KAISER PERMANENTE

Kaiser Permanente Medical Center, San Francisco
Northern California Region
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Work Instruction

Title: TS-Cord Blood Testing		**WI Number** SFOWI-0104 **Revision:** 7
Department: Immunohematology **Area:** 2425 Geary Blvd SFO Hospital Lab	**This document has been edited by: Cara H Lim** ***Modified Document***	
Type of Document: Work Instruction		**Review Period -** 340 **Days**

PURPOSE

To perform ABO group, Rh type and Direct Antiglobulin Test on newborns in suspected cases of hemolytic disease of the newborn (HDN) (generally group A, B or AB babies born of group O mothers) and to determine the mother's candidacy for RhoGAM (Rh positive babies born of Rh negative mothers) and for transfusion purposes.
Anti-IgG is used for testing baby DAT because hemolytic disease of the newborn (HDN) results from immune destruction of the red cells of the fetus/newborn by the coating of these cells with maternal IgG antibody.

REAGENTS

A. Anti-A
B. Anti-B
C. Anti-D
D. Isotonic saline
E. Anti-IgG Antihuman globulin
F. 7% bovine albumin
G. Check cells

EQUIPMENT

A. 12 x 75 mm test tubes
B. Serologic Centrifuge
C. Agglutination viewer
D. Microscope
E. Transfer pipets
F. Cell washer

SPECIMEN

A. All cord blood specimen will be sent from L&D to the Transfusion Service for hold or testing. They are accessioned and filed in the Cord Blood Rack according to the last digit

of the Julian date in the specimen storage refrigerator.

B. Heel stick specimen will be sent when cord blood is not available or for repeat testing. They are filed with the regular specimen in the day's rack.

C. All specimen should be labeled with the baby's:
   1. Last name, male (M/C), female (F/C) or first name
   2. Medical record number
   3. Signature, Initials or NUID of the person who collected the blood
   4. Date and time of collection

D. Refer to **SFOWI-0079 Blood Bank Specimen and Requisition** and **SFOWI-0054 Double Check** for more information.

CONTROL

A. Perform saline control if DAT IgG is positive.

B. Perform D control for AB Pos babies.

PROCEDURE

A. **MD orders cord blood work up on babies born to mothers who have :**
   1. No prenatal care
   2. Positive antibody screen
   3. Group O mother
   4. Rh negative mother
   5. Mothers who have not had an antibody screen for the last 8 months

B. **For testing on ProVue:**
   1. Check and remove clot from cord blood with two applicator sticks.
   2. Load sample on ProVue and run program 'Cord'.
   3. If baby is Rh negative and DAT negative, perform weak D test using tube method.
   4. Refer to SFOWI-0080 ProVue for detailed testing instructions.

C. **For manual tube testing:**

NOTE: For specific testing information, refer to related SOPs listed in the Associated Documents section.

   1. Make a 3-5% cell suspension in saline for testing cord blood.
   2. Label five 12 x 75 mm tubes for each baby
      a. Anti-A
      b. Anti-B
      c. Anti-D
      d. D Control (for AB Pos babies)
      e. DAT (Direct antiglobulin test)
   3. Add one drop of cell suspension to each tube and wash 4 times manually or in the automatic cell washer.
   4. Add anti-A, B and D to the corresponding tubes and add anti-IgG to the DAT tube. There should not be any delay in adding anti-IgG after washing. When a patient types as AB positive, a D control according to manufacturer's instruction must be run in parallel to rule out spontaneous agglutination.
   5. Centrifuge all tubes at HIGH for calibrated time according to the respective SOPs.
   7. If the specimen types as Rh negative and the DAT is negative, perform weak D testing including D control.

8. If DAT IgG is negative, add a drop of Check cells, spin and read for agglutination.
9. If DAT IgG is positive, perform saline control using 2 drops of saline instead of 2 drops of anti-IgG.

D. Results interpretation:

NOTE: For specific interpretation information, refer to related SOPs listed in the Associated Documents section.

1. **Mixed field** reaction **in cord blood** of babies **born to Non-group 'O' mothers** should be interpreted as **ABO** '**Indeterminate**' and the appropriate templates (SF_CD1, SF_MOM) attached.
2. If the **Mother** is of '**O**' **blood group and/or Rh Neg**, refer to the attached **TABLE FOR CORD BLOOD ABORh INTERPRETATION (only for single birth babies born at KPSF from group 'O' and/or Rh Neg Mothers)**.
3. If the saline control is negative and the DAT is positive, report the DAT as a critical result.
   a. Refer to the 'Critical Results' and 'Notification of Critical Values and Early Notification Values' SOPs.
   b. Add **SF_MOM** as Result Note and document Readback (**SF_RB**) as Result Comments in LIS.
4. If the saline control is positive, the DAT result is invalid. Repeat testing. If still invalid, request for peripheral blood.
5. For Rh interpretations, refer to the **TABLE FOR CORD BLOOD ABORh INTERPRETATION**.
6. Elution is only performed when requested by a physician. It can also be a reflex test when further investigation is warranted i.e. in serious cases of HDN.

PROCEDURE NOTE(S)

A. The DAT usually gives a strong positive result in HDN due to anti-D or antibodies in other blood group, reactions are much weaker or negative in HDN due to ABO antibodies.
B. Babies with ABO HDN may have a negative DAT. The DAT is positive in only 20-40% of these infants and typically is only weakly positive even in the case of significant hemolysis. This high false negative rate is felt to be due to a combination of decreased number of antigens on the cells and the distance between antigen molecules on the cells surface.
C. Increase rate of newborn red cell destruction can be caused by passive IgG antibodies from mother directed against antigens on the baby's red cells. The affected baby usually has a positive Direct Coombs test, increase in Bilirubin level and a decreased hematocrit.
D. To investigate HDN, perform type and screen on the mother's specimen, test the baby's eluate against A_1 cells, B cells, screening cells and/or antibody panel.

REFERENCE:

A. AABB, Standards for Transfusion Service and Blood Banks, current edition.
B. AABB, Technical Manual, current edition, Bethesda, MD.

Associated Documents:

External Documents

SFOWI-0081 Reading and Grading Hemagglutination
SFOWI-0082 Performing ABO Grouping & Investigating ABO Grouping Discrepancies
SFOWI-0084 Rho(D) Typing
SFOWI-0118 Direct Antiglobulin Test
SFOWI-0105 Neonatal Transfusion

TABLE FOR CORD BLOOD ABORh INTERPRETATION_SFOWI-0104_v6.doc

Associated Quality System Documents - None

Documents Generated:

Document Revision History:

Revision: 7	**Date Created:** 09/22/2005 **Date of Last Revision:** 04/07/2015	**Last Approval Date:** 05/23/2014

Document Author: Cara H Lim/CA/KAIPERM	**Document Manager:** Patrick K Wong/CA/KAIPERM

Reason for Change:

Revision:	Sec/Para Changed	Change Made:	Date
1	Approver	New Lab Director	1/12/07
1	Procedure Notes	Add HDN investigation	1/14/07
2	Procedure	Enter critical result read back comment.	4/29/07
3	Approver	New Lab Director	7/1/07
4	Control Procedure B. C.2.d. C.4. D. D.2. Associated Documents Approver	Added line B. Deleted retrieval of specimen. New section. Changed 7% albumin control to D Control. Added instructions for AB Pos patients. New section. Added line to refer to associated SOPs. Attached the table for cord blood (born from Group O and/or Rh Neg Mothers) ABORh interpretation. Change Medical Director.	6/17/11
5	Approver Associated Documents Procedure D.3.b. Procedure D.5. Associated Documents	New Lab Director. Attached Table of ABORh Interpretation, version 4 - revised Important Note iv) and Note. Added SF_RB. New. Added SFOWI-0118.	1/21/13
6	Approver Procedure D.1. Procedure D.4.b. Procedure C.9.	New BB Medical Director. Added instructions for interpretation when MF rxn is seen. Added instructions to attach SF_MOM as Result Note for Positive Cord DAT. Deleted repeat DAT.	11/7/13
7	Procedure D.1. and Table Associated Documents	Changed 'Invalid' to 'Indeterminate' for interpretation of unresolved ABO or Rh discrepancy. New.	12/9/13
8	Procedure D.6. Procedure C. & D. Associated Documents	Deleted instructions for Rh 'Indeterminate' interpretation and added reference to the Table for Cord Blood Interpretation. Added NOTE to reference SFOWI-0081, SFOWI-0082, SFOWI-0084 and SFOWI-0111. Revised Table for Cord Blood Interpretation to modify verbiage of SFO templates that now matches the global templates. Added Rh Indeterminate interpretation for cord blood when MF	2/20/15

		is seen in babies born to Rh Pos mothers.	

Notification List:

Approvals:

First Approver's Signature

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Second Approver's Signature

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Title: Chief of Pathology; CLIA Director

Document History Section

TABLE FOR CORD BLOOD ABORh INTERPRETATION (only for single birth babies born at KPSF from group 'O' and/or Rh Neg Mothers)

IMPORTANT NOTE: i) Reaction strength with **Anti-A or Anti-B** must be **3+ to 4+**.
ii) Only apply to babies born at Kaiser SF hospital with **NO history of IUT**.
iii) **Exclude** home births, multiple births, transfer from other facilities – request for peripheral blood samples if MF is seen with these babies.
iv) **Mixed field** reaction **in cord blood of babies born to Non-group 'O' mothers** should be interpreted as ABO '**Indeterminate**' and the appropriate templates (SF_CD1, SF_ MOM) attached.

Mother's ABO	Cord Blood Reactions		Cord Blood ABO Interpretation	Result Comments	Result Note
	Anti-A	Anti-B			
O	MF	Neg	**A**	None	SF_MOM
O	No MF	Neg	**A**	None	None
O	Neg	MF	**B**	None	SF_MOM
O	Neg	No MF	**B**	None	None
O	MF	MF	**AB**	None	SF_MOM
O	No MF	No MF	**AB**	None	None
O	Neg	Neg	**O**	None	None
O	MF	No MF	**Indeterminate**	SF_CD1	SF_MOM
O	No MF	MF	**Indeterminate**	SF_CD1	SF_MOM

Templates: 1. **SF_CD1** Unable to interpret ABO or Rh due to possible contamination with mother's blood. Redraw peripheral sample if necessary. NOTE: RHIG should be given to Rh Negative mother.
2. **SF_MOM** Mother's MR#____; ABORh _____, ABSC _____ on _____ .
3. **SF_NEO** Unable to interpret ABO or Rh due to possible contamination with mother's blood or underdeveloped antigen expression. Recommend redrawing blood in 4-6 months. NOTE: RHIG should be given to Rh Neg mother.
4. **SF_CD2** Unable to interpret Rh due to Positive DAT. NOTE: RHIG should be given to Rh Negative mother. DAT: Test results phoned to and verification read-back made by _ title _, ext _, date/time _ called by _.
5. **SF_DU** Infant is Weak D Positive. Note: RHIG should be given to Rh Neg mother.

CORD BLOOD Rh TYPING

Refer to SFOWI-0084 Rho(D) Typing SOP for detailed instructions and interpretations.

Mother's Rh	Cord Blood Reaction with Anti-D	Cord Blood Rh Interpretation	Result Comments	Result Note
Neg	3+ to 4+ MF	**Pos**	None	SF_MOM
Pos	MF	**Indeterminate**	SF_CD1	SF_MOM

NOTE: i) If the cord specimen types as **Rh negative @IS and the DAT is positive**, interpret as **Rh 'Indeterminate'** and attach **SF_CD2** as Result Comments in the LIS.
ii) If the cord specimen types as **Weak D Positive**, interpret as **Rh 'Indeterminate'** and attach **SF_DU** as Result Comments in the LIS.

NEONATE PERIPHERAL BLOOD

NOTE: i) If **forward reaction is less than 3+** in **peripheral blood** after performing ABO discrepancy workup, interpret ABO as '**Indeterminate**' and add Result Comments, '**SF_NEO**'. Refer to SFOWI-0082 Performing ABO Grouping & Investigating ABO Grouping Discrepancies.
ii) 3+ to 4+ mixed field forward reaction in **peripheral blood** testing can be interpreted as long as there is **NO** history of transfusion (IUT or Exchange) and it is a **single birth**.