killing the individual taking the drugs?
14. What side effects can chemotherapy cause? Explain WHY these are side effects.
15. Is chemotherapy more effective on fast-growing or slow-growing cancers? Explain your answer.

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Modeling Cell Division

HASPI Medical Biology Lab 05a

Modeling Mitosis (Cell Division)



Cells that divide quickly and out of control can lead to the development of tumors and cancer. As a result, understanding how cells divide is crucial to developing treatments and cures for patients with cancer. In the following activity, you will create a series of simple models to simulate the steps involved in cellular division, or mitosis, in order to get a better understanding of the process and to prepare for testing chemotherapy treatments in Part B.



Part A Materials

Cell Division sheets (3 pgs.)	2 Blue pipe cleaners	2 Blue yarn pieces
2 Red pipe cleaners	2 Red yarn pieces	4 White string pieces

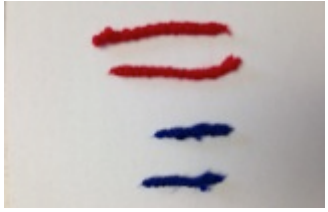
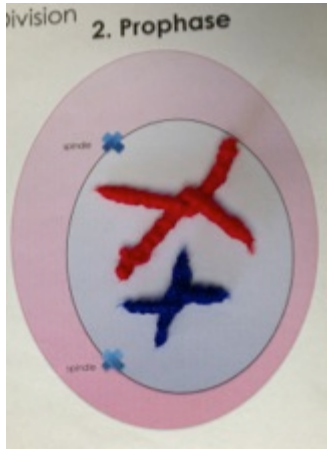
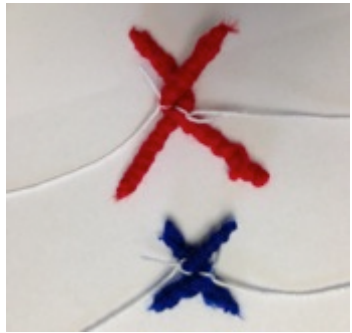
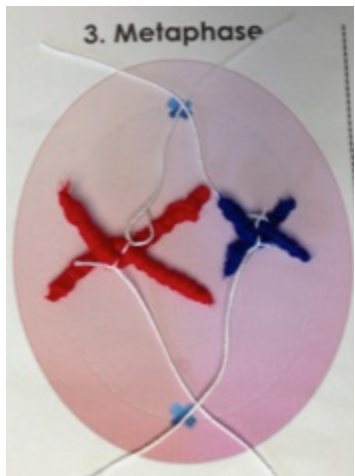
Procedure/Directions

Your lab team will be given tasks, or directions, to perform on the left. Record your questions, observations, or required response to each task on the right.

Part A: Modeling Mitosis

Task	Response
1 Cells must divide to either replace those that have died or to cause an organism to grow. In the following simulation you will model the stages of mitosis to obtain a better understanding of how cells undergo cell division.	
2 Collect a "Cell Division" template (3 pages), 2 red pipe cleaners, 2 blue pipe cleaners, 4 pieces of white string, 2 pieces of red yarn, and 2 pieces of blue yarn. Lay the cell division sheets on your table in order (1-6).	
3 Interphase: The cell spends most of its time in interphase replicating DNA and performing normal cell functions. Once the DNA in the cell has replicated, the cell is ready to divide. There are now 2 copies of all of the genetic information in the nucleus of the cell.	
4 The red and blue yarn represent sections of DNA.	
5 Clump together and place the red and blue yarn in the nucleus of the interphase cell on your "Cell Division" sheet (see image). This "messy" version of DNA is called chromatin .	
6 Take (with a camera) OR draw a picture of your interphase model in Part A of the analysis section and answer the questions. NOTE: If you are taking pictures, you can e-mail and print all of the pictures for this activity to turn in to your teacher. You will still need to answer the questions in Part A of the analysis section.	
7 Prophase: The DNA winds up into organized structures called chromosomes and becomes visible. Each half of a chromosome is called a chromatid and contains the same genetic information. The spindle apparatus begins to form and moves toward the middle.	
8 Take a strand of red yarn and a red pipe cleaner. Tie the yarn to one end of the pipe cleaner (see image). <u>You will need to undo the knot later so do not tie it too tight.</u>	

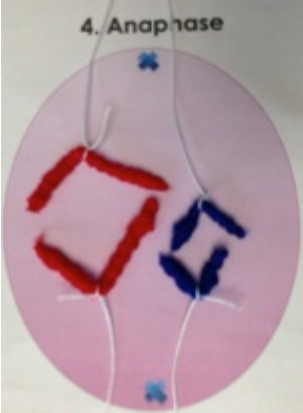
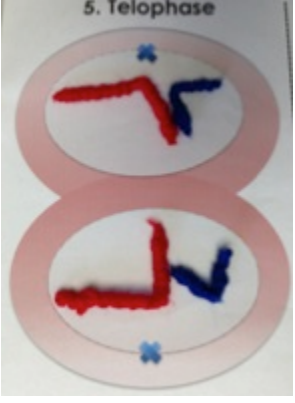

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9	Wrap the yarn around the pipe cleaner tightly until the end of the yarn is reached. <u>Lightly</u> tie off the end of the yarn to the end of the pipe cleaner.	
10	Repeat steps 8 and 9 for the remaining piece of red yarn and red pipe cleaner. Repeat these same steps for the pieces of blue yarn and blue pipe cleaners (see image).	
11	Each yarn/pipe cleaner combination represents how the DNA winds up and organizes into chromatids. Connect the red chromatids and the blue chromatids to each other to form two chromosomes by bending them around each other once (half-wrap; see image). The place where the chromosomes attach is called the centromere .	
12	Place the chromosomes randomly in the nucleus of the prophase cell on your "Cell Division" sheet. Take (with a camera) OR draw a picture of your prophase model in Part A of the analysis section and answer the questions.	
13	Metaphase: The membrane surrounding the nucleus breaks down and the spindle apparatus extends fibers that attach to the center of the chromosomes. As the spindle fibers tighten, the chromosomes line up in the middle of the cell.	
14	The white strings represent spindle fibers . Tie the end of a piece of white string to the center of a chromatid (see image). Repeat this with the 3 other white strings and remaining chromatids. It may be easier to separate the chromatids to tie each string. Make sure to recreate each chromosome after tying the strings (like you did in step 11).	
15	Place the chromosomes in the center of the metaphase cell on your "Cell Division" sheet. The spindle fibers (white string) should be touching the spindle apparatus on each side (see image).	
16	Take (with a camera) OR draw a picture of your metaphase model in Part A of the analysis section and answer the questions.	

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17	Anaphase: The spindle apparatus pulls the spindle fibers toward themselves, causing each chromatid to pull away and separate to opposite sides of the cell.	
18	In this simulation, your hands are going to be performing the action of the spindle apparatus by pulling the spindle fibers. Move the chromosomes and spindle fibers to the center of the anaphase cell on your "Cell Division" sheet with the same set-up as the metaphase cell.	
19	Pull the spindle fibers slowly toward each spindle apparatus. This should cause the chromatids to separate and move toward opposite sides of the cell (see image).	
20	Once the chromatids reach the spindle apparatus on each side, take (with a camera) OR draw a picture of your anaphase model in Part A of the analysis section and answer the questions.	
21	Telophase: Now that the chromatids have separated, the cell will start to reform a nucleus to protect the DNA. The spindle and spindle fibers release the chromatids and begin to be broken down by the cell. The cell begins to split its cell membrane and cytoplasm in half to begin the formation of two cells.	
22	Remove (untie) the spindle fibers from each chromatid.	
23	Move the chromatids to the telophase cell on your "Cell Division" sheet. Place the separated chromatids in both of the reforming nuclei (see image).	
24	Take (with a camera) OR draw a picture of your telophase model in Part A of the analysis section and answer the questions.	
25	Cytokinesis: The cell has separated and formed two completely new cells. The chromatids unwind back into chromatin so the DNA can be used to create proteins. Each cell goes back into interphase, and starts replicating the DNA to prepare for another round of mitosis.	
26	Untie and unwind the red and blue yarn wrapped around each pipe cleaner.	
27	Move the unwound yarn into the nucleus of each cytokinesis cell on your "Cell Division" sheet. Make sure you place one red and one blue yarn in each nucleus to ensure each cell contains the same DNA (see image).	
28	Take (with a camera) OR draw a picture of your cytokinesis model in Part A of the analysis section and answer the questions.	

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Analysis

Part A. Mitosis Steps

<i>Draw your example (or attach printed photos) from Part A for each step.</i>	<i>Answer the following questions related to each cell division step.</i>
Interphase	<i>a. What do the red and blue yarn represent?</i> <i>b. What is the "messy" DNA in the nucleus called?</i>
Prophase	<i>a. Why do you think it is necessary for DNA to wind up into an organized chromosome before it divides?</i> <i>b. What is the relationship between chromatids and chromosomes?</i>
Metaphase	<i>a. Why do you think the nuclear envelope disappears before the cell divides? Explain your answer.</i>
Anaphase	<i>a. What is the purpose of the spindle and spindle fibers?</i>
Telophase	<i>a. Does each side of the cell have the same DNA? Explain your answer.</i>
Cytokinesis	<i>a. Why does the nucleus reform?</i> <i>b. Why does the DNA unwind back into chromatin?</i>

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Cell Division, Cancer, and Chemotherapy

HASPI Medical Biology Lab 05b



Scenario

Three new chemotherapy drugs to treat liver cancer have been developed by HASPI Pharmaceuticals. Before they can be considered for use, they must be extensively researched to determine the toxicity level, side effects, and the effective dosage. This information must be known in advance to make sure the dosage is high enough to be effective, but low enough that it does not inhibit normal cellular processes, or even worse, kill normal cells. In this lab, you will be responsible for choosing and testing one of these chemotherapy drugs to determine the toxicity and highest dosage possible before it starts to destroy the function of healthy cells.

Liver cells contain an enzyme called **catalase** that is responsible for breaking down hydrogen peroxide into oxygen gas and water, which are harmless to the body. If catalase is not effective, hydrogen peroxide would build up in the body and eventually become toxic leading to death. Your team will be looking at how different concentrations of the chemotherapy drugs affect the amount of oxygen gas produced as the liver cells (catalase) break down hydrogen peroxide.



Part B Materials

Test tube rack	Scale	Chemotherapy Drug A
4 Balloons	Weighing boat	Chemotherapy Drug B
Liver solution (cells)	4 Beakers or cups	Chemotherapy Drug C
4 Large test tubes	Distilled water	3% Hydrogen peroxide
Stopwatch	Graduated cylinder	4 Plastic pipettes
Spot plate	Stirrer (spoon)	

Procedure/Directions



Your lab team will be given tasks, or directions, to perform on the left. Record your questions, observations, or required response to each task on the right.

Part B: Chemotherapy Dosage		
Task		Response
1	As a medical research team, choose ONE of the drugs (A, B, or C) to test.	a. What drug are you testing?
2	The amount of drug in grams that is added to water represents the concentration. When diluting the drug into a liquid, it is often represented as a percentage. For example, if 1 gram of drug is added to 9 ml of water, for a total of 10 ml: Divide 1/10, which equals 0.1. Multiply 0.1 x 100 to get the percentage, which is 10%.	b. Determine the concentration (%) of the following amounts of drug added to make 10 ml of solution. 2 g of drug = _____ 0.5 g of drug = _____ 5 g of drug = _____

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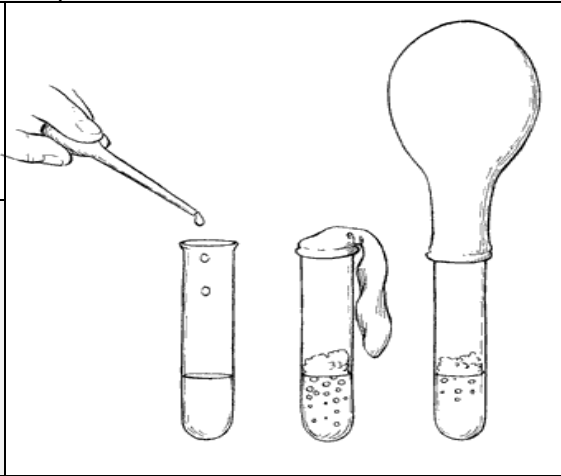
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3	<p>The Problem: What is the highest concentration before liver function is greatly impacted? Discuss with your team and formulate a hypothesis to answer this question.</p>	a. What is your hypothesis?
4	For this experiment, if it takes the liver cells more than 60 seconds to completely fill the balloon with oxygen, then liver cell function is being impacted. You want to get the time as close to 60 seconds as possible, but not over.	a. What 3 concentrations (%) is your team testing?
5	Just as a research team would, it is up to your team to determine what concentrations you are going to test. Discuss and record your concentrations to the right. DO NOT forget that you also need to have a control.	b. What is your control?
6	Calculate the amount of grams of drug you will need to add to water for a total of 10 ml of solution. Record your amounts to the right.	<p style="text-align: center;">Round 1 Concentrations:</p> <p>_____ % = _____ g drug + _____ ml water = 10 ml</p> <p>_____ % = _____ g drug + _____ ml water = 10 ml</p> <p>_____ % = _____ g drug + _____ ml water = 10 ml</p>
7	Using the spoon and a beaker/cup, add approximately 4-5 spoonfuls of the drug your group chose to the beaker/cup.	
8	Label the 3 remaining beakers/cups with the concentrations being tested.	
9	Using the scale and weighing boat, measure out the drug with the amounts needed for each concentration. As you weigh each amount, add it to the beaker/cup labeled with that concentration.	
10	Use a graduated cylinder to measure the amount of water needed to make the 10 ml of total solution for each concentration. As you measure each amount, pour it into the beaker/cup labeled with that concentration. Use the spoon to thoroughly mix each solution. Dry off the spoon with a paper towel between stirs for each beaker/cup. You should have 10 ml of total solution for each concentration.	
11	Add 15 drops of liver solution to 3 wells of the spot plate. Label each of the wells with the concentration to be tested.	
12	If you are wondering why you have 4 test tubes, REMEMBER: You also need to test your control...	
13	Using a pipette, add 15 drops of each drug solution to its respective well.	
14	Allow at least 5 minutes for the drug solution to diffuse and impact the function of the liver cells. While this is happening, measure out and pour 3 ml of hydrogen peroxide into each of the 3 test tubes. Lay the balloons out next to the test tubes.	

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15	On a separate sheet of paper, create a data table that includes the concentrations you are testing and provides space to record your results (time).	
16	Use a plastic pipette to QUICKLY add 5 drops of the liver/drug solution from one of the wells to the <u>first</u> test tube containing 3 ml of hydrogen peroxide. QUICKLY place the balloon over the test tube and start the stopwatch.	
17	The balloon will trap the oxygen gas released as the catalase breaks down hydrogen peroxide. When enough oxygen has been released to cause the balloon to stand completely upright, stop the stopwatch and record the time. If the time reaches 3 minutes, stop the time and write "DNF" (did not finish) in the results on your data table.	
18	Repeat steps 16 and 17 for the remaining drug solutions. Record your results.	
19	Now that you have the results for the 3 concentrations you chose to test, it is time to re-evaluate your hypothesis. Summarize your results for your first round of testing and revise your hypothesis.	a. Revised hypothesis:
20	Clean and rinse out your beakers, test tubes, and pipettes. Repeat the experiment with 3 new concentrations based on your results from the first experiment. Record all of your results in your data table.	<p style="text-align: center;">Round 2 Concentrations:</p> <p>_____ % = _____ g drug + _____ ml water = 10 ml</p> <p>_____ % = _____ g drug + _____ ml water = 10 ml</p> <p>_____ % = _____ g drug + _____ ml water = 10 ml</p>

Analysis

Analysis Questions – answer questions on a separate sheet of paper

1. If you have not already, create a data table that summarizes your results.
2. Were you able to get a good idea of what concentration is safe enough to not impair cell function? Explain why or why not.
3. What was the purpose of your control?
4. What can you conclude from your results?
5. List and explain at least 3 sources of error that could have skewed your results.
6. Are there any additional tests you would like to perform? "YES" or "NO" is not an answer! Explain your answer and/or which additional tests you would like to perform.
7. Why is testing like this important for chemotherapy drugs?
8. Once the exact drug concentration is discovered, what other testing do you think should be completed before it is available to patients? Why?
9. Hypothesize why it is important understand cell division when researching chemotherapy drugs for cancer treatments.

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Connections & Applications

Your instructor may assign or allow you to choose any of the following activities. As per NGSS/CCSS, these extensions allow students to explore outside activities recommended by the standards.

- GRAPHING AND PREDICTING CANCER GROWTH:** When researching cancer growth, it is important to quantify how quickly cells divide. Different cells in the body undergo cell division at different rates. Using an equation, we can predict the rate of cancer growth depending on how quickly the type of cells from which the cancer begins are dividing.

Variable	What it Means	Cell Growth Equation
N_t	Number of cells at time t	$N_t = N_0 2^{tf}$
N_0	Number of cells initially	
t	Time (days)	
f	Frequency of cell cycles	

Example: Breast Cancer

Cell Growth of Luminal Cancer Cells in the Breast

Variable	Amounts	Equation
N_t	?	$N_t = (1)2^{(3)(2.5)} \rightarrow N_t = (1)2^{(7.5)} \rightarrow N_t = (1)181 \rightarrow N_t = 181$
N_0	1	
t	3	
f	2.5	

This means that from a single luminal cancer cell in the breast, there will be a growth of 181 cancer cells after 3 days.

Use the cell growth equation to calculate the growth rate over a 5-day period of three cancers: Lung cancer, leukemia, and basal cell carcinoma. Record your results.

Cancer Cell Type	N_0	f	t (days)	N_t
Lung Cancer Cell Type: small cell	1	3.6	1	
			2	
			3	
			4	
			5	
Leukemia Cell Type: white blood cells	1	2.4	1	
			2	
			3	
			4	
			5	
Basal Cell Carcinoma (Skin Cancer) Cell Type: basal cells	1	0.7	1	
			2	
			3	
			4	
			5	

- Which of these cancer types is the fastest growing? Which is the slowest growing?
- After 30 days, how many cells would be in a cancerous tumor in the lungs? What about 6 months (180 days)?
- Why do you think early detection and treatment of cancer is so important?

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2. **CREATE A MODEL:** Create a 3D visual of the steps of mitosis OR the stages of cancer development. The model can be a physical model or digital/computer-generated. A description of each step/stage needs to be included.
3. **RESEARCH CANCER:** There are many different types of cancer. The severity of a cancer type often depends on the type of tissue the cancer starts within. If the cancer begins in cells that divide quickly, it will grow quickly. Choose and compare 3 different cancer types. Create/label a table that answers the following questions for each cancer type:
 - a. **Cells/Tissue** - what type of cells/tissue does this cancer start developing
 - b. **Symptoms** - what symptoms are associated with this cancer
 - c. **Diagnosis** - how is this cancer diagnosed
 - d. **Treatment** - how can this cancer be treated
 - e. **Risk** - are there any genetic or environmental links that may increase the risk of getting this type of cancer
 - f. **Incidence rate** - how many people have this type of cancer in the United States
 - g. **Mortality rate** - how many people who have this cancer die from it in the United States

Resources & References

- NOVA. 2001. What's The Right Dose? PBS, NOVA Cancer Warriors, <http://www.pbs.org>.
- Goldenberg. L. 2010. Cancer Cells Vs. Normal Cells: What's Different? The Intelligent Patient Guide to Prostate Cancer. MediResource, <http://chealth.canoe.ca/>.

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