# Significance of donor H&I and impact on organ recipients

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**NHS**

**Blood and Transplant**

- What is HLA?

- Why is it important in transplantation?
  - HLA matching
  - Sensitisation to HLA
  - Donor and recipient crossmatching

# What are HLA types?

- HLA types are structures (antigens) found on almost all cells of the body

- Includes cells that make up your tissues and organs

- In blood they are found on white blood cells and platelets, but not on red blood cells

- Play an important role in the immune system
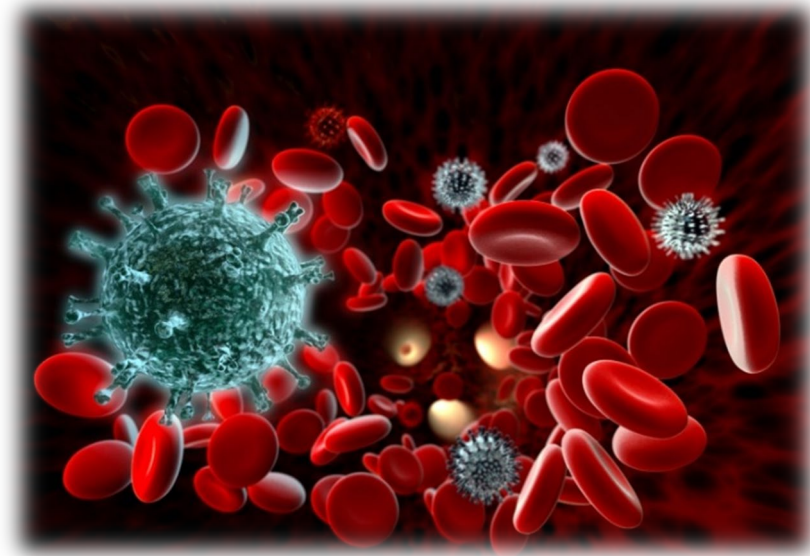
# What are HLA types?

- Acts like a flag on the cell surface

  - Indicate to the immune system whether a response is needed

- Healthy cell - No response

- Infected cell – Immune Response

- Transplanted Cell - Rejection



Healthy
Cell

Infected
Cell

Transplanted
Cell
Mismatched HLA
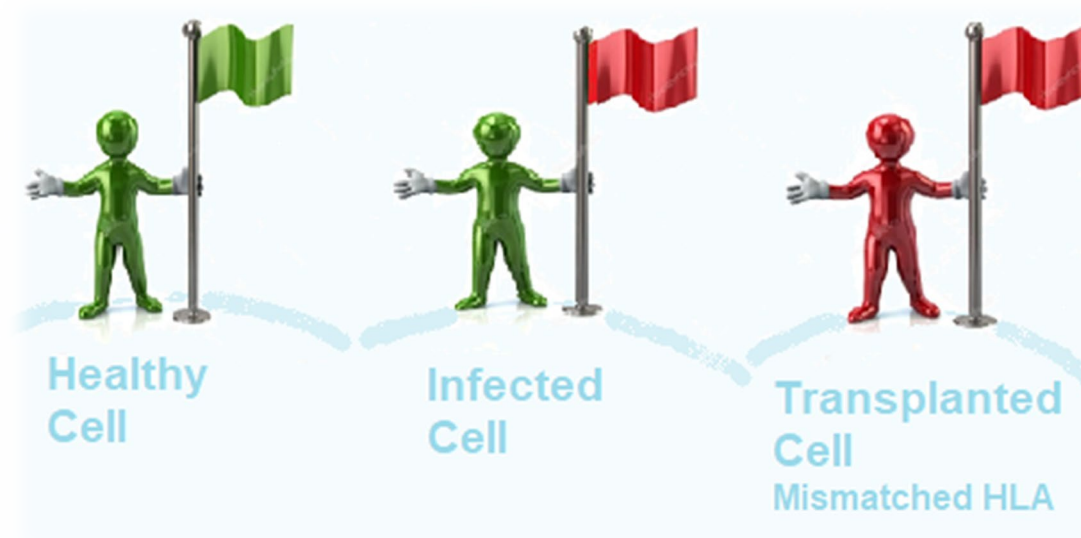
# What are HLA types?

- Everyone has two sets of HLA types

- Inherit 1 set from each parent

- Only a 1 in 4 chance of siblings being matched
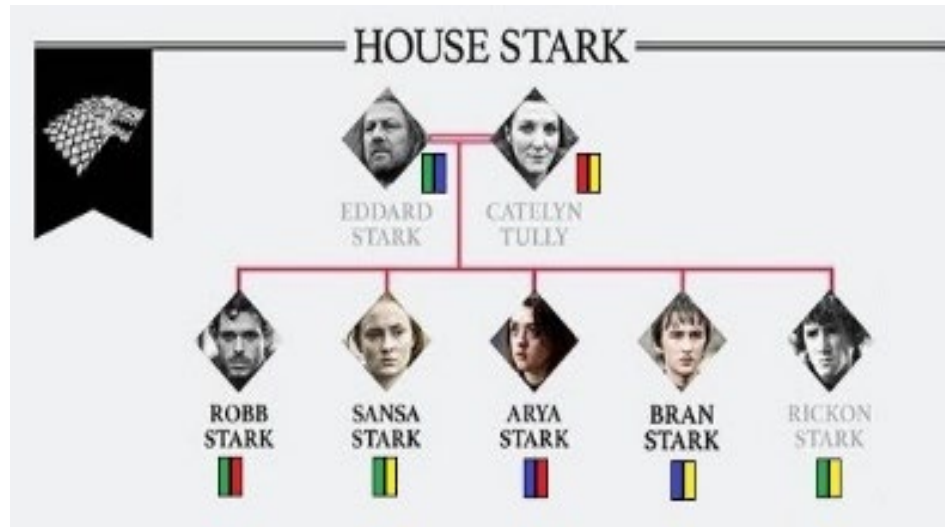
# What are HLA types?



- HLA types can be defined at different levels

  - Resolution

- Dog

- Spaniel

- Cocker Spaniel

- Blue Roan

# HLA Types



Broad Specificity- Dog
   HLA-A2, A3; B5, B15; Cw1, Cw3; DR6, -  DR52, -; DQ1,

Split Specificity - Spaniel
   HLA-A2, A3; B51, B62; Cw1, Cw10; DR13; DR52, -; DQ6,

Allele - Cocker Spaniel
   HLA-A*02:01, A*03:01; B*15:01, B*51:01; C*01:02, C*03:04;
   DRB1*13:01, DRB1*13:02; DRB3:03:01, - DQA1* 01:02
   DQA1*01:03;DQB1*06:04, DQB1*06:03 DPA1*01:03
   DPA1*02:01;DPB1*04:02 DPB1:02:01

# There are many different forms of HLA Types

| Gene | Alleles (Cocker) | Split Antigens (Spaniel) | Broad Antigens (Dog) |
|---|---|---|---|
| HLA-A | 6,192 | 28 | 11 |
| HLA-B | 7,431 | 60 | 30 |
| HLA-C | 6,067 | 18 | 16 |
| HLA-DRB1 | 3,391 | 20 | 11 |
| HLA-DQB1 | 1,232 | 9 | 4 |

Approximated figures

# HLA Matching/ Mismatching

HLA-A,B,DR
Mismatch grade
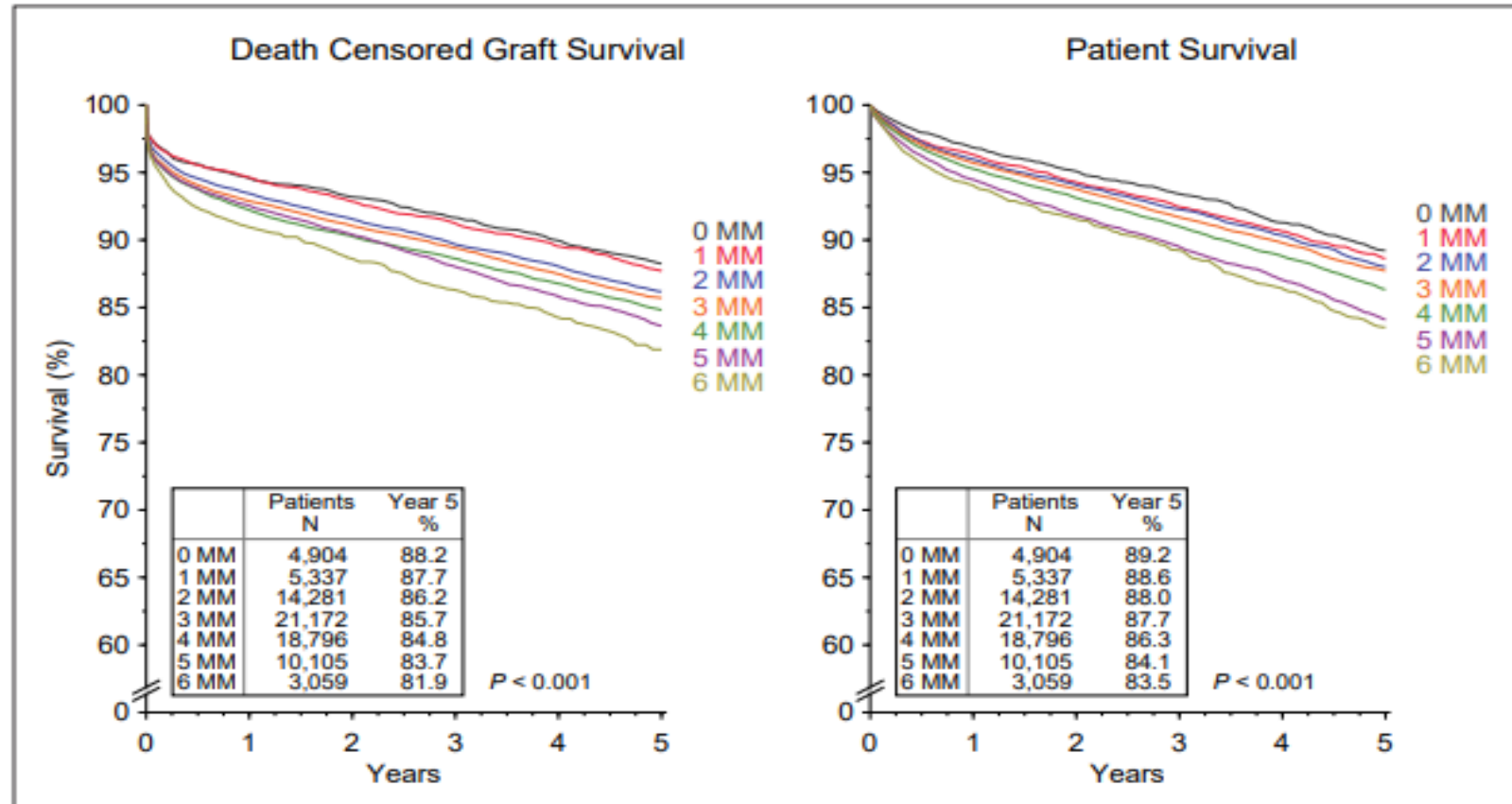
Donor

HLA-A1, A2,  B7, B8,  DR3, DR4

Recipient

A       HLA-A1, A2,  B7, B8,  DR3, DR4          000

B       HLA-A1, A3,  B7, B8,  DR3, DR4          100

C       HLA-A1, A9,  B5, B8,  DR3, DR4          110

D       HLA-A3, A9,  B5, B8,  DR3, DR7          211

# Collaborative Transplant Study  Data –HLA matching 2005-2014

We observed a 71 % increase in death censored graft failures from 0 to 6 HLA mismatches and Cox multivariate analysis showed a mean hazard ratio of 1.09 per HLA mismatch (95% CI 1.07–1.10, $P < 0.001$). The corresponding hazard ratio for patient survival was 1.04 (95% CI 1.03–1.06, $P < 0.001$).

# Why do we HLA type

Assess matching and likelihood of receiving a well matched graft

**Matchability** is a measure of how it difficult it is to match a patient with an organ donor in the UK.
This score takes into account a patient's blood type, HLA type and unacceptable antigens.
A patient with a score = 1 is defined as easy to match and a score = 10 as difficult to match.

Predict graft survival

Prevent sensitisation (antibody production)
    poorly matched graft = high sensitisation
    high frequency single mismatch e.g. HLA-A2

Assist donor selection e.g. in living donor transplant

# HLA Types

Broad Specificity  Used for offering/assigning mismatch grades
000,110,222 etc.

HLA-A2, A3; B5, B15; DR6, DR7;

Deceased donor typing is at this level -

HLA-A*02, A*03; B*15:01 (62), B*51; C*01, C*03; DRB1*13,  DRB1*07;
DRB3:03 (52), DRB4*01; DQA1* 01:02, DQA1*01:03; DQB1*06,
DQB1*02; DPB1*04:02, DPB1:02:01

Enables consideration of recipients antibodies (unacceptable antigens)

B5  - Broad

Splits

B51        B52
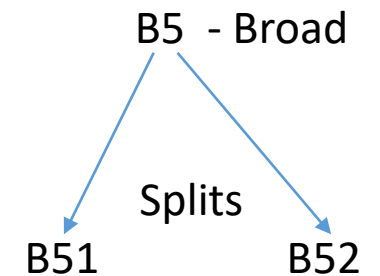
# Antibodies

- Antibodies play a crucial role in our immune system

  - Bind to target

    - Neutralise

    - Recruit other cells to engulf target

    - Start chemical cascade which will kill the target

- Help to destroy bacteria and viruses

- HLA Antibodies are very significant in transplantation

- Antibodies can cause rejection

- In worst case can cause irreversible damage to the transplant

# HLA antibodies

- Patients can make HLA-antibodies in different ways

  - Pregnancy

  - Transplantation

  - Transfusion

- Patients with antibodies are "sensitised"

- If a patient is highly sensitised they become very difficult to transplant

- Calculated Reaction Frequency (cRF) – percentage of donors in pool of 10,000 with whom the donor is antibody incompatible
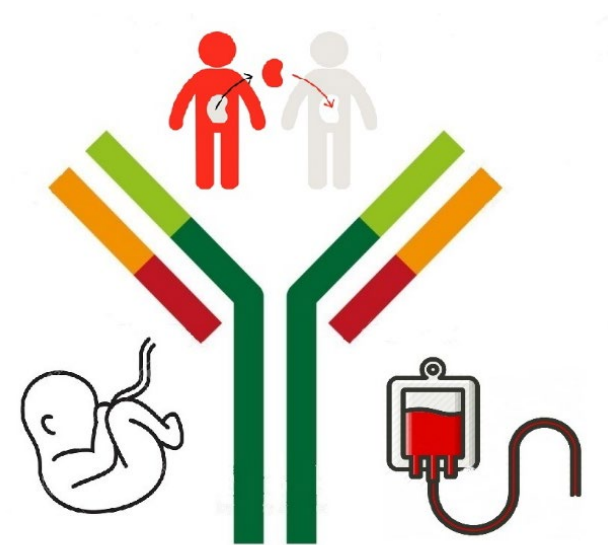
# Laboratory Testing
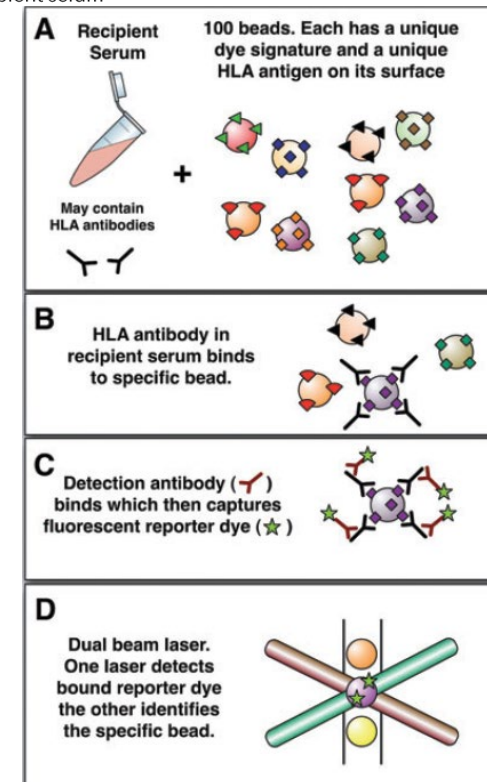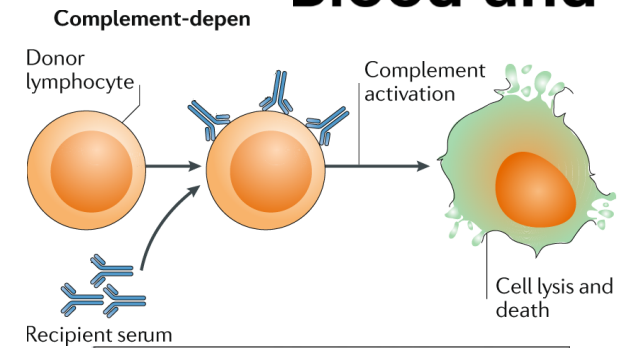
- HLA antibody Detection and Definition
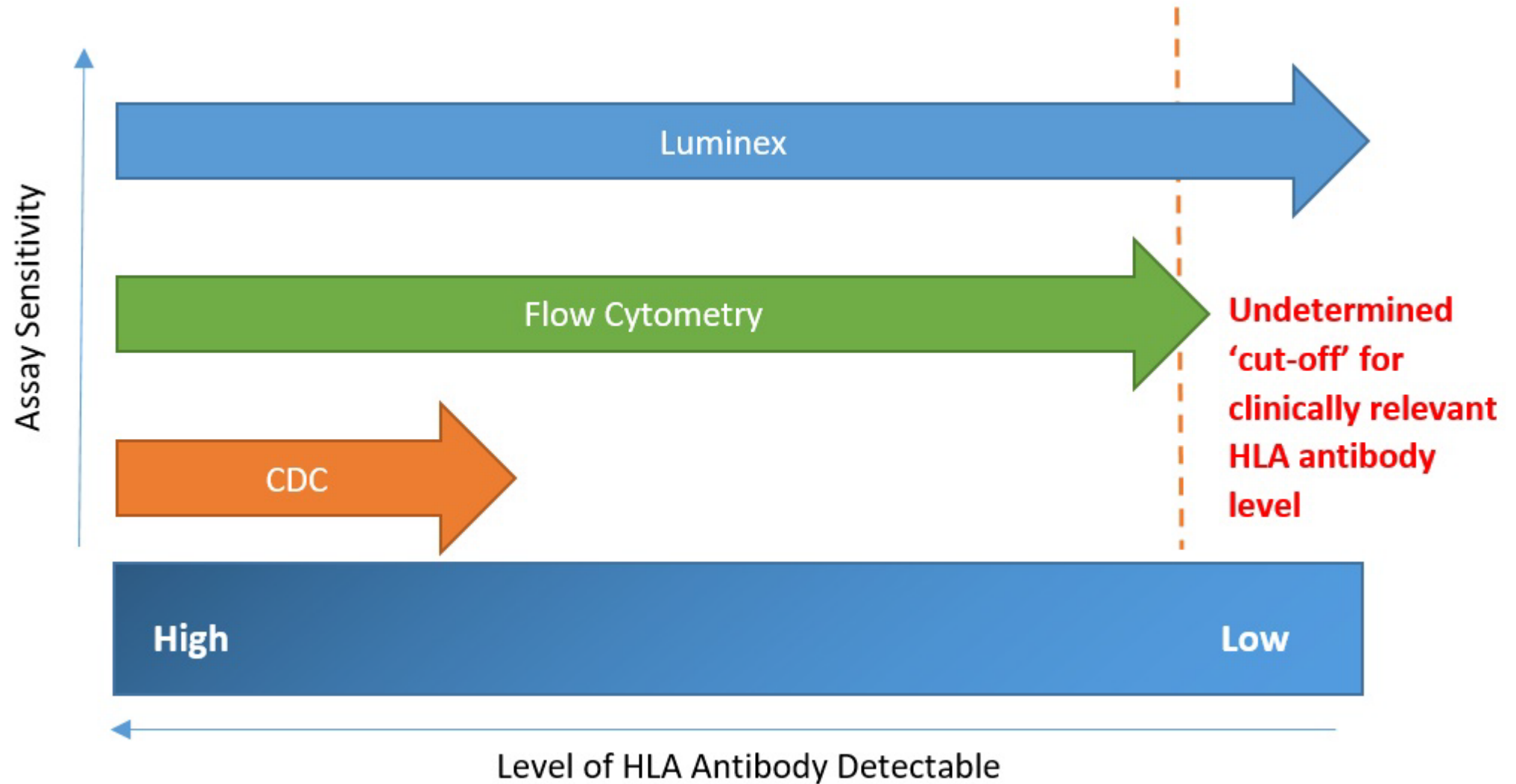
  2 techniques

  - CDC

    - detects "strong" antibodies which would cause instant rejection

  - Luminex

    - Sensitive technique

    - Detects "weak" and "strong" antibodies

    - "Weak" antibodies increase risk of transplant but it may still be possible to transplant

Complement-depen

Donor lymphocyte

Complement activation

Recipient serum

Cell lysis and death

**A** Recipient Serum — 100 beads. Each has a unique dye signature and a unique HLA antigen on its surface

May contain HLA antibodies

**B** HLA antibody in recipient serum binds to specific bead.

**C** Detection antibody (⊦) binds which then captures fluorescent reporter dye (☆)

**D** Dual beam laser. One laser detects bound reporter dye the other identifies the specific bead.

# Antibody Testing – Techniques and Sensitivity

# Why do we test for HLA antibodies

Define HLA antibodies and agree unacceptable antigen listing

Monitor every three months to check for increase or decrease

Check after sensitisation events e.g. transfusion, graft nephrectomy
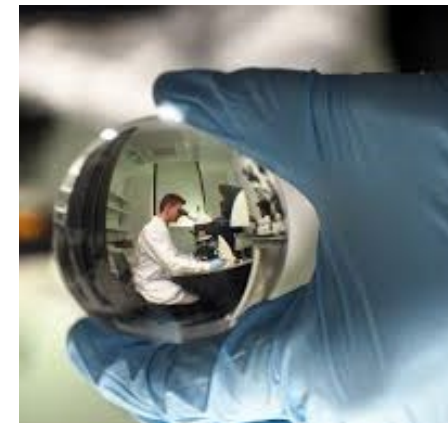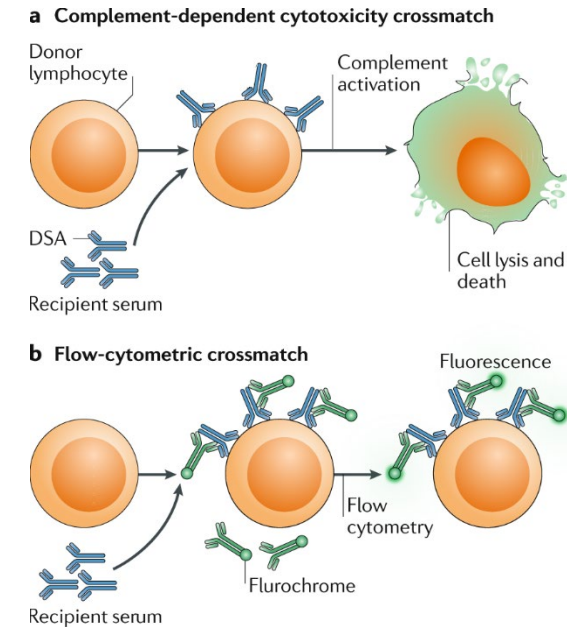
Prevent hyperacute rejection

Avoid <span style="color:red">unexpected</span> positive crossmatches

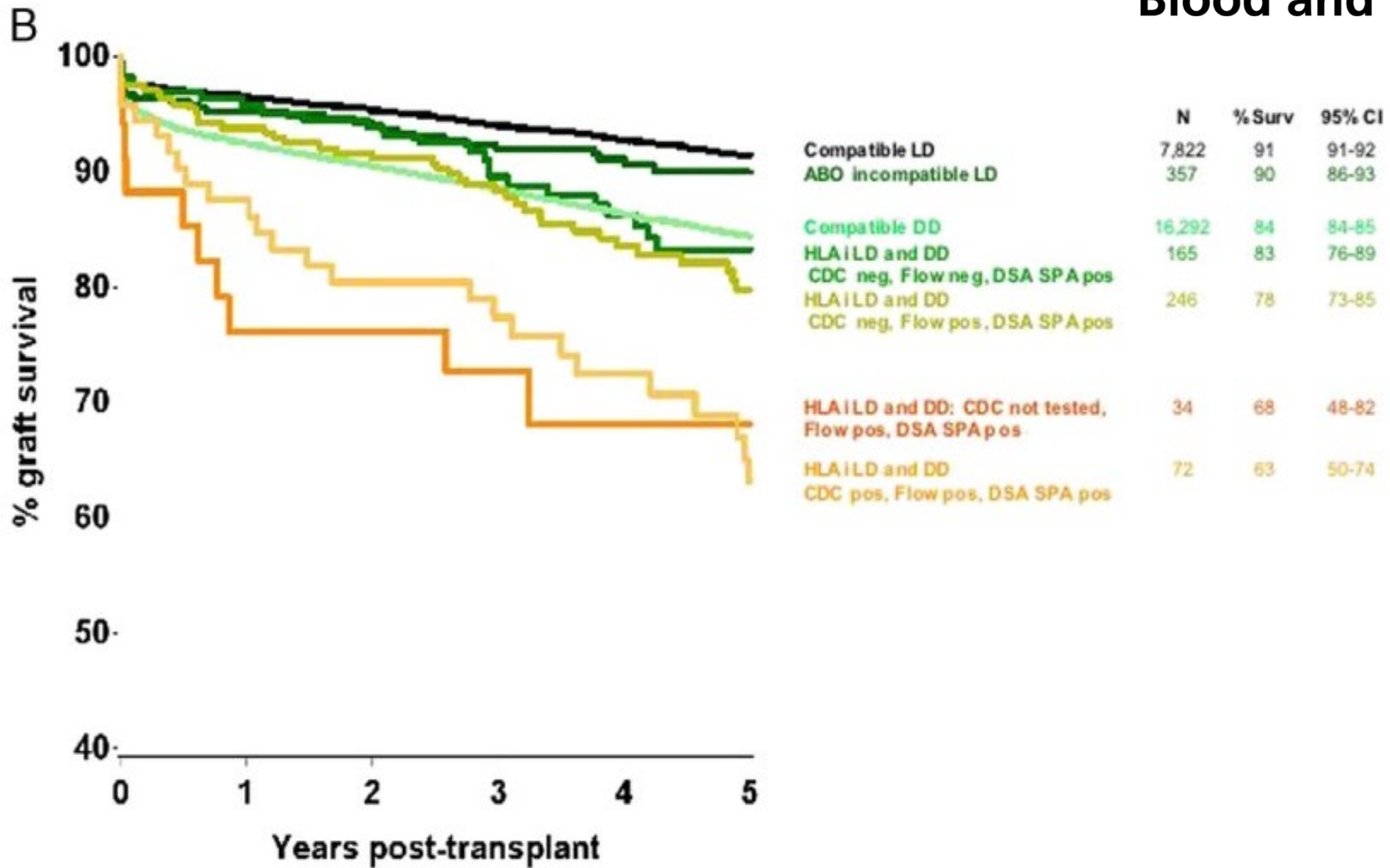Determine cRF (calculated reaction frequency) – predict likelihood of receiving transplant

# Laboratory Testing

**Blood and Transplant**

- Crossmatching
  - Specific test between a recipient and donor
    - Kidney and Pancreas patients
  - Used to determine risk of transplant
  - Decide whether the transplant can go ahead

- Virtual Crossmatch
  - Use donor HLA-type and recipient antibody profile to predict crossmatch result
  - Quicker results
  - Reduces cold ischaemic time
  - Reduces laboratory workload on-call

- Physical Laboratory Crossmatch
  - 2 different techniques
  - CDC XM
    - Detects strong antibodies
  - Flow Cytometry XM
    - Able to detect weak antibodies
    - Not HLA-specific



a Complement-dependent cytotoxicity crossmatch

Donor lymphocyte — Complement activation — Cell lysis and death

DSA

Recipient serum

b Flow-cytometric crossmatch

Fluorescence

Flow cytometry

Flurochrome

Recipient serum

| | N | %Surv | 95% CI |
|---|---|---|---|
| Compatible LD | 7,822 | 91 | 91-92 |
| ABO incompatible LD | 357 | 90 | 86-93 |
| Compatible DD | 16,292 | 84 | 84-85 |
| HLAi LD and DD CDC neg, Flow neg, DSA SPA pos | 165 | 83 | 76-89 |
| HLAi LD and DD CDC neg, Flow pos, DSA SPA pos | 246 | 78 | 73-85 |
| HLAi LD and DD: CDC not tested, Flow pos, DSA SPA pos | 34 | 68 | 48-82 |
| HLAi LD and DD CDC pos, Flow pos, DSA SPA pos | 72 | 63 | 50-74 |

# Assessing Risk

Immunological Assessment

Donor Factors

Transplant

Recipient Factors

Risk appetite

Dr Tracey Rees