



5. The ABO blood group

The discovery of the ABO blood group, over 100 years ago, caused great excitement. Until then, all blood had been assumed to be the same, and the often tragic consequences of blood transfusions were not understood. As our understanding of the ABO group grew, not only did the world of blood transfusion become a great deal safer, but scientists could now study one of the first human characteristics proven to be inherited. A person's ABO blood type was used by lawyers in paternity suits, by police in forensic science, and by anthropologists in the study of different populations.

The ABO blood group antigens remain of prime importance in transfusion medicine—they are the most immunogenic of all the blood group antigens. The most common cause of death from a blood transfusion is a clerical error in which an incompatible type of ABO blood is transfused. The ABO blood group antigens also appear to have been important throughout our evolution because the frequencies of different ABO blood types vary among different populations, suggesting that a particular blood type conferred a selection advantage (e.g., resistance against an infectious disease.)

However, despite their obvious clinical importance, the physiological functions of ABO blood group antigens remain a mystery. People with the common blood type O express neither the A nor B antigen, and they are perfectly healthy. Numerous associations have been made between particular ABO phenotypes and an increased susceptibility to disease. For example, the ABO phenotype has been linked with stomach ulcers (more common in group O individuals) and gastric cancer (more common in group A individuals). Another observation is that individuals with blood type O tend to have lower levels of the von Willebrand Factor (vWF), which is a protein involved in blood clotting.

At a glance

Antigens of the ABO blood group

Number of antigens	4: A, B, AB, and A1
Antigen specificity	Carbohydrate The sequence of oligosaccharides determines whether the antigen is A, B, or A1.
Antigen-carrying molecules	Glycoproteins and glycolipids of unknown function The ABO blood group antigens are attached to oligosaccharide chains that project above the RBC surface. These chains are attached to proteins and lipids that lie in the RBC membrane.
Molecular basis	The ABO gene indirectly encodes the ABO blood group antigens. The ABO locus has three main allelic forms: A, B, and O. The A and B alleles each encode a glycosyltransferase that catalyzes the final step in the synthesis of the A and B antigen, respectively. The A/B polymorphism arises from several SNPs in the ABO gene, which result in A and B transferases that differ by four amino acids. The O allele encodes an inactive glycosyltransferase that leaves the ABO antigen precursor (the H antigen) unmodified.

Frequency of ABO blood group antigens	A: 43% Caucasians, 27% Blacks, 28% Asians B: 9% Caucasians, 20% Blacks, 27% Asians A1: 34% Caucasians, 19% Blacks, 27% Asians Note: Does not include AB blood groups (1).
Frequency of ABO phenotypes	Blood group O is the most common phenotype in most populations. Caucasians: group O, 44%; A ₁ , 33%; A ₂ , 10%; B, 9%; A1B, 3%; A2B, 1% Blacks: group O, 49%; A ₁ , 19%; A ₂ , 8%; B, 20%; A1B, 3%; A2B, 1% Asians: group O, 43%; A ₁ , 27%; A ₂ , rare; B, 25%; A1B, 5%; A2B, rare Note: Blood group A is divided into two main phenotypes, A ₁ and A ₂ (1).

Antibodies produced against ABO blood group antigens

Antibody type	IgG and IgM Naturally occurring. Anti-A is found in the serum of people with blood groups O and B. Anti-B is found in the serum of people with blood groups O and A.
Antibody reactivity	Capable of hemolysis Anti-A and anti-B bind to RBCs and activate the complement cascade, which lyses the RBCs while they are still in the circulation (intravascular hemolysis).
Transfusion reaction	Yes — typically causes an acute hemolytic transfusion reaction Most deaths caused by blood transfusion are the result of transfusing ABO-incompatible blood.
Hemolytic disease of the newborn	No or mild disease HDN may occur if a group O mother has more than one pregnancy with a child with blood group A, B, or AB. Most cases are mild and do not require treatment.

Background information

History

At the beginning of the 20th century an Austrian scientist, Karl Landsteiner, noted that the RBCs of some individuals were agglutinated by the serum from other individuals. He made a note of the patterns of agglutination and showed that blood could be divided into groups. This marked the discovery of the first blood group system, ABO, and earned Landsteiner a Nobel Prize.

Landsteiner explained that the reactions between the RBCs and serum were related to the presence of markers (antigens) on the RBCs and antibodies in the serum. Agglutination occurred when the RBC antigens were bound by the antibodies in the serum. He called the antigens A and B, and depending upon which antigen the RBC expressed, blood either belonged to blood group A or blood group B. A third blood group contained RBCs that reacted as if they lacked the properties of A and B, and this group was later called "O" after the German word "Ohne", which means "without". The following year the fourth blood group, AB, was added to the ABO blood group system. These RBCs expressed both A and B antigens.

In 1910, scientists proved that the RBCs antigens were inherited, and that the A and B antigens were inherited codominantly over O. There was initially some confusion over how a person's blood type was determined, but the puzzle was solved in 1924 by Bernstein's "three allele model".

The ABO blood group antigens are encoded by one genetic locus, the ABO locus, which has three alternative (allelic) forms—A, B, and O. A child receives one of the three alleles from each parent, giving rise to six possible genotypes and four possible blood types (phenotypes).

ABO genotype in the offspring		ABO alleles inherited from the mother		
		A	B	O
ABO alleles inherited from the father	A	A	AB	A
	B	AB	B	B
	O	A	B	O

Nomenclature

- Number of ABO blood group antigens: 4
- ISBT symbol: ABO
- ISBT number: 001
- Gene symbol: ABO
- Gene name: ABO blood group (A transferase, α 1,3-N-acetylgalactosaminyltransferase; B transferase, α 1,3-galactosyltransferase)

Basic biochemistry

ABO phenotypes

The four basic ABO phenotypes are O, A, B, and AB. After it was found that blood group A RBCs reacted differently to a particular antibody (later called anti-A₁), the blood group was divided into two phenotypes, A₁ and A₂. RBCs with the A₁ phenotype react with anti-A₁ and make up about 80% of blood type A. RBCs with the A₂ phenotype do not react with anti-A₁ and they make up about 20% of blood type A. A₁ red cells express about 5 times more A antigen than A₂ red cells, but both types of red cell react with anti-A, and as far as transfusion purposes are concerned, the A₁ and A₂ blood groups are interchangeable.

There are many other subgroups of blood group A in which RBCs tend to weakly express the A antigen,

whereas weak variants of the blood group B phenotype are rare (2).

The immune system forms antibodies against whichever ABO blood group antigens are *not* found on the individual's RBCs. Thus, a group A individual will have anti-B antibodies and a group B individual will have anti-A antibodies. Blood group O is common, and individuals with this blood type will have both anti-A and anti-B in their serum. Blood group AB is the least common, and these individuals will have neither anti-A nor anti-B in their serum.

Blood group	Antigen(s) present on the red blood cells	Antibodies present in the serum	Genotype(s)
A	A antigen	Anti-B	AA or AO
B	B antigen	Anti-A	BB or BO
AB	A antigen and B antigen	None	AB
O	None	Anti-A and Anti-B	OO

ABO antibodies in the serum are formed naturally. Their production is stimulated when the immune system encounters the "missing" ABO blood group antigens in foods or in micro-organisms. This happens at an early age because sugars that are identical to, or very similar to, the ABO blood group antigens are found throughout nature.

The ABO locus has three main allelic forms: A, B, and O. The A allele encodes a glycosyltransferase that produces the A antigen (N-acetylgalactosamine is its immunodominant sugar), and the B allele encodes a glycosyltransferase that creates the B antigen (D-galactose is its immunodominant sugar).

The O allele encodes an enzyme with no function, and therefore neither A or B antigen is produced, leaving the underlying precursor (the H antigen) unchanged. These antigens are incorporated into one of four types of oligosaccharide chain, type 2 being the most common in the antigen-carrying molecules in RBC membranes. Some of the other enzymes involved in the earlier stages of ABO antigen synthesis are also involved in producing antigens of the Hh blood group and the Lewis blood group.

Expression

Although the ABO blood group antigens are regarded as RBC antigens, they are actually expressed on a wide variety of human tissues and are present on most epithelial and endothelial cells.

Each human RBC expresses about 2 million ABO blood group antigens. Other blood cells, such as T cells, B cells, and platelets, have ABO blood group antigens that have been adsorbed from the plasma. In individuals who are "secretors", a soluble form of the ABO blood group antigens is found in saliva and in all bodily fluids except for the cerebrospinal fluid.

A number of illnesses may alter a person's ABO phenotype. Patients can "acquire" the B antigen during a necrotizing infection during which bacteria release an enzyme into the circulation that converts the A1 antigen into a B-like antigen (3). During this time, patients should not receive blood products that contain the B antigen because their sera will still contain anti-B. Once the underlying infection is treated, the patients' blood groups return to normal.

Illness can also cause patients to "lose" ABO blood group antigens. Any disease that increases the body's demand for RBCs may weaken the expression of ABO blood group antigens, e.g., thalassemia. In addition, ABO blood group antigens can be altered by hematological cancers that can modify the sugar chains that bear the ABO blood group antigens, leading to the use of the A and B antigens as tumor markers for acute leukemia, myeloproliferative disorders, and myelodysplasia.

Function of the A and B antigens

The functions of the ABO blood group antigens are not known. Individuals who lack the A and B antigens are healthy, suggesting that any function the antigens have is not important, at least not in modern times.

Diseases associated with ABO blood group antigens

No diseases are known to result from the lack of expression of ABO blood group antigens, but the susceptibility to a number of diseases has been linked with a person's ABO phenotype. Such correlations remain controversial and include the observation that gastric cancer appears to be more common in group A individuals (4), whereas gastric and duodenal ulcers occur more often in group O individuals (5).

A clear correlation has been established between the ABO phenotype and the level of two proteins involved in blood clotting; factor VII (FVIII) and von Willebrand factor (vWF) (6). Blood group O individuals have about 25% less FVIII and vWF in their plasma. It is well established that low levels of FVIII and vWF are a cause of excess bleeding, and therefore it may also be the case that increased levels make clotting more likely, increasing the risk of both arterial (ischemic heart disease) and venous (thromboembolic disease) problems. Indeed, non-group O individuals have been shown to be at an increased risk of both arterial and venous disease (6).

Clinical significance of ABO antibodies

ABO antibodies are of major clinical significance for two reasons: they are naturally occurring and are found universally, and, they are highly reactive.

Transfusion reactions

The routine practice of blood typing and cross matching blood products should prevent adverse transfusion reactions caused by ABO antibodies. However, clerical error can result in "the wrong blood" being transfused into a patient, an error which can result in the death of the patient (7, 8).

If a recipient who has blood group O is transfused with non-group O RBCs, the naturally occurring anti-A and anti-B in the recipient's serum binds to their corresponding antigens on the transfused RBCs. These antibodies fix complement and cause rapid intravascular hemolysis, triggering an acute hemolytic transfusion reaction that can cause disseminated intravascular coagulation, shock, acute renal failure, and death.

Anti-A1 is a less significant cause of transfusion reactions and does not appear to fix complement.

Hemolytic disease of the newborn

Most cases of hemolytic disease of the newborn (HDN) that arise from an ABO incompatibility require no treatment. Cases of severe hemolysis that require exchange transfusions are less common, and fetal hydrops is rare (9).

HDN caused by ABO antibodies occurs almost exclusively in infants of blood group A or B who are born to group O mothers (10). This is because the anti-A and anti-B formed in group O individuals tend to be of the IgG type (and therefore can cross the placenta), whereas the anti-A and anti-B found in the serum of group B and A individuals, respectively, tends to be of the IgM type. Although uncommon, cases of HDN have been reported in infants born to mothers with blood group A2 (11) and blood group B (12).

HDN tends to be relatively mild in nature mainly because fetal RBCs don't express adult levels of A and B antigens. However, the strength of fetal ABO blood group antigens can vary, and therefore the degree of hemolysis and hence the severity of HDN can be unpredictable (13). Early studies suggested that the race of a neonate was a risk factor for developing ABO HDN (14). However, later studies showed that the prevalence of disease that required treatment did not differ significantly among Asian, Black, Hispanic, and Caucasian infants (15).

Molecular information

Gene

The ABO locus encodes specific glycosyltransferases that synthesize A and B antigens on RBCs. For A/B antigen synthesis to occur, a precursor called the H antigen must be present. In RBCs, the enzyme that synthesizes the H antigen is encoded by the H locus (FUT1). In saliva and other bodily secretions, the enzyme that synthesizes the H antigen is encoded by the Se locus (FUT2).

The ABO locus

The ABO locus is located on chromosome 9 at 9q34.1-q34.2. It contains 7 exons that span more than 18 kb of genomic DNA. Exon 7 is the largest and contains most of the coding sequence. Exon 6 contains the deletion that is found in most O alleles and results in a loss of enzymatic activity.

The A and B alleles differ from each other by seven nucleotide substitutions, four of which translate into different amino acids in the gene product (R176G, G235S, L266M, G268A). The residues at positions 266 and 268 determine the A or B specificity of the glycosyltransferase they encode (16).

The O allele differs from the A allele by deletion of guanine at position 261. The deletion causes a frameshift and results in translation of an almost entirely different protein that lacks enzymatic activity (16).

There are many variant ABO alleles that encode a number of variant ABO phenotypes, but they do not encode specific antigens other than the A and B antigens. For example, weak A subgroups, such as A₃, A_x, and A_{e1}, express the A antigen, and weak B subgroups, such as B₃ and B_x, express the B antigen (2).

The H locus (FUT1)

The H locus is located on chromosome 19 at 19q13.3. It contains three exons that span more than 5 kb of genomic DNA, and it encodes a fucosyltransferase that produces the H antigen on RBCs.

Individuals who are homozygous for null alleles at this locus (h/h) do not produce H antigen, and because

the H antigen is an essential precursor to the ABO blood group antigens, they cannot produce A and B antigens. Therefore, their serum contains anti-A and anti-B, in addition to potent anti-H. This rare phenotype of H-deficient RBCs is called the "Bombay phenotype" (O_h) after the city in which it was first discovered. Individuals with the Bombay phenotype are healthy, but if they ever needed a blood transfusion, the antibodies in their serum would place them at a high risk of having an acute hemolytic transfusion reaction. This can be avoided by using only blood products from a donor who also has the Bombay phenotype (usually a relative).

Read more about the Hh blood group in Chapter 6.

The Se locus (FUT2)

The Se locus is located on chromosome 19 at 19q13.3. It contains two exons that span about 25 kb of genomic DNA.

The Se locus encodes a specific fucosyltransferase that is expressed in the epithelia of secretory tissues, such as salivary glands, the gastrointestinal tract, and the respiratory tract. The enzyme it encodes catalyzes the production of H antigen in bodily secretions.

"Secretors" have at least one copy of the Se gene that encodes a functional enzyme—their genotype is Se/Se or Se/se. They secrete H antigen which, depending on their ABO genotype, is then processed into A and/or B antigens.

Non-secretors are homozygous for null alleles at this locus (se/se). They are unable to produce a soluble form of H antigen and hence do not produce A and B antigens.

References

1. Reid ME and Lomas-Francis C. *The Blood Group Antigen Facts Book*. Second ed. 2004, New York: Elsevier Academic Press.
2. Daniels G. *Human Blood Groups*, Second ed. 2002, Blackwell Science.
3. Stayboldt C, Rearden A, Lane TA. B antigen acquired by normal A1 red cells exposed to a patient's serum. *Transfusion* 1987; 27:41-4.
4. Reid ME, Bird GW. Associations between human red cell blood group antigens and disease. *Transfus Med Rev* 1990; 4:47-55.
5. O'Donnell J, Laffan MA. The relationship between ABO histo-blood group, factor VIII and von Willebrand factor. *Transfus Med* 2001; 11(4):343-51.
6. Fuchs CS, Mayer RJ. Gastric carcinoma. *N Engl J Med* 1995; 333:32-41.
7. Sazama K. Reports of 355 transfusion-associated deaths: 1976 through 1985. *Transfusion* 1990; 30:583-90.
8. Williamson LM, Lowe S, Love EM, Cohen H, Soldan K, McClelland DB, Skacel P, Barbara JA. Serious hazards of transfusion (SHOT) initiative: analysis of the first two annual reports. *BMJ* 1999; 319:16-9.
9. Gilja BK, Shah VP. Hydrops fetalis due to ABO incompatibility. *Clin Pediatr (Phila)* 1988; 27:210-2.
10. Ozolek JA, Watchko JF, Mimouni F. Prevalence and lack of clinical significance of blood group incompatibility in mothers with blood type A or B. *J Pediatr* 1994; 125:87-91.
11. Jeon H, Calhoun B, Pothiawala M, Herschel M, Baron BW. Significant ABO hemolytic disease of the

newborn in a group B infant with a group A2 mother. *Immunohematol* 2000; 16:105-8.

12. Haque KM, Rahman M. An unusual case of ABO-haemolytic disease of the newborn. *Bangladesh Med Res Counc Bull* 2000; 26:61-4.

13. Grundbacher FJ. The etiology of ABO hemolytic disease of the newborn. *Transfusion* 1980; 20:563-8.

14. Bucher KA, Patterson AM, Jr, Elston RC, Jones CA, Kirkman HN, Jr. Racial difference in incidence of ABO hemolytic disease. *Am J Public Health* 1976; 66:854-8.

15. Toy PT, Reid ME, Papenfus L, Yeap HH, Black D. Prevalence of ABO maternal-infant incompatibility in Asians, Blacks, Hispanics and Caucasians. *Vox Sang* 1988; 54:181-3.

16. Yamamoto F, Clausen H, White T, Marken J, Hakomori S. Molecular genetic basis of the histo-blood group ABO system. *Nature* 1990; 345:229-33.
