University of Washington Medical Center 1959 NE Pacific Street. Seattle, WA 98195 Transfusion Services Laboratory Policies and Procedures Manual	Original Effective Date: 03-11-16 Revision Effective Date:	Number: PC-0036.03
TITLE: **Testing and Provision of Hemoglobin S Negative Blood**		

PURPOSE:
To provide instructions for performing sickle cell testing on red blood cell components using the SickleDex kit and controls and provide hemoglobin S negative components

PRINCIPLE & CLINICAL SIGNIFICANCE:
Principle
SickleDex is a qualitative solubility test kit used to detect the presence of sickling hemoglobins (HbS) in blood or control material. *SickleDex* uses saponin to lyse the red blood cells. Sodium Hydrosulfite then reduces the released hemoglobin. Reduced HbS is insoluble in the concentrated phosphate buffer and forms a cloudy, turbid suspension.

Clinical Significance
Transfusion of units negative for hemoglobin S reduces tissue hypoxia in patients with Sickle Cells Disease (SCD) and reduces the risk of cellular breakdown from the hemoglobin S positive RBCs in neonates

POLICIES:
- HbS negative red blood cells components shall be provided to patients for the following:
  - Sickle cell disease, sickle cell trait and thalassemia, Diamond Black-fan anemia
  - Red blood cell exchanges (adult and neonatal)
  - Children < 4 months old and intrauterine transfusions
- SCD patients must be flagged in Sunquest with the NHBS attribute to prevent release of untested units. Refer to SOP: *Blood Administrative Data Entry Updates*

SPECIMEN REQUIREMENTS:
Blood from donor segments stored at 1-6°C for up to 45 days

REAGENTS/SUPPLIES/EQUIPMENT:
Reagents:	Supplies:	Equipment:
• SickleDex Kit • Sickle-Chex Controls	• 12 x 75 mm test tubes • Adjustable pipette • Disposable pipette tips • Blood bank transfer pipettes	Test tube rack with lines

QUALITY CONTROL:
Quality control is performed with each batch of testing

TRAINING

INSTRUCTIONS:

TABLE OF CONTENTS:

Working Solubility Buffer Preparation

STEP	ACTION
1	Bring buffer and reagent powder from the SickleDex Kit to room temperature before mixing
2	Add contents of one vial of SickleDex Reagent Powder to one bottle of SickleDex Solubility Buffer
3	Place a white dispenser cap on the bottle of working solubility buffer
4	Agitate the buffer vigorously until the reagent powder is dissolved
5	Label the buffer container with the following after reconstitution: • Reconstitution date • 45-day expiration date • Tech ID

SickleDex Testing

STEP	ACTION
1	Label 12 x 75 mm tubes for each donor unit to be tested, 1 positive control and 1 negative control
2	Allow controls and buffer to warm to room temperature (18°C to 30°C) for approximately 15 minutes before use
3	Place tubes in testing rack and fill each tube with working SickleDex Solubility Buffer to the red line on rack (approximately 2 mL) **NOTE:** Return buffer to refrigerator immediately after use
4	Mix controls by holding vertically between hands and rolling the vials back and forth for 20-30 seconds followed by inverting end over-end 20 times. **NOTE:** Visually inspect the bottom of the vial to ensure all cells are suspended
5	Add 1 drop of each control into the appropriate labeled control tube **NOTE:** The control MUST be inverted and held vertically directly over the test tube to ensure accurate delivery. Wipe threads on each control before returning cap, if necessary
6	Add 20uL of whole blood and swirl the contents to mix **NOTE:** If donor segment does not appear to contain whole blood (due to settling prior to sealing), use 10uL of packed red blood cells for testing
7	Allow tubes to stand in the testing rack at room temperature for at least 6 minutes and no longer than 60 minutes

TRAINING

<table>
<tr><th>STEP</th><th>ACTION</th></tr>
<tr><td>8</td><td>Read reaction macroscopically by looking through the test tubes at black lines on the back of the testing rack
<table>
<tr><td>If result read at 6mins is</td><td>Then</td></tr>
<tr><td>Negative</td><td>Incubate sample for a total of 60 mins to confirm negative result</td></tr>
<tr><td>Positive</td><td>Further incubation is not required</td></tr>
</table></td></tr>
<tr><td>9</td><td>Go to section Result Reporting</td></tr>
</table>

CALCULATIONS/INTERPRETATIONS/RESULTS REPORTING/NORMAL VALUES/CRITICAL VALUES

Interpretation:

If black tube rack lines are	**Interpret as**
CLEARLY VISIBLE thru a transparent suspension	Negative
NOT VISIBLE thru a cloudy, turbid suspension	Positive

Results Reporting in Sunquest

<table>
<tr><th>STEP</th><th>ACTION</th></tr>
<tr><td>1</td><td>Open ‘Blood Order Processing’ function</td></tr>
<tr><td>2</td><td>Assign unit to patient using ‘Blood Inventory Search’ or by scanning unit barcode</td></tr>
<tr><td>3</td><td>QA Warning window will open with warning message about missing NHBS attribute on unit
Check the box on the left to acknowledge QA Warning for each unit
<table>
<tr><th>Acknowledge</th><th>Unit</th><th>Comp Type</th><th>Div #</th><th>Warning message</th><th>Have authority to override?</th></tr>
<tr><td>☐</td><td>W141605200006</td><td>E0382</td><td>00</td><td>Patient/unit attribute incompatibility
Patient's Attribute(s): NHBS
Attribute(s) Missing on Unit: NHBS</td><td>Yes</td></tr>
</table></td></tr>
<tr><td>4</td><td>Order a sickle cell test on the allocated unit
Enter ;SCKL in the ‘Add unit test’ box at the bottom of the screen</td></tr>
<tr><td>5</td><td>Enter the reactions according to the following tables:
<table>
<tr><td></td><td>SKP</td><td>SKN</td><td>SKU</td></tr>
<tr><td>SCKL</td><td>+</td><td>-</td><td>+ or -</td></tr>
</table>
SKP = Sickle Positive Control; SKN = Sickle Negative Control; SKU = Unit Tested</td></tr>
<tr><td>6</td><td>Enter the Interpretation in the interpretation grid:
<table>
<tr><th>Interpretation</th><th>SQ Result</th><th>Sunquest Hot Key</th></tr>
<tr><td>Positive</td><td>POSHBS</td><td>s</td></tr>
<tr><td>Negative</td><td>NGHBS</td><td>n</td></tr>
</table></td></tr>
</table>

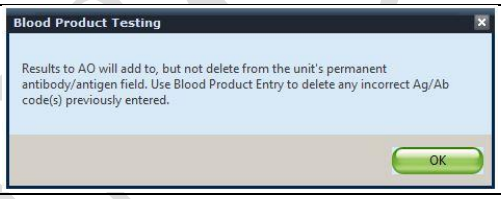
TRAINING

<table>
<tr><td>7</td><td>

Click <Save>
Override QA Failure by adding a ‘TDONE’ comment
In the free text box, add ‘Sickle Cell Testing’
Click <OK> to acknowledge unit failure on the Electronic Crossmatch Eligibility Report

</td></tr>
<tr><td>8</td><td>Open ‘Blood Product Testing’ function</td></tr>
<tr><td>9</td><td>

Scan unit information
Click <Continue>

</td></tr>
<tr><td>10</td><td>Order ‘;AO’ in the ‘Unit Testing’ field
<table>
<tr><th>If SCKL testing is</th><th>Then</th></tr>
<tr><td>POSITIVE</td><td>Result with ;POSHBS</td></tr>
<tr><td>NEGATIVE</td><td>Result with ;NHBS</td></tr>
</table>
</td></tr>
<tr><td>11</td><td>Click <OK> to acknowledge the Blood Product Testing Comment


</td></tr>
<tr><td>12</td><td>Click <Save> and <Exit></td></tr>
<tr><td>13</td><td>Go to ‘Blood Order Processing’
<table>
<tr><th>If SCKL testing on unit is</th><th>Then</th></tr>
<tr><td>NEGATIVE</td><td>Go to next step</td></tr>
<tr><td>POSITIVE</td><td>In the Compatibility Testing area

Uncheck ‘Use reaction grid results’ box
Enter ‘;ND’ in the ‘XM’ field
Enter ‘]’ in the ‘TS’ field for ‘NOK’
Go to step 15

NOTE: Unit will automatically be released from patient</td></tr>
</table>
</td></tr>
<tr><td>14</td><td>Enter comment code on unit to display the status on the Transfusion Record

Enter; CM in the ‘Add unit test’ box at the bottom of the screen
Enter the following code in the CM box; NHBS

</td></tr>
<tr><td>15</td><td>
<table>
<tr><th>If performing</th><th>Then</th></tr>
<tr><td>Electronic crossmatch</td><td>Click <Save></td></tr>
<tr><td>Serologic crossmatch (adult)</td><td>Enter results according to appropriate Crossmatch SOP then click <Save></td></tr>
<tr><td>TNRBC (Neonate crossmatch)</td><td>Answer ‘TS’ Box
Click <Save></td></tr>
</table>
</td></tr>
<tr><td>16</td><td>Continue to Blood Product Issue if issuing unit</td></tr>
</table>

CALIBRATION:
NA

TRAINING

PROCEDURE NOTES AND LIMITATIONS:

- Sickle –Chex controls expire 100 days after open or manufacturer's expiration, whichever date is shorter
- Donors with Hgb S concentrations less than 30.9% may not be detected

REFERENCES:

SickleDex Manufacturer Insert. Omaha, Nebraska: Streck; current version
Sickle-Chek Manufacturer Insert. Omaha, Nebraska: Streck; current version

RELATED DOCUMENTS:

APPENDIX:

NA

TRAINING

UWMC SOP Approval:			
UWMC CLIA Medical Director	Mark H. Wener, MD	Date	
Transfusion Service Manager	Nina Sen	Date	
Compliance Analyst	Christine Clark	Date	
Transfusion Service Medical Director	Monica B. Pagano, MD	Date	
UWMC Biennial Review:			
		Date	
		Date	

REVISION HISTORY:
04/16/18: Updated instructions for mixing controls to match revision to package insert.

06/29/2021: Added incubating negative results for 60 minutes per change in package insert and policy for providing HbS negative RBCs to Diamond-Blackfan anemia patients

TRAINING