**Purpose:**

This procedure provides instructions for how to determine the presence of unexpected antibodies by the tube Indirect Antiglobulin Test (IAT) method.

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

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| --- | --- | --- |
|  | **Action** | **Related Documents**  |
| **1** | * Confirm sample acceptability.
 | * Sample Acceptance Evaluation
 |
| **2** | * Place saline wash bottle in Helmer Plasma Thawer water bath, to warm, using overwrap bag.
 |  |
| **3** | * Label tubes.
* Arrange the tubes in the rack.
 | * Labeling Tubes for Manual Bench
* Manual Bench Set Up
 |
| **4** | * Add **sufficient** drops of patient plasma/serum to complete tests to a labelled tube
 |  |
| **5** | * Add **1** drop of reagent screening cells to respective tubes.
 |  |
| **6** | * Incubate all tubes:
* **10 to 15 minutes** at
* **37 C** incubation
 |  |
| **7** | * + Without removing the tubes from the heatblock, add 2 drops of the prewarmed plasma from step 4, to each of the prewarmed screening cell reaction tubes. Mix gently. Continue to incubate these tubes containing both plasma and reagent cells for 30 to 60 minutes at 37 C.
 |  |
| **8** | * + Remove the tubes from the heatblock, and wash manually four times , using the prewarmed saline from step 2 above.
		- ***NOTE:***  *37 reading is omitted for pre-warm methods*
 | * Washing Red Cell Samples (Manual or Automated Procedure)
 |
| **9** | * + Add **2** drops of anti-IgG.
		- ***Note:*** *Anti-IgG is necessary for the pre-warm method.*
 |  |
| **10** | * + Mix the tubes immediately.
	+ Centrifuge for the posted time in a calibrated serologic centrifuge.
 |  |
|  | **Action** | **Related Documents Title** |
| **11** | * Immediately after centrifugation:
* Resuspend the cells, and
* Read macroscopically and record results.
 | * + Reading and Grading Tube Hemagglutination Reactions
 |
| **12** | * Validate 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.
* Read macroscopically and record the results.
	+ ***Valid control results****: Agglutination of at least grade 2 must be present or the results are invalid and the test must be repeated*.
 |  |
| **13** | * + - Analyze the reactions of the IgG-coated RBCs as follows:
 |
| **If agglutination is** | **Then** |
| * + - * Present
 | * + - * + Test is complete.
 |
| * + - * Absent
 | * + - * Test is invalid:
* Repeat Steps 1-11.
* Consider cell washer problem or inactive AHG.
 |

|  |  |
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| **14** | * Consult the following table to interpret the antibody screen results.
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| If the IAT results show | Then | Report antibody screen as |
| * + No agglutination
 | * + Antibodies were not present or were undetected.
 | * + Negative
 |
| * + Agglutination (any strength)
 | * + An antibody is present
 | * + Positive
 |
| 15 | * Check that the record is complete:
* date and time of completion,
* technologist identification, and
* final clerical check
 |

**References:**

AABB Standards for Blood Banks and Transfusion Services, Current Edition.

AABB Technical Manual, Current Edition.