Lab Cross matching test blood transfusion

Cross matching test is referring to the testing that is performed prior to a blood transfusion in order to determine if the donor's blood is compatible with the blood of a recipient, or to identify matches for organ transplants.

Purposes of this test:

The cross match is routinely used as the final step of pretransfusion compatibility testing. It serves two purposes:

(1) Serve as a final check of ABO compatibility between donor RBCs and patient plasma or serum

(2) To detect clinically significant antibodies that may have been missed by the antibody-screening test.

Types of test:

1- **Major**: using serum of patients with RBC of donor.
2- **Minor**: using RBC of patient with plasma of donor. But it is not use in routine test for all patients.

Materials

1. 5 ml of patient's blood: (2ml) with EDTA tube and (3ml) with plain tube for serum
2. 2 ml of donor blood or blood bottle
3. 4 cane tubes
4. bovine albumin
5. anti-human globulin
6. incubation
7. slides and microscope

1. Why do we use Bovine albumin?

Bovine albumin reduces the zeta potential (the negative surface charge) around red blood cells, allowing them to come closer together and enhancing agglutination if IgG antibodies are present.

2. Why do we use AHG (Anti-Human Globulin)?

IgG antibodies bind to red blood cells but usually do not cause visible agglutination. AHG binds to those IgG molecules attached to RBCs and causes clear, visible agglutination. Converts “invisible reactions” into visible agglutination.

3. How to prepare rbc suspention 5%? By take 2 ml of blood(donor and patient's), washing by normal saline 3 times with each wash must be centrifuge for 3 min finally Fill the tube with **2–3 mL of normal saline**, and you will practically obtain a **5% RBC suspension**.This method is commonly used in **daily routine laboratory practice**.”.

Why the prepare rbc suspention 5% is the essential step ? to remove plasma proteins, antibodies, and other interfering substances.

Procedure of cross matching:

1- Do blood grouping for patient and donor blood (or blood bottle).
2- Collection patient's serum.
3- Prepare RBC suspension (5%) by Taking 2 ml of donor and patient's blood, washing by normal saline.
4- Prepare 3 tubes, two (A, B) for matching, and the other (C) for patient.
5- Add 2 drops of patient's serum to all tubes.
6- Add 2 drops of donor RBC suspension to tube A & B.
7- Add 2 drops of patient's RBC suspension to tube C.
8- Add 3 drops of bovine albumin to all tubes.
9- Keep tube A at room temperature for 45 min : to detect cold-reactive antibodies, such as: Anti-I , Anti-H and Anti-M)These antibodies react best at room temperature (20–25°C) or even lower.
10-Incubate tube B & C at 37c for 30 min.: is the main tube that determines true compatibility between donor RBCs and the patient's serum. At 37°C, we detect: Clinically significant antibodies, mainly IgG (e.g., anti-D, anti-Kell, anti-c) these antibodies only react at body temperature, making this tube the most important.
10- After incubation, washing tubes three times by normal saline.
11- Add 2 drops of anti-human globulin to all tubes and putting it in centrifuge at 1000rpm for 30 sec.

12- Then reading the result:

- Presence agglutination in tube B means incompatible
- Non agglutination in tube B means compatible
- Agglutination in tube **A** only refers to presence cold Ab in patient's blood and the blood can transferred to patient only after warmed to 37c.
- Agglutination in tube **C** means that patient infected with **Autoimmune Hemolytic Anemia** the blood can transferred to patient if tube B is –ve.

Note:

- ✓ Emergency case : Give O -ve RBCs if no time for typing.
- ✓ Universal donor: O -ve → no A/B antigens, safe for all.
- ✓ Universal recipient: AB → no anti-A/B antibodies, can receive any type.
- ✓ Preferred: Use type-specific RBCs if time allows.
- ✓ Crosshatch continues even after emergency transfusion starts.

Cross match procedure

Collect 5 ml of patient blood

2ml of donor blood

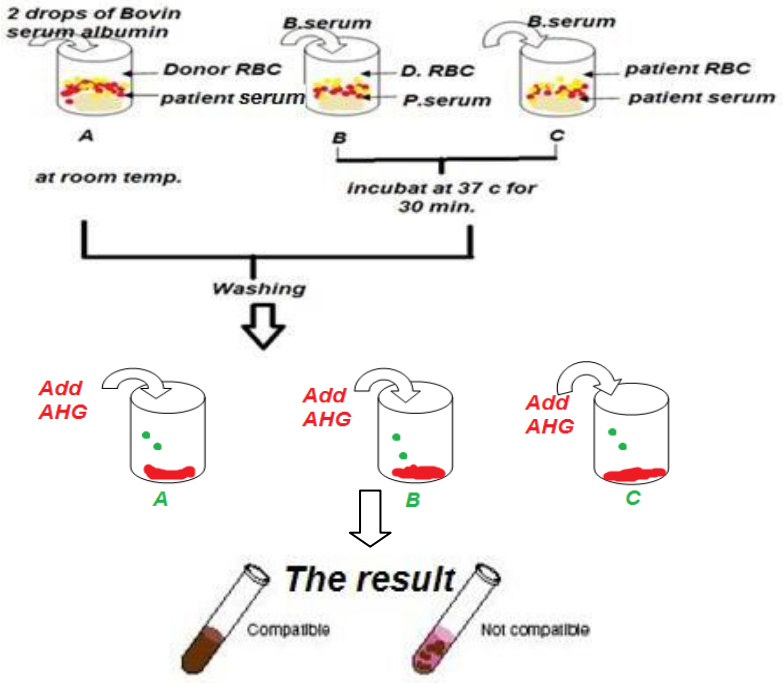
For Prepare 5% RBC as:

3ml for serum collection

2ml in EDTA tube for RBC collection.

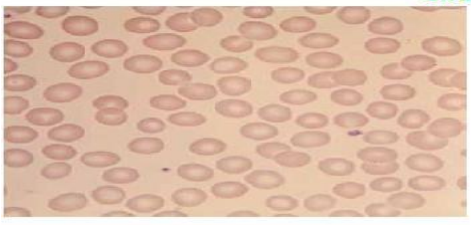
Washing RBC

Prepare 5% RBC suspension (50µl RBC+950µl normal saline)

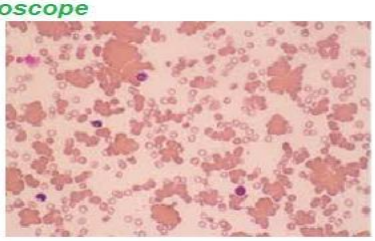


The result of tube

Under microscope



compatible - non agglutination



Non compatible - Agglutination

Types of Anticoagulant/preservative solutions for blood storage:

In blood banking, different anticoagulants are used to preserve whole blood and its components (RBCs, plasma, platelets) during storage. The components of these solution mainly are

- Citrate : chelates calcium thus prevents clotting.
- Dextrose : provides energy to RBCs.
- Adenine : helps maintain ATP levels in RBCs.
- Phosphate : Buffers which also provides phosphate source for metabolism

1. CPDA-1 (Citrate-Phosphate-Dextrose-Adenine) : Used for Whole blood and RBCs storage and Can preserve RBCs up to 35 days at 2–6°C.
2. CPD (Citrate-Phosphate-Dextrose): Used for Whole blood collection. And Supports RBC viability up to 21 days.
3. ACD (Acid-Citrate-Dextrose) :Used for Platelet-rich plasma collection. Used in apheresis. And can RBC preservation for short-term storage (up to 14 days). Summary table below

Anticoagulant / Additive	Storage Use	Shelf Life
CPDA-1	Whole blood / RBCs	35 days
CPD	Whole blood / RBCs	21 days
ACD	Platelets, short-term RBCs	14 days

4. SAG-M (Saline, Adenine, Glucose, Mannitol)

- Saline: Maintains isotonic environment.
- Adenine: Supports ATP synthesis in RBCs.
- Glucose: Energy source for RBC metabolism.
- Mannitol: Protects RBCs from hemolysis by stabilizing the membrane. Mannitol reduces hemolysis during storage and handling, improving post-transfusion survival of RBCs.

    - Use: Added to RBCs after removal of plasma (usually from CPD/CPDA-1 collection) to extend storage life.Shelf-life: RBCs in SAG-M can be stored up to 42 days at 2–6°C.

5. ADSOL (Additive Solution: Adenine, Dextrose, Sorbitol, Sodium Chloride, Mannitol)

Components & Function: Provides excellent preservation of RBC viability and function, suitable for long-term storage up to 6 weeks.

- Adenine: ATP synthesis for RBC energy.
- Dextrose (Glucose): Energy source.
- Sorbitol: Protects RBC membrane, prevents hemolysis.
- Sodium chloride: Maintains isotonicity.
- Mannitol: Further prevents hemolysis and stabilizes RBCs.

- Use: Added to RBC units after removal of plasma to significantly extend shelf life.
- Shelf-life: RBCs in ADSOL can be stored up to 42 days at 2–6°C.
- Special Note:

Summary Table:

Solution	**Components**	**RBC Shelf-life**
SAG-M	Saline, Adenine, Glucose, Mannitol	42 days
ADSOL	Adenine, Dextrose, Sorbitol, NaCl, Mannitol	42 days