**Purpose:**

This procedure provides instructions for reading and grading test tube hemagglutination reactions.

**Policy:** Observations of all tube test results are recorded properly and promptly at the time the test is performed.

**Procedure:**

|  |  |  |
| --- | --- | --- |
|  | **Action** | **Related Documents**  |
| **1** | * Remove a tube(s) from the centrifuge head
* Examine the supernatant for hemolysis over a white background.
	+ ***Note:*** *Plasma/Serum surrounding the centrifuged RBC buttons must be inspected for hemolysis. Hemolysis must be regarded as a positive sign of an antigen-antibody reaction if the pretest serum was not hemolyzed and no hemolytic agent was added to the test.*
 |   |
| **2** | * Record the degree of hemolysis if present.
 |   |
| **3** | * Hold the tube(s) firmly between thumb and forefinger over an illuminated concave mirror.
 |   |
| **4** | * Adjust the concave mirror to eliminate glare.
 |  |
| **5** | * Adjust the tube(s) so that the RBC button is closest to the mirror.
 |   |
| **6** | * Dislodge the cell button gently.
 |  |
| **7** | * Looking into the mirror, observe the way the RBCs leave the cell button.
* The characteristics of the agglutination should be noted.
	+ Loose, “stringy,” mixed field, or refractile agglutinates should be recorded as they provide valuable clues in the investigation of aberrant findings.
	+ Utilize BBCS comments to record atypical findings
 |  |
| **8** | * Continue observation until cell button is completely resuspended.
 |  |

|  |  |  |
| --- | --- | --- |
|  | **Action** | **Related Documents**  |
| **9** | **Interpret reactions as follows:** |  |
|  | * If agglutination and/or lysis are…
 | * Then…
 |  |
| **Not observed** | * The test result is negative; the reaction grade and score are both 0 (zero).
 |  |
| **Observed** | * Grade and score reactions as follows:
 |
| **Atypical Agglutination Observed** | * Record in BBCS comments.
 |
|  | **Grade** | **Appearance** |
|  | **4+** | * Complete Agglutination: No unagglutinated RBCs
 |
|  | **3+** | * Strong Reaction: a few detached masses of agglutinated RBCs; no unagglutinated RBCs
 |
|  | **2+** | * Moderate Reaction: Large agglutinates in a sea of smaller agglutinates; few unagglutinated RBCs
 |
|  | **1+** | * Weak Reaction: Many agglutinates of up to 20 RBCs with smaller agglutinates and unagglutinated RBCs.
 |
|  | **W** | * Trace Reaction: Small agglutinates of 2-4 RBCs with many unagglutinated RBCs
 |
|  | **0** | * No reaction: All cells are smoothly in suspension
 |
|  | **H** | * Hemolysis observed. Objective evaluation of slight, moderate or gross hemolysis should be recorded in BBCS comments
 |
|  | **M** | * Mixed Field observed. Objective grade evaluation and explanation of mixed field investigation should be recorded in BBCS comments
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**References:**

Judd’s Methods in Immunohematology, Current Edition