Beaumont Laboratory
Royal Oak

Effective Date:
Supersedes: 03/30/2020
Related Documents: P060, P061, P101, P120, P122, P205, P340, P515, P605, P606, P623, P623F

SEROLOGIC CONFIRMATION OF ABO GROUP AND Rh(D) TYPE

RC.BB.CT.PR.106.r03.02.00

Purpose

The purpose of this document is to provide the Blood Bank staff with stepwise instructions for confirming the ABO/Rh of Blood Bank specimens by forward typing.

Principle

Serologic confirmation is meant to provide an additional verification of the ABO and Rh(D) of a specimen, as an additional safety measure. Serologic confirmation consists of a forward typing ABO determination and Rh(D) typing; a reverse typing is not performed.

Scope

Indications for Serologic Confirmation of the ABO and Rh(D) type:

- Patient samples, before crossmatching RBCs during computer downtimes.
- Segments from red blood cell (RBC) units that have been washed or deglycerolized.
- Deceased organ donor proxy samples, before transplantation of the corresponding organ.

The scope of this document relating to manual operations is limited to serologic confirmation of the ABO and Rh type. See P120, *Manual Operations*, for additional information.

Serologic confirmation is not required on neonatal patients during a computer downtime as long as group O RBCs are selected for transfusion. If non-group O RBCs are selected:

- Serologic confirmation on the neonatal sample is required.
- Refer also to P515, *Policies for the Selection of Blood Components for Neonatal Transfusion* / to *Policies for Non-Group O Neonates Receiving Non-Group O RBCs.*

Policies

The following policies apply to serological confirmation of the ABO group and Rh(D) type:

- As with all manual tests, batch testing must be limited to 6 tests per batch. If workload becomes excessive, supervisory staff must be notified immediately.

- Pre-transfusion samples must meet the labeling requirements found in P101, *Triaging and Identifying Acceptable Samples for Testing*.

- A confirmatory typing, as described in Table 106-1, should be performed on RBC units that have been washed or deglycerolized if they were not previously confirmed already (i.e. the RBC was received already frozen, and a confirmatory type was impossible until the RBC unit has been deglycerolized), or if multiple RBC units were washed or deglycerolized at the same time. Refer to P205, *Washing and Deglycerolizing Red Blood Cells Using the COBE 2991 Blood Cell Processor*.

- A confirmatory typing must be performed during computer downtimes on every patient sample that is crossmatched. Serologic confirmation must be performed each time that a sample is retrieved from storage for crossmatching during downtime.

- A forward ABO/Rh confirmatory type shall be performed when a deceased organ donor proxy sample is received in the Blood Bank. This is done internally within the Blood Bank, and is not a test of record.

- During computer downtimes, routine ABO and Rh(D) typing (which includes a reverse typing) of all samples must be performed according to Standard Operating Procedures This routine testing must be performed before confirmatory testing is performed.

Policies Relating to ABO and Rh(D) Discrepancies

- Any ABO grouping discrepancy that may be encountered during routine ABO/Rh testing must be resolved before confirmatory testing is performed. If necessary, refer to P623, *Resolution of ABO and Rh(D) Discrepancies*.

- **The following ABO and Rh(D) results must be in agreement when testing patient samples**:
  - ABO/Rh confirmatory testing results (Table 106-1), and
  - Routine ABO and Rh(D) typing results from current sample, and
  - Historical ABO/Rh results, if available.

  If these results are not in agreement, do not crossmatch non-group O RBCs until the discrepancy is resolved. Refer to P623, *Resolution of ABO and Rh(D) Discrepancies*.

- **The following ABO and Rh(D) results must be in agreement when testing segments from washed or deglycerolized RBC units:**
  - The ABO/Rh confirmatory results (Table 106-1), and
  - Historical type of the RBC unit, and
  - The ABO and Rh(D) indicated on the washed or deglycerolized RBC unit face label.

  If these results are not in agreement, do not proceed; do not release the RBC unit to a patient. Consult a manager/supervisor.

- **The following ABO and Rh(D) results must be in agreement when testing a deceased organ donor proxy sample:**
  - ABO/Rh confirmatory results of deceased organ donor proxy sample (Table 106-1), and
  - ABO and Rh(D) of donor records.
- Mixed field reactions causing ABO/Rh discrepancies may exist in the forward typing due to recent transfusions to the donor. These discrepancies may or may not be compatible with the recipient.
  - If an explained discrepancy exists and is compatible with the organ recipient, then
    - Proceed with the Transplant Recipient organ ABO verification
    - Contact tissue coordinator for are any questions regarding the compatibility with the recipient.

- If an unexplained discrepancy exists that is not compatible with the organ recipient, then immediately
  - Repeat the confirmatory typing
    - A reverse typing may be performed on the sample to help investigate the discrepancy if it exists in the ABO.
  - Contact the tissue coordinator
  - Notify the transplant physician
  - Notify inner control
  - Notify Gift of Life
  - Notify Buckeye

Rh(D) Typing Policies

- The Ortho BioClone Anti-D reagent will be used for confirmatory testing. An Rh(D) control is not required when performing serologic testing with the Ortho BioClone Anti-D reagent. However, if a confirmatory type appears to be AB positive, a 6 – 8% bovine albumin control must be tested and must be non-reactive. This control is prepared as described in P122, *Preparing the 6 – 8% Bovine Serum Albumin Control*.

- Valid Rh(D) results must have a graded reaction of 0 or 2-4+ when tested with the Ortho BioClone Anti-D reagent.

Weak D Testing Policies

It is generally not required to perform weak D testing as part of serologic confirmatory testing. However, confirmatory test results must be in agreement with historical test results, as indicated above. In some cases, weak D testing may be required to resolve a Rh(D) Discrepancy. If necessary, refer to ~~P107, *Testing for Weak D* and~~ P623F*, Resolution of Rh(D) Discrepancies.*

ABO Typing Policy

Valid forward ABO results must have a graded reaction of 0 or 3-4+.

Definitions

- **Neonates:** patients from birth through 4 months old.

- ~~**BBCDM:** the Blood Bank Computer Documentation Manual.~~

- **HIS:** Hospital Information Services; the hospital-wide computer system.

- **Routine ABO and Rh(D) testing:** as used in this document, refers to the ABO/Rh testing described throughout the SOPs, and includes both a forward and reverse ABO typing.

Specimen Collection and Handling

- The preferred specimen when confirming the ABO/Rh during computer downtimes is a 6ml EDTA sample with affixed identifying label. See P101, *Triaging and Identifying Acceptable Samples for Testing,* for acceptable alternatives.
- A RBC unit segment will be tested when confirming the ABO/Rh of a washed or deglycerolized RBC unit.

- A deceased organ donor proxy sample will be tested when confirming the ABO/Rh of an organ donation

Reagents, Equipment, and Supplies

- table top centrifuge
- lighted viewing mirror
- Commercial reagents:
  - Ortho Anti-A, colored blue
  - Ortho Anti-B, colored yellow
  - Ortho BioClone Anti-D
- Disposable pipettes
- 10 x 75 mm test tubes

Forms

- F-120a, *Patient Downtime Worksheet*
- F-920, *Organ Transplant Notification*

Procedure

1. Ensure the patient sample or RBC segment is properly labeled.
   - **a.** If a patient sample is being tested, refer to P101, *Triaging and Identifying Acceptable Samples for Testing* for proper labeling requirements.
   - **b.** If a donor segment from a washed or deglycerolized RBC unit is being tested, it must be labeled with the donor unit number.
   - **c.** If an organ proxy sample from a deceased organ donor is being tested, the sample should be labeled with the complete UNOS identification number, date and time drawn, and initials.

Table 106-1: **Confirmatory Testing**

<table>
<tr><th>Step</th><th colspan="2">Action</th><th>Notes</th></tr>
<tr><td rowspan="3">1</td><td colspan="2">Ensure that the patient sample, RBC segment, or deceased organ donor proxy sample is labeled properly.</td><td rowspan="2">Investigate and correct any discrepancy before proceeding.</td></tr>
<tr><td>If the specimen is a:</td><td>Then specimen must be labeled with:</td></tr>
<tr><td>Pre-transfusion sample</td><td>• Medical record number
• Name (spelled correctly)
• Wristband #
• Collection date
• Phlebotomist ID</td><td>• If necessary, refer to P101, Triaging and Identifying Acceptable Samples for Testing.
• Only the bolded information is required to be on the specimen; however, any additional information on the specimen must be accurate.
• The collection date and phlebotomist identification can ~~must~~ be on the sample label or in the computer.</td></tr>
</table>

	Deceased organ donor proxy sample	Complete UNOS identification number, date, time drawn, and initials.	
	Donor segment from a washed or deglycerolized RBC unit	Donor unit number.	
2	Proceed as follows:		
	If the specimen being tested is a:	**Then:**	
	Pre-transfusion sample	Document the technologist's initials, date, time of testing, and patient's name and MRN on F-120a, *Patient Downtime Worksheet*.	The HIS accession label with identifying information (from Step 1, above) should have been placed on this form by triage if one is available.
	Deceased organ donor proxy sample	Document the technologist's initials and date / time of testing on the F-920, *Organ Transplant Notification* that was created for the organ transplant.	This F-920, *Organ Transplant Notification* will also include other information pertaining to the organ transplant such as patient information and the donor UNOS identification number.
	Donor segment from a washed or deglycerolized RBC unit	The confirmatory type will be ordered and resulted in the Blood Bank computer.	The test code is RTDP for Rh(D) positive RBCs and RTDN for Rh(D) negative RBCs.
3	Label four (4) 10 x 75 mm tubes with patient's last name, UNOS number, or donor number. In addition, label tubes as follows:		
	Tube #1 "3%"		For the test RBC suspension.
	Tube #2 "A"		
	Tube #3 "B"		
	Tube #4 "D"		
Step	**Action**		**Notes**
4	Make a 2 – 4% suspension of test sample RBCs in the tube labeled with only the patient's test identification.		Refer to P060, *Making a Test Red Cell Suspension*.
5	Combine 1 drop of the appropriate reagent typing sera and 1 drop of the test RBC suspension to each of the remaining tubes. For example, combine 1 drop of Anti-A and 1 drop of the patient's RBC suspension to the "A" tube.		Typing sera must be added before the RBC suspension. Typing sera should be added just prior to testing.
6	Agitate all tubes to mix. Centrifuge tubes according to calibrated time.		See P340, Calibration of Serologic Centrifuges.
7	Gently re-suspend the cell buttons. Read and grade test reactions.		See P061, *Reading, Grading, and Recording Test Reactions.*
8	Read, grade, and record results.		Results are documented as described in step 2.
9	Interpret the ABO and Rh(D) test results.		See Table 106-2 and following *Interpretation Notes*.

<table>
<tr><td>10</td><td colspan="2">Verify that the confirmatory testing interpretation is in agreement with historical results as follows:</td><td>• See Table 106-2 and following Interpretation Notes.
• Investigate and correct any discrepancy before proceeding.</td></tr>
<tr><td rowspan="4">11</td><td colspan="2">Permanently record the results in the appropriate location.</td><td rowspan="2"></td></tr>
<tr><td>Specimen</td><td>Proceed as follows:</td></tr>
<tr><td>Pre-transfusion sample</td><td>Enter results that were recorded on F-120a into the Blood Bank computer when it becomes available.</td><td></td></tr>
<tr><td>Deceased organ donor proxy sample</td><td>Submit the results on F-920, Organ Transplant Notification to the Tissue Coordinator.</td><td></td></tr>
<tr><td>Donor segment from a washed or deglycerolized RBC unit</td><td>Document the confirmatory ABO/Rh type in the Blood Bank computer system.</td><td></td></tr>
</table>

Interpretation

Table 106-2: Confirmatory Testing Interpretation

If the test results are:			**Then the interpretation is:**
Anti-A	**Anti-B**	**Anti-D**	
3-4+	0	2-4+	A pos
0	3-4+	2-4+	B pos
3-4+	3-4+	2-4+	AB pos
0	0	2-4+	O pos
3-4+	0	0	A neg
0	3-4+	0	B neg
3-4+	3-4+	0	AB neg
0	0	0	O neg

Interpretation Notes:

Before interpretation of test results, correct and investigate any ABO/Rh discrepancies. If necessary, refer to P623, *Resolution of ABO and Rh(D) Discrepancies*. ABO/Rh discrepancies include the following:

- Confirmatory testing results do not correspond to an appropriate interpretation found in Table 106-2
- Confirmatory test results are not valid, graded reactions; see the *Policies* section.
- As described in the *Policies* section, if the following test results are not in agreement **when testing patient samples:**
  - ABO/Rh confirmatory testing results (Table 106-1),
  - Routine ABO and Rh(D) typing results from current sample,
  - Historical ABO/Rh results, if available.
- As described in the *Policies* section, if the following test results are not in agreement **when testing segments from washed or deglycerolized RBC units:**

  - ABO/Rh confirmatory results (Table 106-1),
  - Historical type of the washed or frozen RBC unit,
  - The ABO and Rh indicated on the washed or deglycerolized RBC unit face label.
- As described in the *Policies* section, if the following test results are not in agreement **when testing deceased organ donor proxy samples:**
  - ABO/Rh confirmatory results of deceased organ donor proxy sample (Table 106-1),
  - ABO and Rh(D) of donor records.

References

AABB, *Technical Manual*, current edition.

AABB, *Standards for Blood Banks and Transfusion Services*, current edition.

Authorized Reviewers

Chief, Pathology and Laboratory Medicine
Medical Director and/or Designee, Blood Bank
Manager/Supervisor, Blood Bank

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Document Control

Location of Master: Master electronic file stored on the Beaumont Laboratory server under S:/ Master printed document stored in the *Transfusion Medicine Standard Operating Procedure Manual*.
Number of Controlled Copies posted for educational purposes: 0
Number of circulating Controlled Copies: 0
Location of circulating Controlled Copies: NA

Document History

Signature	Date	Revision #	Modification	Related Documents Reviewed/ Updated
Prepared by: JLSarhan	07/14/2009	r03.00.00	Neonatal and pediatric ABO/Rh moved to P514, *Forward Typing Determination of ABO/Rh*.	
Validated by: Billie Price	01/05/2010			
QA: Louisa Serafimovska	01/30/2010			
Supervisor: Judy Easter	02/04/2010			
Approved by: Peter Millward, MD	06/17/2010			
Reviewed by: (Signature)	**Date**	**Revision #**	**Modification**	**Related Documents Reviewed/ Updated**
Reviewed by: Peter Millward, MD	07/14/2010			
Reviewed by: Peter Millward, MD	07/25/2011			
Reviewed by: Peter Millward, MD	07/24/2012			
Revised by: Jennifer Sarhan	08/16/2012	r03.00.01		
Approved by: Peter Millward, MD	08/16/2012			
r03.00.01 • SOP reformatted onto updated template. • Updated Related Documents and references throughout. • Updated reference to Patient Downtime Worksheet from F-1600 to F-120a. • For deglycerolized units, the confirmatory type will be ordered and resulted in the Blood Bank computer.				
Revised by: Jennifer Sarhan	04/18/2013	r03.00.02	Revised step 12 "Submit results … to the Tissue Coordinator."	
Approved by: Peter Millward, MD	04/22/2013			
Revised by: Jennifer Sarhan	08/19/2013	r03.00.03	Revised the 3rd bullet of the *Scope* section.	
Approved by: Peter Millward, MD	08/20/2013			
Approved by: Peter Millward, MD	07/18/2014			
Approved by: Peter Millward, MD	01/15/2015			
Approved by: Peter Millward, MD	01/25/2017			
Approved by: Elizabeth Sykes, MD	02/23/2018			
Approved by: Peter Millward, MD	02/26/2019			

Document Control, continued

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Reviewed by: (Signature)	Date	Revision #	Modification	Related Documents Reviewed/ Updated
Approved by: Craig Fletcher, MD	03/26/2019			
Revised by: Christopher Ferguson	02/03/2020	r03.01.00	Removed ABO subgrouping information/procedure for donor proxy samples. Adjusted confirmatory testing policy for washed/deglycerolized units. Added bovine albumin control information. Removed QA as authorized reviewer. Updated template.	
Manager: Billie Ketelsen	02/06/2020			
Approved by: Craig Fletcher, MD	02/10/2020			
Approved by: Craig Fletcher, MD	07/15/2020			
Revised by: Samantha Maynard	08/30/2021	r03.02.00	Added information about confirmatory testing of deceased donor proxy samples.	
Manager:				
Approved by: Craig Fletcher, MD				