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DOCUMENT NUMBER:	SCPMG-PPP-0106
DOCUMENT TITLE:	ABO Discrepancy
DOCUMENT NOTES:	

Added instructions for entering results in Cerner and cold agg info, read highlighted areas

LOCATION:	SCPMG-dft	**VERSION:**	02
DOC TYPE:	SCPMG PPP	**STATUS:**	Draft

EFFECTIVE DATE:	02 Nov 2020	**NEXT REVIEW DATE:**	
RELEASE DATE:		**EXPIRATION DATE:**	

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ABO Discrepancy

Purpose

This procedure describes steps and testing to perform when encountering an ABO discrepancy.

- Steps to take when a historical ABO discrepancy (prior result does not agree with current testing) in patient testing is encountered.
- The step by step procedure on how to resolve ABO discrepancies in red cell (forward) typing and plasma/serum (reverse) typing in patient testing.
- How to enter results in Cerner when performing additional testing to resolve discrepancy.
- Transfusion recommendation for patients with ABO discrepancies in testing.
- Steps to take when donor ABO and/or Rh confirmation testing results are discrepant from the product label.

Policy

- Tube method is the routine method for ABO discrepancy testing.
- When a forward/reverse type discrepancy is encountered, the discrepant results must be recorded (performed) in Cerner.
    - After resolution the final interpretation of the ABO group can be verified.
- Before the patient's historical blood type is changed, there must be two matching blood typing results from two different specimen collection events and/or consultation with manager/designee.
    - Refer to procedure *"Hematopoietic Cell Transplantation and Adoptive Cell Transfer: Patient Management"* for additional considerations.
- In the event of massive transfusion, the historical blood type should not be changed.
- In the event of an unresolved ABO discrepancy and transfusion support is required; type O Red Blood Cells and/or type AB plasma will be prepared and/or dispensed until discrepancy resolved. An emergency waiver is required due to incomplete testing.
- Cases may be sent out to the Immunohematology Reference Lab (IRL) for resolution.

ABO Discrepancy, Continued

Definitions

Patient Historical Discrepancy- An ABO discrepancy exists when prior ABO results do not agree with current testing ABO result.

Patient Testing Discrepancy - An ABO testing discrepancy exists when:

- The expected reactions in the forward typing (red cells) do not agree with the expected reactions in the reverse typing (serum/plasma).
- Unexpected weak or mixed-field reaction that cannot be explained with patient condition or history.

Product RBC Testing Discrepancy- A discrepancy exists when the unit ABO and/or Rh confirmation test result does not agree with the unit ABORh face label.

Limitations and procedure notes

- Significant variations in red blood cell suspension (<3% or >5%) may result in false- positive or false negative reactions.
- Reverse grouping tests should never be incubated at 37°C per current manufacturer's instructions for use.
- Anomalous results may be caused by:
  - Incompletely clotted serum
  - Fibrin or particulate matter in serum or plasma or red cells that stick to the sides of the microtube
  - Contaminated specimen
  - Contaminated reagent
  - Wrong/void reagent
  - Specimen error

Equipment and reagents

- 12 X 75 mm test tubes
- Uniform drop transfer pipettes
- Isotonic saline
- ABO typing reagents
- Anti-A1 lectin
- A2 cells
- Anti-A,B
- Cell washer
- Centrifuge
- Agglutination viewer

ABO Discrepancy, Continued

Specimen

- EDTA anticoagulated plasma (preferred)
- Serum, completely clotted

Procedure-Resolving Historical Discrepancy

Follow the steps below to resolve a patient historical discrepancy in ABO/Rh typing.
NOTE: Do not verify result in Cerner until investigation completed.

Step	Action
1.	Perform a clerical check - Verify patient information on tube, paper, electronic records, and repeat testing.
2.	Review patient history (Cerner, Health Connect, contact nursing) for evidence of recent transfusion, stem cell/bone marrow transplant. • If it is discovered that there was a prior admission/treatment at a non-SCAL KP facility it may be advisable to contact the facility for patient testing and transfusion history. Consult with manager/designee if further instruction is needed.
3.	Repeat initial testing with washed patient cells using manual tube method as false positive forward typing reactions may be easily resolved.
4.	**If…** / **Then…** Discrepancy resolved (current results agree with prior results) / Follow separate procedure "*Result Entry: Resulting Patient Tests in Cerner Millennium*" for result entry.

ABO Discrepancy, Continued

Resolving Historical Discrepancy con't

Step	Action
5.	Historical ABO Discrepancy is not resolved…

If…	Then…
Previous sample is still available	Repeat ABO/Rh testing on that sample.
Previous sample is not available	Review the results on previous sample (if available in system) to ensure forward, reverse, and interpretation agreed. Request provider to place an ABORh order to obtain a second sample to retest. • For outpatient specimen, inform the provider's office to ask the patient to return for new specimen collection. **OR** Obtain an additional sample (example: Hematology specimen) from a different date or time of draw for testing. • Order Double Check to retest. • If possible, second sample should be drawn by a different phlebotomist.

ABO Discrepancy, Continued

Resolving Historical Discrepancy con't

Step	Action
6.	Retest new sample. Notify manager/designee if unable to obtain a new sample.

If…	Then…
Repeat specimen sample result matches historical result but does not match 1st current sample.	• Select the correct interpretation in Cerner. • Print unverified results of initial sample and cancel accession (Mislabeled Specimen, Test Not Performed) • Submit QIM for possible mis-identified sample-attach printout of unverified results
New sample result does NOT match the historical result but agrees with the 1st (current) sample.	• Verify results in Cerner. Refer to ABO discrepancy flowchart in procedure *"Result Entry: Resulting Patient Tests in Cerner Millennium"* • Submit QIM for further investigation.

7.	If Rh discrepancy not resolved, follow separate procedure *"Rh Testing"*.

ABO Discrepancy, Continued

Procedure-Resolving Forward typing Discrepancy

Problem with forward/red cell typing: Unexpected, extra, mixed field, missing, weaker than expected reactions
(Expected reaction is less than 2+ reaction grading)

Step	Action
1.	Repeat initial testing with washed patient cells using manual tube method as false positive forward typing reactions may be easily resolved.
2.	Review patient history (Cerner, Health Connect, contact nursing) for evidence of massive transfusion, stem cell/bone marrow transplant. • If it is discovered that there was a prior admission/treatment at a non-SCAL KP facility it may be advisable to contact the facility for patient testing and transfusion history. Refer to Attachment B "Possible Causes of ABO Typing Discrepancies" and/or consult with manager/designee if further instruction is needed.

If…	Then…
Patient received an ABO-mismatched stem cell or bone marrow transplant.	Incubate forward typing tubes at RT for 30 minutes to enhance reactivity. • Mixed field reactions may be observed. • Enter a result note in Cerner "Forward type RT 30'"
Cold agglutinin suspected	Forward typing-Wash cells with warmed saline 4-5 times. • Retype using warmed washed red cells. • Enter a result note in Cerner "Forward type warm washed cells"
Subgroup of type A suspected	Perform A1 lectin testing using **Anti-A1 lectin** procedure below.
Other subgroup suspected	Refer to Attachment A: "Serologic Reactions Observed in A and B subgroups" • Consider send out to IRL
Result notes entered above may also be entered into PPI.	

ABO Discrepancy, Continued

Procedure- Resolving Reverse typing Discrepancy

Problem with reverse cell typing: Unexpected, extra (due to cold antibodies or other issues), mixed field, missing, weaker than expected reactions.

Step	Action
1.	Review patient history. Refer to Attachment B "Possible Causes of ABO Typing Discrepancies" and/or consult with manager/designee if further instruction is needed.

If…	Then…
Patient is elderly, an infant, or immuno-compromised	Follow **reverse type incubation** procedure below.
Patient has disease state such as multiple myeloma, Waldentroms macro-globulinemia,	Read A, B, O, or A2 test tubes microscopically for rouleaux appearance (Stack of coins). If rouleaux is observed, perform **saline replacement** procedure below.
Patient suspected of being subgroup of A or AB, A2 or A2B with anti-A1 (Forward types as A, reverse type is positive with A1 cells)	Test patient rbcs for A2 (optional). See **A2 subgroup** testing procedure above. Refer to Attachment A: "Serologic Reactions Observed in A and B subgroups"
Patient has unexpected reactivity	Probable cold antibody causing discrepancy. See procedure "Antibody Identification- How to interpret test results" See **cold antibody** section below

2.	Proceed to appropriate testing procedure.

ABO Discrepancy, Continued

Procedure- Reverse type incubation Follow the steps below to perform reverse type incubation to enhance weak reactions.

Step	Action
1.	For weak or missing reactivity in reverse typing: In Cerner (DOE) add the ABO Discrepancy test to the accession for the ABO/Rh test to record results for RT and/or 4°C incubations.
2.	Label 2 test tubes for A1 cells, B cell.
3.	Add two drops of patient plasma to each test tube.
4.	Add one drop of appropriate reagent red cells to appropriate tube.
5.	Incubate the A1 and B tube at room temperature for 15-30 minutes.
6.	Mix and centrifuge.
7.	Re-suspend and read macroscopically.

If…	Then…
The reaction is **now positive** with A and/or B cells.	Report result in the ABO discrepancy test with result note of <reverse typing incubated at RT>. Forward and reverse typing agree. Select correct interpretation.
The reaction is **still negative** with A and/or B cells.	Incubate the A1 and/or B tube at 4°C for 15-30 minutes. • Mix, centrifuge re-suspend, and read macroscopically. Report result in the ABO discrepancy test with result note of <reverse typing incubated at 4C>. Forward and reverse typing agree. Select correct interpretation.

ABO Discrepancy, Continued

<table>
<tr><td>7.
(continued)</td><td>Unable to enhance reactions</td><td>Forward and reverse typing do NOT agree.<table><tr><th>If</th><th>Then</th></tr><tr><td>Blood products are required to be dispensed</td><td>Result as “Indeterminate”

Add result comment with forward typing result.</td></tr><tr><td>No Blood products are required to be dispensed</td><td>Leave test as pending</td></tr></table>Refer to appropriate SOPs for product blood type selection and crossmatch testing.

Inform manager/designee of “Indeterminate” typing or pending interpretation.</td></tr>
<tr><td>8.</td><td colspan="2">Enter patient result(s) for testing completed (with result notes above) and interpretation in Cerner for ABO Discrepancy test (procedure).

 
</td></tr>
<tr><td>9.</td><td colspan="2">Refer to Cerner Entry and Final Interpretation sections below.</td></tr>
</table>

ABO Discrepancy, Continued

Procedure- Anti-A1 Lectin testing

Follow the steps below to perform A1 lectin testing using Anti-A1 lectin reagent and patient cells. **(Perform when unexpected reactivity with A1 cells) Perform and record QC on infrequent QC form.**

Step	Action
1.	In Cerner (DOE) add the ABO Discrepancy test to the accession for the ABO/Rh test.
2.	Label 3 test tubes for positive control, negative control, and patient testing using anti-A1 lectin reagent. • Positive control: Reagent A1 red blood cells. • Negative control: Reagent B red blood cells or 3% selectogen/panel cells (Type O cells). Tube 1 Positive Control — Reagent A1 Blood Cells — Anti A1 lectin Tube 2 Negative Control — Reagent B or O Blood Cells — Anti A1 lectin Tube 3 Patient cells (approximately 3-5%) — Patient Cells — Anti A1 lectin
3.	Add one drop of reagent anti-A1 Lectin to each test tube.
4.	Add one drop of reagent red cell to QC tubes.
5.	Add one drop of patient 3-5% washed red cell suspension to patient test tube.
6.	Mix and centrifuge. Examine for agglutination macroscopically • Manually record the required quality control documentation. Record patient results in Cerner **Anti-A1 Lectin** result field found in the ABO Discrepancy test (procedure) 

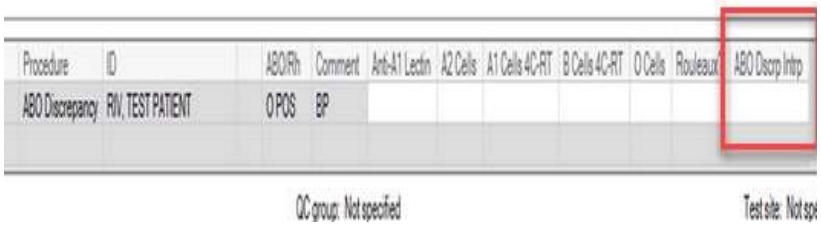
ABO Discrepancy, Continued

Anti-A1 Lectin testing con't

Step	Action
7.	Interpretation

If…	Then…
The patient red cells tested positive with anti-A1 lectin	The patient is group A1 or A1B. Continue discrepancy investigation. Refer to procedure *"Antibody Identification - How to interpret test results"*
The patient red cells tested negative with anti-A1 lectin	Patient is subgroup of A, most likely A2 or A2B. • Assume unexpected reactivity is due to Anti-A1. • Test patient plasma against A2 cells (optional). See **A2 cells testing** below.

Enter ABO Discrepancy Interpretation in Cerner



8.	Refer to **Cerner Entry** and **Final Interpretation** sections below.
9.	Additional Cerner documentation when Anti-A1 detected. (Patient cells are negative with anti-A1 lectin).

In Cerner, launch DOE, Task, Accession Add On, Add Antibody ID. Result Anti-A1.

If patient is	Then
A subgroup (Forward typing for Anti-B is negative)	Add Transfusion Requirement of "Give O RBCs"
A subgroup B (Forward typing for Anti-B is positive)	Consult with manager/ designee as patient may be transfused with either Type O or Type B RBCs and enter appropriate Transfusion Requirement/patient comment.

ABO Discrepancy, Continued

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ABO Discrepancy, Continued

Procedure- A2 cells testing

Follow the steps below to perform A2 cell testing using reagent A2 cells and patient plasma, this is additional confirmation of presence of Anti-A1.

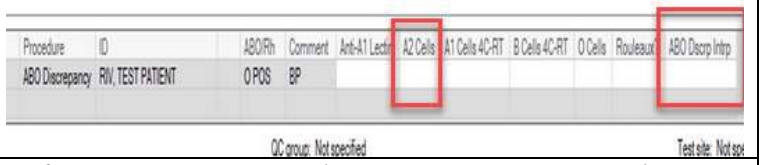
Perform and record QC on infrequent QC form.

NOTE: **Optional** test as A2 cells may not be available at all medical centers.

<table>
<tr><th>Step</th><th>Action</th></tr>
<tr><td>1.</td><td>In Cerner (DOE) add the ABO Discrepancy test to the accession for the ABO/Rh test to record results for A2 cells if not already done when testing with Anti-A1 lectin</td></tr>
<tr><td>2.</td><td>Label 2 tubes for QC or A2 cell reagent.
<table>
<tr><td>Tube 1
Positive Control</td><td>Anti-A reagent</td><td>A2 cells</td></tr>
<tr><td>Tube 2
Negative Control</td><td>Anti-B reagent</td><td>A2 cells</td></tr>
</table></td></tr>
<tr><td>3.</td><td>Label 4 test tubes for A1 cells, A2 cells, O cells, and, auto control (patient cells). (Testing patient plasma)</td></tr>
<tr><td>4.</td><td>Add two drops of patient plasma/reagent control to each test tube.</td></tr>
<tr><td>5.</td><td>Add one drop of appropriate cells to each test tube.</td></tr>
<tr><td>6.</td><td>Mix and centrifuge.</td></tr>
<tr><td>7.</td><td>Re-suspend and read macroscopically.
Record QC results.
For patient testing refer to table below.
<table>
<tr><th colspan="4">If reaction pattern is…</th><th>Then…</th></tr>
<tr><th>A1 cells</th><th>A2 cells</th><th>O cells</th><th>Auto Control</th><th>Conclusion</th></tr>
<tr><td>Pos</td><td>Neg</td><td>Neg</td><td>Neg</td><td>Anti-A1 confirmed</td></tr>
<tr><td>Neg</td><td>Neg</td><td>Neg</td><td>Neg</td><td>Unable to confirm Anti-A1</td></tr>
<tr><td colspan="4">Any other combination of reactions with a positive A2 cell, O cell, and/or positive Auto control</td><td>Probable cold antibody causing discrepancy. See procedure “Antibody Identification- How to interpret test results”</td></tr>
</table></td></tr>
</table>

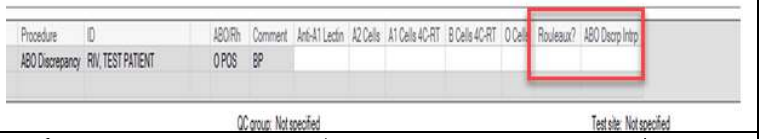
ABO Discrepancy, Continued

Procedure-
A2 cells testing continued

Step	Action
8.	Enter patient A2 cells result and interpretation in Cerner for ABO Discrepancy test (procedure). 
9.	Refer to **Cerner Entry** and **Final Interpretation** sections below.

ABO Discrepancy, Continued

Procedure-Saline Replacement

Step	Action
1.	For extra reactivity in reverse typing: In Cerner (DOE) add the ABO Discrepancy test to the accession for the ABO/Rh test to record results for rouleaux
2.	Re-centrifuge the A1 and B tube.
3.	Remove supernatant leaving the red cell button undisturbed.
4.	Add 2 drops of saline.
5.	Do **not** centrifuge. Re-suspend and read microscopically. **If…** / **Then…** **Positive** (The cells no longer appear like a stack of coins, but appear as agglutination) — The test result is positive for hemagglutination and further testing may be indicated. **Negative** (The agglutination is dispersed- the saline has successfully dispersed the Rouleaux) — The test result is negative. Enter a comment in test results that the sample had rouleaux and was negative with saline replacement.
6.	Enter patient result for testing completed (with result notes above) and interpretation in Cerner for ABO Discrepancy test (procedure). 
7.	Refer to **Cerner Entry** and **Final Interpretation** sections below.

ABO Discrepancy, Continued

Procedure-Cold Antibodies

Follow the steps below to interpret ABO type in the presence of cold auto or allo-antibodies. Unexpected reactivity seen in reverse typing.

The cold antibody must be identified on the sample. If ABO was ordered, you should add the screen/ABID to confirm presence of cold antibodies

Step	Action
1.	Once the presence of cold antibodies are confirmed (refer to *"Antibody Identification - How to interpret test results"* procedure) NOTE: Testing with cold reagents and/or plasma may result in unexpected reactions in reverse typing.
2.	Record reverse typing reactions at room temperature in the Cerner ABO/Rh test
3.	If only an ABO/Rh was ordered and an ABO discrepancy is encountered, an antibody screen and antibody ID may be added in DOE to resolve the ABO discrepancy.
4.	Result may be interpreted on forward type results only once a cold auto/allo antibody is confirmed. • Override exception for interpretation with reason "Not applicable"

Cerner Entry

Unit typing ABO/Rh discrepancy: An ABO/Rh discrepancy exists when the unit ABO confirmation test result does not agree with the unit ABO/Rh face label.

Step	Action
1.	For ABO/Rh test-retain Vision results sent to Cerner
2.	Evaluate additional testing done and recorded with the ABO Discrepancy test.
3.	Enter final ABO/Rh interpretation in Cerner for test • Select manually as expected result pattern match is not found • Override exception for interpretation with reason "Not applicable"
4.	Result to be flagged on exception report and reviewed.

ABO Discrepancy, Continued

Final Interpretation

When testing does not resolve forward and reverse ABO discrepancies follow the steps below to determine the final ABO/Rh interpretation.

If	Then
Patient received an ABO-mismatched **stem cell or bone marrow transplant.**	Refer to *SOP Hematopoietic Cell Transplantation and Adoptive Cell Transfer: Patient Management*
Patient is **elderly, an infant, or otherwise immuno-compromised**	Interpret ABO/Rh based on forward typing only. • Add result comment: Unable to enhance reverse typing
Patient has **confirmed cold antibody** (auto and/or allo)	Interpret ABO/Rh based on forward typing only. • Add result comment: "Unexpected reactivity seen with (A1 cells and/or B cells) due to cold antibody."
Patient has disease state such as **multiple myeloma, Waldentroms macro-globulinemia**	Interpret ABO/Rh based on forward typing only. • Add result comment: "Unexpected reactivity seen with (A1 cells and/or B cells) due to rouleaux."
Patient has been **transfused with RBCs outside of blood group** and is showing mixed field reactivity in forward typing	Interpret ABO/Rh based on forward typing with mixed field reactions. • Add result comment: "Mixed field forward typing due to transfusion of (applicable blood product/type)"
Patient has been **transfused with plasma and/or platelets outside of blood group** and is showing decreased reactivity in reverse typing	Interpret ABO/Rh based on forward typing with missing expected reverse typing. • Add result comment: "Unable to enhance reaction with (A1 cells and/or B cells) due to transfusion of (applicable blood product/type)"

ABO Discrepancy, Continued

Product RBC Testing Discrepancy

Unit typing ABO/Rh discrepancy: An ABO/Rh discrepancy exists when the unit ABO and/or Rh confirmation test result does not agree with the unit ABO/Rh face label.

Step	Action
1.	Perform a clerical check - Verify donor unit information on tube, electronic records, and the blood bag.
2.	Obtain another unit segment.
3.	Repeat testing using washed donor cells.
4.	If the results remain discrepant, immediately quarantine the unit physically and in the Cerner system.
5.	Complete a QIM.
6.	Notify the blood supplier.

Uncontrolled Documents

Fung, Mark K. Ed. Technical Manual, 19th Ed. AABB ,2017
Ortho Clinical Diagnostics, current manufacture insert (Instructions for Use)

Controlled Documents

Attachment A: Serologic Reactions Observed in A and B subgroups
Attachment B: Possible Causes of ABO Typing Discrepancies
Hematopoietic Cell Transplantation and Adoptive Cell Transfer: Patient Management
Antibody Identification - How to interpret test results
Result Entry: Resulting Patient Tests in Cerner Millennium
Rh Testing

MasterControl History of Change:		
Change type: new, major, minor etc.	**Version #**	**Description of Change**
New	1	New SOP
Minor	2	Added instructions for Cerner entry of results. Added sections for cold antibodies and final interpretation. Updated controlled and uncontrolled documents. Updated Attachment A and B (Tables 10-3 and 10-4 from Technical Manual)

Authors:

Transfusion Service Managers
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ABO Discrepancy, Continued
ATTACHMENT A

Serologic Reactions Observed in Selected A and B subgroups

Red Cell Phenotype	Anti-A	Anti-B	A1 cells	B cells
A_1	4+	0	0	4+
A_2	4+	0	0/2+	4+
The following subgroups must be confirmed by Reference Lab				
A_3	3+/mf	0	0/2+	4+
A_x	0/wk	0	0/2+	4+
A_{el}	0	0	0/2+	4+
A_m	0/wk	0	0	4+
B	0	4+	4+	0
B_3	0	3+/mf	4+	0
B_{weak}	0	wk/2+	4+	0
B_{el}	0	0	4+	0
B_m	0	0/wk	4+	0

A1 is most common subgroup of A (80%), A2 is 2nd most common (20%).

Reporting: Cerner does not distinguish group A1 or subgroup A2. These are reported as group A.

Acquired B is usually not demonstrable in mono-clonal Anti-B reagents (check manufacturer insert).

Make sure to add "give O RBC" transfusion requirement if patient is A2. **Do NOT give A RBC!**

ABO Discrepancy, Continued

ATTACHMENT B

Possible Causes of ABO Typing Discrepancies

Category	Causes
Weak/missing red cell reactivity	• ABO subgroup • Leukemia/malignancy • Transfusion (including Intrauterine fetal) • Pregnancy • Transplantation • Excessive soluble blood group substance
Extra red cell reactivity	• Autoagglutinins/excess protein coating rbcs • Unwashed rbcs; plasma proteins • Unwashed rbcs; antibody in patient's serum to reagent constituent • Transplantation • Acquired B antigen or other polyagglutinable conditions • cis AB or B(A) phenomenon • Out of group transfusion
Mixed-field red cell reactivity	• ABO subgroup • Recent transfusion • Transplantation • Fetomaternal hemorrhage • Twin or dispermic (tetragametic) chimerism
Weak/missing serum reactivity	• Age related (<4-6 months old, elderly) • ABO subgroup • Hypogammaglobulinemia • Transplantation • Excessive anti-A or anti-B (prozone) • Hemodilution
Extra serum reactivity	• Cold antibody-allo/auto • Serum antibody to reagent constituent • Excess serum protein • Transfusion of plasma components • Transplantation • Infusion of intravenous immune globulin