

ABO-Rh Blood Typing with Synthetic Blood



TEACHER'S MANUAL
AND STUDENT GUIDE

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TEACHER'S MANUAL

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STUDENT GUIDE

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Photocopy the Student Guide as needed for use in your classroom.

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ABO-Rh Blood Typing with Synthetic Blood

Overview

This kit teaches students about the A, B, O, Rh system of human blood typing. In the process they learn about some of the components of the blood and some basic concepts of immunology. Students test four simulated blood samples to identify their ABO and Rh blood types. The test procedures used in this kit are the same as those used to test real blood. However, the kit contains synthetic blood and synthetic antisera, eliminating any risk associated with exposure to real blood or blood products.

The lab can be completed in two 50-minute class periods. The materials in the standard kit are sufficient for 30 students working in 15 pairs. The materials in the Classroom Kit are sufficient to repeat the lab with four classes of 30 students working in pairs. The Classroom Kit also contains materials for distributing aliquots of the synthetic blood and antisera to avoid classroom bottlenecks.

Correlation to the Next Generation Science Standards*

The activities in this kit address the following dimensions of the Next Generation Science Standards:

HS-LS1-1. Construct an explanation based on evidence for how the structure of DNA determines the structure of proteins which carry out the essential functions of life through systems of specialized cells.

Science and Engineering Practices	Disciplinary Core Ideas	Crosscutting Concepts
<p>Developing and Using Models</p> <ul style="list-style-type: none"> Use a model based on evidence to illustrate the relationships between systems or between components of a system. 	<p>LS1.A: Structure and Function</p> <ul style="list-style-type: none"> Systems of specialized cells within organisms help them perform the essential functions of life. 	<p>Structure and Function</p> <ul style="list-style-type: none"> Investigating or designing new systems or structures requires a detailed examination of the properties of different materials, the structures of different components, and connections of components to reveal its function and/or solve a problem.

To view additional national and local standards met by this kit, visit www.carolina.com/correlations.

*"Next Generation Science Standards" is a registered trademark of Achieve. Neither Achieve nor the lead states and partners that developed the Next Generation Science Standards was involved in the production of, and does not endorse, this product.

Source: NGSS Lead States, 2013. *Next Generation Science Standards: For States, By States*. Washington, DC: The National Academies Press.

Objectives

Students' performance objectives are to

- synthesize information from experiments and simulations into a coherent understanding of the structural and functional components of blood.
- use reasoning to construct an explanation that blood cells are specialized cells with similar structure and functions, yet the specific antigens presented on the cell's surface may interact with specific antibodies as a function of the immune response.
- perform simulations of standard tests used for blood type identification.

Prerequisite Knowledge and Skills

Students should already possess basic familiarity with the fluid mosaic model of the cell membrane. Students should understand that an antigen is any substance to which the immune system might respond. It may be useful to project an image of the fluid mosaic model of the cell in order to review both peripheral proteins and glycoproteins before the lab activity. ABO blood group antigens are *sugars* determined by a person's genes. Rh group antigens are *proteins* determined by a person's genes. Some people possess a version of the Rh gene that does not produce a surface antigen on red blood cells.



Time Requirements

Teacher Preparation	90 minutes
Prelab	50 minutes
Laboratory Investigation	30 minutes
Assessment	20 minutes



Safety

Use this kit only in accordance with established laboratory safety practices, including appropriate personal protective equipment (PPE) such as gloves, chemical splash goggles, and lab coats or aprons. Ensure that students understand and adhere to these practices.

There are no biological materials in the synthetic blood or synthetic antisera that cause any health hazard to students or that affect the method of disposal. Materials in this kit may be discarded after use.

Download Safety Data Sheets (SDS) at carolina.com/sds or scan this code:



Digital Resources

Your kit includes a digital Teacher's Manual with hyperlinks to the following resources. Additional resources may be available. To use these resources, log on to the website below and enter your access code. See the Digital Resource Instruction Card for more information.

<http://www.carolinascienceonline.com>

Digital resources included with this kit:

RESOURCE	DESCRIPTION
Student Guide Copy Master	Student Guide PDF for printing
Fill-in Answer Sheets	A PDF that can be printed out or assigned digitally, with spaces for students to record their data and answers
Editable Assessment Questions	The assessment questions as a Microsoft® Word document
Whiteboard Resources	Color graphics for use with whiteboards

Materials

Your kit includes a digital Teacher's Manual and Student Guide. See the Digital Resource Instruction Card for more information.

Note: Store all the materials at room temperature. The shelf life of the synthetic blood is 1 year or longer. *Shake the blood well before using it.*

Included in both kits 700101 and 700160:

- 30 blood typing slides
- blue mixing sticks
(120 in kit 700101, and 240 in kit 700160)
- yellow mixing sticks
(120 in kit 700101, and 240 in kit 700160)
- white mixing sticks
(120 in kit 700101, and 240 in kit 700160)
- Digital Resource Instruction Card
- Teacher's Manual and Student Guide

Synthetic blood and antisera included in kit 700101 and refill 700102:

- 7-mL vial of Sample 1 (A+ synthetic blood)
- 7-mL vial of Sample 2 (B+ synthetic blood)
- 7-mL vial of Sample 3 (AB- synthetic blood)
- 7-mL vial of Sample 4 (O+ synthetic blood)
- 7-mL vial synthetic anti-Rh (D) serum
- 7-mL vial synthetic anti-A serum
- 7-mL vial synthetic anti-B serum

Synthetic blood and antisera and extra labware included in Classroom Kit 700160 and refill 700161:

- 30-mL vial of Sample 1 (A+ synthetic blood)
- 30-mL vial of Sample 2 (B+ synthetic blood)
- 30-mL vial of Sample 3 (AB- synthetic blood)
- 30-mL vial of Sample 4 (O+ synthetic blood)
- 30-mL vial synthetic anti-Rh (D) serum
- 30-mL vial synthetic anti-A serum
- 30-mL vial synthetic anti-B serum
- 8 empty vials for A+ synthetic blood
(labeled Sample 1)
- 8 empty vials for B+ synthetic blood
(labeled Sample 2)
- 8 empty vials for AB- synthetic blood
(labeled Sample 3)
- 8 empty vials for O+ synthetic blood
(labeled Sample 4)
- 15 empty vials for synthetic anti-Rh (D) serum
- 15 empty vials for synthetic anti-A serum
- 15 empty vials for synthetic anti-B serum
- 30 3-mL bulb pipets

Needed, but not supplied:

- two different colored pencils for each student
- tap water for washing the slides and the mixing sticks
- paper towels for drying the slides
- permanent marker

Preparation

1. Review the content of the Teacher's Manual and the Student Guide. Familiarize yourself with the Prelab and Laboratory Investigation instructions, materials, and assessments. Review the classroom management procedures and the timing of the materials setup steps described below.
2. Gather the materials that are needed, but not supplied. Make certain you have appropriate personal protective equipment for every student in your class.
3. Photocopy or print one copy of the Student Guide for each student.
4. Gather the materials that are needed but not supplied.

Prelab

1. Plan how you will draw (or show) the illustrations modeled in the Prelab activity script. If you intend to do so, plan how you will project the worksheet images—on a chalkboard, whiteboard, or using a document camera.
2. Obtain two colored pencils of two different colors for each student.

Laboratory Investigation

1. Follow the preparation steps below, according to whether you purchased the standard kit or the Classroom Kit.

If you bought the standard kit, 700101:

- a. Set up 15 workstations (one for each group), each stocked with the following:
 - 4 blue mixing sticks
 - 4 yellow mixing sticks
 - 4 white mixing sticks
 - blood typing slide
 - paper towels
- b. Set up a common working area stocked with the following:
 - vial synthetic Sample 1
 - vial synthetic Sample 2

- vial synthetic Sample 3
- vial synthetic Sample 4
- vial synthetic anti-A serum
- vial synthetic anti-B serum
- vial synthetic anti-D (Rh) serum

The groups also will need access to tap water and a sink.

If you bought the Classroom Kit, 700160:

- a. Using the labeled, empty, blood sample vials and the 3-mL bulb pipets included with the kit, create 8 2-mL aliquots of each of the synthetic blood samples—Sample 1, Sample 2, Sample 3, and Sample 4. Shake the blood well before preparing aliquots. Make sure to use a clean pipet for each different blood sample. To avoid cross contamination, label each pipet (1, 2, 3, or 4) for use with only one type of blood sample and then work with the materials for one sample at a time.
- b. Using the labeled, empty, antisera vials and the 3-mL bulb pipets included with the kit, create 8 2-mL aliquots of each of the synthetic antisera—anti-A, anti-B, and anti-D (Rh). Make sure to use a clean pipet for each different antiserum. To avoid cross contamination, label each pipet for use with only one type of antiserum and work with the materials for only one antiserum sample at a time.
- c. Set up 15 workstations (one for each group), and stock each with the following materials:
 - 2-mL aliquot of synthetic anti-D (Rh) serum
 - 2-mL aliquot of synthetic anti-A serum
 - 2-mL aliquot of synthetic anti-B serum
 - blood typing slide
 - 4 blue mixing sticks
 - 4 yellow mixing sticks
 - 4 white mixing sticks
 - paper towels

Science Background

Multiple Alleles and Codominance

The multiple alleles considered in the ABO blood typing system do not follow basic Mendelian principles of complete dominance. Instead, the expression of the surface antigen trait in the ABO blood typing system is governed by the genetic principle of codominance, where both A and B are codominant alleles. Therefore, a person inheriting a Type A allele from their mother and a Type B allele from their father would express the codominant phenotype, Type AB. Because the only recessive allele in the ABO system is the O allele, only homozygous recessive individuals express the Type O phenotype, which is characterized by a lack of A and B surface antigens on the red blood cell. As the following table illustrates, it is possible to express Type A and Type B phenotypes in the heterozygous condition.

Blood Type Chart

Blood Type (Phenotype)	Surface Antigen Protein(s)	Possible Alleles in Genotype
Type A	A	AA or Ao
Type B	B	BB or Bo
Type AB	A and B	AB
Type O	none	oo

The Rh Factor

Early studies using rhesus monkeys as test subjects allowed scientists to discover what is now understood as the Rh factor. The expression of the Rh family of blood cell surface antigens is binary. That is, a person either presents Rh antigens (Rh positive phenotype; homozygous or heterozygous) or does not (Rh negative phenotype; homozygous negative).

Rh Factor Chart

Blood Type (Phenotype)	Surface Antigen Proteins	Possible Alleles in Genotype
Rh+	+	+ + or + -
Rh-	none	-

A Rh+ individual will not produce Rh antibodies. However, a Rh-negative individual may produce Rh+ antibodies upon exposure to the Rh antigen. As the following Punnett square demonstrates, it is possible that a Rh- mother and a Rh+ father may produce a Rh+ baby during a fertilization event.

Parental Genotypes: ♂ + - × ♀ - -

	+	-
-	+ -	- -
-	+ -	- -

Placental transfer of maternal antibodies to a developing fetus is an essential mechanism that serves to protect a developing fetus *in utero*. However, students may not understand that this placental transfer mechanism is unidirectional, exclusively from mother to fetus. Because the blood of a fetus and that of the mother do not mix, a

Rh⁻ mother should not be exposed to her child's potential Rh⁺ blood antigens, and will therefore not initiate a specific Rh⁺ antibody driven immune response that may attack her unborn child.

However, there are circumstances in which a mother could initiate an immune response against a fetus during a subsequent pregnancy. For instance, a Rh⁻ mother and Rh⁺ infant's blood may mix during childbirth, an event which may trigger the production and accumulation of Rh⁺ antibodies by the mother's immune response. During subsequent pregnancies, a Rh⁻ mother will send antibodies through the placental barrier that have the potential to harm a Rh⁺ fetus. As a result, many developed countries give Rh⁻ mothers an injection during the 28th week of pregnancy that acts to suppress the maternal immune system from attacking potential Rh⁺ cells in her fetus.

Rh-negative Blood: An Evolutionary Disadvantage?

Students may wonder why the Rh-negative genotype occurs at all if there is the potential for a mother to harm her unborn child. It is likely that Rh-negative blood originated from a mutation in the RHD gene, which results in the lack of a specific functional protein product being "translated" or made. Although individuals with Rh-negative blood are rare, the trait may confer some advantages to those carrying the Rh⁻ allele. It is hypothesized that the heterozygous condition may confer some protection against the most severe effects of parasitic threats such as *Toxoplasma gondii*, and therefore may be selected for by evolutionary mechanisms.

Teaching Guide

Overview

During this fun and simple pre-laboratory guided inquiry activity, students are introduced to the human ABO blood typing system, surface antigens, antibodies, and the mechanisms that govern blood type compatibility. A fictional tale of four planets, each representing a blood type, is used to introduce the ABO blood typing system. Working in groups of 2 or 3, students draw surface antigens and antibody defense mechanisms as they learn about some of the components of blood and the basic concepts of immunology. Students demonstrate understanding by applying the fictional tale to determine which blood type acts as the universal donor, and which blood type acts as the universal receiver. A student worksheet and script is provided for the teacher. The lesson assumes that students have little or no prior knowledge of the ABO blood typing system, antigens, antibodies, or the humoral immune response.

Background

Specific antigens on red blood cells determine whether a human has type A, B, AB or O blood. As B cells generate antibodies to establish humoral immunity, antibodies to “non-self” antigens regularly circulate in the blood stream. In this manner, a person with type A surface antigens (type A blood type) would generate antibodies specific to foreign antigens, such as type B antigens (found on both type B and type AB blood). In response to a foreign threat presented by non-self antigens, a full-scale humoral immune response may be initiated to help attack, and ultimately neutralize, the invading antigen presenting blood cell. Due to this defense mechanism, a life-threatening situation may occur if a type A individual receives a blood transfusion from an incompatible donor.

Procedure

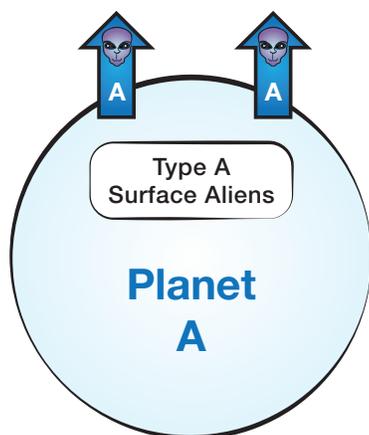
1. Distribute the Student Guide to each student, and direct students to the Prelab worksheet titled “Interplanetary Battle for the Blood Types.” (Students can work independently or in groups of 2 or 3 depending on your preference.)
2. Read the following **Teacher Script** aloud and draw with students at each step. You may choose to simply recreate the images on a board, project them on a smartboard, or use a document camera to project the blank student worksheet. Check students’ progress and drawings at each step.
3. Guide students through the Prelab Analysis and Prelab Questions. By the end of the exercise, students should recognize the fictional tale as analogous to the basic mechanisms of the ABO-Rh blood typing system.

Teacher Script: Interplanetary Battle for the Blood Types

1. Read aloud the following:

Once upon a time in a galaxy fairly close to our own, four planets orbited a sun. They were known as Planet A, Planet B, Planet AB, and Planet O. On the surface of Planet A, all of the Type A surface aliens looked like arrows. Please draw them with me.

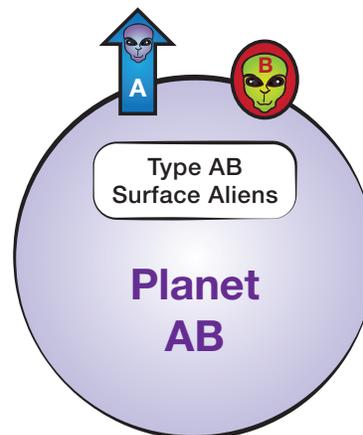
Action: Draw two arrow-shaped Type A aliens on Planet A. Have students do the same. (An example is shown below.)



3. Read aloud the following:

On the third planet, Planet AB, both types of surface aliens live in perfect harmony. When visiting other planets, one surface alien A always travels with one surface alien B. Please draw both Type A and Type B surface aliens on Planet AB.

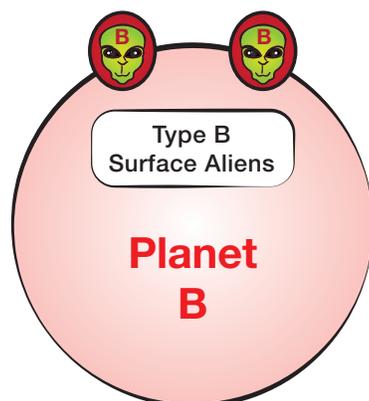
Action: Draw one Type A and one Type B alien on the surface of Planet AB. Have students do the same. (An example is shown below.)



2. Read aloud the following:

As you might expect, only Type B aliens live on the surface of Planet B. The Type B surface aliens have a very different shape than the Type A surface aliens. Please draw the egg-shaped Type B surface alien.

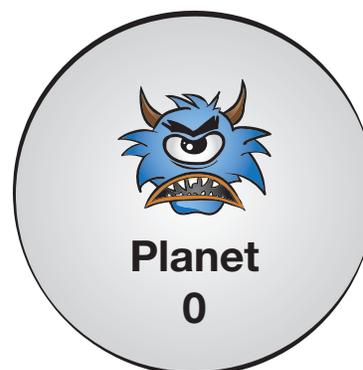
Action: Draw two egg-shaped Type B aliens on Planet B. Have students do the same. (An example is shown below.)



4. Read aloud the following:

Absolutely no surface aliens live on the surface of the fourth planet, Planet O. Instead, a horrible ogre lives deep inside the planet. Draw the ogre. Notice that the surface of Planet O is barren.

Action: Draw the Ogre within Planet O. Have students do the same. (An example is shown below.)



5. Read aloud the following:

Being a horrible kind of guy, the ogre decides to set up a defense system that orbits around his barren planet. His Anti-A defense system is perfectly shaped to trap the heads of the Type A surface aliens, the first step in their neutralization and ultimate demise! His Anti-B defense system works in a similar manner, recognizing the specific shape of the Type B surface alien's head and attaching to it. Please draw the Ogre's defense systems with me. Let's assume that one Type A and one Type B surface alien are attempting to land on Planet O.

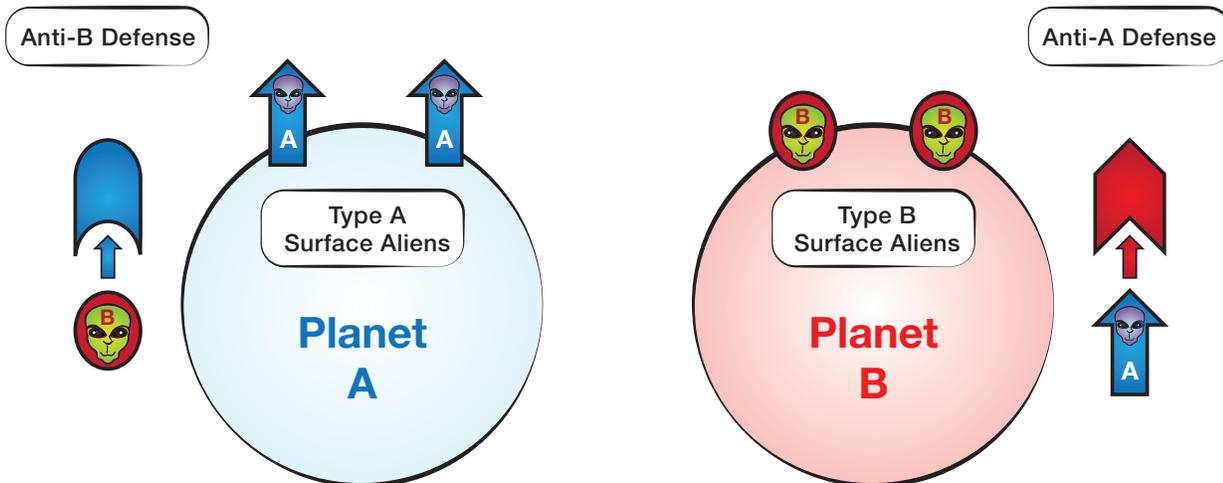
Action: Draw the defense system of Planet O. Demonstrate how the specific shape of the Anti-A and the Anti-B defense mechanisms bind to the specific shape of the surface alien attempting to visit the planet. Be sure to indicate that the defense mechanism is found **surrounding**, rather than **on**, Planet O. Have students draw the active defense mechanisms of Planet O. (An example is shown below.)



6. Read aloud the following:

One day, a spacecraft from Earth carrying three kittens was discovered by the inhabitants of planets A, B, and AB. Now, these weren't just any kittens—these kittens were the cutest kittens in the entire universe. The inhabitants of Planet A and Planet B fought so intensely over who should get to keep the kittens that the two planets placed a permanent ban on interplanetary visitation. Planet A built an Anti-B defense system to prevent any Type B surface aliens from visiting. Incensed by this belligerent act, Planet B built an Anti-A defense system to prevent any Type A surface aliens from visiting. Let's illustrate the outcome of this conflict.

Action: Draw the defense system of Planet A and the defense system of Planet B. Demonstrate how the specific shape of the Anti-A and Anti-B defense mechanisms to bind to the specific shape of the surface alien attempting to visit. Be sure to indicate that the defense mechanism is found **surrounding**, rather than **on**, each planet. Have students draw the active defense mechanisms of Planet A and Planet B. (An example is shown below.)



7. Read aloud the following:

Unfortunately for its peaceful inhabitants, the surface aliens found on Planet AB are caught in the interplanetary crossfire. As surface aliens on Planet AB travel together, an attack on surface alien A will cause its partner, surface alien B to suffer, and vice versa. The universe has become very dangerous for the inhabitants of Planet AB.

Appalled by the conflict, Planet AB abandons all defensive action, choosing instead to remain neutral. Planet AB continues to be a place of harmony to this day, allowing all interplanetary visitors access to its surface. The inhabitants of Planet AB are perfectly content with their own kittens, which are of average cuteness.

Action: Students' drawings are complete. Point out to students that there is absolutely no Anti-O defense system, and that this observation will come in handy shortly.

Analysis

1. In small groups, allow students to use their knowledge from the storyline to fill in the table on the worksheet. It may be helpful to remind students that each surface alien always can visit its own planet. For example, Type A surface aliens can visit Planet A.
2. Ask students which planet is the universal receiver. Students should determine that Planet AB is the universal receiver. Have students write "universal receiver" in the column cell for Planet AB.
3. Ask students which planet is the universal donor. In other words, which planet's inhabitants may visit any other planet without fear of attack? Students should determine that Planet O is the universal donor. Ask students to write "universal donor" on the column cell for Planet O.
4. A summary of the completed table is provided on page 15. Review students' answers as a class discussion, and address any misconceptions students might have.
5. By this point, students should have figured out that this activity focuses on blood types rather than planets. Ask groups to read the student background and then as a class, determine the correct identity of each of the following characters/events in the storyline. It may be helpful for students to cross out the following words and replace them with ABO blood typing terminology, as follows:

Replace	planet	with	blood type (specifically, the red blood cell belonging to a blood type)
Replace	surface alien	with	blood surface antigen
Replace	defense system	with	antibody (found in plasma surrounding the red blood cell)

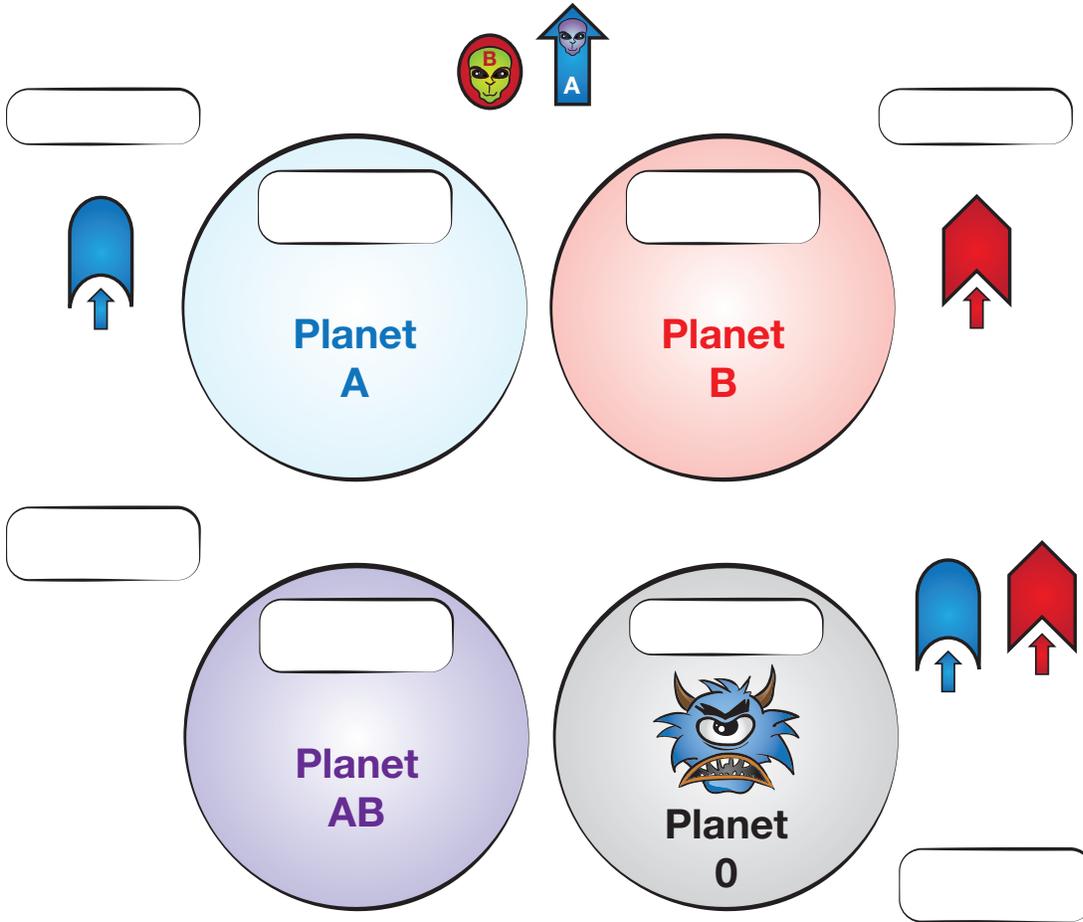
6. Have students answer the Prelab Questions, filling in the tables individually or as a class discussion. Answers are provided in the next section.

Prelab

STUDENT GUIDE

Name _____ Date _____

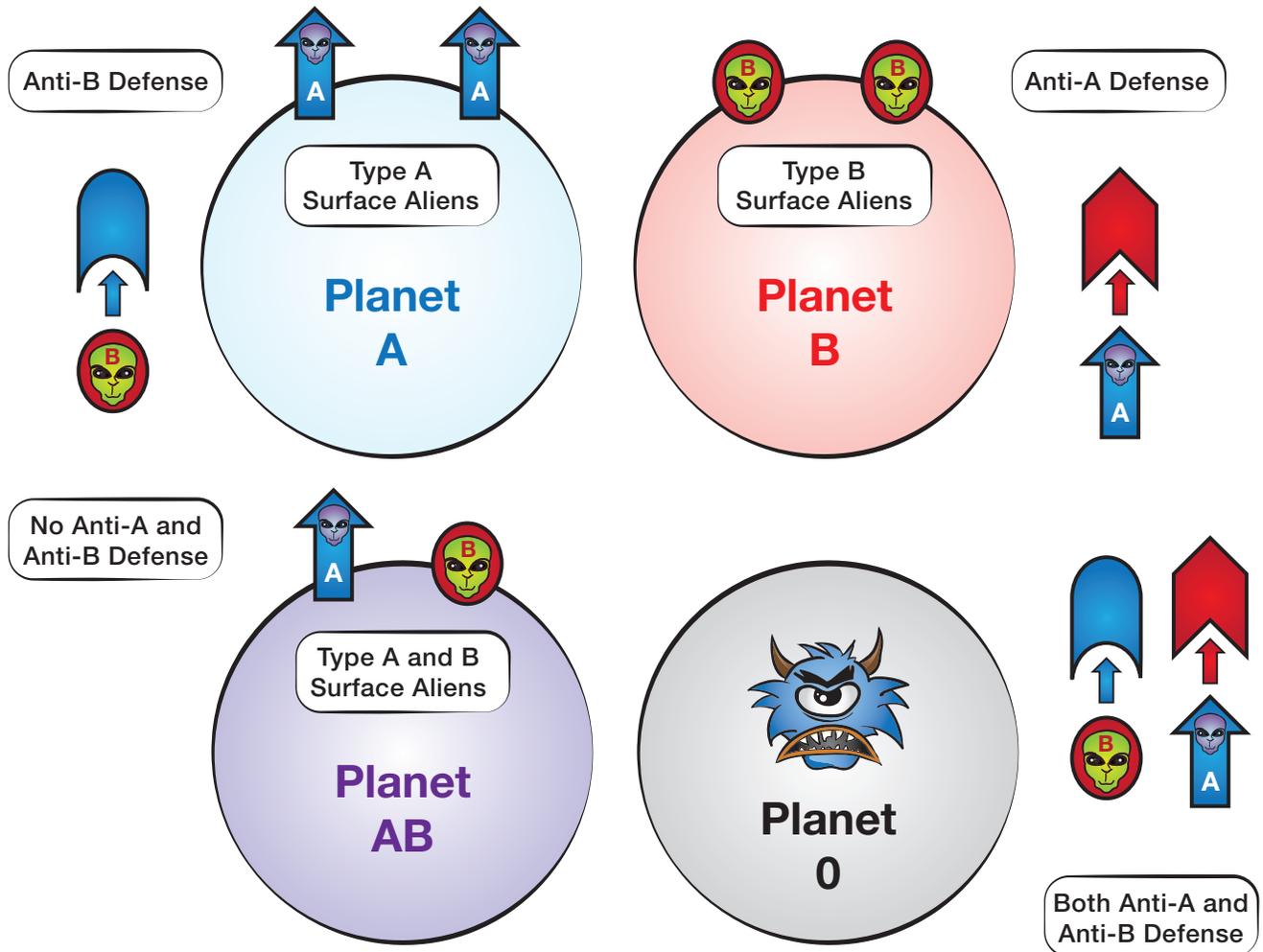
Interplanetary Blood Typing Battle



Prelab Analysis

Planet	Surface Alien(s)	Defense System	Aliens from this planet can visit:	This planet can receive visitors from:
A				
B				
AB				
O				

Answers to Interplanetary Battle for the Blood Types



Planet	Surface Alien(s)	Defense System	Aliens from this planet can visit:	This planet can receive visitors from:
A	Type A	Anti-B	Planet A or Planet AB	Planet A, Planet O
B	Type B	Anti-A	Planet B or Planet AB	Planet B, Planet O
AB	Type A and Type B	None	Planet AB only	all planets (universal receiver)
O	None	Anti-A and Anti-B	all planets (universal donor)	Planet O only

Prelab Questions

- Shown in the table below are the antigens present on the blood cells of four different people. Fill in the corresponding ABO blood type for each person and the antibodies expected to be found in each person's blood.

Blood Type	Red Blood Cell Antigen	Antibody in Blood Plasma
	A	
	B	
	AB	
	Neither	

- For each blood type in the table below, fill in the expected agglutination result from mixing the blood with each of the antibodies.

Blood Type	Anti-A	Anti-B	Anti-Rh
A+			
A-			
B+			
B-			
AB+			
AB-			
O+			
O-			

Answers to Prelab Questions

1. Shown in the table below are the antigens present on the blood cells of four different people. Fill in the corresponding ABO blood type for each person and the antibodies expected to be found in each person's blood.

Blood Type	Red Blood Cell Antigen	Antibody in Blood Plasma
A	A	Anti-B
B	B	Anti-A
AB	AB	Neither
O	Neither	Anti-A, Anti-B

2. For each blood type in the table below, fill in the expected agglutination result from mixing the blood with each of the antibodies.

Blood Type	Anti-A	Anti-B	Anti-Rh
A+	Yes	No	Yes
A-	Yes	No	No
B+	No	Yes	Yes
B-	No	Yes	No
AB+	Yes	Yes	Yes
AB-	Yes	Yes	No
O+	No	No	Yes
O-	No	No	No

Real-World Connections

Students will be interested in what happens in a real-life scenario if someone receives a transfusion from an individual with an incompatible blood type. For instance, allow students to use their worksheet and associated drawings to predict what would happen if a person with Type A blood has given Type B blood in a transfusion. Students should predict that Type A blood carries Anti-B plasma antibodies that will recognize and then attempt to destroy the Type B blood cells that carry the foreign Type B surface antigen.

Using the ABO blood typing slides, allow students to observe a couple drops of Type B synthetic blood (Sample 2) placed in each of the three wells. Ask students what surface antigens are present on Type B blood cells. Add Anti-A antibody serum to the Anti-A well. As you mix the sample, ask students if type Anti-A antibody serum recognizes and destroys Blood Type A surface antigens or Blood Type B surface antigens. Remind them that the answer is in the name of the serum used. Students should conclude that Anti-A antibody serum recognizes and attacks Type A blood. Demonstrate that Type B blood has no reaction with Anti-A serum, as it has no Type A surface antigens to attack.

Ask students to choose the antibody serum that is most likely to recognize and attack Type B blood. As student conclude that Anti-B antibody serum attacks Type B surface antigens, allow students to observe the agglutination event resulting from mixing Type B Blood with Anti-B serum.

Finally, relate that an agglutination event is related to a positive result. In other words, Anti-A serum only reacts with A antigens, Anti-B with B antigens, and Anti-Rh with Rh+ antigens. Perform the final anti-Rh test and verify that students understand the results.

Laboratory Investigation

STUDENT GUIDE

Name _____ Date _____

Background

By volume, blood tissue is approximately 55% plasma and 45% cells. Plasma is a straw-colored liquid consisting of 90% water and a variety of salts and proteins important for maintaining osmotic balance, buffering against pH changes, maintaining blood viscosity, transporting certain materials, and for blood clotting when a blood vessel is injured.

There are three major types of **blood cells**—red blood cells (erythrocytes), white blood cells (leukocytes), and platelets.

Red blood cells, the most numerous cells in the blood, carry oxygen from the lungs to all parts of the body. A red blood cell is a biconcave disk with a thin center. This shape provides a large surface area for diffusion of oxygen. Red blood cells contain the protein hemoglobin. Iron is incorporated into the hemoglobin molecule. When blood travels through the lungs, the oxygen in the lungs combines with the iron in hemoglobin. When the blood moves through the body's capillary system, the oxygen carried in the red blood cells is released from the iron in hemoglobin to the other cells of the body. Red blood cells' flexibility allows them to pass through even the smallest blood vessels.

White blood cells make up only about 1% of the blood volume. They are an important part of the **immune system**. Their primary function is to provide defense against invaders in the body, which may include bacteria, parasites, fungi, and viruses. White blood cells may attack a foreign body directly, they may produce antibodies that identify, attach to, and neutralize a foreign body, or they may trigger other cells to act in destroying the foreign body.

Platelets perform a vital function in the process of coagulation, or blood clotting, which occurs when a blood vessel is injured.

Blood Types

Although the basic composition and function of blood in each of us is the same, there are different human blood types. The cell membrane of red blood cells, like that of other cells, has molecules that project from its surface. Some of the molecules function as identification badges, allowing the immune system to recognize the cell as a normal component of an individual's body. If blood from a person whose red cells have different surface molecules is injected into someone, those molecules are recognized as **antigenic**, or foreign to the body. The immune system attacks the antigens and attempts to destroy them and the cells that carry them. This is why transfusion with an incompatible blood type is harmful. The recipient's body recognizes the antigens on the transfused red blood cells as foreign and attacks and destroys the cells. For that reason, donated blood is thoroughly tested for A, B, O, and Rh antigens and is transfused only into compatible recipients.

The ABO Blood Groups

The ABO blood groups (types) result from the presence or absence of two antigens, A and B, on the surface of the red blood cells. If antigens are present very early in life, the immune system recognizes those antigens as "self" and will not generate an immune response to them. As a result, the body does not generate antibodies to any A and B antigens present on its own blood cells. However, the immune system does produce antibodies to any A and B blood antigens not present on the organism's own cells. Type A blood has the A antigen on its red blood cells and anti-B antibodies in the plasma. Type B blood has the B antigen on its red blood cells and A antibodies in the plasma. Type AB blood has both A and B antigens on the red blood cells and no antibodies in the plasma. Finally, Type O blood has neither A nor B antigens on the red blood cells and both A and B antibodies in the plasma.

(Student Guide pages S-4–S-6 not shown)

Using the Key Question

The Key Question may be used to engage students in the laboratory investigation and to help them identify what they learned from the experience. Before starting the investigation, have your students read the question and respond to it in their notebook. After completing the investigation, have them return to the Key Question and revise their answer on the basis of their observations and conclusions. You may wish to center a class discussion on the question both before and after the laboratory investigation.

Procedure

- Review relevant safety protocols with students.
- Before beginning the lab, review with students the designations of the blood groups (A+, A-, etc.) and their agglutination reactions.
- Have students conduct the lab according to the instructions in the Student Guide. Monitor the class to ensure that students are using safe laboratory techniques and wearing appropriate PPE.
- Remind students to type all four synthetic blood samples.
- Make sure that students sufficiently rinse the blood typing slide in between testing the different blood samples.
- The blood typing slides and mixing sticks can be reused. After students have completed the lab activities, have them use water to clean the blood slides and mixing sticks thoroughly.
- Have the students compare their results. If a student has a result that differs from that obtained by the rest of the class, discuss what may have happened. Possible causes:
 - The blood sample of antiserum was contaminated.
 - The blood sample was not sufficiently mixed before drops were placed in the wells.
 - The blood sample was not sufficiently mixed with the antiserum.
 - Insufficient time was allowed before viewing the reaction.
 - The wrong antiserum was added to the well.
 - The wrong blood sample was added to the well.
- Have students answer the Assessment Questions in the Student Guide.

Expected Results

	Sample 1	Sample 2	Sample 3	Sample 4
Anti-A	Yes	No	Yes	No
Anti-B	No	Yes	Yes	No
Rh	Yes	Yes	No	Yes
Blood Types	A+	B+	AB-	O+

Assessment

STUDENT GUIDE

Name _____ Date _____

Questions

1. At 1:00 a.m., someone breaks a window in the back of a store and robs the safe. On the way out, the thief is cut on a piece of broken glass. A forensic team collects and tests a sample of blood left behind by the thief. It is O-. The police bring in a suspect with a cut forearm who was arrested just three blocks from the store. The person resembles someone seen leaving the store at the time of the robbery. A sample of the suspect's blood is taken and tested for blood type. Once the suspect's blood is mixed with anti-A serum, it is immediately clear that the suspect is not the person who was cut on the broken glass in the store. How did the test indicate that fact?
2. Suppose the same suspect's blood does not agglutinate when tested with anti-A or anti-B, but does agglutinate when tested with anti-Rh. Would this connect the suspect with the crime scene? Explain your answer.
3. Tom and Jane participate in a Red Cross blood drive. Both are first-time donors. As part of the screening process, their blood is typed. Tom is A+. Jane is AB+.
 - a. What ABO antibody is found in Tom's blood?
 - b. What ABO antigens are found in Jane's blood?
4. The same Tom and Jane's blood donations are sent to a processing center where the blood cells are separated from the plasma in each of the two samples. The separated cells and plasmas are then sent to a hospital. A blood researcher wishes to use Tom's blood in an attempt to extract and identify the A antigen. Should she attempt the extraction process on his blood cells or on his plasma?

Answers to Assessment Questions

1. The blood agglutinated, indicating the presence of the A antigen. This would not have happened if the suspect had type O blood. Note that this does not mean that the suspect was not involved in the break-in. It does mean that the blood collected at the crime scene could not have come from the suspect.
2. No. The blood left at the crime scene was O⁻. The suspect's blood tested O⁺.
3. a. Anti-B.
b. A, B, and Rh.
4. The blood cells. The antigens are part of the cell membranes of the blood cells.

Extension Activities

1. You may want students to observe the different kinds of blood cells. This is easily done with compound microscopes and prepared blood smears. Carolina's prepared blood smear slide with an information card (Discovering Blood, item 313100) can be used for this activity. Have the students examine a normal human blood film under 400–450× magnification. They will immediately see the red blood cells. With careful examination, the students should be able to infer the cells' basic shape—biconcave disks—and to see red blood cells in various degrees of flexure. White blood cells are much less numerous and require some searching to find. With Wright stain, several types of white blood cells can be distinguished. Students can simply observe that there are different types of white blood cells, or they can try to identify the types, using textbook or online references. Basophils have cytoplasmic granules that stain purplish blue. Eosinophils' granules stain pinkish red. Neutrophils' granules stain a paler shade than those of basophils. Because of their cytoplasmic granules, basophils, eosinophils, and neutrophils are collectively called granulocytes. In both lymphocytes and monocytes, the nucleus stains purplish blue. Platelets are much smaller than any of these cells and stain blue.
2. Students can be assigned to research and report on disorders that cause changes to the form or the relative abundance of the different blood cells (e.g., sickle-cell anemia or human acute granulocytic leukemia).
3. Have students research the history of blood antigens, e.g., their discovery and the early development of blood typing.
4. Have students research more about the importance of blood antigens with respect to the relationship between a pregnant mother and her fetus.

Name _____ Date _____

Interplanetary Blood Typing Battle

Prelab Analysis

Planet	Surface Alien(s)	Defense System	Aliens from this planet can visit:	This planet can receive visitors from:
A				
B				
AB				
O				

Prelab Questions

1. Shown in the table below are the antigens present on the blood cells of four different people. Fill in the corresponding ABO blood type for each person and the antibodies expected to be found in each person's blood.

Blood Type	Red Blood Cell Antigen	Antibody in Blood Plasma
	A	
	B	
	AB	
	Neither	

2. For each blood type in the table below, fill in the expected agglutination result from mixing the blood with each of the antibodies.

Blood Type	Anti-A	Anti-B	Anti-Rh
A+			
A-			
B+			
B-			
AB+			
AB-			
O+			
O-			

Name _____

Date _____

Background

By volume, blood tissue is approximately 55% plasma and 45% cells. Plasma is a straw-colored liquid consisting of 90% water and a variety of salts and proteins important for maintaining osmotic balance, buffering against pH changes, maintaining blood viscosity, transporting certain materials, and for blood clotting when a blood vessel is injured.

There are three major types of **blood cells**—red blood cells (erythrocytes), white blood cells (leukocytes), and platelets.

Red blood cells, the most numerous cells in the blood, carry oxygen from the lungs to all parts of the body. A red blood cell is a biconcave disk with a thin center. This shape provides a large surface area for diffusion of oxygen. Red blood cells contain the protein hemoglobin. Iron is incorporated into the hemoglobin molecule. When blood travels through the lungs, the oxygen in the lungs combines with the iron in hemoglobin. When the blood moves through the body's capillary system, the oxygen carried in the red blood cells is released from the iron in hemoglobin to the other cells of the body. Red blood cells' flexibility allows them to pass through even the smallest blood vessels.

White blood cells make up only about 1% of the blood volume. They are an important part of the **immune system**. Their primary function is to provide defense against invaders in the body, which may include bacteria, parasites, fungi, and viruses. White blood cells may attack a foreign body directly, they may produce antibodies that identify, attach to, and neutralize a foreign body, or they may trigger other cells to act in destroying the foreign body.

Platelets perform a vital function in the process of coagulation, or blood clotting, which occurs when a blood vessel is injured.

Blood Types

Although the basic composition and function of blood in each of us is the same, there are different human blood types. The cell membrane of red blood cells, like that of other cells, has molecules that project from its surface. Some of the molecules function as identification badges, allowing the immune system to recognize the cell as a normal component of an individual's body. If blood from a person whose red cells have different surface molecules is injected into someone, those molecules are recognized as **antigenic**, or foreign to the body. The immune system attacks the antigens and attempts to destroy them and the cells that carry them. This is why transfusion with an incompatible blood type is harmful. The recipient's body recognizes the antigens on the transfused red blood cells as foreign and attacks and destroys the cells. For that reason, donated blood is thoroughly tested for A, B, O, and Rh antigens and is transfused only into compatible recipients.

The ABO Blood Groups

The ABO blood groups (types) result from the presence or absence of two antigens, A and B, on the surface of the red blood cells. If antigens are present very early in life, the immune system recognizes those antigens as "self" and will not generate an immune response to them. As a result, the body does not generate antibodies to any A and B antigens present on its own blood cells. However, the immune system does produce antibodies to any A and B blood antigens not present on the organism's own cells. Type A blood has the A antigen on its red blood cells and anti-B antibodies in the plasma. Type B blood has the B antigen on its red blood cells and A antibodies in the plasma. Type AB blood has both A and B antigens on the red blood cells and no antibodies in the plasma. Finally, Type O blood has neither A nor B antigens on the red blood cells and both A and B antibodies in the plasma.

These antibodies are present even if the person has not had any foreign blood introduced into their body. It is hypothesized that the antibodies are present because of similarity between the A and B blood antigens and other antigens present in the environment. If two antigens are similar enough, the antibodies generated to one antigen will also recognize the other.

The relationships of the ABO blood types to the presence of antigens and antibodies in the blood are summarized in the table that follows.

The Rh Blood Groups

Another important antigen found on the surface of blood cells is the Rh factor. The Rh antigen is actually a whole group of closely related antigens. Blood containing an Rh antigen is said to be Rh positive (Rh+); blood lacking the antigen is said to be Rh negative (Rh-). Unlike the case for the ABO antigens, the production of Rh antibody requires prior exposure to the antigen, such as would occur in an Rh- pregnant woman carrying a fetus that was Rh+.

Blood Group	Red Cell Antigen Present on Cells	Antibody that Recognizes Cells
A	A	anti-B
B	B	anti-A
AB	A and B	neither
O	neither	anti-A and anti-B

Blood Type Incompatibility: Using Agglutination to Determine the Blood Type of a Sample

Agglutination is the clumping of particles. If a blood sample interacts with a specific antibody (i.e., blood type A interacting with Anti-A antibody serum), then agglutination will occur. In today's lab, you will use the presence or absence of agglutination to determine an unknown blood type in a sample of synthetic blood.

To begin, you will add one drop of a blood sample in each well of a blood typing slide, labeled A, B, and Rh.

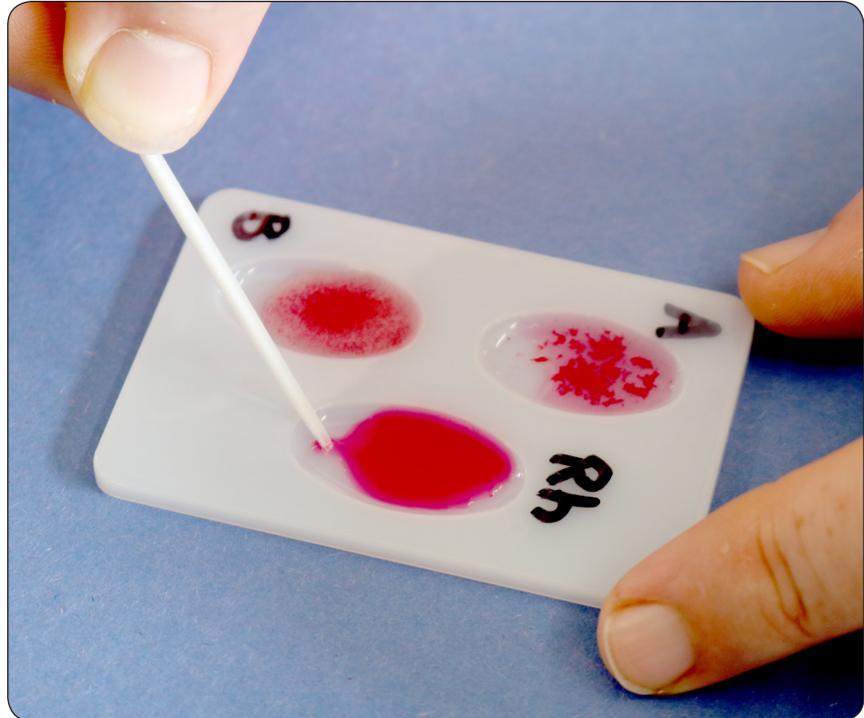
You will then add a synthetic antibody serum to each well as follows:

Labeled Well on Blood Typing Slide	Synthetic Antibody Serum Added
A	Anti-A
B	Anti-B
Rh	Anti-Rh

Interpreting the Results

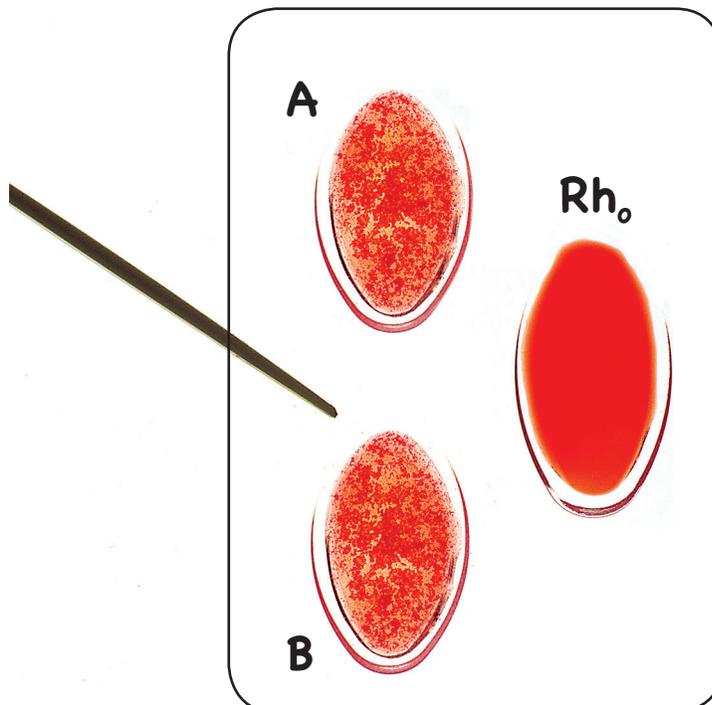
After mixing the specific serum with the blood sample, you will observe the presence or absence of agglutination in each well.

Clumping, or agglutination indicates that the blood has reacted with a specific antibody serum (Anti-A, Anti-B or Anti-Rh) and therefore has antigens that are incompatible with that type of antibody. In the example photo shown here, the blood has A antigens (reacted with Anti-A serum) and B Antigens (reacted with Anti-B serum). *The blood type is identified as AB.*



If the blood does not agglutinate when mixed with a specific antibody serum, the results indicate that the blood does not have the antigens binding the specific antibody in the reagent. In the example photo, the blood *did not react* with the Anti-Rh serum (negative result). The blood type tested negative for Rh antigens.

The blood type may now be classified as **AB negative**.



Name _____

Date _____



Materials

vial synthetic anti-D (anti Rh) serum
 vial synthetic anti-A serum
 vial synthetic anti-B serum
 blood typing slide
 4 blue mixing sticks
 4 yellow mixing sticks
 4 white mixing sticks
 vial of Sample 1 synthetic blood
 vial of Sample 2 synthetic blood
 vial of Sample 3 synthetic blood
 vial of Sample 4 synthetic blood
 paper towels
 access to tap water and a sink

Key Question

How is blood type determined?

In this lab, you will determine the blood type of four different synthetic blood samples using antisera to the A, B, and Rh (D) antigens that exist on human red blood cells. The procedure for the blood test is the same that would be used for a real blood test, but, for convenience and safety, the blood and antisera are synthetic and contain no biological materials.

Procedure

- Using the dropper vial, place a drop of the Sample 1 synthetic blood sample in each well of the blood typing slide. Close the cap on the dropper vial. To prevent cross contamination, always close the cap on one vial before opening the next vial.
- Add a drop of synthetic anti-A serum (blue) to well A. Close the cap.
- Add a drop of synthetic anti-B serum (yellow) to well B. Close the cap.
- Add a drop of synthetic anti-Rh serum (clear) to well Rh. Close the cap.
- Using a different-colored mixing stick for each well (blue for anti-A, yellow for anti-B, white for anti-Rh), gently stir the synthetic blood and antiserum drops for 30 seconds. Remember to use a new mixing stick for each sample to avoid contamination of your samples.
- Examine the resulting films of liquid mixture in the well. If a film is uniform in appearance, there is no agglutination. If the sample is granular, agglutination has occurred.
- Fill in the column for Sample 1 in the following Data Table, answering *yes* or *no* as to whether agglutination occurred with each antiserum.
- Thoroughly rinse the blood typing slide and then repeat steps 1 through 7 for synthetic blood Samples 2, 3, and 4.

Data Table

	Sample 1	Sample 2	Sample 3	Sample 4
Anti-A				
Anti-B				
Rh				
Blood Types				

Name _____

Date _____

Questions

1. At 1:00 a.m., someone breaks a window in the back of a store and robs the safe. On the way out, the thief is cut on a piece of broken glass. A forensic team collects and tests a sample of blood left behind by the thief. It is O⁻. The police bring in a suspect with a cut forearm who was arrested just three blocks from the store. The person resembles someone seen leaving the store at the time of the robbery. A sample of the suspect's blood is taken and tested for blood type. Once the suspect's blood is mixed with anti-A serum, it is immediately clear that the suspect is not the person who was cut on the broken glass in the store. How did the test indicate that fact?
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3. Tom and Jane participate in a Red Cross blood drive. Both are first-time donors. As part of the screening process, their blood is typed. Tom is A⁺. Jane is AB⁺.
 - a. What ABO antibody is found in Tom's blood?
 - b. What ABO antigens are found in Jane's blood?
4. The same Tom and Jane's blood donations are sent to a processing center where the blood cells are separated from the plasma in each of the two samples. The separated cells and plasmas are then sent to a hospital. A blood researcher wishes to use Tom's blood in an attempt to extract and identify the A antigen. Should she attempt the extraction process on his blood cells or on his plasma?

ABO-Rh Blood Typing with Synthetic Blood • Teacher's Manual and Student Guide
700101 Kit • 700102 Refill • 700160 Classroom Kit • 700161 Classroom Kit Refill



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