Introduction to the immune system and immunology

Overview

This lesson introduces basic concepts related to the human immune system and immunology for high school students. It includes an instructional worksheet with student questions, an interactive activity demonstrating a technique commonly used in immunology research, and a guided experimental design.

Grade level

Introductory to advanced high school biology (grades 9-12), after some cell biology and human health content has been introduced

Learning objectives

• Students explain key features that distinguish the human innate vs. adaptive immune system and name some important cell types in the immune system
• Students explain the concept of homeostasis as it applies to the immune system, and describe some consequences of when homeostasis is lost
• Students construct a model that demonstrates what a flow cytometer does
• Students describe what flow cytometry does, how it is useful in studying the immune system, and its limitations
• Students design an experiment using flow cytometry to study an immune system function or disorder of their choice
• Students conduct independent research into the immunology literature

Equipment

• Laptop or desktop computers
• Internet access
• For flow cytometry activity in Part 2: see worksheet for detailed list of common classroom and household supplies
Outline and contents

Section 1 – Introduction to the immune system

• Explanatory handout introduces fundamental concepts in the immune system, including
  • Innate vs. adaptive immune system
  • The immune response to a threat
  • Balance of the immune system (homeostasis vs autoimmunity vs susceptibility)
• Students answer questions reinforcing their understanding of fundamental concepts in immune function

Section 2 – Flow cytometry, an important method in immunology

• Students learn what a flow cytometer is and how it can be used in immunology research
• Instructions for how to build a model flow cytometer from common classroom/household supplies that sorts marbles, candies, or other small round objects
• Students answer questions about their observations of the model flow cytometer
• Students answer questions about interpreting flow cytometry data, applications of flow cytometry data

Section 3 – apply your knowledge

Students design their own experiment that uses flow cytometry to investigate a disease or immune system function of their choice

Expected student knowledge

Students should already be familiar with:

• The idea that there are many different types of cells in the body
• The scientific method and how to develop a research question

Students do not need to already be familiar with:

• Any detailed knowledge about the immune system
• Types of cells in the immune system and how they work
• Any methods used for studying the immune system
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Teachers are welcome to adapt the lesson to suit their classes and curriculum, but may not share modified lessons. If you develop your own lesson plan using Allen Institute resources, we invite you to share your experience with us at communications@alleninstitute.org.
## Standards alignment

### Next Generation Science Standards

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<td>HS-LS1: From Molecules to Organisms</td>
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About the Allen Institute for Immunology

The Allen Institute is a nonprofit biomedical research institute located in Seattle, Washington. Our four divisions – Allen Institute for Brain Science, Allen Institute for Cell Science, Allen Institute for Immunology, and The Paul G. Allen Frontiers Group - are dedicated to answering some of the biggest questions in bioscience and accelerating research worldwide. We share all of our data and research findings with the scientific community and general public. Launched in 2003 by founder Paul G. Allen, the Allen Institute is supported by government, foundation, and private funds to enable its projects.

The Allen Institute for Immunology works to understand the dynamic balancing act of the human immune system, how it senses friend from foe and what goes wrong when we’re ill. This will help us to improve immune health and how we diagnose, treat and prevent immune-related diseases. Everything we do begins with patients who are living with and suffering from these diseases. We believe that by unlocking the mysteries of the immune system, we can make a significant improvement in patients’ health and well being.

To learn more, visit immunology.alleninstitute.org.
Introduction to the immune system and immunology:
Student worksheets

Your immune system protects you from threats to your health, from outside of your body (like viruses) and within your body (like cancer). In this lesson, you’ll learn about some basic properties of your immune system and about an important method for studying the immune system called flow cytometry. Then you’ll apply your knowledge to design your own study of the immune system.

Resources to learn more about the immune system

It can be difficult to find reputable scientific information about the immune system online. Allen Institute scientists recommend these sources:

- MedlinePlus immunology index
- National Institute for Allergy and Infectious Diseases (NIAID)
- NIAID disease profiles
- PubMed (a database of journal articles produced by scientists on research in immunology and many other areas of biological and medical science)
Section 1: Introduction to the immune system

Learning goals

• Explain key features that distinguish the human innate vs. adaptive immune system
• Briefly describe what happens in the immune system when a threat is detected
• Name some important cell types in the immune system, describe their roles, and describe how they interact
• Describe major categories of immune disorders and their relation to immune homeostasis

Overview of the immune system

The job of the immune system is to identify threats to your health, defend against them, and remember what they looked like for the future in case they return. Your immune system has many different types of cells, so it is constantly ready to respond to threats in a few minutes or hours after they enter your body.

Read:
• Overview of the Immune System (niaid.nih.gov/research/immune-system-overview), Immune Cell Types (niaid.nih.gov/research/immune-cells), and The Features of An Immune Response (niaid.nih.gov/research/immune-response-features) from NIAID
• Five Cool Things Your Immune System Can Do (alleninstitute.org/what-we-do/immunology/news-press/articles/5-cool-things-your-immune-system-can-do) from the Allen Institute for Immunology

When we talk about how the immune system identifies threats, one major division in immune function is between the innate and adaptive immune system.
The **innate** immune system consists of rapid responder cells that recognize pre-programmed threats.

- These cells recognize pathogens and other substances based on their molecular signature. These signatures are inherited and have evolved over many thousands of years.
- Example cell types in this category include macrophages, NK cells, and neutrophils. Visit the Allen Institute website to see videos of these cells under the microscope ([alleninstitute.org/about/education-outreach/introduction-to-immunology/cell-videos](http://alleninstitute.org/about/education-outreach/introduction-to-immunology/cell-videos)).

The adaptive immune system consists of cells that can respond to any pathogen, not just the ones you’ve evolved to recognize, and if you encounter it again, you’ll retain your immunity from the first exposure.

- The **adaptive** immune system is the main target of vaccination, to help your body learn to recognize unfamiliar pathogens and develop immunity without ever having been infected. This protection can last many years or even your whole life.
- These cells can even recognize novel (unfamiliar) and mutated pathogens as long as they are similar enough to something they have seen before. For example, there are multiple, similar viruses that cause the common cold, and exposure to one can cause some immunity to others.
- Example cell types in this category include B and T cells.

Pathogens are bacteria, viruses, or other microorganisms that cause diseases. The innate and adaptive immune system work together to respond to a pathogen. The amount of time it takes to respond, the specific cell types from the immune system that are involved, and other details vary depending on the pathogen.

For example, when a person is infected by a virus, the innate immune system recognizes general features of viruses that cause them to activate and travel to the site of infection. The T and B cells are then “trained” to target the virus itself or infected cells, which leads to the virus being eliminated. In contrast, if you are infected with bacteria, the innate immune system recognizes the bacteria and works to engulf and degrade it. These innate cells then “train” adaptive T and B cells to target the bacteria, which then release chemicals and antibodies to target these bacteria for recognition and destruction.

In either a viral or a bacterial infection, the adaptive immune system recognizes specific proteins on the surface of a pathogen, called epitopes, to help your body figure out the pathogen’s identity and that it is a threat. The immune system can also recognize other triggers, such as foreign genetic sequences or unique sugars, to create a response to some pathogens. Once the pathogen is eliminated, the activated T and B cells die off and leave some long-lived memory cells that remember the specific pathogen to fight it again in the future.
While scientists have a very good understanding of the components of the immune system and how they respond to pathogens, the Allen Institute for Immunology is working on collecting this data in order to create a complete model of healthy human immune function. Understanding the healthy immune system is also important to understanding diseases and disorders of the immune system, so we understand how the disease is different from healthy function.
Student questions

Name three things a healthy immune system might recognize as a threat.

1. _______________________________________________________________________________________
2. _______________________________________________________________________________________
3. _______________________________________________________________________________________

Summarize the role of the innate immune system in your own words, in two sentences or less.

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Summarize the role of the adaptive immune system in your own words, in two sentences or less.

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Name three ways that the innate and adaptive immune system are different. Hint: consider the cells that are part of each system and their jobs. Think about the details you learned in the readings.

1. _______________________________________________________________________________________
2. _______________________________________________________________________________________
3. _______________________________________________________________________________________
Name two ways that the innate and adaptive immune system are similar.

1. _______________________________________________________________________________________

2. _______________________________________________________________________________________

Choose one immune cell type and read at least two more articles about it online. See the top of this packet for a list of recommended reputable resources to find articles.

Cell type: ________________________________________________________________________________

Cite article #1: ____________________________________________________________________________
__________________________________________________________________________________________

Cite article #2: ____________________________________________________________________________
__________________________________________________________________________________________

Is this cell type part of the adaptive or innate immune system? _________________________________

Summarize the role of this cell type. What kind of threats to your health does it respond to? What does it do?
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What is at least one other interesting piece of information you learned about this cell type and its role?
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Disorders of the immune system

The immune system’s job is to defend your body against threats like bacteria and viruses. It has to strike a balance between identifying and responding to all threats, but not responding to harmless substances like pollen. When this balancing act called homeostasis becomes unbalanced, it causes a disorder of the immune system.

Immune disorders can be divided into a hyperactive immune response (leading to autoimmune and allergy conditions) or a hypoactive response (leading to chronic infections or cancer, among others).

Conditions that cause too little immune response include:

- **Severe Combined Immunodeficiency (SCID):** A genetic disorder that affects a gene needed to produce T and B cells, which recognize and respond to pathogens. People with SCID are vulnerable to many different infections because their bodies do not recognize the pathogens. They can also be reinfected by diseases that people usually only catch once and then become immune to because their immune system does not develop a memory for them.

- **HIV:** A virus that infects immune cells (CD4+ T cells, monocytes, macrophages) and establishes lifelong infection by inserting itself into DNA, termed latency. Acquired immunodeficiency syndrome (AIDS) is the end-result of uncontrolled HIV, particularly through disabling or killing CD4+ T cells. This causes a condition similar to SCID, with increased susceptibility to infections and AIDS-related cancers such as Kaposi’s sarcoma. Advances in antiretroviral therapies, aka drugs that block the virus, have significantly improved outcomes. Where treatment is affordable and available, HIV has become a chronic disease and AIDS a rarity.

- **Multiple myeloma:** Cancer of antibody-producing B cells (aka plasma cells). The plasma cell population grows out of control and begins crowding out healthy cells in the bone marrow. This causes fatigue and an inability to fight infections since your healthy blood cells are not developing normally. These plasma cells also create many abnormal antibodies that don’t fight disease, but just clog up the blood and can create problems with the kidneys. As with most cancers, the tumor cells release chemical messengers and interact with other immune cells to “turn off” the immune response. This causes an insufficient activation of the immune system and the body can’t kill the cancer cells on its own.
Conditions that cause too much immune response include:

- **Type 1 diabetes**: An autoimmune disease caused by an immune response to insulin and other proteins, which is produced by our own bodies. The T cells treat insulin-producing cells as invaders and kill them, leaving the person without the ability to produce insulin.

- **Crohn's Disease**: A type of inflammatory bowel disease, Crohn's Disease is an autoimmune condition that results in injury to parts of the intestine by the immune system. While the cause for Crohn’s disease is generally unknown, several factors (environmental, bacterial, immune) are believed to increase the chances of occurrence in genetically susceptible individuals. Crohn’s disease may be caused by a defective innate immune system. Inflammation and scarring are exacerbated by the adaptive immune system as it attempts to compensate for this deficient innate immunity.

- **Allergies**: For reasons that scientists don’t fully understand, a person’s immune system may treat a harmless substance, such as pollen or tree nuts, as a target and generate a special kind of antibody called IgE specific to that substance. Allergies occur at places where your body is exposed to substances in the environment, such as the surfaces of your lung and intestine. IgE antibodies can arm specialized innate immune cells to activate when that substance is encountered again, inducing these cells to release molecules like histamine that cause the common symptoms of allergies: sneezing, runny nose, congestion, and others.

Read: [Disorders of the Immune System](https://niaid.nih.gov/research/immune-system-disorders) from NIAID to learn more about some specific disorders.

Read: the Allen Institute for Immunology is studying healthy immune systems and immune-related diseases. Read the [research overview](https://alleninstitute.org/what-we-do/immunology/disease-research) to learn about their approach and why it’s needed.

Listen: the episode [Starved but Unable to Eat](https://alleninstitute.org/news-press/articles/starved-unable-eat-life-crohns) from the Allen Institute’s podcast Lab Notes to hear about one person’s experience of having an autoimmune condition.
Student questions

Explain the two main roles of the immune system that are in balance when homeostasis is maintained.

1. _______________________________________________________________________________________
   _______________________________________________________________________________________

2. _______________________________________________________________________________________
   _______________________________________________________________________________________

Summarize what is happening in the immune system when it has too little response, in four sentences or less. Do not describe a specific disorder - explain generally what is happening in these conditions.

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________________________________________________________________________________________
________________________________________________________________________________________
________________________________________________________________________________________

Name two things an unhealthy immune system might not recognize as a threat that it should. (Hint: these are things that the immune might not respond to that it should.)

1. _______________________________________________________________________________________
   _______________________________________________________________________________________

2. _______________________________________________________________________________________
   _______________________________________________________________________________________

14
Summarize what is happening in the immune system when it has too much response, in four sentences or less. Do not describe a specific disorder - explain generally what is happening in these conditions.

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Name two things an unhealthy immune system might recognize as a threat that it should not. (Hint: these are things that the immune might respond to that it should not.)

1. _______________________________________________________________________________________
__________________________________________________________________________________________

2. _______________________________________________________________________________________
__________________________________________________________________________________________
Choose one immune disorder from this list in order to answer the following questions.

- Leukemia
- Lymphoma
- SCID
- HIV
- Lupus
- For an added challenge: IBD

Flow cytometry is useful for studying and/or diagnosing these conditions. You’ll learn more about this method in the next section of this assignment.

Describe this disorder’s effects. What parts or systems of the body are affected?

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Where does it fall on the spectrum from no immune response to too much immune response?

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What cell types are involved in this disorder?

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__________________________________________________________________________________________

What are those cell types doing, or not doing, in this disorder compared to a healthy immune system?

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Section 2: Flow cytometry
A method for studying the immune system

Learning goals

• Explain what a flow cytometer does and how it does it
• Construct a flow cytometer model
• Describe why the output of a flow cytometer is useful for immunology research
• Describe some limitations of flow cytometry outputs and data
• Interpret flow cytometry data
• Reflect on what further experimental steps could follow use of a flow cytometer
• In the next section, design your own experiment that uses flow cytometry to study the immune system

A flow cytometer is a machine that detects specific properties of cells. It can be used to complete tasks such as sorting cells into types, counting cells by type, detecting the presence of cell types or pathogens, and detecting the presence of other biomarkers such as specific proteins. The Allen Institute for Immunology uses a flow cytometer to separate cell types, usually from blood samples, in order to then separately study each cell type further.

Often, fluorescent antibodies or tags are added to the sample to be tested, which attach themselves to the different cells to be counted. The flow cytometer then uses a laser to identify which colored antibody is attached to each cell and sorts them. For an average blood sample, scientists will analyze about ¼ of a teaspoon of blood (1.25ml), containing about 1.25 million cells. The flow cytometer processes about 3000-5000 cells per second, and processing a sample takes about 2 minutes. In one day, the Allen Institute for Immunology can generate 72 million data points from flow cytometry.

Sometimes the flow cytometer provides data that is the main goal of the analysis: for example, if you want to know if there is too little of one cell type in a blood sample compared to all other cell types. But often, once the cells have been sorted and counted, they will go through further analysis, such as single-cell RNA sequencing, which tells you what makes each cell unique, or cell culture, to grow more of a specific cell type. Clinical uses include bone marrow transplants or CAR-T therapy.

In this activity, we are interested in the use of this technique to study the immune system, usually using a blood sample, but flow cytometers have many applications outside of immunology too, such as determining the size of a plant genome, analyzing phytoplankton abundance in ocean samples, checking the safety of pharmaceuticals, and sorting different kinds of neurons for further analysis.

You will work in teams of three or four to complete this activity. Follow the instructions on the next page to build your model, then to collect data.
Activity: Build your own flow cytometer model

Supplies:

• “Cells:” Round objects in about 3-4 different colors to sort. Assorted marbles, multicolored candy, or any other small, spherical object that comes in multiple colors.
• Paper towel or wrapping paper tubes, PVC pipe, drinking straws, rolled up paper, or any other material you can use to make a sturdy tube about 6ft (2m) long and wider than the object you’re sorting.
• As many cups as there are colors of your objects.
• A craft knife or scissors.
• A sheet of cardboard or other stiff backing to attach your cups to.
• A textbook or other object about an inch (2cm) tall.
• A table.

Assemble your flow cytometer:

1. Assemble your tubes into a long, straight track. About 1/3 of the way down, cut a window in the tube that is about the size of your thumb.
2. Attach your cups to the cardboard. Label each cup with one of the colors of your “cells.”
3. Using the textbook, prop up the end of the track that the window is closer to. Position the other end so it hangs over the end of the table.

Team members and “flow cytometry” operation:

• Flow cytometry technician: positioned at the top of the track. Rolls the “cells” (marbles or other objects) to be sorted down the track one at a time, at least 5 seconds apart.
• Detector: Positioned by the window cut into the track. Looks through the window and tells the counter what color the cell is.
• Counter: Positioned at the bottom of the track: Catches the cells in the cups based on what color the detector says they are.

Rotate team roles so each person gets to sort at least 20 cells in each job. Use new candies, marbles, or whatever material you are counting for each person. Collect the data from the runs of the flow cytometer in the chart below.

For each run, in the box labeled “correct,” mark the number of cells of color #1 that you caught in the cup for color #1. In the box labeled “incorrect,” mark the number of cells that were not color #1 that you caught in the cup for color #1. For example, say you set color #1 to be red. At the end of the first run, you count the cells in cup #1 and find 8 red cells and 1 blue cell. The cell for that run would look like this:

<table>
<thead>
<tr>
<th>Cell type A: Red</th>
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</thead>
<tbody>
<tr>
<td>Correct: 8</td>
</tr>
<tr>
<td>Incorrect: 1</td>
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</tbody>
</table>

Record your data in the table on the next page.
## Model flow cytometer data collection

Group names: ___________________________________________________________

<table>
<thead>
<tr>
<th>Counter’s name</th>
<th>Cell type 1 (color #1) Color: _______</th>
<th>Cell type 2 (color #2) Color: _______</th>
<th>Cell type 3 (color #3) Color: _______</th>
<th>Cell type 4 (color #4) Color: _______</th>
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Total: correctly sorted

Total: incorrectly sorted
Using the total of only **correctly** sorted cells from all runs, create a histogram of your data.

Could you have created this histogram of the distribution of colors without using your model flow cytometer? Why or why not?

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Could you have created this histogram of the distribution of colors without using your model flow cytometer? Why or why not?
What do you observe about the distribution of “cells” in the histogram?

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Imagine you saw a histogram like this for data from real cells. How would you interpret the data?
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Describe your experience being the flow cytometer “technician” for this model.
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Imagine what you think the process of being a real flow cytometry technician would be like. Describe the process of acquiring a sample, running the machine, and acquiring results.
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Describe your experience being the detector and compare it to the job of a real flow cytometry machine’s detector. In your answer, think back to what the real detector is sensing on the cells.

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Think about the properties of the objects you were sorting. In this case, you were sorting based on color. If you wanted to automate sorting candies or marbles, what kind of equipment and software would you need? What do you need your equipment to do?

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Think about the cells that were sorted into one color (a “cell type”) in your model flow cytometer. What conclusions can you draw about this one “cell type” based on this sorting exercise? What further analysis could be interesting? (Hint: think about other properties of your sorted objects besides color.)

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After the cells have been sorted, they can be sent off for further analysis. Based on your reading about the immune system, what is one type of cell you might like to study further, and what would you like to measure about this cell type?

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We can study the human immune system with flow cytometry to measure the amounts of different types of cells, and then study each of those types individually to learn more about how they are functioning. Why do we need to study a healthy human immune system using this technology in order to also study immune disorders?

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Section 3: Apply your knowledge

You investigated a disease or disorder that impacts the immune system at the end of Section 1 of this activity. Design an experiment that uses flow cytometry to study this condition. (Flow cytometry is not useful for studying all aspects of the immune system or all conditions. The Allen Institute scientists picked the diseases you selected from in Section 1 because flow cytometry is useful for studying them.)

As you design your experiment, think about these guiding questions:

• What do you want to measure about the immune system in this condition? Think about the capabilities of flow cytometry, and note that you can conduct further analysis steps after using flow cytometry to sort cells by type.
• What is the link between the biology of the condition you chose and the symptoms? For example, in SCID, a lack of B and T cells leads to an inability to recognize pathogens, which leads to repeated infections.
• Once you have collected data using flow cytometry, and potentially other methods if you choose, how would you interpret your findings?

You will need to do some additional reading to learn about a specific function of the immune system, immune system disorders, and/or other laboratory methods in immunology besides flow cytometry. See the list of reputable resources for research at the beginning of this packet for recommendations.

Disease or disorder that you examined in Section 1 of this activity: ______________________________

Symptoms: ______________________________________________________________________________
________________________________________________________________________________________
________________________________________________________________________________________
Cell types affected (which cell types, and how):
________________________________________________________________________________________
________________________________________________________________________________________
________________________________________________________________________________________
Immune system function affected (which function or functions, and how it is affected):

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Research question:

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__________________________________________________________________________________________
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How will you apply flow cytometry: That is, which cell type(s) do you want to isolate to quantify them or study them further?

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What additional methods will you use, if any? How will you apply them?

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Predicted results, and why you made this prediction:

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Citations: