**Purpose:** This procedure provides instructions for how to determine the presence of unexpected antibodies by the tube Indirect Antiglobulin Test (IAT) method using Low Ionic Strength Solution (LISS).

* ***Note:*** *A patient antibody screen test may be performed at the same time as the patient ABO/D test, per established procedure.*

**Procedure:**

|  |  |  |  |
| --- | --- | --- | --- |
|  | **Action** | | **Related Documents** |
| **1** | * Confirm sample acceptability. | | * Evaluating Patient Samples and Request Forms |
| **2** | * Label tubes for 3 test cell antibody screen panel. * Arrange the tubes in the rack. * Ensure reagents are well mixed and at room temperature before use. | | * Labeling Tubes Procedure * Rack Set-Up Procedure |
| **3** | * Add **2** drops of patient plasma/serum to each tube. | |  |
| **4** | * Add **1** drop of reagent screening cells to respective labelled tubes. * Mix gently. | |  |
| **5** | * Add **2** drops of LISS reagent to each tube. * Mix gently. * Inspect each tube for comparable appearance and volume. | |  |
| **6** | * Incubate at 37°C incubation per manufacturer’s package insert instructions. (Minimum of 10 min at 37°C; can be extended to 30 min.) | |  |
| **7** | * Centrifuge for the posted time in a calibrated serologic centrifuge. | |  |
| **8** | * Read and record macroscopic readings. | | * Reading and Grading Tube Hemagglutination Reactions |
| **9** | * Wash the tubes four times with saline. | | * Washing Red Cell Samples (Manual or Automated Procedure) |
| **10** | * Add **2 drops** of anti-IgG. | |  |
|  | **Action** | | **Related Documents** |
| **11** | * Mix the tubes **immediately.** * Centrifuge for the posted time in a calibrated serologic centrifuge. | |  |
| **12** | * Immediately after centrifugation: * Resuspend the cells, and observe macroscopic agglutination. * Record results. | | * Reading and Grading Tube Hemagglutination Reactions |
| **13** | * Validate all weak and negative antiglobulin results: * Add **1** drop of IgG-coated control cells to each tube with a weak or negative antiglobulin result. * Centrifuge for the posted time in a calibrated serologic centrifuge. * Resuspend the cells, and observe macroscopic agglutination. * Record results.   ***Valid control results****: Agglutination of at least grade 2+ must be present or the test results are invalid and the test must be repeated*. | |  |
| **14** | * Analyze the reactions of the IgG-coated RBCs as follows: | | |
| **If agglutination is…** | **Then…** | |
| * Present | * Test is complete. | |
| * Absent | * Test is invalid: * Repeat Steps 1-13. * Consider cell washer problem or inactive AHG. | |

|  |  |  |  |  |
| --- | --- | --- | --- | --- |
| **15** | Consult the following table to interpret the antibody screen results. | | | * Antibody Identification Process |
| If the IAT results show | Then | Report Antibody screen as |
| * No hemolysis (at 37°C) and no agglutination | * Antibodies were not present or were undetected. | Negative |
| * Hemolysis (at 37°C) or agglutination (any strength) | * An antibody is present. * Proceed to antibody identification procedures. | Positive |
| 16 | * Check that the record is complete: * Date and time of completion, * Technologist identification, and * Final clerical check | | |  |

Reference:

Current version of reagent manufacturer’s package insert instructions.

AABB Technical Manual, Current Edition