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Resolution of ABO/Rh Discrepancies Caused by Cold Reacting Antibodies - Blood Bank

Document Type: Procedure

I. PURPOSE AND OBJECTIVE:

This document will provide the Blood Bank staff with guidance for the resolution of ABO/Rh discrepancies caused by cold reacting antibodies.

II. CLINICAL SIGNIFICANCE:

- A. Cold reacting antibodies can cause unexpected agglutination in the forward or Rh typing, or in the reverse typing. When observed in the forward typing, this agglutination may often be dispersed by washing the patient's cells multiple times with saline, or with warm saline if necessary.
- B. If unexpected agglutination is observed in the reverse typing, a tube panel will be performed to determine whether the unexpected antibody is a cold reacting antibody and to attempt to identify the specificity. If a specificity is identified in the tube panel, then the reverse typing will be performed with reverse cells that are negative for the antigen corresponding to the cold reacting antibody that was identified. See policies Reverse Typing Patients with Identified Cold Reacting Antibodies and Obtaining Antigen Negative Reverse Cells listed below.
- C. Note that in some cases, it may not be possible to identify the unexpected antibody or to resolve the ABO/Rh discrepancy.

III. DEFINITIONS:

A. ABO discrepancy: Generic term for a variety of situations in which the interpretation of a patient or donor's ABO grouping results is unclear.

- B. Rh discrepancy: Generic term for a variety of situations in which the interpretation of a patient or donor's Rh results are unclear.
- C. GND (Group not determined): If the forward typing and the reverse typing do not agree, an ABO discrepancy has occurred. If the discrepancy cannot be resolved at that time, then the patient's group is considered "Not Determined".
- D. RND (Rh not determined): If the Rh typing discrepancy of a patient cannot be resolved at that time, then the patient's Rh type is considered "Not Determined".
- E. Mixed field (MF): A sample that contains two (2) distinct populations of red cells, usually as a result of recent RBC transfusions of a dissimilar ABO or Rh type as the patient. Mixed-field reaction results when one population is agglutinated in testing, while the other population is not. Referred to as "dual population" (dp) by the VisionTM.
- F. Rouleaux: Red cells that assume a stacked-coin formation in testing due to an abnormality with the patient's serum protein. Rouleaux is most readily observed on microscopic examination.
- G. Hemolytic Disease of the Newborn (HDN): Also called erythroblastosis fetalis, is a blood disorder that occurs when the blood types of a mother and baby are incompatible.
- H. Unexpected antibody: Any antibody (other than naturally occurring anti-A or anti-B that is regularly found in normal serum or plasma) that is currently or was historically present in a patient's sample.
- Clinically significant antibody: An antibody that is known to cause Hemolytic Disease of the Newborn (HDN) or shortened survival of antigen positive RBCs, and requires transfusion of antigen negative red cells, and is usually IgG and best detectable with antihuman globulin (AHG).
- J. Clinically insignificant antibody: An antibody that does not cause shortened red cell survival of antigen positive RBCs, does not require transfusion of antigen negative red cells, and is usually IgM and reacts best below 37°C. Antibodies that are usually considered clinically insignificant include anti-IH, anti-H, auto-anti-I, anti-Le^a, anti-Le^b, anti-P₁, anti-M, anti-N, and anti-A₁.
- K. WBIT: Wrong blood in tube collection error is when a sample is drawn from the wrong patient, so that the identifying information of the patient on the label does not correlate with the patient from whom the blood in the tube was drawn.
- L. Internal Variance: Report made internally in the Blood Bank for documentation of an incident such as error detected, accident, complaint, unplanned deviation, or incident for review, evaluation, investigation, and correction.
- M. QSR (Quality Safety Report): Report made in the hospital incident reporting system (i.e. RL Solutions) regarding any process/incident inconsistent with the routine operation of the hospital or the routine care of patients in any setting. This includes errors that result in actual or potential injury to a patient or visitor, including near misses or unsafe conditions.
- N. Designee: Any Blood Bank technical director, or transfusion medicine fellow.

IV. POLICIES:

A. Frequency and Purpose of Tube Antibody Panels

1. Frequency:

a. If unexpected antibody reactivity is observed in the <u>tube</u> reverse typing, a <u>tube</u> panel will be performed every 90 days. The reaction strength (of the unexpected reactivity in the reverse typing) will not be considered when determining the frequency at which to perform tube panels to resolve ABO discrepancies.

2. Purpose:

- a. To determine whether the unexpected reactivity in the reverse type is due to a cold reacting antibody. If the patient has a history of a cold reacting antibody of known specificity, then 3 test cells that are positive for the corresponding antigen should be included in the tube panel; this will help prove whether the unexpected reactivity in the reverse type is due to the previously identified cold reacting antibody.
- b. To attempt to identify an antibody specificity.

B. Reverse Typing Patients with Identified Cold Reacting Antibodies

- 1. If specificity is identified in the tube panel, then the following apply:
 - a. If anti-M or anti-P₁ is identified, then the reverse typing will be performed with A and B reverse cells that are negative for the corresponding antigen.
 - b. If anti-I is identified, then the reverse typing will be performed with reverse cells from group A and B neonatal samples. Neonatal red cells are I negative; the I antigen is not well-formed at birth. Note that cord blood samples may also be used.
 - c. If anti-A₁ is suspected, refer to Transfusion Medicine policy, Resolution of ABO Discrepancies for A Subgroups and Patients with Anti-A₁. In this case the A reverse typing will be performed using an A₂ test cell.

C. Obtaining Antigen Negative Reverse Cells

- Reverse typing cells that are negative for the antigen corresponding to the cold reacting antibody that was identified may potentially be obtained from several different sources.
 - a. Manufacturers preferentially select M-negative donors for their pooled reverse cells but do not guarantee that the reverse cells will be M negative; therefore the Blood Bank must type each lot of these reverse cells for the M antigen as needed.
 - b. Neonatal heel stick samples that have been previously ABO/Rh typed are the best source of I-negative reverse cells.
 - c. In some cases, it may be necessary for the Blood Bank to antigen type donor units in order to find suitable antigen negative reverse cells.
 - d. Antigen typings of donor units shall be performed as described in Transfusion

- Medicine policy, Antigen Typing.
- e. Antigen negative reverse cells may be obtained from a patient sample that was previously antigen typed. For example: A patient has a history of anti-M and was previously typed as M-negative. If the patient is group A or B and has not recently been transfused, the patient's cells may be used as a source of M-negative reverse cells.
- Once a donor unit is found to be antigen negative, several segments can be saved in the reagent refrigerator. The segments should be labeled with the donor number and expiration date. These segments may be used to reverse type patient samples until the original expiration date of the unit.
- 3. The Log of M and P_1 Negative Cells used for Reverse Typing may be used to document M and P_1 antigen typing results of tests cells that may be suitable for reserve typing.

D. Crossmatching Red Blood Cells for Patients with Cold Reacting Antibodies

Antibodies that react only at temperatures less than 37°C are usually considered clinically insignificant. Refer to Transfusion Medicine policy, *Policies for Providing Red Blood Cells to Patients with Unexpected Antibodies*.

E. Prewarm Technique for Reverse Typing

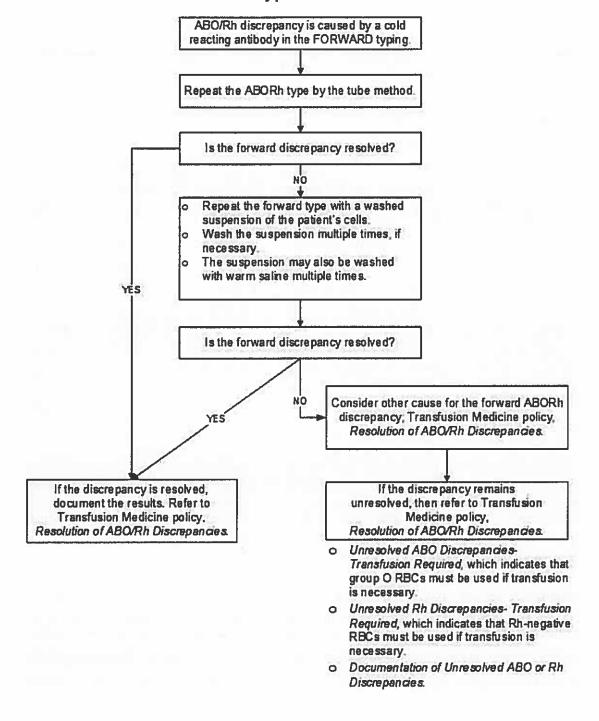
- As indicated in Transfusion Medicine policy, Prewarm Technique, the reverse typing may be performed using the prewarm technique only if specifically directed by procedure or the Medical Director or designee.
- 2. As described throughout this document, the reverse typing may be performed by the prewarm technique only if:
 - a. A tube panel has been performed, if indicated and
 - b. A cold reacting antibody with no specificity was detected.
 - c. If anti-M, anti-P, anti-I, or anti-A₁ is identified, then a prewarmed reverse should not be performed. The reverse typing should be performed with antigen negative reverse cells.

F. Saline Replacement for Reverse Typing

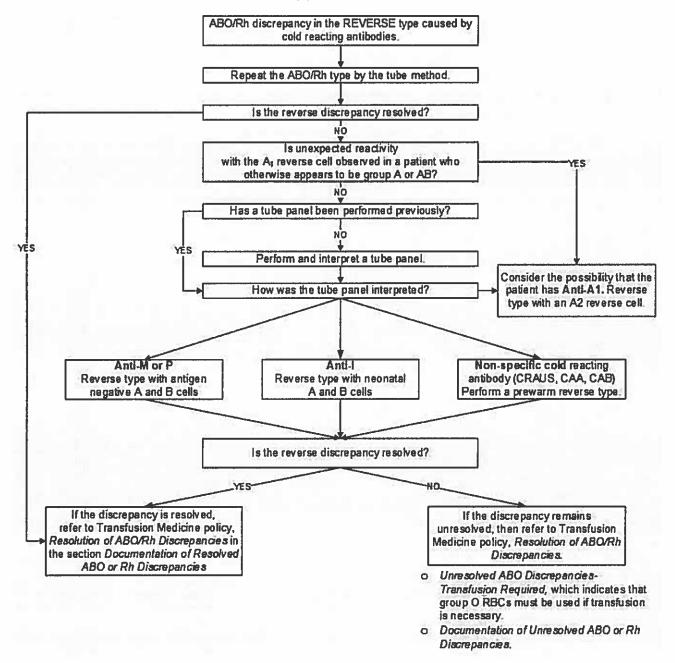
 Rouleaux may cause extra reactivity in forward and reverse typings, if this is suspected, refer to Transfusion Medicine policy, Compatibility Testing for Patients with Rouleaux – Saline Replacement Technique.

V. PROCEDURE:

A. Resolution of ABO/Rh Discrepancies Caused by Cold Reacting Antibodies in the Forward Type



B. Resolution of ABO/Rh Discrepancies Caused by Cold Reacting Antibodies in the Reverse Type



VI. INTERPRETATIONS:

A. Adding Comments to the ABO/Rh Test

1. Observations of all test results must be recorded properly at the time the test is performed. Therefore, when an ABO or Rh discrepancy is observed, all results (including discrepant results) must be documented at the time the test is performed. The ABO/Rh canned message may be used for this purpose. For example:

- a. If anti-I was detected, the Note field may be documented with the MRNs of the neonates from whom the I-negative A and B reverse cells were obtained. The Method field may be documented as "Reverse with I-negative neonatal cells".
- b. If anti-M was detected, the Note field may be documented with the donor numbers of the A and B units used for the reverse type. The Method field may be documented as "Reverse with M-negative reverse cells."
- 2. The results should not be interpreted until the investigation is complete.
- 3. If rouleaux is identified microscopically, this must be documented as an internal comment under the ABO/Rh test prior to performing a saline replacement.

VII. NOTES:

- A. If a tube panel is non-reactive at the immediate-spin phase, then it may be helpful to incubate the test tubes at room temperature and/or 4°C to enhance the reactivity of the unexpected antibody, which may aid in its identification.
- B. Some cold reacting antibodies may appear as mixed-field reactivity in gel method.
- C. Anti-I is a common antibody present in the serum of normal healthy individuals, therefore it may be helpful to include at least one test cell that is negative for the I antigen in the tube panel. Stronger examples of anti-I may react at room temperature; however, anti-I is best identified at 4°C.
 - 1. Anti-I may be identified by its reactivity with most adult cells, and its non-reactivity with most neonatal cells as the I antigen is not well formed at birth.

VIII. REFERENCES:

- 1. AABB, Technical Manual, current edition.
- 2. AABB, Standards for Blood Banks and Transfusion Services, current edition.
- 3. College of American Pathologists, Transfusion Medicine Checklist, current edition.

Attachments

Log of M and P1 Negative Cells used for Reverse Typing

Approval Signatures

Step Description Approver Date

Kelly Sartor: Supv, Laboratory Pending

Beaumont Laboratory

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Typing
Reverse
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Cells
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Log

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Exp. Date of Test		:							
	Test Cell ID • Mfg / lot# / cell# • donor unit								
	Tech/ Date						:		

Refer to Transfusion Medicine procedure, Resolution of ABO/Rh Discrepancies Caused by Cold Reacting Antibodies. Save segments of M or P1 negative units in the appropriate designated rack/container for future retrieval.

page 1 of 1