# Applicable Laboratory(s)):

North Carolina Baptist Hospital (NCBH)

Lexington Medical Center (LMC)

Davie Medical Center (DMC)

Wilkes Medical Center (WMC)

High Point Medical Center (HPMC)

Westchester

Clemmons

# Policy Purpose

The ABO blood group system is the most important blood group system in transfusions and organ transplants. The ABO antigens are found on red blood cells, platelets and other proteins found in the blood. It is the only system where antibodies are naturally formed (believed to be due to exposure to gut and environmental bacteria. i.e. Enterobacteriaceae) to the missing RBC antigen. Due to these antibodies the transfusion of incompatible red cells can cause immediate red cell lysis (acute intravascular hemolysis) which can lead to renal failure and death. One of the leading causes of death as reported to the FDA is due to transfusion of ABO incompatible blood.

The Rh system is very complex consisting of over 50 different antigens. It was first discovered in Hemolytic Disease of the Newborn that was caused by a paternal antigen inherited by the newborn. These antigens are found on red cells but not platelets. These antigens are very immunogenic with the D antigen being the most immunogenic of the system. Thus the need for routine testing of the D antigen.

# Scope

This policy applies to blood bank staff and management

# Definitions

1. Policy: As defined in the Policy on Creating and Amending Policy, a statement of principle that is developed for the purpose of guiding decisions and activities related to governance, administration, or management of care, treatment, services or other activities of WFBH.  A policy may help to ensure compliance with applicable laws and regulations, promote one or more of the missions of WFBH, contain guidelines for governance, and set parameters within which faculty, staff, students, visitors and others are expected to operate.
2. WFBH Lab System: Wake Forest Baptist Lab System is a health system that includes Wake Forest Baptist Medical Center and all affiliated organizations including Wake Forest University Health Sciences (WFUHS), North Carolina Baptist Hospital (NCBH), Lexington Medical Center (LMC), Davie Medical Center (DMC), Wilkes Medical Center (WMC), High Point Medical Center (HPMC), Lab at Westchester and Lab at Clemmons.
3. **NTD:** Unable to Determine (NTDoup), unable to determine the ABO group
4. **UTD:** Unable to determine the Rh (D) type
5. **Sunquest:** Blood Bank Information System
6. **BAD file:** Blood Bank Administrative Data file
7. List of Sunquest tests that contain ABORh testing:

|  |  |  |  |
| --- | --- | --- | --- |
| **LAB Code** | **Name** | **Sunquest Code** | **Testing** |
| **LAB2609** | ABO/Rh type < 4 months | **NEABRH** | ABO/Rh (forward) |
| **LAB895** | ABO/Rh type | **ABRH** | ABO/Rh |
| **LAB647** | Transfusion Reaction Workup | **TRXI** | ABO/Rh, DAT  Post Transfusion Reaction |
| **LAB276** | Type and Screen | **TYHD** | ABO/Rh, Antibody Screen |
| **LAB892** | Cord Blood Workup | **CDBLD** | ABO/Rh, DAT IgG |
| **LAB5047** | Prenatal Testing | **PREN** | ABO/Rh, Antibody Screen |
| **LAB5161** | Crossmatch Neonatal Red Cells | **TNRCC** | ABO/Rh |
| **LAB5045** | RhoGam Antenatal Evaluation | **RAE** | Rh(D) Factor |
| **LAB6626** | Patient ABO/Rh Recheck | **PRCXM** | ABO/Rh |
| **LAB7537** | PUBS Type and Screen |  | ABO/Rh, DAT IgG |
| **LAB7409** | Type and Screen BMT donor |  | ABO/Rh, Antibody Screen |
| **LAB762** | Rhogam Workup | **TRHIG** | ABO/Rh, Fetal Screen |

# Policy Guidelines

1. Protocol
2. **ABO Forward typing:** in the forward ABO typing, the presence or absence of ABO antigens is determined by testing cells with anti-A, anti-B, and/or anti-A,B and observing for agglutination. Absence of agglutination is a negative test result which indicates the absence of the corresponding antigen on the red cells. Agglutination of ≥2+ with a given reagent is a positive test result, which indicates the presence of the antigen.
3. **ABO Reverse Typing:** reverse typing is demonstrated by the presence or absence of agglutination of the expected, reciprocal ABO antibodies in the plasma or serum of A, B, and O patients with reagent A1 and B cells. Absence of agglutination is a negative test result which indicates that the antibody corresponding to the red cell is not present. Agglutination of ≥2+ with a given cell is a positive test result, which indicates the presence of the antibody in the patient’s plasma/serum.
4. **Rh typing:** Rh typing tests for the presence or absence of the D red cell antigen. Rh typing is demonstrated by the presence or absence of agglutination when testing cells with reagent anti-D antibody. Absence of agglutination is a negative test result while agglutination of ≥2+ is a positive test result.
5. **Weak D testing:** Weak D phenotypes are defined as having a reduced amount of D antigen, requiring an indirect antiglobulin test (IAT) for detection. Red blood cells that appear to be D negative or are reacting <2+ by direct test methods may be further tested for the weak D antigen by the weak D test. Weak D testing is performed on all OB patients, Rh Immune Globulin candidates, Bone Marrow and organ donors and recipients, and recipients of directed donor units.

**Weak D testing on Vision:** All *Positive* Weak D tests performed on the Vision must have a DAT performed on the sample before resulting the Weak D. The Weak D result is invalid if the DAT is positive.

RhD typing by direct agglutination and/or Rh weak D testing reactions must show >2+ reactivity to be considered positive.

* 1. Rh weak D reactions showing <2+ reactivity should be examined for mixed field.
  2. Patients demonstrating reactivity <2+ with weak D testing will be interpreted as Unable to determine (UTD), will have a specimen sent for Rh DNA genotyping and will receive Rh negative products until confirmation of RhD status is received.
  3. Rh weak D testing will be performed on all the following patients:
     1. Patients demonstrating <2+ reactivity with RhD direct agglutination testing
     2. Rh negative neonates up to 4 months of age
     3. Rh negative obstetrics patients
     4. Rh negative bone marrow and organ donors
     5. Rh negative patients with Rh positive directed donor units to determine the patient’s true Rh type.
     6. Those patients previously determined to be weak D positive
     7. Rh negative patient who could potentially receive Rh positive products (due to low inventory)-Pre transfusion sample
     8. As requested by physician
  4. **Weak D testing on Vision:** All *Positive* Weak D tests performed on the Vision must have a DAT performed on the sample before resulting the Weak D. The Weak D result is invalid if the DAT is positive.

1. RhD typing by direct agglutination and/or Rh weak D testing reactions must show >2+ reactivity to be considered positive.
2. Rh weak D reactions showing <2+ reactivity should be examined for mixed field.
3. Patients demonstrating reactivity <2+ with weak D testing will be interpreted as unable to determine (UTD), will have a specimen sent for Rh DNA genotyping and will receive Rh negative products until confirmation of RhD status is received.
4. Rh weak D testing will be performed on all the following patients:
   * + Patients demonstrating <2+ reactivity with RhD direct agglutination testing
     + Rh negative neonates up to 4 months of age
     + Rh negative obstetrics patients
     + Rh negative bone marrow and organ donors
     + Rh negative patients with Rh positive directed donor units to determine the patient’s true Rh type.
     + Those patients previously determined to be weak D positive
     + Rh negative patient who could potentially receive Rh positive products (due to low inventory)-Pre transfusion sample
     + As requested by physician
   1. Positive weak D results of any strength on an OB patient should be sent for weak and partial D analysis at Blood Center of Wisconsin.
   2. The tests that were performed by BCW will be charged under the appropriate previously tested specimen.
      1. Order BCW tests: Weak RHD analysis and Partial RHD analysis.
      2. Complete the Reference Lab Tracking Form.
   3. The RH interpretation (Rh Geno Interp) should be added to the BAD file via comment and then resulted based on the BCW genotype results.
5. **Frequencies** of ABO and Rh in the population:

*Refer to Attachment 1: ABO Frequencies*

1. **Interpretation of ABO and Rh results**

*Refer to Attachment 2: Interpretation of ABO and RH Results*

* 1. In Sunquest, if forward type is mixed field result as indicated in table below. An investigation shall follow to determine the source of MF results.
     1. Mixed field reaction choices in Sunquest are as follows:

|  |  |
| --- | --- |
| 1. **Reaction** | 1. **Sunquest Result** |
| 1. Weak mixed field | MW |
| 1. 1+ mixed field | M1 |
| 1. 2+ mixed field | M2 |
| 1. 3+ mixed field | M3 |
| 1. 4+ mixed field | M4 |

* 1. **NTDoup (NTD)**: When the forward ABO typing does not match the reverse ABO typing the interpretation must be Unable to Determine (NTD). When the Rh results are invalid such as when the saline control or Weak D control is positive the Rh interpretation must be Unable to Determine (UTD). When the Rh results are valid but the ABO results do not correlate, the Rh interpretation may be entered as Positive or Negative.
  2. **NTDoup (NTD):** If the ABO forward and reverse types do not match and there is no prior history or the patient is a kidney or solid organ transplant, then a green sheet should be complete to flag management for review.
     1. If the NTDoup is due to a missing forward or reverse then test with anti-A,B and anti-A1 Lectin
     2. If there is a discrepancy on a **Kidney patient**- notify management.
        + If the forward type is >2+ and the back type doesn’t match-result the ABO interpretation based on the forward type. Do Not result as NTD.
        + This will require a supervisor override. If no one is available to do this leave for management to result.
  3. Check the patient BAD file in Sunquest. Management needs to remove any prior ABO if present since the ABO does not automatically update to NTD. The same is true when the Rh is interpreted as unable to determine (UTD). Notify management to update. Technologist should enter a comment in BAD file that the patient is NTD or UTD.
     1. When an interpretation of NTD is entered the patient must receive the following component blood types, see following chart.

Patients on BMT protocol who type NTD or UTD may have specific product ABO/Rh requirements noted in the BAD file.

*Refer to* *BB-POL-0055: Transplant Testing Protocol*

|  |  |  |  |  |
| --- | --- | --- | --- | --- |
| 1. ABO interp | Rh interp | 1. Platelets to give\* | Plasma to give | RBCs to give\*\* |
| NTD | POS | AB Pos or Neg | AB | 1. O Pos or Neg |
| NTD | NEG | AB Neg | AB | O Neg |
| NTD | UTD | AB Neg | AB | O Neg |

\*Refer to Platelet Protocols for options if AB Neg platelets are not available.

**\*\*NOTE:** Group AB patients that have received multiple out of group red cells and meet the criteria for NTDoup should continue receiving Group A red cells. Management may approve the interpretation for AB with an additional ABO recheck (ABO2) so that group A red cells can be issued without a supervisor override in Sunquest.

1. **Quality control**

*Refer to BB-POL-0064: Daily Quality Control Protocols*

*Refer to BB-SOP-0063: QC-Daily Quality Control*

1. The Blood Bank Work Organization Protocols must be followed throughout all steps of testing.

*Refer to BB-POL-0026: Blood Bank Work Organization Policy*

1. An ABO/Rh typing performed within the last 12 months is required for any patient receiving non-red cell containing blood products. If products are needed emergently, issue components on Blood Product Release form.
2. **ABO Recheck (PRCXM)**: Patient specimens with no previous ABO/Rh and/or those with interpretations on file prior to February 2006 must have an ABO Recheck performed on a separate specimen.
3. Results of ABO recheck must be identical to initial results
   1. **When historical or present ABO/Rh does not match the current specimen, a new suspension from the same source specimen tube must be tested by a second technologist.**

|  |  |  |  |  |  |
| --- | --- | --- | --- | --- | --- |
| **If Historical ABO/Rh is** | **and Current is** | **Comment** | **RBCs to give\*\*** | **Platelets to give\*** | **Plasma to give** |
| Present | NTD UTD,  NTD NEG |  | O Neg | AB Neg | AB |
| Present | NTD POS |  | O Pos or Neg | AB Pos or Neg | AB |
| Present | ABO/Rh different than historic | Obtain another blood specimen and  notify management | O Neg | AB Neg | AB |
| NTD UTD,  NTD NEG  NTD POS | Conclusive ABO/Rh | Current ABO and Rh type must be 2+ or greater | Group Specific | Group Specific | Group Specific |

\*Refer to Platelet Protocols for options if AB Neg platelets are not available.

\*\*Refer to NOTE in section 6.4a for AB patients.

* 1. Testing for patient ABO rechecks include forward ABO and RhD testing and reverse type.
     1. Refer to BB-POL-0074: Second ABO for No History Patients
  2. When unable to perform ABO recheck:
     1. For Pediatrics: give group O neg red cells, AB Neg platelets and AB plasma products.
     2. For Adults: give group O red cells and request the Blood Product release form be signed.
  3. It is not required to repeat weak D testing as part of an ABO recheck.
  4. If the historical ABORh is NTD UTD, NTD NEG or NTD POS while the current sample is conclusive or the historical does not match the current ABORh **and** the patient has had a BMT transplant in the past, the patient’s historical blood type may be changed. However, this must be reviewed and OK’d by the Medical Director or management of the Blood Bank.

1. Performance of electronic crossmatch requires that there are concordant results of at least two determinations of the recipient's ABO type on record, one of which is from a current sample.
2. **Discrepancies:**
   1. All forward and reverse reactions must be ≥2+ positive in order to be valid. Reactions which are <2+ must be investigated further.
   2. All forward and reverse reactions must agree before an interpretation of ABO group can be made.
   3. If an interpretation cannot be made after investigation the interpretation should be resulted as: NTD (NTDoup) for ABO and UTD for Rh type.
      1. Refer to Steps 6.2 and 6.4.
      2. Check the patient BAD file in Sunquest. Management needs to remove any prior ABO if present since the ABO does not automatically update to NTD. The same is true when the Rh is interpreted as UTD. Notify management to update. Technologist should add a comment in the BAD file that the patient is NTD or UTD.
      3. The exception to this is ABORhs performed on Neonates (0-6 months) when the patient is too young to have developed antibodies yet. Neonates do not require reverse testing. The ABO interpretation is resulted based on forward typing only.
      4. If there is a discrepancy on a Kidney patient- notify management.
   4. All ABO discrepancies (Forward and Reverse) must be resolved and documented before an ABO interpretation can be made.
   5. RH results are currently positive with historical negative results must be investigated.
      1. Review patient testing history and methodology used to obtain previous results.
      2. Repeat ABO and RH testing using tube method
         * A weak D test should be performed if patient types D negative or reaction results are <2+.
      3. Weak D positive, female patients who are of childbearing age and/or pregnant must be sent for Weak and Partial D analysis
         * Result as **UTD** until reference lab testing is completed
         * Document on green Antibody Summary form for management review.
      4. Male patients or female >50 years old may be resulted as Rh positive.
         * Document on green Antibody Summary form for management review.
   6. Patients who have cold (RT) reactive allo- or naturally occurring antibodies should have reverse typing performed with reagent reverse cells (A1 cells and B cells) found to lack the offending antigen for which the patient has the antibody.
3. **ABO/Rh reconfirmation of donor units:**
   1. For incoming donor units only the forward typing is required for ABO reconfirmation of donor units. Testing with Anti-A,B is acceptable to reconfirm ABO type on O donor units. The Rh must be tested on all Rh negative units. ABO/Rh reconfirmation must be performed on the following:
      1. Red blood cells
      2. Granulocyte products
      3. Bloody platelets that contain more that 2mls of red blood cells
      4. The following Red cells that have been processed at WFBMC Blood Bank:

* Deglyced red cells
* Washed red cells
* The RETYP test must be manually ordered on the washed red cell in Inventory > Orders > New add

1. **Cord blood specimens** received for ABO testing should be reamed for clots prior to testing on instruments. If all results are positive including control then cells should be washed thoroughly (≥6 times) to remove Wharton’s Jelly from the red cells. Reverse typing is not performed.
2. **Post-delivery and OB patients:** weak D reactions showing agglutination <2+ or showing a mixed field reactions may indicate a mixture of Rh negative maternal blood with Rh positive fetal blood. Consult with BB medical director and/or management to determine if performance of a quantitative fetal hemoglobin test is indicated. (Refer to Weak D typing above).
3. Manufacturer’s directions and package inserts should be consulted for performance characteristics and limitations of all reagents used.

# Literature References:

AABB Technical Manual. Revised periodically.

Reid, Marion E. et al. THE BLOOD GROUP ANTIGEN FACTS BOOK.

Blood Grouping Reagents [package insert]. BIO-RAD Medical Diagnostics GmbH.

Blood Grouping Reagents [package insert]. Immucor Gamma

Modern Blood Banking and Transfusion Practice, Harmening; revised periodically.

# Related Policies/Procedures in Navex: NA

# Attachments/Linked Documents in Title 21:

BB-SOP-0003: ABO Testing Manual

BB-POL-0026: Blood Bank Work Organization Policy

BB-POL-0004: Grading of Positive and Negative Reactions

BB-POL-0052: Selection of Blood and Blood Components

BB-POL-0053: Specimen Labeling Requirements and BBID Numbers

BB-POL-0055: Transplant Testing Protocols

BB-POL-0064: Daily Quality Control Protocols

BB-SOP-0063: QC-Daily Quality Control

# Revision Dates: Review Change Summary as Represented in Title 21.

**Attachment 1: ABO and Rh(D) Frequencies**

|  |  |  |  |  |
| --- | --- | --- | --- | --- |
| **ABO (% Occurrence)** | | | | |
| **Phenotype** | **Caucasians** | **Blacks** | **Asian** | **Mexican** |
| **A1** | **33** | **19** | **27** | **22** |
| **A2** | **10** | **8** | **Rare** | **6** |
| **B** | **9** | **20** | **25** | **13** |
| **O** | **44** | **49** | **43** | **55** |
| **A1B** | **3** | **3** | **5** | **4** |
| **A2B** | **1** | **1** | **Rare** | **Rare** |
| **Rh (D) (% Occurrence)** | | | | |
| **Phenotype** | **Caucasians** | **Blacks** | **Asian** | **Native American** |
| **D** | **85** | **92** | **99** | **99** |

**Attachment 2: Interpretation of ABO Results**

|  |  |  |  |  |  |  |
| --- | --- | --- | --- | --- | --- | --- |
| **Reactions with Red Cells:**  **Forward Type** | | | **Reactions with Plasma:**  **Reverse Type\*\*** | | **ABO Interpretation** | |
| **Anti-A** | **Anti-B** | **SC** | **A1 cells** | **B cells** |
| **>2+** | **0** | **ND** | **0** | **>2+** | **A** | **Conclusive interpretation** |
| **0** | **>2+** | **ND** | **>2+** | **0** | **B** |
| **0** | **0** | **ND** | **>2+** | **>2+** | **O** |
| **>2+** | **>2+** | **0** | **0** | **0** | **AB** |
| **0-1+** | **0** | **ND** | **0** | **>2+** | **NTD** | **Inconclusive interpretation** |
| **>2+** | **0** | **ND** | **0** | **0-1+** | **NTD** |
| **0** | **0-1+** | **ND** | **>2+** | **0** | **NTD** |
| **0** | **>2+** | **ND** | **0-1+** | **0** | **NTD** |
| **0** | **0** | **ND** | **0-1+** | **0-1+** | **NTD** |
| **0-1+** | **0-1+** | **ND** | **0** | **0** | **NTD** |
| **1+** | **1+** | **ND** | **>2+** | **>2+** | **NTD** |
| **1+-4+** | **1+-4+** | **1+-4+** | **0-4+** | **0-4+** | **NTD** |

**SC= saline control NTD= No ABO Group Type**

**Attachment 3: Interpretation of RH Results**

|  |  |  |  |  |  |
| --- | --- | --- | --- | --- | --- |
| **Immediate spin** | | **AHG** | | **Rh (D) Interpretation** | |
| **Anti-D** | **SC** | **Wk D** | **Wk D control** |
| **>2+** | **ND** | **ND** | **ND** | **POS** | **Conclusive interpretation** |
| **0-1+** | **ND** | **>2+** | **0** | **POS** |
| **0** | **ND** | **0** | **0** | **NEG** |
| **0** | **ND** | **ND** | **ND** | **NEG** |
| **1+-4+** | **1+-4+** | **ND** | **ND** | **UTD** | **Inconclusive interpretation** |
| **0-1+** | **ND** | **0-1+** | **0** | **UTD** |
| **0-1+** | **ND** | **0-4+** | **1-4+** | **UTD** |

**SC= saline control**

**NTD= Unable to Determine (No ABO Group Type)**

**UTD= Unable to Determine (No Rh Type (Rh Unknown))**