"The Doctor's Corner"

An Introduction to Flow Cytometry

Grades: 6-8

Subjects: Science; Immunology

Concepts: Understanding flow cytometry; learning about white blood cells; basic immunology background

Materials:
-Computer and projector
- (optional) cards with either "Dog" "Cat" "Fish" or "Other" printed on them
- (optional) large grid on the ground with rope or tape

Time Considerations
Preparation: 10 minutes
Activity: 30-40 minutes

Preparation

Part A

Set-up: Using a computer hooked up to a projector, pull up the Flow Cytometry powerpoint. If performing the optional activity, mark out a large grid using either rope or tape. Make it large enough to hold all your students.

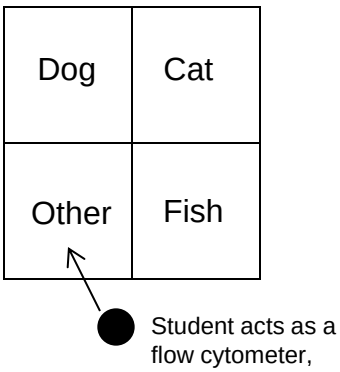
Part B

Go through the doctor simulation.

Part C

Discuss how flow cytometry can be used to diagnose disease. Talk about the importance of the immune system.

Diagram of Flow Cytometry Grid used in Part A (optional)



Objectives

1. To learn about different types of immune cells
2. To understand basic flow cytometry
3. Learn how flow cytometry can be used to diagnose different diseases

Background

Flow cytometry is a technique used largely in research science, but also in hospitals to diagnose different diseases and illnesses. Flow cytometry uses fluorescence to sort cells. It is called flow cytometry because cells 'flow' in a stream, and cytometry means to measure a parameter of a cell – in this case the fluorescence. Cells are tagged with fluorescent markers (Green, blue, red, etc.) and then sorted based on their color. In this way, we can stain for three different proteins expressed on a cell. More information is available by following the link below, along with an interactive display: http://www.unsolvedmysteries.oregonstate.edu/flow_06

Immunology is the study of the immune system. There are many types of immune cells, but this activity focuses on white blood cells. Those are comprised of B- and T-cells. Typically, T-cells will respond and fight off an invading bacterium or virus, while B-cells will produce antibodies specific to the invader. However, some diseases are mediated predominantly by B-cells, or by T-cells.

Epstein Barr virus (mononucleosis) is caused by the virus infecting B-cells, which then expand. Symptoms are synonymous with strep throat – fever, swollen lymph nodes, sore throat, and white spots in the throat and on the tongue. However, strep throat is bacterial in origin, and T-cells respond to that bacterium. Thus, a patient with increased B-cells and those symptoms may have the Epstein Barr virus.

By tagging B-cells with one color, and T-cells with another color, we can use flow cytometry to quantify the percent total of these cells.

Guiding Questions: When we get a cold, and we have the sniffles, is that an immune response? Why do we take antibiotics?

Hint: What is the function of an antibiotic? Is it similar to that of a T-cell? Do they work together?

Why do we have to use fluorescence? Hint: can we see differences between B-cells and T-cells just by using a microscope?

Part A – Flow Cytometry Sorting Activity

A flow cytometer ultimately sorts cells based on what they are. Have the students stand in front of the grid – one student will be the flow cytometer. Have that student decide which grid is for dogs, cats, fish and other. Give the remaining students the prepared cards. Have them line up in front of the 'flow cytometer' who must 'sort' them into their square. When they are done, point out that a real flow cytometer can sort over 4,000 cells per second.

To show how the sorting by fluorescence works, use the animated powerpoint slide (Flow Cytometry Four Square). Have the students guess where the cells go.

Part B – Diagnosing Patient X

In this activity, students are given a case history. The patient is symptomatic for both mononucleosis and strep throat. The first test shows that they have increased white blood cells, indicating that they have a strong immune response.

Flow cytometry is used to determine how many B-cells and how many T-cells are present in Patient X. Using that data, students must determine what illness is afflicting Patient X.

This activity is designed to help students understand the practical uses for flow cytometry.

Guiding Questions: What are other uses for flow cytometry? What if both illnesses were mediated by T-cells? How would we determine the type of illness then?

Hint: Would the cells have different proteins? Could we tag the proteins with different colors? (*Answer – yes*)

What are other tests that could be run to validate the flow cytometry results?

Fun Facts

- The average flow cytometer can sort over 4,000 cells per second.
- Some cytometers can 'see' up to 14 different colors.
- A high speed cell sorter flow cytometer will not just sort the cells, but will isolate them into a tube so that you can use those cells for other experiments.