Processing, testing and selecting blood components

In this article...
- How blood components should be stored
- Which blood groups are incompatible with each other
- Methods used to match recipients with suitable components

Keywords: Blood/Transfusion/Safety

This article has been double-blind peer reviewed

Transfusing patients with incompatible blood components can be fatal. Understanding the testing and transfusion process can help reduce errors.

Whole blood given at donation is separated into three constituent parts for therapeutic use: red blood cells; platelets; and plasma. These are known as blood components. They are usually transfused independently of each other according to the clinical requirements of the patient. All three components can be collected individually from whole blood by a process called apheresis, also referred to as component donation. Apheresis involves the donor’s circulation being connected to a machine in a closed circuit; blood is taken from the donor, separated into component parts and the desired component collected and removed, with the rest returned to the donor. All blood donations are tested to determine the ABO and RhD blood group; this is explained in further detail below.

During whole blood donation, around 475ml of blood is drawn off (MacLennan, 2013), to be processed. White blood cells are filtered out during processing (leu-codepletion), to reduce the risk of transmission of variant Creutzfeldt-Jakob disease (vCJD) (Cardigan and Thomas, 2013). All donations are tested for HIV, hepatitis B and C, human T-cell lymphotrophic virus (HTLV) and syphilis. The blood is then centrifuged so it separates into three layers:
- Red blood cells at the bottom;
- Plasma at the top;
- A buffy coat (containing platelets) in the middle.

These layers are pressed out into separate bags. To reduce the risk of contamination, separation of blood components is carried out using an entirely closed system, so they are never exposed to open air. Every whole-blood and apheresis donation is given a unique donation number, which is then applied to all components derived from that donation, allowing every component to be traced from donor to recipient (MacLennan, 2013).

Red cells
Once separated, red blood cells are resuspended in an additive solution (for example SAG-M, a mix of saline, adenine, glucose and mannitol), intended to provide energy and stability for the cells. This

5 key points

1. Death or severe harm due to an ABO-incompatible transfusion is classified by the Department of Health as a “never event”
2. All blood components and laboratory testing procedures must comply with Blood Safety and Quality Regulations 2005 requirements
3. The ABO and RhD blood group systems are the most clinically significant
4. All blood components have strict storage conditions to minimise bacterial growth and ensure clinical efficacy
5. Red cell antibodies in a patient’s plasma may delay the provision of red cells

All blood donation samples must be tested...
results in a unit of “red cells in additive solution” (also simply called “red cells”), which has a mean volume of 274ml and may still contain up to 30ml of residual plasma (Kosmirak, 2014).

The red cells component from a donation may be further split into smaller packs to be used for neonates; they are never mixed with blood from other donations – one unit of red cells of whatever size will only ever come from one donor.

Red cells are stored at 2-6°C to minimise bacterial growth and slow cell metabolism (to maximise cell life) for up to 35 days (MacLennan, 2013). It is vital that monitored cold storage conditions (known as “cold chain”) are maintained to optimise the red cells and meet regulatory requirements (tinyurl.com/BloodSafetyRegulations). Red cells can only be outside temperature-controlled conditions for up to 30 minutes – they must then go back into cold storage, be used or be returned to the transfusion laboratory to be discarded. Transfusion should be completed within four hours of removal from cold storage.

It is common to transfuse over 90-120 minutes per unit (British Committee for Standards in Haematology, 2009) but caution must be exercised when it comes to the red cell transfusion rate in patients with increased risk of circulatory overload. Blood components should be given only one unit at a time, except when managing a massive haemorrhage. Red cells may be irradiated for patients who are immunocompromised to reduce the risk of transfusion-associated graft versus host disease; this will also reduce the life of the cells, so they are issued with a reduced expiry time.

Plasma

The plasma part of a donation is frozen to make a unit of fresh frozen plasma (FFP) or processed to make cryoprecipitate, which has a higher concentration of fibrinogen and factor VIII, and then frozen (Norfolk, 2013). Freezing allows for long-term storage – up to 36 months at -25°C (MacLennan, 2013). When needed, these components can be thawed to 37°C in 15-30 minutes, depending on the thawing system used by the transfusion laboratory. Once thawed, FFP and cryoprecipitate can be kept for up to four hours at 20-24°C and should be transfused over 30 minutes per pack; alternatively, thawed FFP can be stored for up to 24 hours at 2-6°C. Plasma from male donors only is used to produce FFP and cryoprecipitate, as this is considered to have less risk of triggering a transfusion-related acute lung injury (TRALI) reaction in the recipient (Norfolk, 2013).

Platelets

Platelets can be produced in two ways:

- The buffy coat of a donation may be pooled with those from three other donations to make a single platelet dose for transfusion (Norfolk, 2013);
- They can be acquired from a single donor by apheresis.

However platelets are made, an individual dose is called an adult therapeutic dose. Smaller packs of platelets are split from single apheresis donations and are called paediatric therapeutic doses. Platelets are resuspended either in plasma alone or in 70% platelet additive solution and 30% plasma; pooled platelets use only plasma from one male donor of the “pool” to reduce the risk of TRALI (Norfolk, 2013).

Platelets are stored at room temperature (20-24°C) on an agitator to prevent them aggregating (cells “clumping”); they expire after five days, or seven days if the unit undergoes bacterial screening (MacLennan, 2013). The shelf life of platelets is much shorter than that of red blood cells, which reflects the increased risk of bacterial growth at room temperature. Platelets can be irradiated, but this does not reduce the short shelf life. Both red cells and platelets are living cells and, as such, appropriate care should be taken in their handling and transport.

Red cell antigens and antibodies

All cells have molecules on their surface (antigens), which can stimulate an immune response in patients. Those present on the surface of red blood cells, known as red cell antigens, can react with antibodies called red cell antibodies. More than 300 red cell antigens have been classified into different “systems”, which give rise to the different blood groups.

The red cell antibodies produced by the immune system each react with a specific red cell antigen. Red cell antibodies can be split into two notable types:

- Naturally occurring antibodies to the A and B antigens of the ABO system;
- Acquired antibodies to all other red cell antigens.

Naturally occurring antibodies are produced in early life and are found in everyone after the first three months of life. Acquired antibodies are produced by exposing the immune system to foreign red cell antigens, most commonly by blood transfusion or pregnancy. Red cell antibodies are only produced against those antigens that a person’s red blood cells do not express.

The ABO system

The most well-recognised and clinically important blood group system is the ABO system. The ABO groups are determined by whether red blood cells express A, B, A and B, or neither antigen on their surface; these correspond to the A, B, AB and O blood groups respectively. Incidence of each group varies by ethnic population, with group O being the most common in the UK donor population (Norfolk, 2013).

Antibodies to ABO antigens naturally occur and can be detected in plasma after three months of age. The antibody formed will react with the antigen not present on the patient’s red cell. For example, a patient who has the B antigen on the red cell surface (blood group B) will have antibodies that react with the A antigen called anti-A. The antibodies and antigens of the ABO blood group system form the basis of transfusion incompatibility.

<table>
<thead>
<tr>
<th>TABLE 1. ABO ANTIGENS AND ANTIBODIES</th>
</tr>
</thead>
</table>

<table>
<thead>
<tr>
<th>ABO group</th>
<th>A</th>
<th>B</th>
<th>AB</th>
<th>O</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>Red cell antigens</strong></td>
<td>![A]</td>
<td>![B]</td>
<td>![AB] and ![B]</td>
<td>![None]</td>
</tr>
<tr>
<td><strong>Red cell antibodies</strong></td>
<td>![Anti-B]</td>
<td>![Anti-A]</td>
<td>![None]</td>
<td>![Anti-A and anti-B]</td>
</tr>
<tr>
<td>Can receive RCCs group</td>
<td>A, O</td>
<td>B, O</td>
<td>AB, A, B, O</td>
<td>O</td>
</tr>
<tr>
<td>Can receive FFP group</td>
<td>A, AB</td>
<td>B, AB</td>
<td>AB</td>
<td>O, A, B, AB</td>
</tr>
<tr>
<td>Can receive platelets group¹</td>
<td>A, AB, B, O²</td>
<td>B, AB, B, O²</td>
<td>AB, A, B, O²</td>
<td>O, AB, A, B</td>
</tr>
</tbody>
</table>

¹Order of preference: High-titre anti-A/anti-B negative. FFP = fresh frozen plasma. RCC = red cell component.
The ABO blood group is important because ABO-incompatible red cell transfusions can be fatal. During an ABO-incompatible transfusion, anti-A and/or anti-B in the patient’s blood binds to the transfused red cells, leading to their destruction (haemolysis) and an inflammatory response that can cause shock, renal failure and disseminated intravascular coagulation. An ABO-haemolytic reaction can occur after transfusion of only a very small volume of incompatible cells. Transfusion of ABO-incompatible plasma containing anti-A and/or anti-B can cause destruction of the patient’s red cells, especially in neonates. Haemolytic reactions can occur with other red cell antibodies/antigens, but these are not likely to be as severe.

Ensuring the patient receives the right blood component is the most important step in clinical transfusion practice: the recipient and donor unit should ideally be ABO identical, and must always be ABO compatible. Death or severe harm as a result of the inadvertent transfusion of ABO-incompatible blood components is listed as one of the Department of Health’s “never events” (DH, 2012).

The Rh system

The Rh system comprises five main antigens, for which people are positive or negative: C/c, E/e and D. Of these, RhD is the most clinically important. Antibodies to RhD are formed only in RhD-negative people after exposure to the D antigen via transfusion of RhD-positive red cells or pregnancy with an RhD-positive baby.

The antibodies produced (called anti-D) can trigger a haemolytic transfusion reaction if RhD-positive red cells are transfused and can cause haemolytic disease of the foetus and newborn in later pregnancies. It is crucial therefore, to transfuse only RhD-negative red cells and platelets to girls and women with childbearing potential who are RhD negative; the only exception is in an emergency where no RhD-negative component is available in time, when prophylactic anti-D immunoglobulin cover must be given (Norfolk, 2013).

Compatibility procedures in the transfusion laboratory

To ensure the patient receives compatible blood components, a venous blood sample will be tested in the laboratory.

Group and screen testing

A “group and screen” test will determine the ABO and RhD group by assessing the red cell antigens present, then screen the plasma for any red cell antibodies that can cause a transfusion reaction (such as anti-D) (British Committee for Standards in Haematology, 2012). If a screen is positive, further testing will be done; this may affect how long it takes for blood to be available.

Pre-transfusion blood samples (group and screen and/or crossmatch) must be labelled with the patient’s first name, last name, unique identification number (for example NHS number) and date of birth. In Wales, the first line of the patient’s address is also required (BCSH, 2009), while in Scotland gender must be specified.

ABO grouping is the most important test performed on pre-transfusion samples. To help minimise ABO-incompatible transfusions, national guidelines now recommend taking a second sample and testing it to confirm the ABO group in patients where there is no record of a previous test result (BCSH, 2012). This is intended to prevent potential errors caused by “wrong blood in tube” incidents.

Selection methods

When a red cell transfusion is requested, the laboratory selects units that are ABO and RhD compatible and negative for antigens corresponding to any red cell antibodies that might have been detected in the plasma. Depending on the group and screen result, two main methods can be used to issue blood: electronic issue or serological crossmatch.

Electronic issue can be used if the patient’s ABO and RhD group has already been established and the patient’s plasma does not contain red cell antibodies (currently or historically). ABO and RhD-compatible red cell units are selected by laboratory staff, then a computer is used to check them against the patient’s results before they are issued. Electronic issue is quicker than crossmatching and can allow issue of red cell units to be managed remotely using computer-controlled satellite fridges that may be located in the clinical area. This method also helps with stock control as units do not become tied up being “reserved” for specific patients.

If red cell antibodies have been detected in the patient’s plasma, a serological crossmatch will be performed. The patient’s plasma will be directly tested against a sample from the unit of red cells at 37°C to look for a reaction indicating incompatibility. Only units that are compatible (have no reactions) with the patient’s plasma will be issued (Klein and Anstee, 2005). Units crossmatched for patients upon request are usually held in reserve for that patient for a given period of time (usually 24-48 hours, depending on local transfusion policy).

Conclusion

The ABO and RhD blood groups are the most clinically significant in blood transfusion medicine. It is important to ensure the patient’s ABO and Rh blood group is determined before blood is issued. If no red cell antibodies are detected, blood will be matched for ABO and RhD groups. If red cell antibodies are detected, a sample from the blood component will be tested against the patient’s plasma to make sure it is compatible. Blood components must be handled with care and stored correctly before use to ensure they are safe to transfuse.

References

British Committee for Standards in Haematology (2012) Guidelines for Pre-Transfusion Compatibility Procedures in Blood Transfusion Laboratories. tinyurl.com/PreTransfusionGuidelines


Kosmirek L (2014) NHSEBT Portfolio of Blood Components and Guidance for their Clinical Use. tinyurl.com/blood-comp-guide

For more on this topic go online...

- Blood transfusions 1: How to monitor for adverse reactions
- Bit.ly/NTTransfusionReaction