WARNING — This set contains chemicals that may be harmful if misused. Read cautions on individual containers carefully. Not to be used by children except under adult supervision.

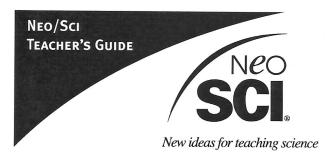


UNIT 3: What's your blood type? Using Simulated Neo/BLOOD

INVESTIGATIONS

OBJECTIVES

- Determine the ABO and Rh blood type of unknown simulated blood samples.
- *Calculate* the frequency distribution of each blood group in a given population.
- Prepare a simulated blood smear.
- Examine a prepared blood smear under the microscope to locate and identify red blood cells, white blood cells, and platelets.
- Estimate the number of simulated red blood cells in a given area.



Science Concepts

- · ABO and Rh blood type system
- · Red and white blood cells
- Hemoglobin
- Hematocrit
- Platelets
- Antibodies
- Antigens
- Serum

Kit Materials List

30 mL	Anti-A Simulated Serum
30mL	Anti-B Simulated Serum
30 mL	Anti-Rh Simulated Serum
40	Blood Typing Trays
25 mL	Patient #1, Simulated Neo/BLOOD Sample
25 mL	Patient #2, Simulated Neo/BLOOD Sample
25 mL	Patient #3, Simulated Neo/BLOOD Sample
25 mL	Patient #4, Simulated Neo/BLOOD Sample
40	Stirring Sticks, blue
40	Stirring Sticks, green
40	Stirring Sticks, yellow

Materials Needed But Not Provided:

Compound microscope

Coverslips

Microscope slides

Paper towels

Time Requirements

Pre-lab preparation

5 minutes

Lab Activity 1

30 minutes

Lab Activity 2

15 minutes

Safety & Disposal

The simulated Neo/BLOOD and sera samples provided in this kit contain no biological components and are therefore safe from any potential biological hazards. However, students should always wear safety goggles, gloves and a lab apron to protect the eyes and clothing when working with any chemicals. The dye in the simulated Neo/BLOOD solution will stain your skin and clothing. Be sure that students wash their hands before leaving the laboratory.

Any simulated Neo/BLOOD waste from this lab may be disposed of by pouring it down the drain with copious amounts of water.

Pre-lab Setup

Enough materials are provided for up to 40 students to type all four unknown simulated Neo/BLOOD samples provided in the kit. Provide each student with one blood typing tray and a set of blue, yellow and green stirring sticks. Set up a lab station with the simulated Neo/BLOOD samples and the simulated blood typing sera for students to dispense the samples on to their trays. Alternatively, the samples may be circulated until every student types all four patient blood samples.

As an alternate way to conduct this investigation, you may have students compare the frequency of each blood group in a given population by distributing the simulated Neo/BLOOD samples to students as follows:

Provide 42% of your students with "Patient 1" blood, 10% of your students with "Patient 2" blood, 4% of your students with "Patient 3" blood and 44% of your students with "Patient 4" blood. Have all the students pool their results and determine the percent blood type in a given population. The percentages that students determine should be the actual results in the US population. You may also want to vary the percentages to match a specific population of your choice.

Lab Activity Procedures & Notes

ACTIVITY



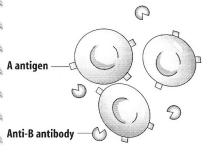
Determining the ABO-Rh Blood Type of Simulated Blood Samples

Surface proteins on red blood cells determine an individual's blood type. These surface proteins are called "antigens."

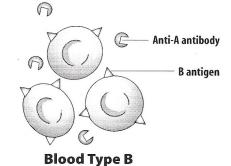
The system used to classify human blood is called the "ABO system." Dr. Karl Landsteiner, an Austrian physician, received the Nobel Prize in physiology for this discovery in 1930.

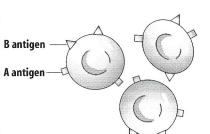
With the ABO system, the kinds of antigens present on red blood cells determines the blood type. An individual with A antigens has blood type A, one with B antigens has blood type B, one with both A and B antigens has blood type AB, and one with no antigens on the surface of his/her red blood cells has type O. Blood typing is performed using "antiserum" - blood that contains specific antibodies. "Anti-A Serum," which contains anti-A antibodies, and "Anti-B Serum," which contains anti-B antiAnother important antigen on the surface of red blood cells is the Rh protein, named for the *Rhesus* monkey.

Blood plasma has circulating proteins called "antibodies". For example, individuals with A surface antigen have anti-B antibodies; those with B surface antigen have anti-A antibodies. Those with both A and B surface antigens have no antibodies. Individuals with no surface antigens have both anti-A and anti-B antibodies.

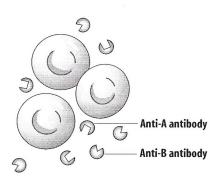


Blood Type A





Blood Type AB

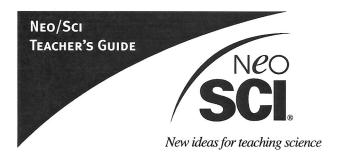


Blood Type O

Data Table 1 ABO Blood Types Summary

Blood Type	Antigen on Red Blood Cells	Antibodies in Plasma	Can Receive Blood From	Can Donate Blood To
Α	A	В	0, A	A, AB
В	В	A	О, В	B, AB
AB	A and B	None	O, AB, A, B	AB
0	None	A and B	0	O, A, B, AB

Unit 3 What's your blood type? Using SimulatedNeo/BLOOD 1-800-526-6689



Blood typing is performed using "antiserum" - blood that contains specific antibodies. "Anti-A Serum," which contains anti-A antibodies, and "Anti-B Serum," which contains anti-B antibodies, are used in ABO blood typing.

To perform a blood typing test, anti-A and anti-B sera are each separately mixed with a drop of sample blood and observed for "agglutination" or clumping.

Data Table 2 Agglutination Reactions in the ABO System

ABO Agglutina		
Anti-A Serum	Anti-B Serum	Blood Type
Agglutination	No Agglutination	A
No agglutination	Agglutination	В
Agglutination	Agglutination	AB
No Agglutination	No Agglutination	0

Another important antigen on the surface of red blood cells is the Rh protein, named for the rhesus monkey in which it was first studied.

People who have this protein are "Rh-positive," and those who lack it are "Rh-negative."

Rh-negative individuals who have been transfused with Rh-positive blood can produce Rh antibodies. They may develop a transfusion reaction, during which agglutination may occur, if they are transfused again with Rh-positive blood. Rh compatibility is tested when the ABO blood type is determined.

Data Table 3 Rh Agglutination Reactions

Rh Agglutination Reaction	Rh Factor
Agglutination	+
No agglutination	

Data Table 4 Frequency of ABO Blood Types and Rh Factor in the U.S.

Blood Type	Frequency Percentage	Blood Type & Rh Factor	Frequency Percentage
A	42	A +	34
		A -	8
В	10	B +	8
	10	В —	2
AB	А	AB +	3
AD	*	AB –	1
0	44	0 +	35
	77	0 –	9

What you need

Per Student

Anti-A Serum (simulated)

Anti-B Serum (simulated)

Anti-Rh Serum (simulated)

1 Blood typing tray

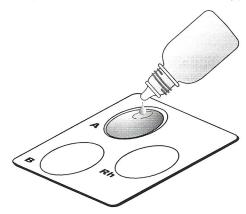
Paper towels

4 Patient Blood Samples (simulated)

1 set Stirring sticks (blue, green and yellow)

What to do...

Have your students place 5 drops of Patient 1 Simulated Blood Sample in each well on their blood typing tray.

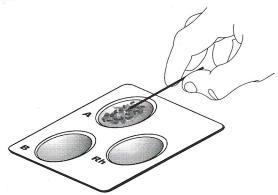


Next, the students should place 3 drops of Anti-A Simulated Serum in Well A.

Step 3 The students should now place 3 drops of Anti-B Simulated Serum in Well B.

Step 4 Finally, have your students place 3 drops of Anti-Rh Simulated Serum in Well Rh..

Have students use a separate stirring stick to mix the simulated blood and serum in each well for about 10 seconds.



Step 6 Have your students carefully examine each well to determine if the simulated blood in each well has clumped or agglutinated. Have them record their results and observations in Data Table 1.

Step 7 The students should thoroughly rinse the tray and stirring sticks and repeat Steps 1-6 to type the remaining, simulated blood samples.

> Unit 3 What's your blood type? Using SimulatedNeo/BLOOD 1-800-526-6689



Understanding Your Results:

Clumping indicates that the simulated blood sample contains antigens that reacted against the antibodies in the typing serum that the students mixed it with.

Type A

If the blood in Well A is the only blood that agglutinates or clumps, then the blood sample you tested is Type A blood.

Type B

If the blood in Well B is the only blood that agglutinates or clumps, then the blood sample you tested is Type B blood.

Type AB

If the blood in both Well A and Well B agglutinates or clumps, then the blood sample you tested is type AB blood.

Type 0

If the blood in both Well A and Well B does not agglutinate or clump, then the blood sample you tested is Type O blood.

Rh

If the blood in Well Rh agglutinates or clumps, then the blood sample you tested is Rh Positive blood.

Data Table 5

Simulated Blood Sample	Agglutination in Well A (+/-)	Agglutination in Well B (+/-)	Agglutination in Well Rh (+/-)	Blood Type	Observations
Patient 1	+	-	+	A +	
Patient 2	-	+	-	В-	Students should describe the
Patient 3	+	+	+	AB+	
Patient 4	-	_	-	0-	

Questions

1. If your blood type is B, which antigens are present on your red blood cells? What if your blood type is A, type AB, or type O?

Individuals with blood type B have type B antigens on the red blood cells; type A individuals have A antigens; type AB individuals have A and B antigens and type O individuals have none.

2. Based upon your results, which ABO blood type, can Patient 1 receive safely? Patient 2? Patient 3? And Patient 4?

Patient 1's blood type is A and therefore can receive type A and O blood.

Patient 2's blood type is B and therefore can receive type B and O blood.

Patient 3's blood type is AB and is considered a universal recipient and can receive type AB, A, B or O blood.

Patient 4's blood type is O and can only receive type O blood.

3. Which patient is considered a universal donor?

Patient 4's blood type is O and is considered a universal donor, since there are no antigens on the red blood cells.

4. How could you determine if a blood sample is compatible to transfuse from one individual to another in an emergency situation, if blood typing serum is not available?

A small amount of blood can be drawn from each individual and mixed together to determine if the red blood cells will agglutinate or clump together. If there is no agglutination, then the blood types are compatible and can be transfused from one individual to the other.

5. How is this simulated blood typing activity similar to actual human blood typing?

The blood typing procedure for simulated blood is similar to actual blood typing. Simulated blood agglutinates in a similar fashion to actual blood when mixed with the appropriate antiserum.

6. What would happen to a type O patient if he receives type A or B blood?

The antibodies in the plasma of a type O patient would attach to the antigens that are found on the red blood cells of the A or B blood and cause the red blood cells to clump together. Transfusion of type A or B blood type to a type O patient would produce potentially fatal complications.

7. What are the consequences of Rh incompatibility?

Rh-negative individuals who have been transfused with Rh-positive blood can produce Rh antibodies. They may develop a transfusion reaction, during which agglutination may occur, if they are transfused again with Rh-positive blood. Usually Rh compatibility is tested when the ABO blood type is determined.

Unit 3 What's your blood type? Using SimulatedNeo/BLOOD 1–800–526–6689



Lab Activity Procedures & Notes

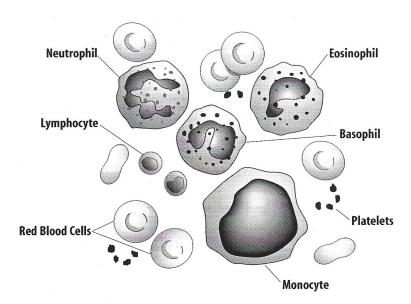
ACTIVITY



Taking a Closer Look at Blood

Physicians and other health care professionals regularly examine blood under the microscope to identify infections, blood cell abnormalities and to count the various types of cells.

The cells of the blood are of two classes: red blood cells (RBCs), or erythrocytes; white blood cells (WBCs), or leukocytes, which in turn are of many different types. Platelets, or thrombocytes, are also present as are cell fragments.



Red Blood Cells (RBCs)

The red blood cells are tiny, round, biconcave disks, without nuclei, that average about 7.5 microns (0.003 in) in diameter. Red blood cells, as well as most white cells and platelets, are made by the bone marrow. The main function of the red blood cells is to transport oxygen from the lungs to the tissues. A healthy 70kg (154lb) man has about 5 L (5.3 qt) of blood in his body, containing more than 25 trillion RBCs. The normal life span of RBCs in the circulation is only about 120 days. Worn out RBCs are removed by the spleen and liver where hemoglobin is recycled.

A number of conditions can be diagnosed based upon the red blood cell count. A high RBC level, a condition called "erythrocytosis," can be caused by smoking, living at high altitudes, or by disease. Low red blood cell levels, a condition called "anemia," can be due to a loss of blood, loss of iron, a vitamin deficiency, or other disease conditions.

Data Table 6
Normal and Abnormal Red Blood Cell Counts

RED BLOOD CELLS			
	MALE	FEMALE	
Normal (at birth)	5.1 million cells per µl	4.5 million cells per µl	
Normal (adult)	5.4 million cells per µl	4.8 million cells per µl	
Anemia (low RBC count)	< 4.5 million cells per µl	< 4 million cells per µl	
Erythrocytosis (high RBC count)	> 6.8 million cells per µl	>6 million cells per µl	

What to do...

•••••

•••••

••••••

White Blood Cells (WBCs)

Leukocytes, or white blood cells, are considerably larger than red cells, have nuclei, and are much less numerous; only one or two exist for every 1,000 red blood cells, and this number increases in the presence of infection. There are three types of leukocytes, all involved in defending the body against foreign organisms: granulocytes, monocytes, and lymphocytes. There are three types of granulocytes: neutrophils (the most abundant), eosinophils, and basophils.

Platelets

Platelets (thrombocytes) are tiny bits of cytoplasm, much smaller than the red blood cells, which also lack nuclei. They are normally about 30 to 40 times more numerous than the white blood cells. They are produced as fragments of the cytoplasm of the giant cells of the bone marrow-the megakaryocytes. The platelets' primary function is to stop bleeding. When tissue is damaged, the platelets aggregate in clumps as part of the clotting process.

What you need

Blood sample (simulated)
Compound microscope
Coverslip
Microscope slide

Step 1

Have your students place a minute drop of simulated blood on a microscope slide.

Step 2

They should then place a coverslip on the simulated blood and place the slide under a compound microscope.

Step 3

The students should examine the simulated blood smear at 430x magnification and note the various types of simulated cells. Red blood cells will appear pink, white blood cells are stained blue, and platelets will be seen as amorphous shaped cell fragments.

Step 4

Have the students use the following simplified procedure to estimate the number of simulated red blood cells in 1 μ l of volume of blood in a given blood sample.

The students should count the number of simulated red blood cells seen in their field of view. Assuming that the field of view at 430x magnification represents 0.01 μ l of blood, and that the dilution factor is 1000x, then the number of blood cells that would be found in 1 μ l of whole blood can be calculated as follows:

(Cells in field of view) x (Dilution factor) = Number of Red Blood Cells in 1 µl volume of blood

Example: Suppose you count 45 simulated red blood cells in the field of view, then,

 $\frac{45 \times 1000}{0.01 \, \mu l} = 4,500,000 \, \text{red blood cells per } \mu l \, \text{of blood}$

Step 5

Have students select several other fields of view and repeat Step 4. Students average their results to obtain a closer representative number of red blood cells per µl of blood.

Unit 3 What's your blood type? Using SimulatedNeo/BLOOD 1–800–526–6689



- 1. Based upon your results, was the red blood cell count within the accepted normal range for an adult male?
 - Individual student answers will vary. Generally, students should obtain between 4.5 million and 5.0 million per μ l of blood.
- 2. What does a high number of red blood cells indicate? A low level?
 - A high red blood cell count is a condition known as erythrocytosis which may be caused by smoking, living at high altitudes or by disease. A low red blood cell count is an indication of anemia, which may be due to loss of blood, iron deficiency or other conditions.

3. What does a high number of white blood cells indicate?

A high white blood cell count indicates an infection or a blood disorder. An increase in number of select white blood cells indicates specific disorders, ranging from mononucleosis to leukemia and other conditions.

Cross-Curricular Integration

Technology

Discuss with your students the various advances that are being made to develop artificial blood which would eliminate the need of human blood transfusion in emergency situations.

Health

Relate blood analysis to various health conditions which can be diagnosed based upon blood cell counts and morphology. Discuss with your students the various blood-borne pathogens that are carried in blood and the diseases that they cause.

Social studies

Discuss with your students the process of blood donation and the crucial function that it serves in saving people's lives in a medical emergency. Discuss the facts and myths of blood donation.

Mathematics

Various math concepts can be incorporated in this investigations in which students determine the percent frequency of specific blood types in a given population.

Assessment

The questions included in the Questions section are provided as a means for you to check your students' understanding of the concepts, whether they can apply them to everyday situations, and also their ability to think critically and visually.

There are certain ideas that your students should be able to understand after completing this activity. They should be able to understand the ABO and Rh blood typing system, the blood typing procedure and interpret blood typing results. Students should also be able to understand and define terms such as antigens, antibodies, serum, red and white blood cells, hemoglobin, hematocrit and other terms presented in this investigation.

Additionally, you could set up a teacher-designed laboratory practical examination which displays the lab setup and asks questions relating to it.

Students could be required to make an oral presentation to their class who would ask selected questions to evaluate student learning of blood typing and blood cell structure and function.

Students could demonstrate their learning by conducting the activities in the Going Further section or completing an Internet search and presentation of what they found.

Unit 3 What's your blood type? Using SimulatedNeo/BLOOD 1–800–526–6689