

# HUMAN BLOOD TYPE: TESTING FOR ABO AND Rh FACTORS



Westminster College

## STANDARDS

- 3.3.7.B, 3.3.7.C
- 3.3.10.B, 3.3.10.C








## INTRODUCTION

Blood is a living tissue which circulates through the human body providing oxygen and nutrients and removing carbon dioxide and other waste products. Blood cells are produced in the bone marrow and an average adult has about 5 liters of blood in their body. There are four different components of blood, **red blood cells** (erythrocytes), **white blood cells** (leukocytes), **platelets** (thrombocytes) and **plasma**. The predominant cells are the **red blood cells** (RBCs); there can be as many as one billion RBCs in 2 to 3 drops of blood. The RBCs contain a protein, hemoglobin, whose major function is to transport oxygen (O<sub>2</sub>) to all parts of the body.

**White blood cells** (WBCs) are part of the immune system and are important for fighting diseases. **Plasma** is the yellowish liquid portion of the blood which contains nutrients, vitamins, hormones, and antibodies (part of the immune system). The **platelets**, clotting factors that help blood coagulate at the site of a wound, are also carried in the plasma.

The ABO-blood typing system is based on two protein molecules that can be present on the surface of the red blood cells. These protein molecules, or **agglutinogens**, are named **Type A** and **Type B**. If neither of these proteins is present on your red blood cells, you are considered **Type O**. The pictures in *Figure 1* (Row 1) show how your blood cells would appear for all the possible human blood types. In the case of Type AB, both

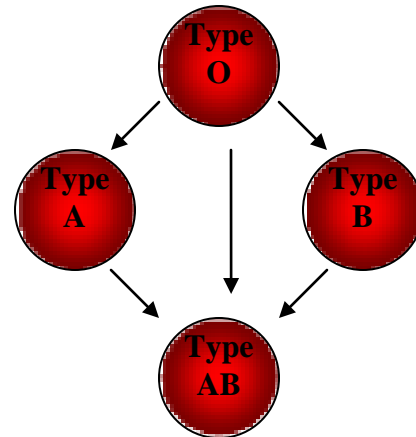
*Figure 1.* Graphic of Different Human Blood Types

<b>The ABO Blood System</b>				
Blood Type (genotype)	Type A (AA, AO)	Type B (BB, BO)	Type AB (AB)	Type O (OO)
Red Blood Cell Surface Proteins (phenotype)	 A agglutinogens only	 B agglutinogens only	 A and B agglutinogens	 No agglutinogens
Plasma Antibodies (phenotype)	 b agglutinin only	 a agglutinin only	NONE No agglutinin	 a and b agglutinin

<http://learn.genetics.utah.edu/units/basics/blood/types.cfm>

proteins can be expressed on the surface of the blood cell at the same time. Type O is the absence of either protein on the blood cell surface. Another protein on the surface of the red blood cells is known as the Rh factor. The name Rh is from Rhesus monkeys, where this factor was discovered and later found to be present in humans as well. The presence of the Rh antigen makes a person Rh+ (positive); absence of the antigen makes a person Rh- (negative).

Figure 2. Compatible blood transfusions



Within the blood plasma, a person also develops **agglutinins** (or antibodies) to blood types other than their own (Fig. 1, Row 2). For example, if you are Type B, then in your plasma you carry antibodies against Type A antigens (anti-A). If you are Type AB, you have no antibodies to A or B. Type O has both anti-A and anti-B antibodies. Knowing your blood type is medically important. In the case of blood transfusions and organ transplants, it is critical that the blood type and Rh factor match. If a Type A person were given blood from a Type B person, the anti-B agglutinins in their own plasma would attack the Type B red blood cells resulting in clumping of the cells and hemolysis (bursting of the cells). Hemolysis can trigger a toxic and sometimes fatal reaction, particularly if a lot of blood was transfused. Rather than helping, the blood transfusion could make the recipient worse. The diagram in Figure 2 shows which blood types can be given and received for a blood transfusion. Type O is called the **universal donor** because it can donate to any of the ABO blood types. Type AB is the **universal recipient** because it can receive any of the other blood types. The Rh-factor must also be taken into account; if you are Rh<sup>+</sup>, you may receive Rh<sup>+</sup> or Rh<sup>-</sup> blood; Rh<sup>-</sup> may only receive Rh<sup>-</sup>.

**Genetics of Blood Type:** Your blood type is inherited; you receive a total of two genes for your blood type from your parents, one gene from your father and one gene from your mother. The genes can be designated as A (AA, AO) for Type A blood, B (BB, BO) for Type B blood and O (OO) for Type O (no proteins present). An example of how you can inherit your blood type is shown in Figure 3. In this particular example, the mother is Type A and the father is Type B. It is assumed that we know that they have only one copy of the gene for either A or B. Using a table called a Punnett Square, you can predict what possible blood types their children

Figure 3. Example of Blood Type Inheritance

		<u>Mom</u>	
		A	O
<u>Dad</u>	B	AB	BO
	O	AO	OO

will have. These parents would have children that could have 4 possible blood types, AB, A, B, and O, depending on the combination of genes that each child receives.

The following activity will **simulate** a standard ABO and Rh blood typing. You will be given “blood” from four individuals and asked to determine each blood type using high levels of **agglutinins** (antisera) against the A, B, and Rh protein factors. Several drops of each kind of antiserum are added to the separate samples of blood. If agglutination occurs only in the suspension to which the anti-A serum was added, the blood type is A. If agglutination occurs only in the anti-B mixture, the blood type is B. Agglutination in both samples indicates that the blood type is AB. The absence of agglutination in any of the samples shows the blood type is O. The procedure with the Rh antiserum is the same; agglutination suggests Rh-positive. No agglutination indicates an individual is Rh-negative.

### GUIDING QUESTIONS

- What are the major components of human blood?
- What do the blood types A, B, AB and O represent?
- What is the Rh-factor?
- What does the term agglutination mean?
- How is agglutination used to determine blood type?
- How is blood type inherited?

### MATERIALS

4 blood typing trays  
4 simulated blood samples  
Simulated anti-B serum  
Marking pen

12 plastic toothpicks  
Simulated anti-A serum  
Simulated anti-Rh serum

### SAFETY

**None** of the samples are real blood or blood products. This is a simulation of what a blood-typing test would look like.

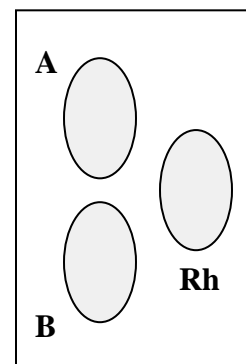
### PROCEDURE

1. There should be 4 plastic blood typing trays at each lab station and 4 unknown blood samples. Each tray has 3 wells which have the markings **A**, **B** and **Rh** (Fig. 4).

2. Use a marking pen to label one tray for each blood sample; Ms Brown, Mr. Green, Mr. Jones and Mr. Smith.

**NOTE:** *In Steps 3-7, it is very important to get the correct person and correct anti-sera onto the blood-typing trays. Read through the following directions first before starting to add the blood samples and anti-sera to the trays.*

**Figure 4.** Blood-typing tray

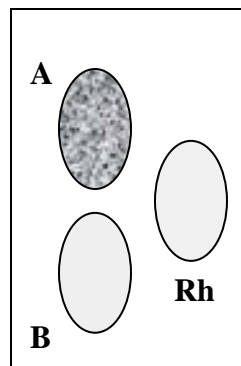


3. Place **two to three drops** of Ms. Brown’s “blood” in each of the three wells on the tray labeled Ms. Brown.
4. Repeat Step 3 for the other trays. It is very important to get the right person’s blood sample on the correct tray.
5. Obtain the bottle of simulated anti-A serum. On each of the four trays, place **2-3 drops** of the anti-A serum in the well marked A only.
6. Obtain the bottle of simulated anti-B serum. On each of the four trays, place **2-3 drops** of the anti-B serum in the well marked B only.
7. Obtain the bottle of simulated anti-Rh serum. On each of the four trays, place **2-3 drops** of the anti-Rh serum in the well marked Rh only.
8. Place three clean toothpicks by each tray. Stir each well gently with a separate, clean toothpick for 30 seconds. It is important to not splatter the simulated blood, as you can contaminate the other wells on the tray. This will affect the final results.

9. Even after stirring the wells for 30 seconds, it may take a full minute for the components to react. After a minute, observe the results. Agglutination usually makes the mixed components cloudy and there may be opaque clumps in the well (Fig. 5).

**Note:** The agglutination of the B blood type is the most difficult to discern and may take up to a minute for the clotting reaction to complete.

**Figure 5.** Example of a blood type result



In this example of a blood type result, only well A shows agglutination, indicating the presence of the A-antigen. The blood type of this person is A-negative.

10. Observe each slide and record your observations in the Data Analysis section of your lab worksheet. To confirm agglutination try reading text through the mixed sample. If you cannot read the text, assume you have positive agglutination reaction.
12. Once the results from all the trays are recorded, please rinse the trays and the toothpicks and return them to your teacher.

## **REFERENCES**

Simulated ABO & Rh Blood Typing Lab Activity. (2002) Ward’s Natural Science Establishment, Inc.

Dr. Jennifer Doherty and Dr. Ingrid Waldron. (2010) Teacher Preparation Notes for Using Blood Tests to Identify Babies and Criminals. Department of Biology, University of

Pennsylvania.

[http://serendip.brynmawr.edu/sci\\_edu/waldron/pdf/BloodTypeGeneticsTeachPrep.pdf](http://serendip.brynmawr.edu/sci_edu/waldron/pdf/BloodTypeGeneticsTeachPrep.pdf)

T. Trimpe. (2006) Blood Basics: Notes and Powerpoint and Ernie's Exit: Blood Typing Lab Activity. Science Spot.net.

<://sciencespot.net/Media/FrnsScience/bloodbasicswkst.pdf>

Graphic of ABO Blood Type in Figure 1:

<http://learn.genetics.utah.edu/units/basics/blood/types.cfm>

## **CREDITS**

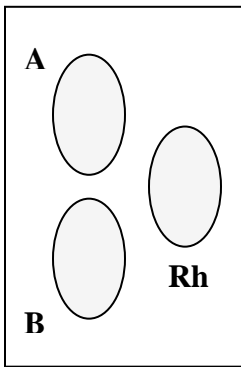
This lab design was revised and adapted by Dr. Stephanie Corrette-Bennett using the above references.

**DATA SHEET**

Name: \_\_\_\_\_  
Group: \_\_\_\_\_  
Date: \_\_\_\_\_

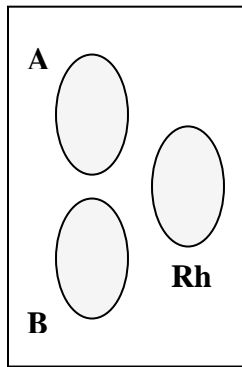
**DATA ANALYSIS**

Record the results of the blood type for each person. If there is agglutination in a well, mark it “Ag”; otherwise, leave the well blank. Agglutination indicates that a particular antigen is present in the blood (see Fig. 2). Based on the agglutination pattern, determine the blood type and Rh factor and write the results on the line provided below each person’s name.



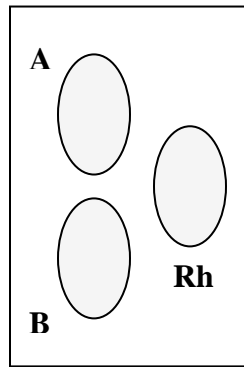
Ms. Brown

Type: \_\_\_\_\_  
Rh: \_\_\_\_\_



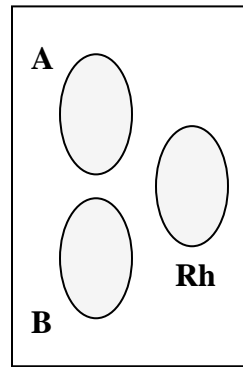
Mr. Green

Type: \_\_\_\_\_  
Rh: \_\_\_\_\_



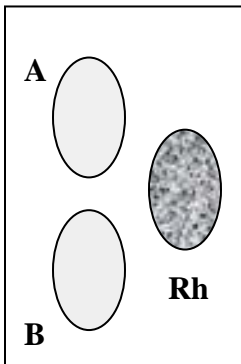
Mr. Jones

Type: \_\_\_\_\_  
Rh: \_\_\_\_\_



Mr. Smith

Type: \_\_\_\_\_  
Rh: \_\_\_\_\_



**Example:** In the sample to the left, there is no agglutination in wells A or B. The person would have type O blood. Agglutination in the Rh well of the same person would make them O+ (positive).

Use the following table to help determine the blood type of each person.

<b>Analysis of Blood Types</b>		
<b>Sample Well</b>	<b>Agglutination</b>	<b>Blood Type</b>
<b>A</b>	Yes	A
	No	(no A-antigen)
<b>B</b>	Yes	B
	No	(no B-antigen)
<b>Rh</b>	Yes	Positive (+)
	No	Negative (-)

## QUESTIONS

1. List the four major blood components and why they are important?
  - a.
  - b.
  - c.
  - d.
2. What are agglutinogens? Why are they important in ABO blood typing?
3. An emergency room patient has lost a lot of blood. Their blood type is AB+. There is a shortage of blood for transfusions, and all the ER staff can find is O<sup>-</sup> blood. Do you think it will be safe to give the patient a transfusion with this blood? Why or why not?

4. A father and mother have the following blood types, AO and BB, respectively. Use the Punnett square shown to determine the possible blood types that their children might have.

a. What are the possible blood types of the children?

b. Which parent could donate blood to any of the children?

		<u>Mom</u> = AO	
		<u>Dad</u> = BB	
<u>Dad</u>			<u>Mom</u>
		<b>B</b>	<b>B</b>
<b>A</b>			
<b>O</b>			