

STANDARD OPERATING PROCEDURE

(Name of the Blood Centre)

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LOCATION	SUBJECT
Red Cell Serology Laboratory	Antiglobulin Cross-match
FUNCTION	DISTRIBUTION
Detection of incompatibilities caused by warm complete antibodies	- Supervisor in charge of Red Cell Serology Laboratory - Master File

1. SCOPE & APPLICATION

This procedure applies to compatibility testing of all multi-transfused patients and transfusion recipients who currently demonstrate or have a history of clinically significant antibodies.

2. RESPONSIBILITY

It is the responsibility of the technician in the cross match facility of the red cell serology laboratory to perform the anti-globulin cross match using quality controlled reagents and proper cell concentrations. One technician performs the tests and another checks it. If any unexpected blood group antibody is detected, inform the staff of Advanced Red Cell Serology to carry out further investigations.

3. REFERENCES

1. Technical Manual of the American Association of Blood Banks 13th Edition, 1999. Pages 380-381, 383-384, 258-262, 668-671.
2. Procedures in Blood Banking and Immunohaematology; H.M. Bhatia, 1977. Pages 72-75.
3. Introduction to Transfusion Medicine Zarin Bharucha & D.M. Chouhan, 1st Edition, 1990. Pages 69-71, 74-76, 85.

4. MATERIAL REQUIRED

4.1 Equipment:

- Refrigerator to store samples & reagents at 2- 6°C.
- Table top centrifuge.
- Automated Cell Washer.

- Microscope.
- Dri bath.

4.2 Specimen:

- Clotted blood sample of patient.
- Segment from donor unit.
- Donor red cells suspended in saline.

4.3 Reagents:

- 22% bovine albumin.
- Antihuman globulin reagent(anti-IgG+anti-C3d).
- IgG sensitised control cells.
- 0.9% Saline.
- Distilled water.

4.4 Glassware:

- Serum tubes.
- Coombs' tubes.
- Pasteur pipettes.
- Glass slides.

4.5 Miscellaneous:

- Rubber teats.
- Disposal box.
- 2 plastic beakers.
- Aluminium racks to hold serum and coombs' tubes.

5. PROCEDURE

5.1 Principle:

The cross match through the anti-globulin phase permits detection of clinically significant incompatibilities caused by incomplete antibodies that sensitise cells at 37°C, but do not directly cause agglutination.

5.2 Anti-Globulin Cross-Match:

1. Label tube with patient/unit and test identification.
2. Add two drops of patient serum to each tube.
3. Prepare a 5% cell suspension in saline from each donor unit segment. (Sp015).
4. Add 1 drop of donor's 5% red cell suspension to the tube.
5. Add 2 drops of 22% bovine albumin and mix well.
6. Incubate at 37°C for minimum 15 minutes. (Follow manufacturer's directions when using commercial reagents).
7. Wash the cells a minimum of 3 times with saline. Decant completely after last

- wash. (washing can be done manually or in automated cell washer).
8. Add two drops of antihuman globulin reagent to the dry cell button.
 9. Mix well and centrifuge at 1000 rpm for 1 minute.
 10. Resuspend and read for agglutination. Grade and record test results immediately.
 11. To all negative antiglobulin tests add 1 drop of IgG-sensitised control cells. Centrifuge, resuspend and read for agglutination. Grade and record test results. After the addition of IgG-sensitised control cells to a negative test, the presence of agglutination indicates that the AHG serum added was capable of reacting and that the negative antiglobulin test is valid.

5.3 Interpretation:

1. Hemolysis or agglutination indicates the presence of a serologically incompatible cross-match. This result is interpreted as **Incompatible**.
2. Absence of agglutination and hemolysis is a negative test result and indicates a serologically compatible crossmatch. This result is interpreted as **Compatible**.

If the IgG-sensitised control cells added to confirm the activity of the polyspecific reagent show only weak or no agglutination the test is invalid and must be repeated.

5.4 Limitations:

The anti-globulin cross- match will not:

1. Detect error in Rh typing.
2. Prevent isoimmunisation of the recipient.
3. Ensure normal red blood cell survival.
4. Detect some weakly reactive antibodies.

6. DOCUMENTATION

Enter all results on the transfusion record card and OT/Ward transfusion register. Enter only the results of compatible units in the blood compatibility form. The technician who performed the test and the one who checked the results sign all records.

7. END OF DOCUMENT