PROCEDURE

Title: Antibody Screen-N-HANCE			
Procedure #: 2015BLOODBANK68			
Institution: Highlands Regional Medical Center			
Address: 3600 Highlands Avenue, Sebring Florida 33870			
Prepared by: Anita Smith Date: 6/12/2015			
Title: Laboratory Administrative Director			
Accepted by: Date: KIZIT			
Title: Laboratory Medical Director			
Date Patient Testing Implemented: 4/1/2015			
Review of procedure every two years			
Reviewed by: Date:			
Discontinued testing date:			



Policy Name: Antibody Screen – N-HANCE Department: Blood Bank-Lab

Departmental Review: Policy #: B1.7

INITIATE DATE DATE REVIEWED/REVISED PAGE 1 of 4
4/2015

PURPOSE:

The detection of unexpected antibodies in the serum or plasma of a patient or donor is accomplished by testing against selected red blood cells possessing between them the common inherited blood group antigens (C, c, D, E, e, M, N, S, s, P₁, K, k, Le^a, Le^b, Fy^a, Fy^b, Jk^a, Jk^b). The addition of a polyethylene glycol/low-ionic-strength solution both enhances the sensitivity of the test and enables the incubation time to be reduced to ten minutes. The antibody detection procedure is based on the principle of agglutination.

SPECIMEN:

No special preparation of the patient is required before specimen collection. Blood should be collected by an approved technique, with or without an anticoagulant. If not tested immediately, the sample should be stored at 2° to 8° C. Antibodies dependent for their detection upon the binding of complement may not be detected if plasma from an anticoagulated sample is used for antibody detection tests.

REAGENTS:

- 1. Reagent Red Blood Cells for Detection of Unexpected Antibodies,
- 2. N-HANCE for Antibody Detection Tests
- 3. Anti-Human Globulin Anti-IgG.-C3d; poly specific or Anti-IgG
- 4. Coombs Control Cells (ElgG, Strong or ElgG, Weak)

Store all reagents at 2° to 8°°C. May be left at room temperature (up to 30°C) while in use. Do not use beyond expiration date.

QUALITY CONTROL:

Reagent Red Blood Cells must be examined daily for hemolysis or color change and tested daily with appropriate controls.

PROCEDURE:

- 1. Label an appropriate number of test tubes for each reagent cell. An autologous control is not required in antibody screening, but can be set up optionally.
- 2. Thoroughly mix each of the vials of antibody screening cells in turn, and add one drop of the suspended cells to each appropriate tube in sequence.
- 3. Deliver 2 drops of patient serum or plasma to each tube.
- 4. Centrifuge and resuspend the red blood cells by gently shaking and examine for agglutination. Record results.
 - **Note:** The addition of gamma N-Hance may be postponed until immediately before the incubation at 37°C. This is recommended for crosmatching since anti-A and anti-B may not be detected at 37°C in a low-ionic strength medium.
- 5. To all negative tubes add two drops of N-HANCE
- 6. Mix all tubes thoroughly and incubate at 37°C for 10 to 15 minutes. (Incubation may be extended to 30 minutes).
- 7. Examine for hemolysis without centrifugation. (Hemolysis may be an indication of a positive reaction. Antibodies of the Lewis, P, Kidd or Vel systems may cause hemolysis of incompatible cells when the serum being teted is freshly drawn. Hemolysis will probably not be seen if plasma is being tested.)



Policy Name: Antibody Screen - N-HANCE

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Blood Bank-Lab

Departmental Review:

Policy #: B1.7

INITIATE DATE

DATE REVIEWED/REVISED 4/2015

PAGE 2 of 4

- 8. Wash the cells in all tubes at least three times. The tubes must be filled with saline, the saline must be carefully decanted after each wash, and the cells must be thoroughly resuspended when adding saline for the next wash. Decant the saline completely after last wash.
- 9. Add one or two drops of Anti-Human Globulin in accordance with manufacturer's directions.
- 10. Mix well and centrifuge.
- 11. Resuspend the cells by gently shaking the tube.
- 12. Examine the agglutination and record test results.
- 13. Interpret test results immediately upon completion of the test.
- 14. To all tests interpreted as negative, add one drop of Coombs Control Cells.
- 15. Centrifuge and examine again for agglutination.

REPORTING RESULTS:

- 1. Hemolysis or agglutination of one or more individual screening cell suspensions is a positive test result and indicates the presence of an antibody directed against an antigen present on the cells.

 NOTE: Antibody identification panel must be performed to confirm a positive antibody screen. SoftBank will automatically change the immediate spin crossmatch to AHG crossmatch.
- 2. No agglutination or hemolysis or any screening cell suspension is a negative test result and indicates that no antibody directed at an antigen present on the cells is being detected.
- 3. Notify nursing of positive screen and delay in testing for compatible units. Notification must be documented in SoftBank (added in the comment box).

PROCEDURE NOTES:

- 1. Agglutination occurring after the addition of Coombs Control Cells to a test interpreted as negative assures that active Anti-Human Globulin is present in the test mixture, and that the negative test result is valid.
- 2. No agglutination after the addition of Coombs Control Cells to a test interpreted as negative indicates the Anti-Human Globulin was either inactivated or omitted. The negative test result is invalid and the test must be repeated.
- 3. The identity of an unexpected antibody detected during screening may be determined by testing the serum or plasma against a panel of red blood cells and matching the reaction pattern of the serum or plasma against the antigen profiles of the panel.
- 4. Complement-binding antibodies may not be detected if plasma is used for the test, or if Anti-IgG is used instead of Anti-IgG,-C3d; Poly specific.
- 5. Antibodies may be present in a serum or plasma but not be detected if their level of potency is below the threshold of detectability by the test procedure used.



Policy Name: Antibody Screen - N-HANCE

Department:

Blood Bank-Lab

Departmental Review:

Policy #: B1.7

INITIATE DATE

DATE REVIEWED/REVISED 4/2015 PAGE 3 of 4

6. An Autologous control or DAT is not required or recommended as part of routine pretransfusion testing. An Autologous control, however, is of value when performing antibody identification. Autologous control is not the same as DAT.

REFERENCES:

- 1. AABB Technical Manual
- 2. Immucor, Inc, Norcross, Ga, Package Inserts:
 - a. Anti-Human Globulin Serum, (Anti-IgG-C3d), green
 - b. Anti-IgG (Murine Monoclonal)(Green or Uncolored) Gamma-clone
 - c. Gamma PeG Polyethylene Glycol Additive for Antibody Detection Tests



$\begin{array}{c} \text{HIGHLANDS REGIONAL MEDICAL CENTER} \\ \text{Sebring, FL} \\ \textit{Laboratory} \end{array}$

DOCUMENT CHANGE RECORD

Document Name:	Blood Bark	
Document Section:	Blod Barle	
Author: Meri	tel Porofule	
Please circle one of the	following: NEW	EVISION ARCHIVE
Effective		
Description of docume	ent, changes, and rationale: Roca	hural sits added (#16)
Formal Training of sta	off required: No	
Attach email sent to st	aff about new procedures or changes t	o procedure if applicable
Method Validation req	uired (attach documents):	
List any changes to the	Lab Information system: the	
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Review and	Signature	Date
Approval		
Author	Spritt Proful	\$ -30.2015
Chief Technologist	U	4
Admin. Lab Director		

Laboratory Director

Implementation occurs after signature by Laboratory Director.



Policy Name: Antibody Screen - N-	HANCE Department	: Blood Bank-Lab
Departmental Review:	Poli	cy #: B1.7
INITIATE DATE	DATE REVIEWED/REVISED 4/2015	PAGE 4 of 4
Reviewed by Reviewed Da	te Reviewed by	
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Initial Implementation Date:	•	p
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Reason:		
Reviewed by:Sign	atures on file	Date:
Department Supervi	sor	-
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Department Adm. D	irector	
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