# Significance of Antibodies and appropriate selection of red cells for transfusion
## Chris Elliott
## Haematology Directorate manager

# Over the next 30 minutes

- Bit of basic immunology
- Serological red cell transfusion reactions
- Testing and sample timing
- Problems      - multiple antibodies
                – rare antibodies
                - haemolytic anaemias
- Coping strategies

# An Antigen.

- An antigen can be defined as a substance that, when introduced into the circulation of an individual lacking that antigen, can stimulate the production of a specific antibody.

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# Blood Group Antigens

- Substances found on red cells that can cause an immune response. They are not there to make our life difficult even though sometimes they do.

- Why have blood groups ?
  - Developed over 1000's of years in response to environmental factors, including bacteria, parasites, food substances

- –Hundreds of RBC antigens known

- –Important in recognising 'self'/ 'non-self'

- –May confer protection from disease ($Fy^a$-)

- –May enable attack by disease ($Le^b$)

- –May be needed for RBC membrane structure and function (Rh)

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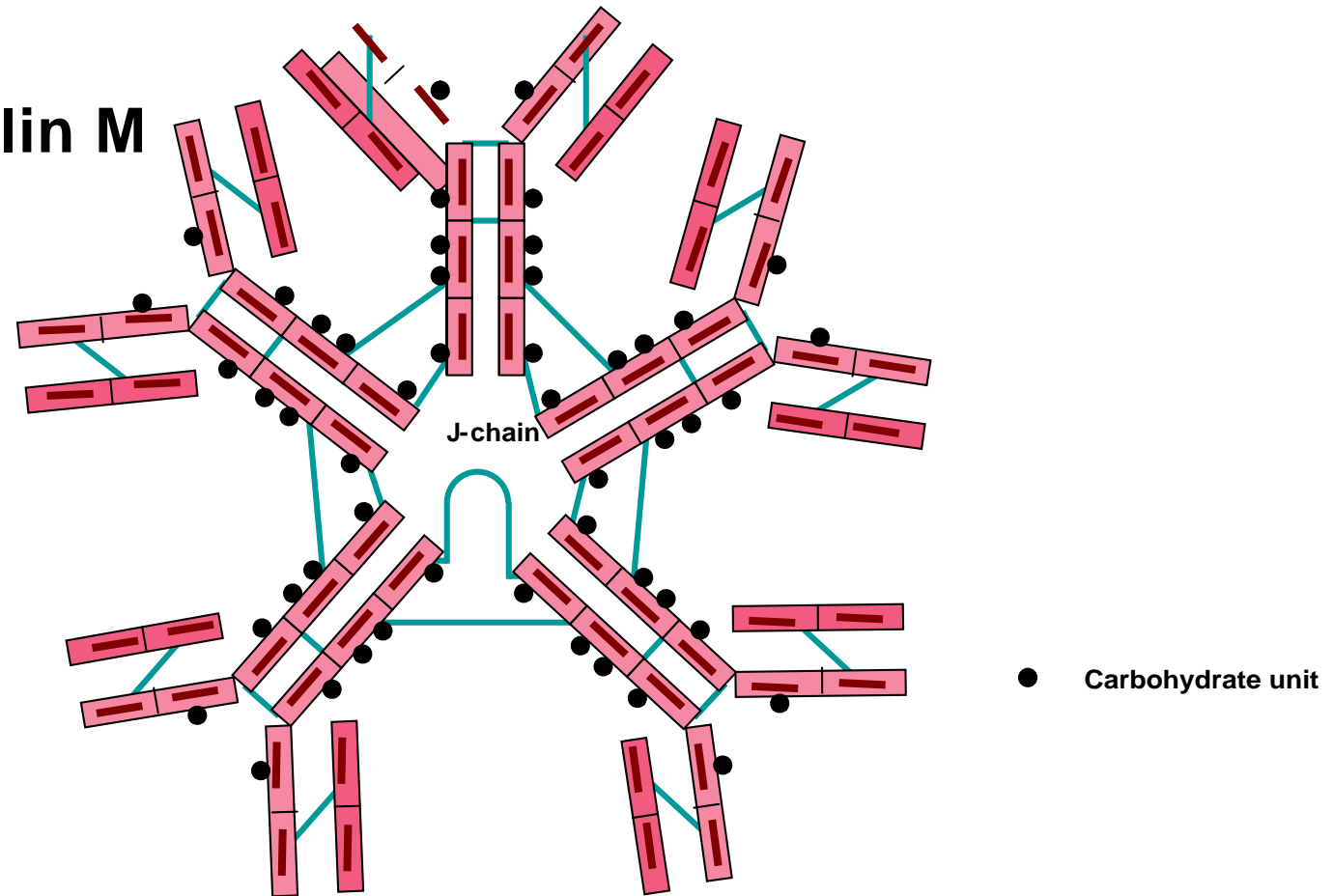
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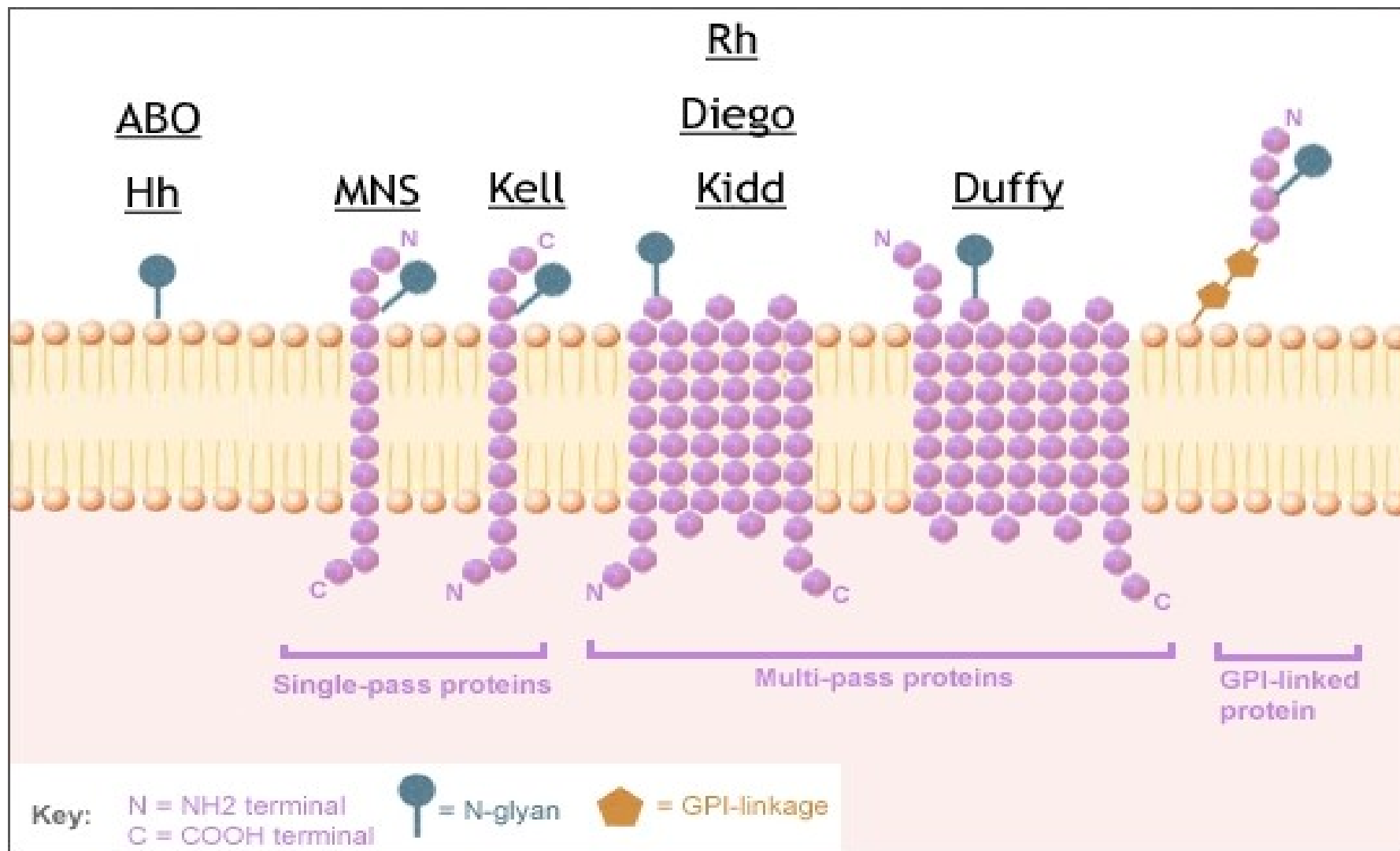
# An Antibody.

- An antibody can be defined as a protein (*i.e.* an immunoglobulin with specific antigen binding sites) produced as a result of the introduction of a foreign antigen, that has the ability to combine with (and, in many cases, destroy) the cells carrying the antigen that stimulated its production.
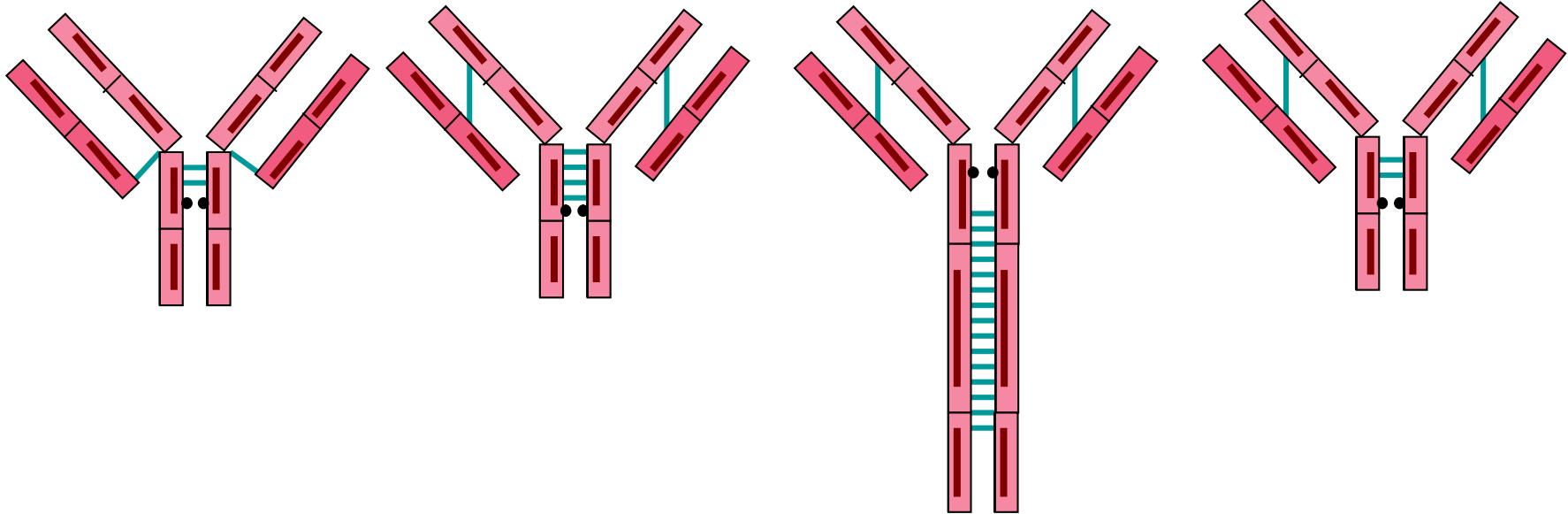
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# Antibodies

**Immunoglobulin M**



J-chain

● Carbohydrate unit

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# Antibodies

## Immunoglobulin IgG
## subclasses



**IgG1**          **IgG2**          **IgG3**          **IgG4**

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# Gel test
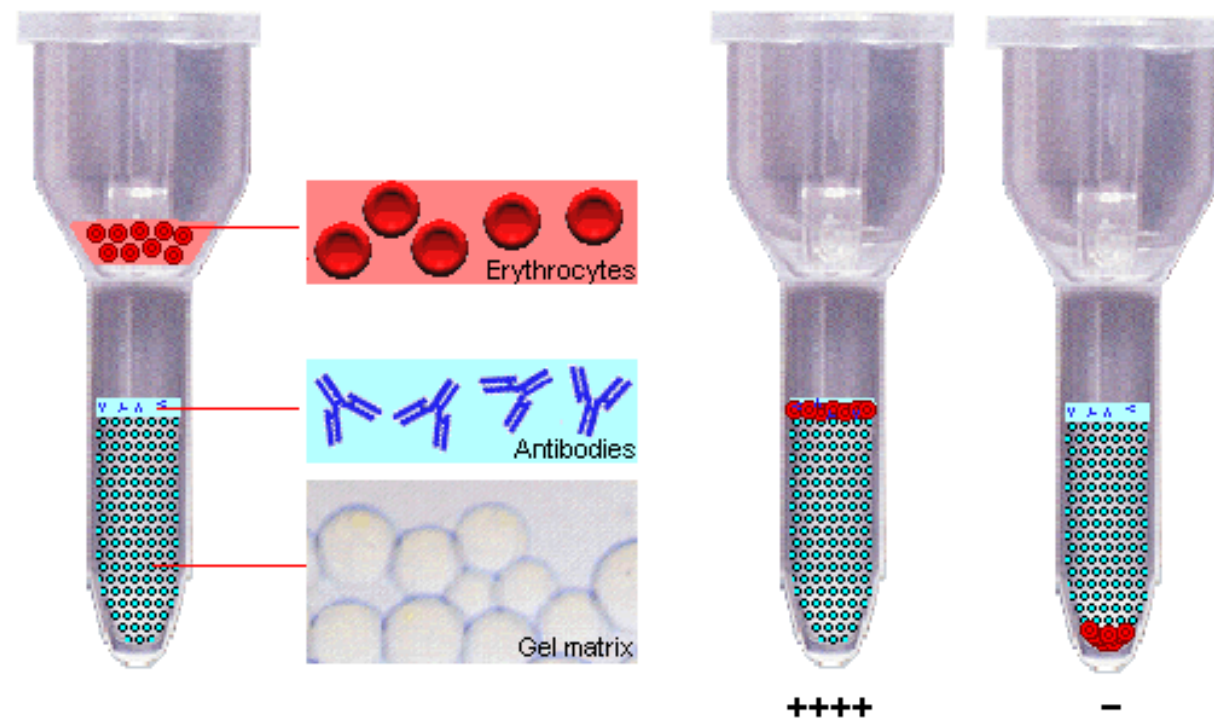
# How the Gel Test Works.
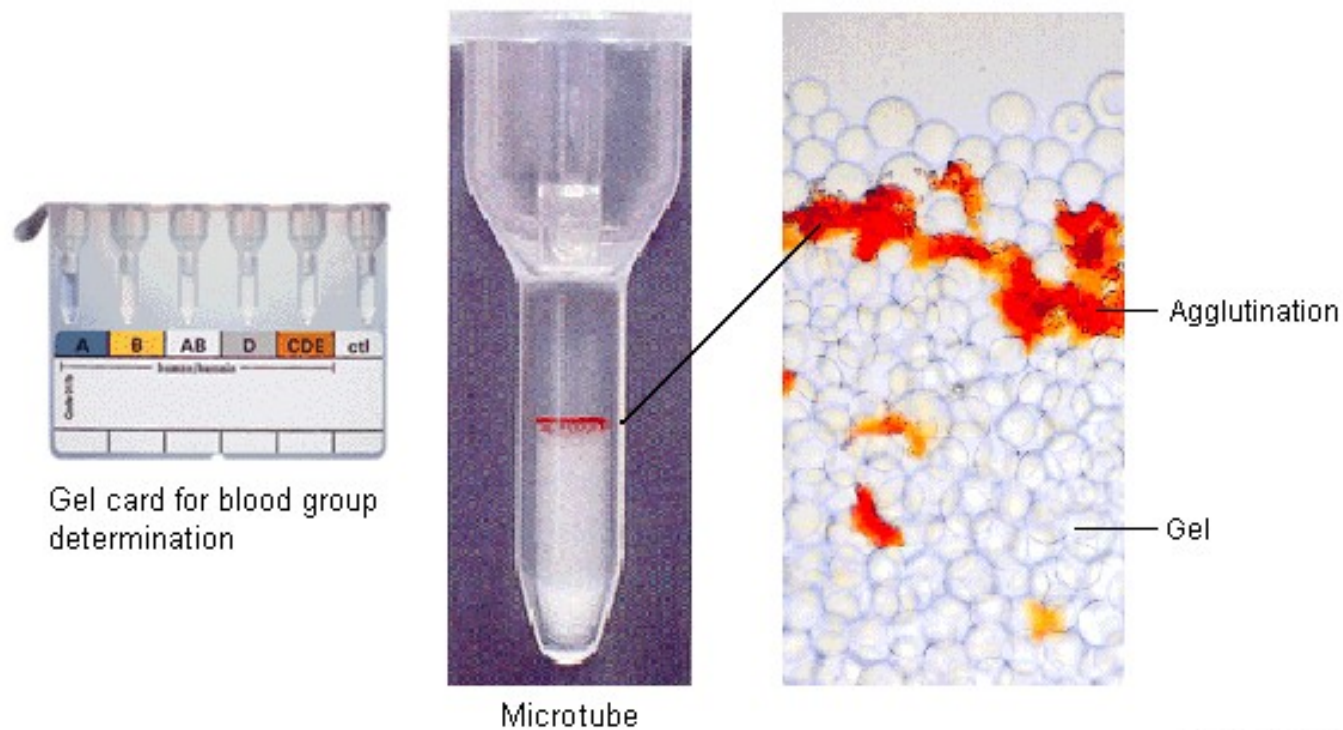
# IMMUNE DESTRUCTION OF CIRCULATING BLOOD CELLS

- Intravascular complement-mediated destruction usually initiated by antibody

- Extravascular macrophage-mediated destruction: antibody (IgG, IgA), complement (C3b, iC3b), antibody + complement
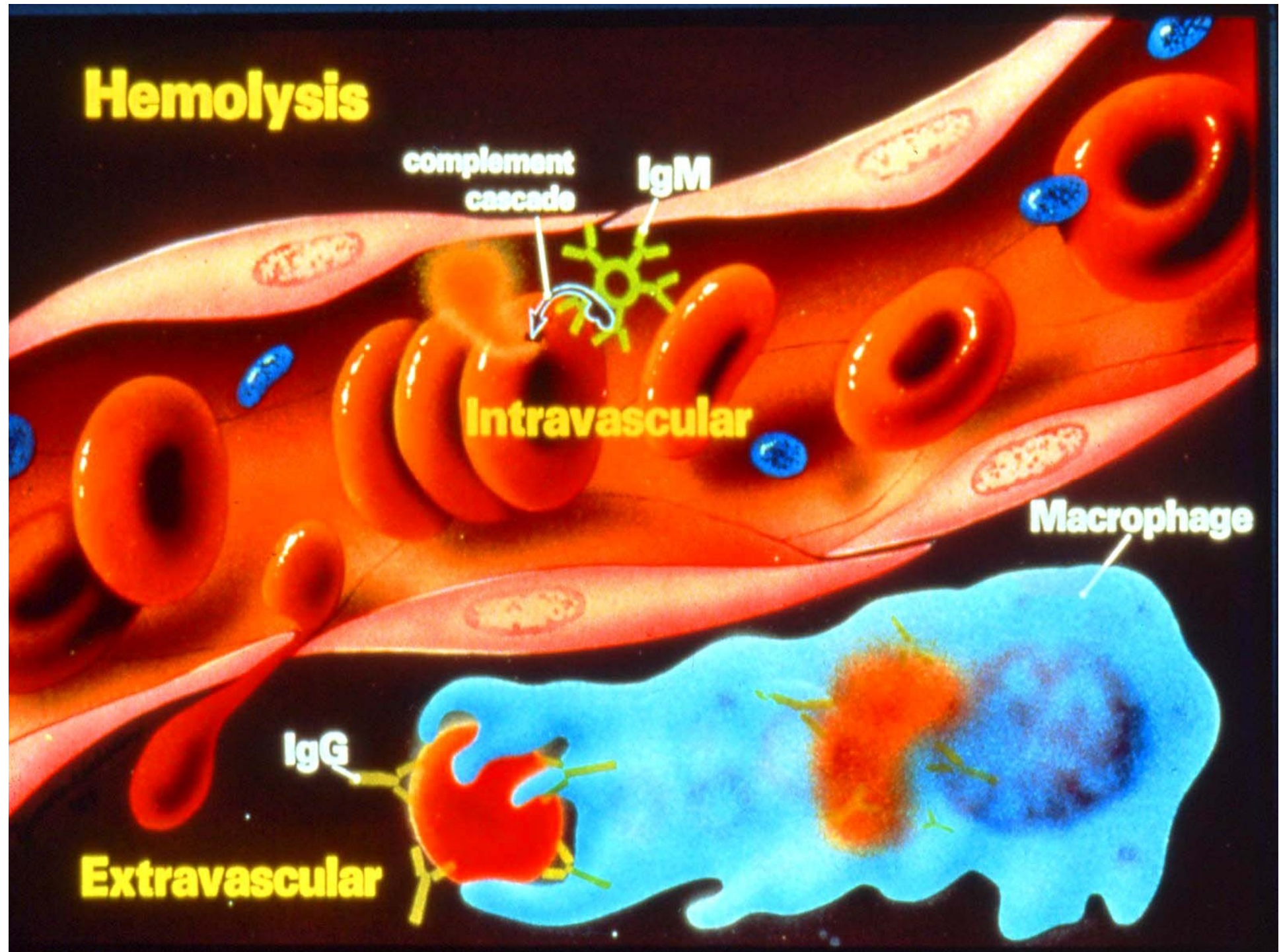
# Intravascular Haemolysis

**Neutrophils**

**Complement**

**Macrophages**

**Cytokines**

**Activates clotting factors**

**Thrombosis**

**Consumption of clotting factors**

**Organ failure**

**Bleeding**
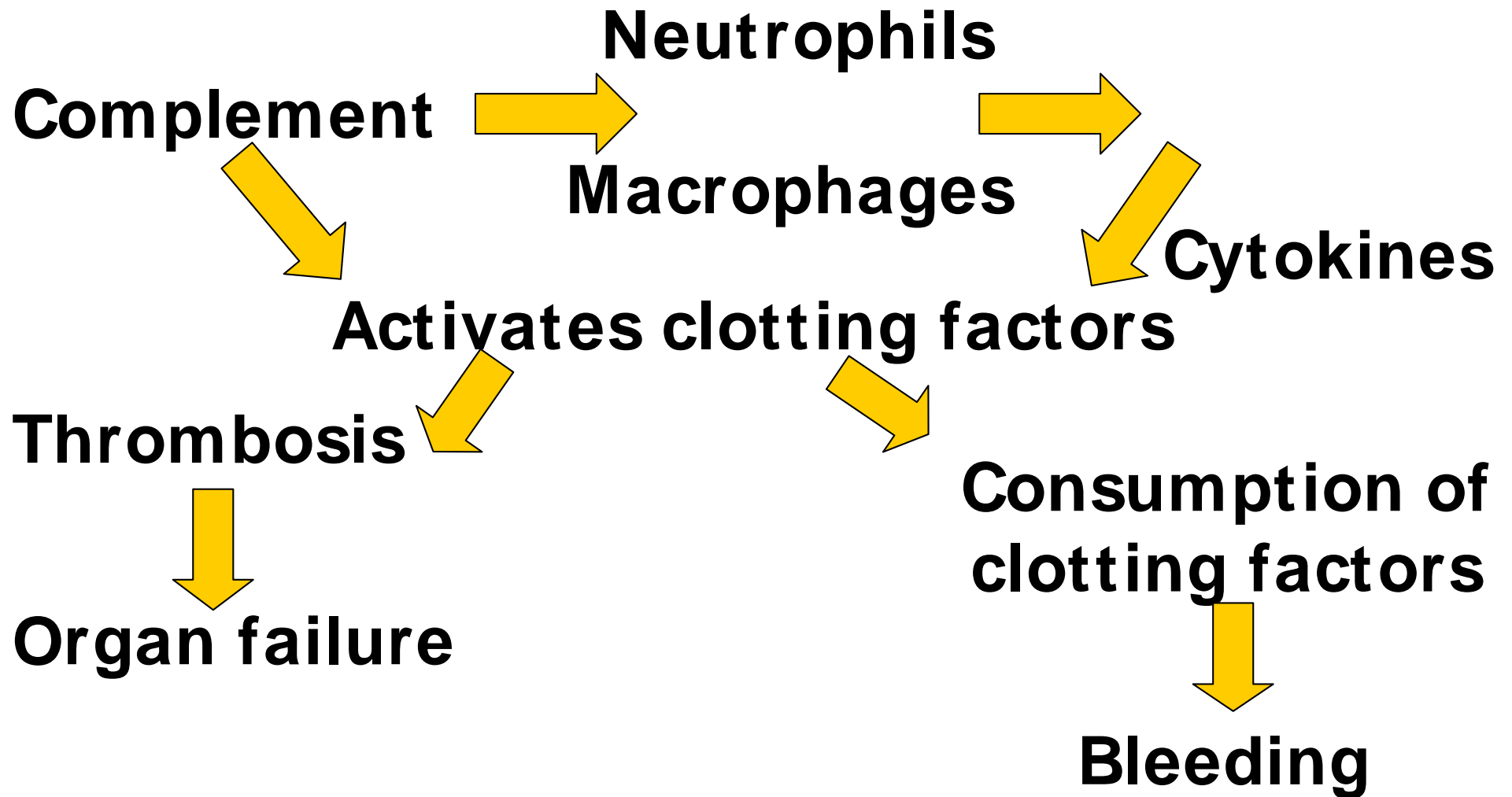
# Renal Failure.

- Hypotension, Shock.

- DIC.

- Free haemoglobin binds nitric oxide.

- Causes excessive vasoconstriction.

# Delayed Haemolytic Transfusion Reaction.

Clinical features.

- Fever.

- Fall in haemoglobin over time (several days).

- Jaundice.

- Haemoglobinuria.

- Renal failure (very rarely).

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# SPECIFICITY AND CLINICAL SIGNIFICANCE OF 37C REACTIVE ANTIBODIES

| Usually Significant | Not or Rarely Significant | Sometimes Significant |
|---|---|---|
| ABO | Bg | Cartwright |
| Rh | Le$^b$ | Gerbich |
| Kell | Ch/Rg | Dombrock |
| Kidd | Knops | Lutheran |
| Duffy | JMH | Lan |
| S, s, U | Cs$^a$ | LW |
| Vel | Sd$^a$ | Jr$^a$ |
| | Xg$^a$ | |

# Timing of the immune response
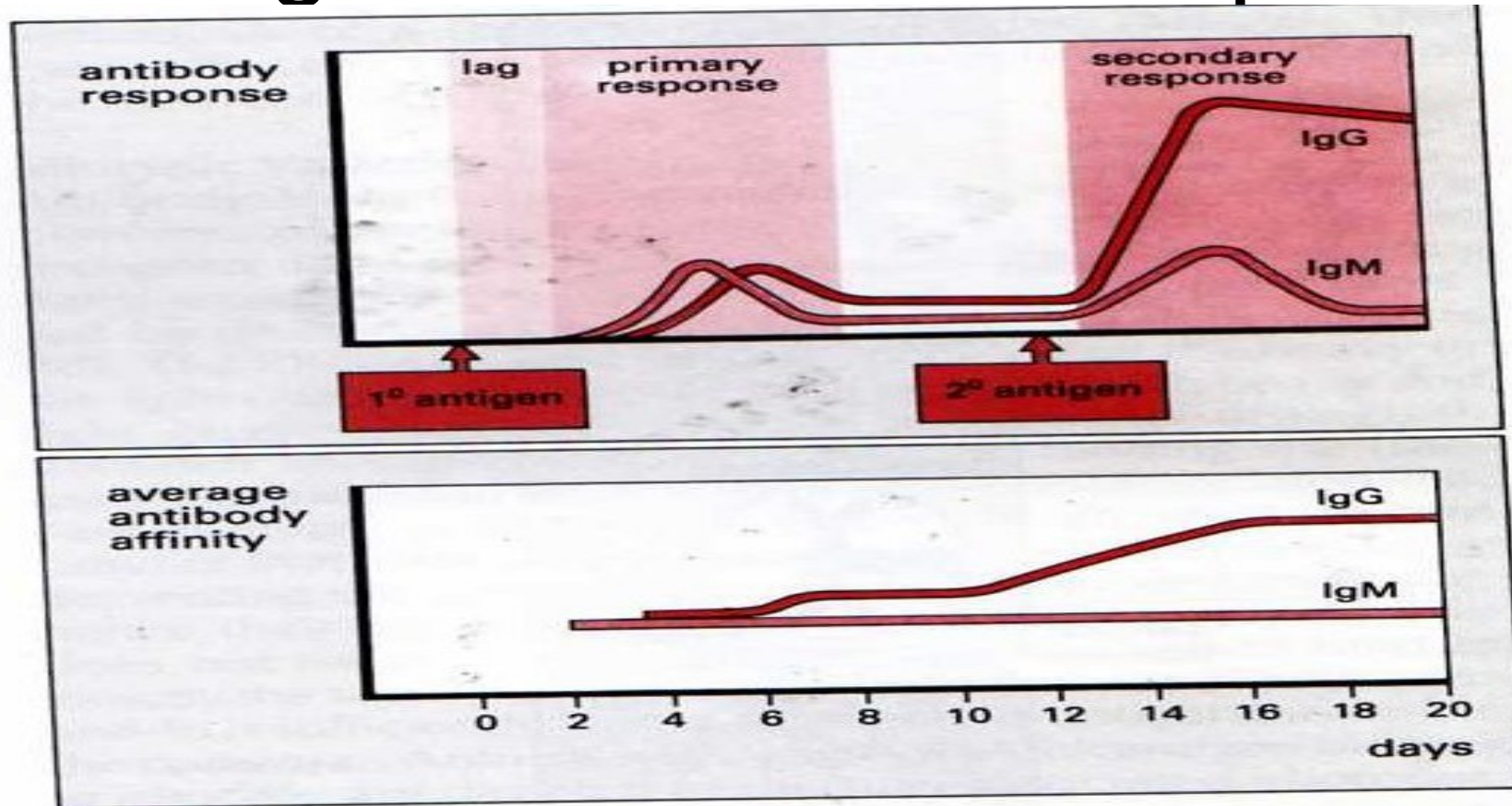


- ABO antibodies are mainly IgM and are formed in response to analogous bacterial antigens early in life, no red cell transfusion required for antibodies to be present
- Other red cell antibodies are mainly IgG and most HTR problems are caused following 2ndry sensitisation

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# When do non ABO red cell allo-antibodies form?

- There is a dearth of published data regarding when non ABO red cell allo-antibodies form and are first detectable following a stimulating event (be it a primary or secondary response).

- Of the papers available for review it is clear that only a very small percentage of antibodies which are below detectable level pre transfusion become detectable in the first 72 hours, estimated at 2.3% (Schonewille 2006), and supported by SHOT data (SHOT annual reports).

- Red cell destruction does not begin before the 4th day post transfusion (Mollison 2005, pg 477). Following this time, most developing antibodies will manifest themselves within the next 30 days (there are occasional stragglers), and by 3 months post transfusion very few antibodies will develop.

- SHOT data shows that the majority of delayed haemolytic transfusion reactions are noted 3-14 days post transfusion.

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# Timing of samples for red cell issue

**Current BCSH guidelines**

- 72 hour viability for 3 month post transfusion – but based on red cell availability not sample viability
- Patients undergoing repeated transfusions do not need daily samples.
- Regularly transfused patients with no allo-antibodies could undergo regular (e.g. annual) assessment to elongate viability e.g. extend to 7 days.
- Ante natal patients samples have a 7 day viability
- Need to consider intervening transfusion or pregnancy when using stored samples for crossmatch.

# Frequency of antigen negative blood

| ANTIBODY | % Compatible (Caucasian) |
|---|---|
| Anti-A | 56 |
| Anti-B | 86 |
| antiA,B | 42 |
| Anti-D | 15 |
| Anti-C | 30 |
| Anti-E | 70 |
| Anti-c | 20 |
| Anti-e | 2 |
| Anti-K | 91 |
| Anti Jka | 23 |
| Anti-Jkb | 27 |
| Anti-Fya | 34 |
| Anti-Fyb | 20 |
| Anti-S | 45 |
| Anti-s | 11 |
| Anti-M | 22 |

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# Multiple antibodies

- More difficult to secure compatible units the more antibodies a patient has.
  e.g. Patient is A Pos with anti-c, anti-K, anti-S and anti Fya
  % compatible = $0.56 \times 0.2 \times 0.91 \times 0.45 \times 0.34$
  $$= 0.016 \text{ or } <2\%$$
  This means only 2 units in every 100 would be compatible

- Have to get NHSBT red cell reference laboratory to screen units

# Rare antigen negative blood

- Problem when a patient who is negative for an antigen that over 98% of the population is antigen positive (so less than 2% of blood is compatible if they make an antibody)

- Examples:
  Anti-k - <0.2% Caucasians compatible
  Anti-Coa - <1% Caucasians compatible
  Anti–U – 0% Caucasians compatible, <3% blacks compatible
  Anti-Tja - <0.1% compatible, antigen negative donors restricted to Scandinavia

- Need to go to National/International rare blood banks, frozen blood supplies

# Nuisance antibodies

- Antibodies that while they themselves maybe of little clinical significance react against significant numbers of screening/panel cells and may mask presence of clinically significant antibodies
e.g. anti P1, anti-Leb, anti –N, cold reacting anti-M

- Delays occur while lab ensures no further antibodies present and then serologically crossmatch

# Haemolytic Anaemias

- AIHA
  - Warm type
  - Cold-type
  - Paroxysmal cold haemoglobinuria
  - Paroxysmal nocturnal haemoglobinuria
  - Drug induced
  - Combined warm/cold

- HDN/HDF

- HTR

# Diseases Associated With Warm AIHA.

- Reticulo-endothelial neoplasms (CLL and lymphoma).
- Myelodysplastic syndromes (MDS).
- Systemic lupus erythematosus (SLE).
- Infection (especially post-viral in children).
- Immunological disease.

Often, in cases of AIHA, the auto-antibody is not only bound to the red cells, but is also free in the plasma. Under this free auto-antibody, however, 32% of cases have alloantibodies present.

If free antibody is detected in the plasma, is it an autoantibody, an alloantibody, or are both present?

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# Coping strategies

- Ensure patients have a viable group and screen

- Schedule regular transfused patients for day clinics so that G&S can be processed and remain viable

- Consider using outreach nurses for regularly transfused day patients

# TRANSFUSING INCOMPATIBLE BLOOD

- Even though not perfect, historical data relating to specificity provides a basis for decision-making [see Daniels et al. The clinical significance of blood group antibodies. Transfus Med. 2002;12:287-95].

- Although RBC survival may not be normal, in presence of antibodies usually thought to be insignificant and also those known to cause delayed transfusion reactions, overt HTRs almost never occur.

- May be better to give antigen positive blood and risk a delayed HTR + give treatment for the potential HTR up front

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# Clinical team must be counseled so that risk of waiting can be balanced with clinical significance of antibody

"OBTAINING COMPATIBLE BLOOD FOR A CORPSE IS

NOT A THERAPEUTIC TRIUMPH "

Ed Snyder, MD (Yale University)

ISBTS meeting (Edinburgh)

September 6, 2002

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