|  |  |  |  |
| --- | --- | --- | --- |
|  | **Titrations**  BB.SP.1004.11 | **Dept:**  | 324311 |
| **Dept Name** | Blood Bank |
| **Effective Date:** | 8/4/10 |
| **Revised Date:** | Title 21 |
| **Name & Title**: CLIA Laboratory Medical Director | **Contact:** | JHSimmons/CS Warren |
| **Signature:**  | Refer to Title 21 | **Date:** | **Title 21** |

****

**1. General Procedure Statement:**

1. **Purpose:**

This procedure explains the proper way to make serial twofold dilutions necessary to perform antibody titrations, how to properly perform prenatal, parallel prenatal, transplant, QC, isohemagglutinin, CAP, and HTLA titrations using both tube and gel media.

 **B.** **Responsible Department/Scope:**

 i. Procedure owner/Implementer: Julie H. Simmons/Christina S. Warren

 ii. Procedure prepared by: Julie Jackson

 iii. Who performs procedure: Department staff/management

  **C. Definitions:**

Titration: Semi quantitative method using serial twofold dilutions of serum/plasma to determine the antibody concentration

AHG: Anti-human globulin

Coombs: Either IgG or polyspecific sera for AHG testing

HTLA: High titer low avidity antibody

LISS: Low ionic strength solution

PEG: Polyethylene glycol

DTT: Dithiothreitol

QC: Quality control

RT: Room temperature

Kidney Desensitization: Lowering the isohemagglutinins titer of patient to accept an incompatible donor kidney

BMT: Bone Marrow Transplant

ABOi: ABO Incompatible (referring to kidney transplants)

PUBS: Percutaneous Umbilical Cord Blood Sampling (also called cordocentesis), is a blood sample collected on an unborn infant.

SCC: Soft Computer Company; WFBMC’s Blood Bank computer system.

 For a list of tests see *Attachment 9: Titer Tests in SCC*

**D. Sections:**

1. Protocols:
	1. General Titration Protocol Statement
	2. Prenatal Titrations Protocol
	3. HTLA Titration Protocol
	4. Kidney Donor and Kidney Patient
	5. BMT PATIENT AND DONOR PROTOCOL
	6. QC antiserum
	7. CAP Surveys
	8. Internal assessment
	9. PUBS Titers
2. Preparation of Master Dilutions
3. Gel Testing Serum/Plasma Dilutions IgG Routine
4. Gel Testing Serum/Plasma Dilutions IgM Kidney
5. DTT Treatment of Plasma/Serum for IgG Kidney
6. Tube Testing Serum/Plasma, HTLA and QC Dilutions
7. Interpretation of Titer Results
8. Computer Entry for Antibody Titer (excluding BMT and Kidney ABO titers)
9. Computer Entry for Antibody Titer (BMT sample ABO titers)
10. Computer Entry for Antibody Titer (ABOi Kidney sample ABO titers)
11. Preparation of 0.01M DTT for Distinguishing IgM and IgG Antibodies

**Section I. Titration Protocols**

1. **General Titration Protocol Statement:**
2. Technical variables greatly affect the results and caution should be exercised to achieve the most uniform possible practices. Variables can occur with the below:
* Pipetting – use disposable tips that are changed after each dilution.
* Media – continue testing any future titration tests on patient with the original media started.
* Time of incubation – saline is 60 minutes, PEG/LISS is 30 minutes, gel is 40 minutes, solid phase automated 30 minutes test.
* Temperature of incubation is:

IgG: 35.5C to 38.5C.

IgM: 20-24 C (Kidney Patients)

* Time and Force of centrifugation – HIGH at 3600RPM (3500-3700 RPM) Time based on QC
* Cells used for titering – choose the same donor ID number, cells concentration, phenotype,

 as fresh as possible for all titrations.

1. All titrations are entered into SCC except HTLA and QC titers.
2. During downtime, titrations will be recorded on the following forms until computer system is back up and then results will be entered:

***Attachment 4: Antibody Titer Form*** or ***Attachment 8: Kidney Antibody Titer Form***

* Report any previous titer on same episode, date tested, antibody, media, and titer.
1. Do not report any scoring
2. Do not test any parallel titers unless results are more or less than 1 tube dilution difference from previous titer.
3. Titers must be reviewed by Medical Director.
	1. A consultation fee is charged by management with the Action BOUT.
4. In cases of cold antibodies and antibodies with complement antibody activities, titrations will be a guide to determining the severity of the outcome at 4C.
5. Clinical findings indicate hemolysis
6. Thermal amplitude test is negative.
7. Cold auto antibody detected optimally at 4C.
8. **All** antibody titer samples must have all other clinically significant antibodies ruled out each time a titer is requested.
	1. Perform a selected screen/panel to rule out all other antibodies.
	2. If there are multiple antibodies the cells used in the titer must be positive (homozygous) for the antigen being tested and negative for all other antigens to the associated antibody(ies).
		1. Example: Anti-A is being titered. The patient has an Anti-K.
			* The cells used to titer the anti-A must be A positive and Kell negative.
		2. Example: Anti-D and anti-K are being titered.
			* The cells for titering anti-D must be D positive and Kell negative.
			* The cells for titering anti-K must be Kell positive and D negative.
9. **Prenatal Titration Protocol**
10. When an antibody known to cause Hemolytic Disease of the Newborn is identified, titration studies may contribute to the decision when to perform more complex invasive investigation of fetal condition such as an Intrauterine Transfusion.
11. Titers are not repeated on previously titered specimens with one exception:
12. If the current titer and the last previous titer is more or less than 1 dilution tube titer difference.
13. Examples:

|  |  |  |  |
| --- | --- | --- | --- |
| **Current titer** | **Previous titer** | **Conclusion:** | **Action:** |
| 4 | 1 | difference is 2 titer tubes | 1. Repeat current and previous titers
2. Review with medical director on what to report
3. Review with management on how to report in SCC
 |
| 8 | 16 | difference is 1 titer tube | 1. Report the current titer
 |

1. To determine the correct test to order in SCC refer to *Attachment 9: Titer Tests in SCC*
2. Cell Selection
	1. Use homozygous expression for antibody to be titered. Example: anti-D use R2R2 cells for titer.
	2. Identify cells by manufacturer, lot number and cell number.
	3. Attempt to select the same cell if in dated.
	4. Search in Antigen Plus for cell number.
	5. Use only select cells that are in dated.
	6. If same cell number is not available or expired, select another cell with same homozygous expression needed
	7. Rare low frequency antigens may necessitate using the father’s red cells in titration if it is confirmed that he carries the antigen to the mother’s antibody.

**NOTE:** Freeze the father’s RBCs for subsequent titers ordered.

1. Titer all antibodies detected in prenatal women by **GEL METHOD** using 40 minutes at 37C incubation and carry to IgG coombs.
	1. DO NOT use ficin treated cells in gel for titer unless directed by management.
	2. Do not titer antibody in another media (use Gel only) even though the antibody may react optimally in another media.
2. Do not titer the antibodies currently detected in prenatal women below unless directed:
	1. RHIG preparations
	2. Lewis antibodies
	3. P 1 antibodies
	4. cold auto-antibodies
	5. HTLA antibodies
	6. Warm auto-antibodies with undetermined specificity
	7. DAT testing is:
		1. Not required on obstetric patients that have new antibodies unless transfused.
		2. Required if there is a previous history of a warm or pathological cold autoantibody.
	8. Do not titer antibodies
		1. Previously detected during pregnancy episode and now are not detectable in gel method.
		2. Antibodies previously identified or new antibody identified at time of hospital admission to Labor & Delivery.
	9. For patients with previous titers during same pregnancy, do the following:
		1. Look up the previous titers along with dates and record on the Antibody Titer form.
		2. Perform titration on the current blood specimen.
		3. If the current titer is within + 1 dilution tube with the last previous titer or there is no previous titer, report current titer.
		4. If the current titer is NOT within + 1 dilution tube with the last previous titer, repeat both the current and previous titer. For current titer, prepare master dilutions again.
	10. Record titer endpoint:
3. Record whole number - not number 1 and semicolon (**1:)**

 Example: Correct titer interpretation: 8

 See example below in 2.10.b

 Example: Incorrect titer interpretation: 1:8

1. Record titer endpoint as **Negative** when serological reactions at 1:1 are not present or Wk+.
	1. The titer endpoint is the last reaction at 1+. Wk+ is considered negative.

Example:

|  |  |  |  |  |  |  |  |
| --- | --- | --- | --- | --- | --- | --- | --- |
| **1:1** | **1:2** | **1:4** | **1:8** | **1:16** | **1:32** | **1:64** | **Interp** |
| 0 | 0 | 0 | 0 | 0 | 0 | 0 | NEG |
| Wk+ | 0 | 0 | 0 | 0 | 0 | 0 | NEG |
| 2+ | 1+ | 1+ | 1+ | Wk+ | 0 | 0 | 8 |

1. **Note:** a significant difference in titers is three or more dilutions.
	1. Freeze aliquots of serum/plasma at -25C to -35C in the appropriate storage box to be retain 1 yr.
	2. Storage box labeled: OBX.
	3. Samples are saved in order by month of collection as labeled on storage box.

**3.0** **HTLA Titration Protocol**

1. SCC Order: TOAB1, TOAB2 or TOAB3
	1. Refer to ***Attachment 5: Antibody Titer Flowchart***
2. Most weakly reactive antibodies lose reactivity when diluted even modestly; however, some antibodies that give weak reactions when undiluted continue to react at high dilutions (i.e. 1:2048.)
	1. Examples of some antibodies: anti-Ch, -Rg, -Csa, -Yka, -McCa, -JMH
3. Select cells in the media of testing that indicate that a “high titer low avidity” antibody may exist.
4. For HTLA titers: observe the highest dilution that produces any agglutination MICROSCOPICALLY.
5. The media that demonstrates the reaction that is suspected of HTLA should be selected to perform the titer. The routine method is incubation at 37C for 60 minutes followed by AHG if not specified.
6. Use **Poly Antiglobulin coombs**
7. **ONLY READ ENDPOINT MICROSCOPICALLY** for tube testing.
8. HTLA= High Titer Low Avidity:

Endpoint titer will titer consistently the same grade 3-5 or more tubes out, often 1+ or weaker to microscopically. The last tube giving a positive reaction *microscopically* is the endpoint.

1. The titration is part of the antibody identification.
2. HTLA Titers are not resulted in SCC. To charge for the titer order and complete the following Action:
	1. HTLAT (HTLA Titer Charge)

**4.0 Kidney Donor and Kidney Patient**

*Refer to Protocols: Transplant Testing Protocols, Section I: Kidney Transplant Testing*

**5.0 BMT Patient and Donor Protocol**

5.1 BMT Titrations

*Refer to Protocols: Transplant Testing: Section II*

5.2 BMT DONOR PROTOCOL

*Refer to Protocols: Transplant Testing: Section II*

5.3 BMT PATIENT (RECIPIENT) PROTOCOL

*Refer to Protocols: Transplant Testing: Section II*

**6.0 QC antiserum**

 Dilution of antibody for Daily QC-

 *BB.Forms.2079: Blood Bank – Preparation of Diluted Antisera for Daily QC*

*BB.Forms.1002: Antibody Titer Form*

1. Methods
2. For 2-5% testing, use PEG at 37C for 15 minutes and carry to AHG.
3. For 0.8% testing, use gel at 37C for 15 mins.
4. For all test cells, use appropriate cell suspension for test method
5. Endpoint for Daily QC antisera is 2+ not 1+.
6. Definition of 2+ macroscopic tube reactions: medium sized aggregates with a clear back ground.

**7.0 CAP Surveys**

1. All CAP survey testing in ABT surveys will be **40 minutes GEL phase at 37C.**
	1. Do not use ficin treated cells

**8.0 Internal assessment- HTLA antibodies**

1. Internal assessment of the ABT will be tested: Saline 60 minutes at 37C and proceed to AHG (macroscopic and microscopically) using poly specific AHG.
2. Read results at AHG phases for any microscopic reactions
3. If the CAP ABT survey is used for internal assessments, it will be performed AFTER the due date of the CAP survey.

**9.0 PUBS titers**

1. The need for a PUBS titer will be determined by Blood Bank management or Blood Bank Medical Director.
2. The titer will be added onto the PUBS TSX sample in SCC
	1. Patient > Orders > Modify
		1. For test name refer to *Attachment 9: Titer Tests in SCC*
3. Titer all antibodies by **GEL METHOD** using 40 minutes at 37C incubation and carry to IgG coombs.
	1. DO NOT use ficin treated cells in gel for titer unless directed by management.
	2. Do not titer antibody in another media (use Gel only) even though the antibody may react optimally in another media.
4. If the mother has had previous titers during same pregnancy, do the following:
	1. Test a selected screen/panel to rule out other antibodies present.
	2. Look up the previous titers along with dates and record on the

*Antibody Titer form: BB.Forms.1002*.

* 1. Perform titration on the current PUBS blood specimen.
1. Cell Selection
	1. Use homozygous expression for antibody to be titered. Example: anti-D use R2R2 cells for titer.
	2. Identify cells by manufacturer, lot number and cell number.
	3. Search in Antigen Plus for cell number.
	4. Use only select cells that are in dated.
	5. Attempt to select the same cell used for mother’s last titer if in dated.
		1. If same the cell number (from mother’s last titer) is not available or expired, select another cell with same homozygous expression needed
	6. Rare low frequency antigens may necessitate using the father’s red cells in titration if it is confirmed that he carries the antigen to the mother’s antibody.

**NOTE:** Freeze the father’s RBCs for subsequent titers ordered.

1. Record titer endpoint:
	1. Record whole number - not number 1 and semicolon (**1:)**

 Example: Correct titer interpretation: 8

 See example below in 9.6.b

 Example: Incorrect titer interpretation: 1:8

* 1. Record titer endpoint as **Negative** when serological reactions at 1:1 are not present or Wk+.
		1. The titer endpoint is the last reaction at 1+. Wk+ is considered negative.

Example:

|  |  |  |  |  |  |  |  |
| --- | --- | --- | --- | --- | --- | --- | --- |
| **1:1** | **1:2** | **1:4** | **1:8** | **1:16** | **1:32** | **1:64** | **Interp** |
| 0 | 0 | 0 | 0 | 0 | 0 | 0 | NEG |
| Wk+ | 0 | 0 | 0 | 0 | 0 | 0 | NEG |
| 2+ | 1+ | 1+ | 1+ | Wk+ | 0 | 0 | 8 |

* 1. **Note:** a significant difference in titers is three or more dilutions.

**2. Procedure:** **Section II: Preparation of Master Dilutions**

Chemical Risk Assessment: none

Biological Risk Assessment: low

Protective Equipment: Lab coat, gloves

Supplies: 12x75mm and 10x75mm test tubes, MLA pipette tips, permanent marker,

Reagents: 0.85% Saline, 3-5% red cell suspension, 0.8% red cell suspension,

Equipment: 250ul MLA pipette, cell washer, agglutination viewer, microscope

Special Requirements: Plasma from EDTA pink top tube, serum from red top tube, plasma from donor unit drawn in CPDA1 unit diluted with preservative

| **STEPS** | **INSTRUCTIONS** | **CHANGE/****APPROVAL** |
| --- | --- | --- |
| **1.0** | **Label eleven (11) 12 x 75 mm test tubes.*** 1. Record patient last name.
	2. Consecutively label tubes 1 through 11.
	3. Label 11th tube “SAVE”—to be used for further dilutions if required
	4. For titers requiring readings after RT incubation and after the addition of AHG (such as isohemagglutination titrations) two separate sets of tubes must be set up for testing—one for each phase of reactivity. DO NOT CARRY TITER FROM ROOM TEMPERATURE TO 37ºC → AHG.
 |  |
| **2.0** | **Retrieve MLA pipette and set the gauge on pipette to the 250ul setting.**1. When plasma is insufficient to make master titer with 250ul of plasma at 1:1 and 1:2, determine the volume available and review with management.
 |  |
| **3.0** | **Add 250ul saline to tubes 2 thru 10 with an MLA pipette.** |  |
| **4.0** | **Add 250ul of serum/plasma to tube (1:1) and tube 2 (1:2) using an MLA pipette.**1. Wipe pipette tip before adding serum/plasma to tube 2.
2. Dispense directly into bottom of tube when adding plasma/dilutions to tubes, do not allow plasma to run down the side of the tube.
 |  |
| **5.0** | **Mix tube 2 very well by shaking and discard pipette tip.** |  |
| **6.0** | **Using a clean pipette tip, transfer 250ul from tube 2 (1:2) to tube 3(1:4).**6.1 Wipe pipette tip after aspiration from tube 2 before dispensing carefully into bottom of tube 3.  |  |
| **7.0** | **Mix tube 3 very well by shaking and discard pipette tip.** |  |
| **8.0** | **Using a clean pipette tip, transfer 250ul from tube 3 (1:4) to tube 4 (1:8).**1. Wipe pipette tip after aspiration from tube 3 before dispensing carefully into bottom of tube 4.
 |   |
| **9.0** | **Mix tube 4 very well by shaking and discard pipette tip.** |  |
| **10.0** | **Using a clean pipette tip, transfer 250ul from tube 4(1:8) to tube 5 (1:16).**10.1 Wipe pipette tip after aspiration from tube 4 before dispensing carefully into bottom of tube 5.  |  |
| **11.0** | **Mix tube 5 very well by shaking and discard pipette tip.** |  |
| **12.0** | **Using a clean pipette tip, transfer 250ul from tube 5(1:16) to tube 6 (1:32).**12.1 Wipe pipette tip after aspiration from tube 5 before dispensing carefully into bottom of tube 6.  |  |
| **13.0** | **Mix tube 6 very well by shaking and discard pipette tip.**  |  |
| **14.0** | **Using a clean pipette tip, transfer 250ul from tube 6 (1:32) to tube 7 (1:64).**14.1 Wipe pipette tip after aspiration from tube 6 before dispensing carefully into bottom of tube 7.  |  |
| **15.0** | **Mix tube 7 very well by shaking and discard pipette tip.** |  |
| **16.0** | **Using a clean pipette tip, transfer 250ul from tube 7 (1:64) to tube 8 (1:128).**16.1 Wipe pipette tip after aspiration from tube 7 before dispensing carefully into bottom of tube 8.  |  |
| **17.0** | **Mix tube 8 very well by shaking and discard pipette tip.** |  |
| **18.0** | **Using a clean pipette tip, transfer 250ul from tube 8 (1:128) to tube 9 (1:256).**18.1 Wipe pipette tip after aspiration from tube 8 before dispensing carefully into bottom of tube 9.  |  |
| **19.0** | **Mix tube 9 very well by shaking and discard pipette tip.** |  |
| **20.0** | **Using a clean pipette tip, transfer 250ul from tube 9(1:256) to tube 10(1:512).**20.1 Wipe pipette tip after aspiration from tube 9 before dispensing carefully into bottom of tube 10.  |  |
| **21.0** | **Mix tube 10 very well by shaking and discard pipette tip.** |  |
| **22.0** | **Using a clean pipette tip, transfer 250ul from tube 10(1:512) to a labeled tube for future dilutions.**22.1 Wipe pipette tip after aspiration from tube 10 before dispensing carefully into bottom of “SAVE” tube.  |  |
| **23.0** | **Set “Save” tube (11th tube) aside.**23.1 Should additional dilutions be necessary, add 250ul of saline to the “SAVE” tube. a. Dilution is now 1:1024.23.2 Set up additional labeled tubes for further dilutions: 2048, 4096, 8192 and >81921. Label with patient’s last name and dilution

23.3 Add 250ul saline to each tube.23.4 Proceed with serial titration procedure as in previous steps.  MC900056715[1] |  |
| **24.0** | **Ready for testing.**24.1 For IgG gel testing (BMT, OB, Kidney, PUBS): Go to *Section III: Gel Testing Serum/Plasma Dilutions IgG*24.2 For IgM gel testing (Kidney) Go to *Section IV: Gel Testing Serum/Plasma Dilutions IgM Kidney.*24.3 For DTT Treatment of Plasma/Serum to differentiate IgG and IgM: Go to *Section V: DTT treatment of Plasma/Serum for IgG Kidney.***NOTE: When testing with DTT Treated Plasma/Serum – the initial tube is a dilution of 1:2. Record results on correct section of Kidney Antibody Titer form.** 24.4 For tube testing: *Go to Section VI: Tube Testing Serum/Plasma, HTLA and QC Dilutions.* |  |

**Section III: Gel Testing Serum/Plasma Dilutions IgG Routine**

Chemical Risk Assessment: none

Biological Risk Assessment: low

 Protective Equipment: Lab coat, gloves

Supplies: MLA pipette tips, permanent marker, Ortho IgG gel cards, micropipette tips

Reagents: 0.8% red cell suspension,

Equipment: 250ul MLA pipette, cell washer, agglutination viewer, microscope, micropipette, Ortho Workstation

Special Requirements: Plasma from EDTA pink top tube, serum from red top tube, plasma from donor unit drawn in CPDA1 unit diluted with preservative

| **STEPS** | **INSTRUCTIONS** | **CHANGE/****APPROVAL** |
| --- | --- | --- |
| **1.0** | ***Gel is the routine method that is used for titers.*** |  |
| **2.0** | **Label eleven (11) microtube wells appropriately.**1. Record patient last name.
2. Well 11 is the saline control well.
 |  |
| **3.0** | **Remove foil seal on wells 1 through 11. Leave unused wells covered.**  |  |
| **4.0** | **Using the appropriately calibrated micropipette, dispense 50ul of 0.8% suspension cells into each well.** 1. Selection of cells

a. For BMT transplants, use fresh A1 and/or B cells.b. For incompatible kidney recipients (Group B or Group O), use A2 cellsc. For QC refer to procedure *Daily QC:BB.QC.1006* d. See Section I. Protocols |  |
| **5.0** | **Using a different clean micropipette tip for each master dilution, add 25ul of each dilution into the appropriately labeled well and 25ul of saline into well 11 (saline control.)** 1. Refer to *section II: Preparation of Master Diluents*
 |  |
| **6.0** | **Incubate at 37°C ± 1.5°C for 40 minutes.** |  |
| **7.0** | **Centrifuge the required 10 minutes in the Ortho Workstation centrifuge.** |  |
| **8.0** | **Read the gel card titer starting with well 11.** 8.1 Read the front only of each well. |   |
| **9.0** | **Record results on the correct Antibody titer form:**9.1

|  |  |
| --- | --- |
| **Titer Type** | **Form** |
| OB, PUBS, BMT, CAP, QC | Antibody Titer form*Refer to Attachment 4: Antibody Titer form.* |
| Kidney Patient | Kidney Antibody Titer form *Refer to Attachment 8: Kidney Antibody Titer form*  |

* 1. Go to *Section VII for Interpretation of Titer Results.*
	2. Exception: For QC refer to the procedure *Daily QC:BB.QC.1006*

**MC900056715[1]** |  |

**Section IV: Gel Testing Serum/Plasma Dilutions IgM Kidney**

Chemical Risk Assessment: none

Biological Risk Assessment: low

 Protective Equipment: Lab coat, gloves

Supplies: MLA pipette tips, permanent marker, **Ortho NEUTRAL** gel cards, micropipette tips

Reagents: 0.8% red cell suspension,

Equipment: 250ul MLA pipette, micropipette, Ortho Workstation

Special Requirements: Plasma from EDTA pink top tube, serum from red top tube, plasma from donor unit drawn in CPDA1 unit diluted with preservative

| **STEPS** | **INSTRUCTIONS** | **CHANGE/****APPROVAL** |
| --- | --- | --- |
| **1.0** | ***Gel is the routine method that is used for titers.***  |  |
| **2.0** | **Label eleven (11) microtube wells appropriately.**1. Record patient last name.
2. Well 11 is the saline control well.
 |  |
| **3.0** | **Remove foil seal on wells 1 through 11 of Neutral Gel card. Leave unused wells covered.**  |  |
| **4.0** | **Using the appropriately calibrated micropipette, dispense 50ul of 0.8% suspension cells into each well.** 4.1 Selection of cellsa. For incompatible kidney recipients (Group B or Group O), use fresh A2 cellsb. See Section I. Protocols |  |
| **5.0** | **Using a different clean micropipette tip for each dilution, add 25ul of each dilution into the appropriately labeled well and 25ul of saline into well 11 (saline control.)**  |  |
| **6.0** | **Centrifuge the required 10 minutes in the Ortho Workstation centrifuge to get an immediate spin reading.**  |  |
| **7.0** | **Read the gel card titer starting with well 11.** 7.1 Read the front of each well. |   |
| **9.0** | **Record results on Antibody titer form.** 9.1

|  |  |
| --- | --- |
| **Titer Type** | **Form** |
| Kidney Patient | Kidney Antibody Titer form *Refer to Attachment 8: Kidney Antibody Titer form*  |

* 1. Go to *Section VII for Interpretation of Titer Results*
	2. Determine next step.

|  |  |  |
| --- | --- | --- |
| Titer | Go to: | Perform |
| Group O Recipient evaluation for A2 kidney | Go Section V | DTT Treatment of Plasma/Serum for IgG Kidney |
| Group B Recipient evaluation for A2 kidney | No further testing | No further testing |

**MC900056715[1]** |  |

**Section V: DTT Treatment of Plasma/Serum for IgG Kidney**

Chemical Risk Assessment: none

Biological Risk Assessment: low

 Protective Equipment: Lab coat, gloves

Supplies: MLA pipette tips, permanent marker, Ortho IgG gel cards, micropipette tips

Reagents: 0.8% red cell suspension, 0.01M DTT for Plasma/Serum treatment (Refer to Section IX on preparation)

Equipment: 250ul MLA pipette, micropipette, Ortho Workstation

Special Requirements: Plasma from EDTA pink top tube, serum from red top tube, plasma from donor unit drawn in CPDA1 unit diluted with preservative

| **STEPS** | **INSTRUCTIONS** | **CHANGE/****APPROVAL** |
| --- | --- | --- |
| **1.0** | **Obtain 0.01M DTT.*** 1. Check **Freezer 12** to see if some is available frozen.
	2. Prepare if none available. ***Refer to Section XI: Preparation of 0.01M DTT for Distinguishing IgM and IgG antibodies.***

**NOTE: This is NOT the DTT that is used to treat red cells. This should be 0.01M DTT.** |  |
| **2.0** |

|  |  |  |
| --- | --- | --- |
| **a. Label Tube** | **b. Pipet to labeled tube** | **c. Pipet** |
| MRN+Name+DTT | 1ml of Patient plasma/serum | 1 ml of 0.01M DTT |

**Label tube as indicated in the table below. Tubes will contain 1ml of Patient plasma/serum and will have 1 ml of 0.01M DTT added.** |  |
| **3.0** | **Mix the tubes and incubate at 37C for 30 to 60 minutes.** |  |
| **4.0** | **Prepare a Master Dilution for DTT treated sample.**4.1 Refer to *Section II: Preparation of Master Dilutions.*NOTE: The tubes you are starting with are already diluted 1:2 before beginning  the master dilutions.  *Refer to Attachment 8: Kidney Antibody Titer form.* |  |
| **5.0** | ***Gel is the routine method that is used for titers.*** |  |
| **6.0** | **Label eleven (10) microtube wells appropriately.*** 1. Record patient last name and DTT.
	2. Well 1 is the 1:2 dilution.
	3. Well 11 is the saline control well.
 |  |
| **7.0** | **Remove foil seal on wells 1 through 10 of IgG Gel card. Leave unused wells covered.**  |  |
| **8.0** | **Using the appropriately calibrated micropipette, dispense 50ul of 0.8% suspension cells into each well.** 8.1 Selection of cellsa. For incompatible kidney recipients (Group O and B), use fresh A2 cellsb. See *Section I. Protocols* |  |
| **9.0** | **Using a different clean micropipette tip for each dilution, add 25ul of each dilution into the appropriately labeled well and 25ul of saline into well 10 (saline control.)**  |  |
| **10.0** | **Incubate at 37°C ± 1.5°C for 40 minutes.** |  |
| **11.0** | **Centrifuge the required 10 minutes in the Ortho Workstation centrifuge.** |  |
| **12.0** | **Read the gel card titer starting with well 10.** 12.1 Read the front of each well.**NOTE:** Titer begins at 2 due to addition of DTT prior to master dilution.  |   |
| **13.0** | **Record results on antibody titer form.** 13.1

|  |  |
| --- | --- |
| **Titer Type** | **Form** |
| Kidney Patient | Kidney Antibody Titer form *Refer to Attachment 8: Kidney Antibody Titer form*  |

NOTE: The tube you are starting with is diluted 1:2. Make sure you record reactions in the correct result box on the Kidney Antibody Titer form. * 1. Go to *Section VII for Interpretation of Titer Results*

**MC900056715[1]** |  |

**Section VI: Tube Testing Serum/Plasma, HTLA and QC Dilutions**

Chemical Risk Assessment: none

Biological Risk Assessment: low

 Protective Equipment: Lab coat, gloves

Supplies: 12x75mm and 10x75mm test tubes, MLA pipette tips, permanent marker,

Reagents: 0.85% Saline, 3-5% red cell suspension, LISS, anti-globulin Coombs sera, Coombs check cells,

Equipment: 250ul MLA pipette, cell washer, agglutination viewer, microscope, micropipette, incubator, serofuge

Special Requirements: Plasma from EDTA pink top tube, serum from red top tube, plasma from donor unit drawn in CPDA1 unit diluted with preservative

| **STEPS** | **INSTRUCTIONS** | **CHANGE/****APPROVAL** |
| --- | --- | --- |
| **1.0** | **Label 10 x 75 mm test tubes appropriately.*** 1. Record patient last name or QC name
	2. Consecutively label tubes 1 through 11.
1. Tube 11 is the saline control tube.
	1. If RT Isohemagglutination titration is required, prepare two separate sets of 11 tubes.
2. One will be used for the RT testing.
3. The second is to be used for the 37ºC AHG testing.
 |  |
| **2.0** | **Using a different clean disposable pipette for each dilution, add two (2) drops of each dilution to the appropriately labeled test tubes and two (2) drops of saline in tube 11 (saline control).**  |  |
| **3.0** | **Add one (1) drop of 3-5% suspension of cells.**3.1 Selection of cells:1. For HTLA, select cell with “weakest” reaction on panel already tested.
	1. See Section I:HTLA Titration Protocol
2. For QC refer to the procedure *Daily QC:BB.QC.1006*
 |  |
| **4.0** | **Incubate titer tubes and control tube.**4.1 If you are asked to do a **saline room temperature** isohemagglutinin titer, incubate at RT for 30 minutes ± 1 minute. Proceed to steps 7.0-9.0a. Routinely saline room temperature isohemmaglutinin titers are not ordered4.2 For HTLA titrations, incubate 37ºC ± 1.5C for 60 minutes. Proceed to steps 5.0-14.0.4.3 Do not use enhancement techniques on patient titers (albumin, PEG, LISS, red cell solid phase, or enzyme-treated rbcs) because falsely elevated titers may be obtained.4.4 When testing QC titers for Daily Rack QC refer to:1. Procedure *Daily QC:BB.QC.1006*
2. Section I: *QC Antisera/CAP Internal Assessments*
 |  |
| **5.0** | **Wash three (3) times with saline.** |  |
| **6.0** | **Add the proper antiglobulin Coombs sera to each 10 x 75 mm test tube.**6.1 For QC testing use IgG Coombs6.2 For HTLA titers, use polyspecific monoclonal Coombs sera.  |  |
| **7.0** | **Centrifuge the required seconds for Coombs phase.** 7.1 Look at label on centrifuge for coombs phase time. |  |
| **8.0** | **Starting with the last tube, read the titer.**8.1 Read QC MACROSCOPICALLY ONLY8.2 Read HTLA titers MACROSCOPICALLY AND MICROSCOPICALLY. |   |
| **9.0** | **If you are asked to perform a saline room temperature titer, the tubes should be discarded after reading and recording.**9.1 If you need to test at 37ºC to AHG phase, set up the titration again.  See *Section II: Preparation of Master Dilution**DO NOT CARRY TITER FROM ROOM TEMPERATURE TO 37ºC→AHG PHASE.* |  |
| **10.0** | **Add one (1) drop of Coombs check cells to all negative tubes.** |  |
| **11.0** | **Mix and centrifuge at 3500-3700rpm the required number of seconds for Coombs phase.**11.1 Look on label on centrifuge for Coombs phase time. |  |
| **12.0** | **Read all tubes appropriately using either magnifying mirror or microscope.**1. Coombs check cells must yield at least a 2+ reaction.
2. If less than a 2+ reaction, repeat steps 1.0-12.0.

a. Failure to receive a ≥2+ reaction indicates that the washing was inadequate and/or antiglobulin sera was not added. |  |
| **13.0** | **Record results on titer form.** 13.1

|  |  |
| --- | --- |
| **Titer Type** | **Form** |
| HTLA, QC | Antibody Titer form*Refer to Attachment 4: Antibody Titer form.* |

MC900056715[1] |  |
| **14.0** | **HTLA and QC titers are not recorded in SCC.**1. HTLA Titers will be charged using an Action code.
	1. Add on to the original test
		1. Go to Patient > Orders > Modify
		2. Add Action: HTLAT
	2. Confirm the action
		1. Go to Patient > Orders > Actions
 |  |

**Section VII: Interpretation of Titer Results**

Chemical Risk Assessment: none

Biological Risk Assessment: none

 Protective Equipment: Lab coat, gloves

Supplies: none

Reagents: none

Equipment: none

| **STEPS** | **INSTRUCTIONS** | **CHANGE/****APPROVAL** |
| --- | --- | --- |
| **1.0** | **Interpretation of Results:**1. Is titer valid?

|  |  |
| --- | --- |
| **For titer to be valid, saline control MUST be negative. If** | **Action** |
| Saline control positive | Discard dilutionObtain new saline sourceRepeat master titration |
| Saline control negative | Proceed |

1. Is there agglutination in the tube containing the most dilute serum/plasma (tube/microtube well 10)?

|  |  |
| --- | --- |
| **If** | **Action** |
| Agglutination presentHTLA: M+ or strongerGel/QC titer: 1+ or stronger | Carry the dilutions out further (use reserved aliquot in tube 11 labeled “SAVE”) |
| Agglutination not present (or Wk+)HTLA: No reactionGel/QC titer: Wk+ or No reaction | Proceed |

1. Determine the titer endpoint.

|  |  |
| --- | --- |
|  **If** | **Then** |
| Prenatal(Gel) | Look up the last previous reported titer for pregnancy.Titer endpoint: Highest dilution that gives a **1+** reaction.  Reporting titer: report as reciprocal of dilution.  *Example: 1:32 dilution, report as 32.***Note:** if current titer is within + 1 dilution tube of the previous titer or if no previous titer, report the current titer.If current titer is NOT within + 1 dilution tube of previous titer, repeat previous and current titers.Prepare new master dilution for current titration. |
| CAP, BMT, PUBS, Kidney(Gel)QC (Gel or tube) | Titer endpoint: Highest dilution that gives a **1+** reaction.Reporting titer: report as reciprocal of dilution.  *Example: 1:32 dilution, report as 32.* |
| HTLA(tube) | Titer endpoint: Highest dilution showing **micro**scopic reactions.Reporting titer: report as reciprocal of dilution. *Example: 1:32 dilution, report as 32.* |
| Titer does not show any reactions  | Report titer endpoint: negative(in Gel Wk+ is considered negative) |

MC900056715[1] |  |
| **2.0** | **Record interpretation on antibody titer form:**

|  |  |
| --- | --- |
| **Titer Type** | **Form** |
| OB, BMT, HTLA, CAP, QC | Antibody Titer form*Refer to Attachment 4: Antibody Titer form.* |
| Kidney Patient | Kidney Antibody Titer form *Refer to Attachment 8: Kidney Antibody Titer form*  |

 |  |
| **3.0** | **Enter titer results and interpretation in computer:**3.1

|  |  |  |
| --- | --- | --- |
| **For**  | **Go to**  | **Refer to** |
| OB, PUBS, other gel titers as requested by management | Section VIII: Computer Entry for Antibody Titer (excluding BMT and Kidney ABO titers) | *Attachment 5: Antibody Titer Flow Chart* |
| BMT ABO titers | Section IX: Computer Entry for Antibody Titer- BMT sample ABO titers | *Attachment 6: Resulting ABO Titers for BMT and Kidney samples* |
| Kidney ABO titers | Section X: Computer Entry for Antibody Titer- ABOi Kidney sample ABO titers | *Attachment 6: Resulting ABO Titers for BMT and Kidney samples* |
| HTLA titers | Next step, 4.0 |  |
| QC titers | NAQC titers are not recorded in SCC |  |

 |  |
| **4.0** | **HTLA titer results are not recorded in SCC.**1. HTLA Titers will be charged using an Action code.
	1. Add on to the original test
		1. Go to Patient > Orders > Modify
		2. Add Action: HTLAT
	2. Confirm the action
		1. Go to Patient > Orders > Actions
 |  |

**Section VIII: Computer Entry for Antibody Titer- Excluding BMT and Kidney ABO titers**

Chemical Risk Assessment: none

Biological Risk Assessment: none

 Protective Equipment: Lab coat, gloves

Supplies: none

Reagents: none

Equipment: none

| **STEPS** | **INSTRUCTIONS** | **CHANGE/****APPROVAL** |
| --- | --- | --- |
| **1.0** | **Order Titer in SCC**1. In SCC, go to Patient > Orders > Modify and add proper titer test.
2. Refer to ***Attachment 5: Antibody Titer Flow Chart***
 |  |
| **2.0** | **Go to SCC Patient > Orders > Results** |  |
| **3.0** | **Enter Results**1. To determine antibody associated with tests***refer to Attachment 5: Antibody Titer Flow Chart***
2. To enter the results for the following titer tests:

**TAB1-TAB 5, TOAB1-TOAB3** and **PTAB1-PTAB3**

|  |  |
| --- | --- |
| **Results to enter** | **Actions** |
| **Titration Results** | 1. Double click on the **Antibody Titer test** to enter the reactions
2. Result reactions under each dilution
	1. Remember W+ is considered negative
	2. See NOTE on next page
3. Verify that the SC (saline control) is negative.
4. Always result the SC
5. Titer is invalid if the SC is positive
6. F12 twice to Accept and save
 |
| **Antibody being Titered** | 1. Double click on the **Antibody Titered** or **Oth Ab Titered test** to enter the antibody. Enter antibody in both columns.
2. The first column is two characters and the second column expands to three characters.
	1. Example: anti-Fya

* 1. If the antibody is not found in the dropdown menu see next step 3.3.

Refer to***Attachment 7: Antibody Codes for Titer Tests***1. F12 twice to Accept and save.
 |

1. For **TOAB1- TOAB3** titers:
2. If the antibody is not on the dropdown list, select OT and Other
3. F12
4. Type in antibody in the short comment box:

 1. F12 to Accept and save.

**NOTE:** The titers are built as two sections: 1-512 and 1024->8192

|  |  |  |
| --- | --- | --- |
| **Results 1-512** | **Results 1024->8129** | **SC** |
| Always result 0-4+ or H |  | Always result, test is valid only if result is 0 |
| If 512 is positive\* then → | Result all with 0-4+ or H |
| If 512 is negative then → | Result all with NT |

\* 1+ or stronger. All titers in gel have an end point of 1+. |  |
| **4.0** | **Select interpretation from the drop down box that corresponds with the Highest Dilution that reacts 1+.*** 1. The Interpretation in SCC is called **TINT** (**T**iter **Int**erp)

Refer to:***Section VII: Interpretation of Titer Results******Attachment 5: Antibody Titer Flow Chart*** |  |

**Section IX: Computer Entry for Antibody Titer- BMT sample ABO titers**

Chemical Risk Assessment: none

Biological Risk Assessment: none

 Protective Equipment: Lab coat, gloves

Supplies: none

Reagents: none

Equipment: none

| **STEPS** | **INSTRUCTIONS** | **CHANGE/****APPROVAL** |
| --- | --- | --- |
| **1.0** | **Order Titer in SCC**1. In SCC, go to Patient > Orders > Modify and add proper titer test.
2. Refer to ***Attachment 5: Antibody Titer Flow Chart***
3. Refer to ***Attachment 6: ABO Titers for BMT and Kidney Samples***
 |  |
| **2.0** | **Go to SCC Patient > Orders > Results.** |  |
| **3.0** | **Select Titer to result.**1. Double click on test to open
2. Select rack if necessary
3. Below is a list of the titers to be found within each test:

|  |  |
| --- | --- |
| **TBMTD****(Titer BMT Donor)** | **TBMTR****(Titer BMT Recipient)** |
| Donor < A Titer | Recipnt < A Titer |
| Donor < A1 Titer | Recipnt < A1 Titer |
| Donor < B Titer | Recipnt < B Titer |

< = Antibody3.4 Cancel any test not neededa. Go to Patient > Orders > CancelRefer to ***Attachment 6: ABO Titers for BMT and Kidney Samples*** |  |
| **4.0** | **Results reactions under each dilution referring to table below.**1. Double click on the Titer test to enter the reactions
2. Result reactions under each dilution
	1. Remember W+ is considered negative
	2. The titers are built as two sections: 1-512 and 1024->8192

|  |  |  |
| --- | --- | --- |
| **Results 1-512** | **Results 1024->8129** | **SC** |
| Always result 0-4+ or H |  | Always result, test is valid only if result is 0 |
| If 512 is positive\* then → | Result all with 0-4+ or H |
| If 512 is neg or W+ then → | Result all with NT |

\* 1+ or stronger All titers in gel have an end point of 1+.1. Verify that the SC (saline control) is negative.
	1. Always result the SC
	2. Titer is invalid if the SC is positive
2. F12 twice to Accept and save
 |  |
| **5.0** | **Select interpretation from the drop down box that corresponds with the Highest Dilution that reacts 1+.**1. The Interpretation in SCC is called **TINT** (**T**iter **Int**erp)

Refer to:***Section VII: Interpretation of Titer Results******Attachment 6: Resulting ABO Titers for BMT and Kidney samples Flow Chart*** |  |

**Section X: Computer Entry for Antibody Titer- ABOi Kidney sample ABO titers**

Chemical Risk Assessment: none

Biological Risk Assessment: none

 Protective Equipment: Lab coat, gloves

Supplies: none

Reagents: none

Equipment: none

| **STEPS** | **INSTRUCTIONS** | **CHANGE/****APPROVAL** |
| --- | --- | --- |
| **1.0** | **Review Titer Order in SCC**1. The provider will order the appropriate test in Epic.
2. The test will be ordered based on the blood type of the recipient:

|  |  |
| --- | --- |
| **Test ordered** | **Recipient’s blood type** |
| **Group O** | **Group B** |
| **SCC Code** | TKDIO | TKDIB |
| **Test name** | Kidney O Patient ABOi Titer | Kidney B Patient ABOi Titer |

1. Refer to ***Attachment 6: ABO Titers for BMT and Kidney Samples***
 |  |
| **2.0** | **Go to SCC Patient > Orders > Results.** |  |
| **3.0** | **Select Test to result.**1. Double click on test to open
2. Select rack if necessary
3. Below is a list of the tests to be found within each order:

|  |  |
| --- | --- |
| **TKDIO** | **TKDIB** |
| Recp< A Titer IgG | Recp< A Titer IgG |
| Recp< A Titer IgM |  |
| DTT Tr IgG Titer |  |

< = Antibody1. Refer to ***Attachment 6: ABO Titers for BMT and Kidney Samples***
 |  |
| **4.0** | **Results reactions under each dilution referring to table below.**1. Double click on the **Titer test** to enter the reactions
	1. Recp<A Titer IgG
	2. Recp<A Titer IgM
2. Result reactions under each dilution
	1. Remember W+ is considered negative
	2. The titers are built as two sections: 1-512 and 1024->8192

|  |  |  |
| --- | --- | --- |
| **Results 1-512** | **Results 1024->8129** | **SC** |
| Always result 0-4+ or H**Exception:** DTT treated IgG 1:1, result DT |  | Always result, test is valid only if result is 0 |
| If 512 is positive\* then → | Result all with 0-4+ or H |
| If 512 is neg or W+ then → | Result all with NT |

\* 1+ or stronger All titers in gel have an end point of 1+.1. Verify that the SC (saline control) is negative.
2. Always result the SC
3. Titer is invalid if the SC is positive
4. F12 twice to Accept and save
 |  |
| **5.0** | **Document when the DTT Treatment of the IgG Titer has been performed.**1. Double click on **DTT Tr IgG Titer** to enter the reactions
2. Select Y for Yes in first drop down box
3. Select Yes in the 2nd drop down box.

1. F12 to accept and save.
 |  |
| **6.0** | **Select interpretation from the drop down box that corresponds with the Highest Dilution that reacts 1+.**1. The Interpretation in SCC is called **TINT** (**T**iter **Int**erp)

Refer to:***Section VII: Interpretation of Titer Results******Attachment 6: ABO Titers for BMT and Kidney Samples***  |  |

**Section XI: Preparation of 0.01M DTT for Distinguishing IgM and IgG Antibodies**

Chemical Risk Assessment: low

Biological Risk Assessment: low

 Protective Equipment: Lab coat, gloves

Supplies: Blood Bank Phosphate buffered saline, pH paper, parafilm, Reagent Label

Reagents: powered DTT

Equipment: scale, weigh boat, graduated cylinder, Freezer 12

| **STEPS** | **INSTRUCTIONS** | **CHANGE/****APPROVAL** |
| --- | --- | --- |
| **1.0** | **Obtain Blood Bank Phosphate buffered saline.**1.1 Confirm pH is 7.3 with pH paper. |  |
| **2.0** | **Measure 100mL of 7.3pH PBS into a graduated cylinder.** |  |
| **3.0** | **Go to BMT lab to use the Sartorius analytical scale.** |  |
| **4.0** | **Obtain a weigh boat** |  |
| **5.0** | **Place on scale and tare to 0** |  |
| **6.0** | **Open the powered DTT** 6.1 Using the scale, add small amounts to get 0.154g of DTT |  |
| **7.0** | **Add to the 100mL of PBS saline** |  |
| **8.0** | **Cover with parafilm and mix gently till dissolved** |  |
| **9.0** | **Freeze any not used at -18C or colder in Freezer 12.** 9.1 Can be kept frozen for 1 year |  |
| **10.0** | **Complete Reagent Label:**Reagents Name: 0.01M DTTPreparation Date: Expiration Date: (1 year from Preparation Date)Storage Temperature:Tech’s initials: Manufacturer/Lot number of DTT |  |

 **3. Review/Revised/Implemented:**

All procedures must be reviewed per document control protocol.

All new procedures and procedures that have major revisions must be signed by the CLIA Director.

All reviewed procedures and procedures with minor revisions can be signed by the designated section medical director

**4. Related Procedures:**

Preparation of Master Dilutions

Routine Testing of Plasma/Serum Dilution

Gel testing of Plasma/Serum Dilution

**5. References**:

Technical Manual. American Association of Blood Banks, revised periodically

SCC System

Finck, Rachel, Carrie Lui-Deguzman, Shih-Mao Teng, Rebecca Davis, and Shan Yuan. *“Comparison of a gel microcolumn assay with the conventional tube test for red blood cell alloantibody titration.”* Transfusion, April 2013

Duez, Alexis Francoise Flourie, Olivier Garraud*. “Antibody titration for immunized pregnant women: conventional tube test or gel microcolumn assay.”* Transfusion, April 2014.

**6. Attachments/Links**:

Attachment 1: Incompatible ABO Kidney Blood Transfusion Protocol

Link: Isohemagglutinin Titrations for Incompatible Kidney Transplants Card

Attachment 2: Titration for BMT Recipients and Donors

Link: Antibody Titer Form

Attachment: Master Antibody Titer flow chart

Attachment 6a & 6b: Resulting ABO Titers for BMT and Kidney samples

Attachment 7: Antibody Codes for Titer Tests

Link: Kidney Antibody Titer Form

Attachment 9: Titer Tests in SCC

**7. Revised/Reviewed Dates and Signatures:**

Refer to archive history/title21

**Incompatible ABO Kidney Blood Transfusion Protocols**

**ATTACHMENT 1**

|  |  |  |  |  |  |  |
| --- | --- | --- | --- | --- | --- | --- |
| **Donor** | **Patient’s Group Type** | **Donor Group Type** | **Patient Antibody titered****< = antibody** | **Blood****W= washed** | **Plasma** | **Platelets****PAS preferred****W=washed** |
| Deceased | B | A2 | <A | B/Ow | AB/\* | AB/Aw/Bw/Ow |
| B | A2B | <A | B/Ow | AB/\* | AB/Aw/Bw/Ow |
| LIVING | O | O | - | O | ANY | O |
| O | A2 | <A | O | ANY | A/AB/Bw/Ow |
| O | A2B | <A | O | ANY | A/AB/Bw/Ow |
| A | A | - | A | A | A |
| A | O | - | A | A | O |
| B | B | - | B | B | B/Ow |
| B | O | - | B | B | B |
| AB | AB | - | AB | AB | AB |

Abbreviations:

\*Discuss with Medical Director

< antibody symbol

W washed product

**Titration Protocol for BMT recipients and donors**

**ATTACHMENT 2**

|  |  |  |  |
| --- | --- | --- | --- |
| Recipient | Donor | Titer Recipient | Titer Donor |
| A | O | No titer | Titer anti-A |
| B | O | No titer | Titer anti-B |
| AB | O | No titer | Titer anti-A and anti-B |
| O | O | No titer | No titer |
| A | A | No titer | No titer |
| B | A | Anti-A | Anti-B |
| AB | A | No titer | Anti-B |
| O | A | Anti-A | No titer |
| A | B | Anti-B | Anti-A |
| B | B | No titer | No titer |
| AB | B | No titer | Anti-A |
| O | B | Anti-B | No titer |
| A | AB | Anti-B | No titer |
| B | AB | Anti-A | No titer |
| AB | AB | No titer | No titer |
| O | AB | Anti-A and anti-B | No titer |
| NOGR | Any | Anti-A and anti-B | Anti-A and anti-B |







**ANTIBODY CODES FOR TITER TESTS**

**ATTACHMENT 7**

| **TITER TEST** | **CODES** | **CODE EXPANDED** | **DESCRIPTION** |
| --- | --- | --- | --- |
| TAB1TAB2TAB3TAB4TAB5PTAB1PTAB2PTAB3 | D  | D  |  |
| C | C |  |
| E | E |  |
| LC | LC | Little C |
| LE | LE | Little E |
| A | A |  |
| A1 | A1 |  |
| B | B |  |
| FA  | FYA | Duffy A |
| FB  | FYB | Duffy B |
| K | K | Kell |
| JA  | JKA | Kid A |
| JB | JKB | Kid B |
| M | M |  |
| N | N |  |
| S | S |  |
| TOAB1TOAB2TOAB3 | V | V |  |
| CW | CW |  |
| GO | GOA |  |
| OT | OTHER |  |
| LK | LK | Cellano |
| KP | KPA |  |
| JS | JSA |  |
| U | U |  |
| LS | LS | Little S |

**ATTACHMENT 9**

|  |
| --- |
| **Titer Tests in SCC** |
| **Prenatal Titers or as requested by management** |
| **SCC Test:** | **For:** | **Antibody:** |
| TAB1 | 1st antibody | C, E, D, LC, LE, K, FYA, FYB, JKA, JKB, S, M, N, andA, A1, B (not for kidney or BMT ABO titers) |
| TAB2 | 2nd antibody |
| TAB3 | 3rd antibody |
| TAB4 | 4th antibody |
| TAB5 | 5th antibody |
| TOAB1 | 1st antibody | V, CW, GOA, LK, KPA, JSA, U, LS, OTHER (Rare antibody) |
| TOAB2 | 2nd antibody |
| TOAB3 | 3rd antibody |
| **PUBS Titers** |
| **SCC Test:** | **For:** | **Antibody:** |
| PTAB1 | 1st antibody | C, E, D, LC, LE, K, FYA, FYB, JKA, JKB, S, M, N, andA, A1, B (Consult management if the antibody is not one of the above) |
| PTAB2 | 2nd antibody |
| PTAB3 | 3rd antibody |
| **Kidney Titers** |
| **SCC Test:** | **For:** | **Antibody:** |
| TKDIB | Kidney B Patient ABOi Titer | Anti-A IgG |
| TKDIO | Kidney O Patient ABOi Titer | Anti-A IgG Anti-A IgM |
| **BMT Titers** |
| **SCC Test:** | **For:** | **Antibody:** |
| TBMTD | BMT Donor ABO Titer | A, A1, B |
| TBMTR | BMT Recipient ABO Titer | A, A1, B |
| **HTLA Titers** |
| **SCC Action:** | **Note:** |
| HTLAT | HTLA results are not recorded in SCC; patient is charged with this Action code |