Learning Objectives

Upon completing the exercise, each student should be able:

- to explain the concept of blood group antigens;
- to list the genotypes and phenotypes of the ABO blood groups in humans;
- to list possible blood transfusion donors and recipients for each of the ABO blood types;
- to determine the possible ABO blood group genotypes and phenotypes for the children of a couple, given the blood groups of the parents.

Background

Antigens are substances that can stimulate a response in the immune system of an animal. For example, if a rabbit is injected with albumin from a cow, the rabbit’s immune system will recognize the cow protein as “non-self” and will make antibodies specific to the cow albumin. If you collect blood from the rabbit, you can isolate “anti-cow albumin antiserum” from the rabbit blood. When the anti-cow antiserum is mixed with cow albumin, the mixture will clump together (agglutinate) because the antibody proteins in the antiserum bind to the cow albumin.

Around 1900, Karl Landsteiner discovered that blood from different people could be divided into four major groups based on its immunological compatibility. Later, it was discovered that these blood groups differed because they had different antigens of the surface of their erythrocytes (red blood cells). The antigens are designated A and B, and their production is regulated by three different alleles of a single gene.

<table>
<thead>
<tr>
<th>Allele</th>
<th>Antigen Produced</th>
<th>Dominance Relationships</th>
</tr>
</thead>
<tbody>
<tr>
<td>I^A</td>
<td>A antigen</td>
<td>Codominant to I^B; completely dominant to i^o</td>
</tr>
<tr>
<td>I^B</td>
<td>B antigen</td>
<td>Codominant to I^A; completely dominant to i^o</td>
</tr>
<tr>
<td>i^o</td>
<td>None</td>
<td>Recessive to both I^A and I^B</td>
</tr>
</tbody>
</table>

There are six possible genotypes and four possible phenotypes.

<table>
<thead>
<tr>
<th>Genotypes</th>
<th>Phenotype</th>
<th>Agglutination with Anti-A antiserum?</th>
<th>Agglutination with Anti-B antiserum?</th>
<th>Can donate blood to:*</th>
<th>Can receive blood from:*</th>
</tr>
</thead>
<tbody>
<tr>
<td>I^A I^A and I^A i^o</td>
<td>Type A</td>
<td>Yes</td>
<td>No</td>
<td>A or AB</td>
<td>A or O</td>
</tr>
<tr>
<td>I^B I^B and I^B i^o</td>
<td>Type B</td>
<td>No</td>
<td>Yes</td>
<td>B or AB</td>
<td>B or O</td>
</tr>
<tr>
<td>I^A I^B</td>
<td>Type AB</td>
<td>Yes</td>
<td>Yes</td>
<td>AB</td>
<td>A, B, AB, or O</td>
</tr>
<tr>
<td>i^o i^o</td>
<td>Type O</td>
<td>No</td>
<td>No</td>
<td>A, B, AB, or O</td>
<td>O</td>
</tr>
</tbody>
</table>

* There are several other blood group antigens other than the ABO group (for example, the Rh factor and the M-N group) that can affect compatibility during transfusions. This table only shows compatibility relationships in the ABO groups.
Persons with type O blood blood are sometimes called “universal donors” because they do not have the A or B antigens in their blood; therefore, they can donate blood to persons of the other three blood types as well as type O. Persons with blood type AB are “universal recipients;” this means that they can receive blood from the other three blood types as well as type AB. Of course, it is important to note that there are several other blood group antigens other than the ABO group (for example, the Rh factor and the M-N group) that can affect compatibility during transfusions.

The ABO alleles are inherited just like any other alleles. It is important to remember that it is the allele that is inherited from the parent, not the phenotype. The phenotype is determined by the combination of alleles inherited from both of the parents. Here are some sample problems:

1. A woman with type A blood is married to a man with type O blood. No other information is available about this couple. What are the possible blood group phenotypes of their children?

Solution:
The man must be homozygous $i^o$ since he is type O. The woman could be heterozygous $I^A i^o$. If this is the case, then the cross is just like a testcross (heterozygous X homozygous recessive).

\[
I^A i^o \times i^o i^o \rightarrow \frac{1}{2} I^A i^o, \frac{1}{2} i^o i^o
\]

Therefore, this couple could possibly have children with either type A or type O blood.

2. A woman with type AB blood is married to a man with type O blood. She has a child with type A blood. In a divorce suit, the man claims that the child cannot be his biological child because neither he nor the woman have type A blood. Based solely on the information stated here, does the science of genetics support the man’s claims?

Solution:
In this problem, we know the exact genotype of both the man and the woman: he is homozygous type O, and she is heterozygous AB. Once again, the cross is analyzed just like a testcross.

\[
I^A I^B \times i^o i^o \rightarrow \frac{1}{2} I^A i^o, \frac{1}{2} I^B i^o
\]

Notice that for this couple, the parents are AB and O, but their children will either be type A or type B. None of their children will be type AB or type O, because each child must inherit one allele from one parent, and one allele from the other parent. So in this case, the science of genetics does not support the man’s claim that the child is not his.
**Procedure**

In this lab exercise, you will carry out a blood typing exercise using simulated blood and antiserum. The appearance of the simulated agglutination is approximately the same as that seen with real blood. Here is the scenario: two children, born in a hospital at about the same time, may have been “mixed up” in the nursery. By determining the blood types of the parents and the children, try to determine which child belongs to which couple.

Here is the procedure.

1. Label each blood typing slide:
   Slide #1: Mr. Smith  
   Slide #2: Mrs. Smith  
   Slide #3: Mr. Jones  
   Slide #4: Mrs. Jones  
   Slide #5: Child 1  
   Slide #6: Child 2
2. Place two to three drops of Mr. Smith’s blood in each of the A and B wells of Slide #1.  
3. Place two to three drops of Mrs. Smith’s blood in each of the A and B wells of Slide #2.  
4. Place two to three drops of Mr. Jones’ blood in each of the A and B wells of Slide #3.  
5. Place two to three drops of Mrs. Jones’ blood in each of the A and B wells of Slide #4.  
6. Place two to three drops of Child 1’s blood in each of the A and B wells of Slide #5.  
7. Place two to three drops of Child 2’s blood in each of the A and B wells of Slide #6.  
8. Add two to three drops of the simulated anti-A serum in each A well on the six slides.  
9. Add two to three drops of the simulated anti-B serum in each B well on the six slides.  
10. Stir each mixture with a different clean toothpick. Use only one toothpick per well to avoid cross contamination.  
11. Examine each well for agglutination. Agglutination indicates a positive test result. Record results (positive or negative) in Table 1 on the lab worksheet. Use the results to determine the blood type for each person.  
12. Determine possible genotypes of all six individuals. Record results in Table 2 on the lab worksheet.  
13. Based on your results, answer the questions given in the worksheet.

**Reference**

Analysis

Table 1
Agglutination reactions

<table>
<thead>
<tr>
<th>Slide #</th>
<th>Blood Type</th>
</tr>
</thead>
<tbody>
<tr>
<td>#1: Mr. Smith</td>
<td>Anti-A Serum</td>
</tr>
<tr>
<td>#2: Mrs. Smith</td>
<td></td>
</tr>
<tr>
<td>#3: Mr. Jones</td>
<td></td>
</tr>
<tr>
<td>#4: Mrs. Jones</td>
<td></td>
</tr>
<tr>
<td>#5: Child 1</td>
<td></td>
</tr>
<tr>
<td>#6: Child 2</td>
<td></td>
</tr>
</tbody>
</table>

Table 2
Possible Genotypes

<table>
<thead>
<tr>
<th>Blood Type</th>
<th>Possible Genotypes</th>
</tr>
</thead>
<tbody>
<tr>
<td>Mr. Smith</td>
<td></td>
</tr>
<tr>
<td>Mrs. Smith</td>
<td></td>
</tr>
<tr>
<td>Mr. Jones</td>
<td></td>
</tr>
<tr>
<td>Mrs. Jones</td>
<td></td>
</tr>
<tr>
<td>Child 1</td>
<td></td>
</tr>
<tr>
<td>Child 2</td>
<td></td>
</tr>
</tbody>
</table>
Questions

1. Could Child 1 have been born to Mr. and Mrs. Smith? Why or why not?

2. Could Child 2 have been born to Mr. and Mrs. Smith? Why or why not?

3. Could Child 2 have been born to Mr. and Mrs. Jones? Why or why not?

4. Could Child 1 have been born to Mr. and Mrs. Jones? Why or why not?

5. What conclusions can you draw from the above evidence?

6. What possible blood types could the offspring of Mr. and Mrs. Smith have?

7. What possible blood types could the offspring of Mr. and Mrs. Jones have?