

## BLOOD BANK ANTIBODY IDENTIFICATION

Test Code: **ABID**

### I. PRINCIPLE

The subsequent testing of plasma found to contain unexpected antibodies, using a panel of typed red blood cell suspension, enabling the specificity of the antibody to be determined, in order to select donor blood negative for the appropriate antigen(s) for transfusion.

### II. CLINICAL SIGNIFICANCE

UnityPoint Pekin blood bank personnel will systematically test patient plasma against Biotestcell-8 and/or Biotestcell-11 reagent red blood cells. The pattern of reactivity obtained with these cells is compared with the antigen profile Master List to determine the specificity of any antibody present.

### III. SPECIMEN

- A. Plasma from a pink top tube is preferred.
- B. Samples with the proper labeling including the patient's first and last name, date of birth, or admission number, blood bank armband number, collection date, collection time, and initials of phlebotomist, may be used up to three (3) days after collection. (The day of sample draw is day zero.) Exception: Outpatient presurgical specimens may be used up to seven (7) days, if the patient has not been pregnant or transfused within the previous three months.
- C. Blood specimens are stored at 2 to 8° C if not used immediately.

### IV. REAGENT

- A. Reagent kits:
  1. Biotestcell® 11 Plus and Biotestcell® 8 are Reagent Red Blood Cells with polyvalent antigens. Both panels are also suitable for use with enzymes or enhancement reagents. The life span of enzyme treated Reagent Red Blood Cells is listed in the instructions for use of the respective enzymes
  2. Biotestcell® 11 Plus and Biotestcell® 8 are suspended 3.0 to 3.4% in a modified Alsevers solution and can be used immediately after careful resuspension. They are produced every 4 weeks.
  3. Preservative: 0.01% Neomycin, 0.033% Chloramphenicol, 5ppm Amphotericin
- B. Handling and Storage of Reagents:
  1. For in vitro diagnostic use only.
  2. No US standard of potency.
  3. Suspend the red cells before use by gently inverting each vial several times.
  4. Caution: This product contains natural rubber latex which may cause allergic reactions.
  5. Store at 2-8° C when not in use.

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6. Do not freeze or expose to elevated temperatures.
7. Avoid contaminating this product during use. Contamination will adversely affect the product's performance during its shelf life.
8. Do not use contaminated reagents.
9. Do not use reagents beyond the expiration date.
10. Do not use leaking or damaged vials.
11. Do not use unlabeled vials.
12. Reagent red cells should not be used if the cells darken, spontaneously clump or if there is significant hemolysis. Slight hemolysis may occur with age. In this instance, the red cells may be washed and suspended in saline immediately prior to use.
13. Handle and dispose of reagents as if potentially infectious.

## V. PROCEDURE

- A. Label one 12 x 75 mm test tube for each Biotestcell® vial to be used and one additional tube for an autologous control.
- B. Label each tube with first and last initial of patient being tested. (Lengthen the minimum letters to differentiate patients with the same initials, if necessary.)
- C. Place 2 drops of plasma to be tested into each of the tubes.
- D. Gently invert all Biotestcell® vials several times to achieve a complete suspension of the cells.
- E. Add one drop of each Biotestcell® reagent to the appropriately labeled tubes. If an autologous control is to be run in parallel, add one drop of 2-4% saline suspension of autologous cells to the appropriate tube.
- F. Mix the contents of each tube thoroughly.
- G. Centrifuge for 20 seconds at 3400 rpm (1000 rcf).
- H. Gently resuspend cells and examine macroscopically for agglutination or hemolysis.
- I. Add two drops of LISS to each tube.
- J. Shake to mix.
- K. Incubate tubes at 37° C for 10 minutes.
- L. Centrifuge for 20 seconds at 3400 rpm (1000 rcf).
- M. Gently resuspend cells and examine macroscopically for agglutination or hemolysis.
- N. Wash tubes with saline three times (an automatic cell washer may be used).
- O. Add two drops IgG Coombs serum, shake to mix, and centrifuge for 20 seconds at 3400 rpm (1000 rcf)
- P. Gently resuspend cells and examine macro and microscopically for agglutination or hemolysis.
- Q. Add one drop of Coombs control cells to all tubes with negative results. Centrifuge for 20 seconds, read and record results. If the cells are now agglutinated, the negative result is valid.



## VI. INTERPRETATION OF RESULTS

### A. Possible Results:

1. Positive:
  - a. Agglutination of any of the Biotestcell® red cells at any phase.
  - b. Hemolysis at the saline or potentiated phases of testing.
2. Negative:
  - a. Absence of agglutination and hemolysis throughout the test procedure indicates that the test serum does not contain detectable antibodies to any of the antigens present in the reagent.
3. Identifying unknown antibody:
  - a. Review the reactions obtained with the autologous control to determine if the antibody is allo or auto in nature.
  - b. Delete all antigens present on the red cells that are non-reactive at all phases of testing by drawing a slash through the particular antigen at the top of the Biotestcell® Master List.
  - c. Compare the pattern of agglutinated cells with the profiles of antigens not deleted from the Biotestcell® Master List.
  - d. If only one antigen remains after deletion of antigens present on nonreactive panel cells and the pattern of the antigen matches the pattern of reactivity obtained, then the antibody is tentatively identified.
  - e. If more than one antigen remains following the deletion procedure, steps must be taken to identify the multiple antibodies that might be present.
4. Positive and negative results that do not fit any of the established patterns for any antigens can indicate the presence of multiple antibodies or antibodies to unspecified antigens.
  - a. For multiple suspected antibodies, review the phases when agglutination occurred and the strengths of the reactions.
    - 1) The pattern of reactions obtained at each test phase, when considered independently, may match the profile of an antigen on the Master List, thus giving a clue to the specificity of at least one of the antibodies that may be present.
    - 2) If all reactions occur at the same phase or phases, differences in strengths of reactions might also give a clue to the antibodies present.
  - b. Test the patient's own red cells for antigens corresponding to antibodies suspected.
    - 1) If the patient's red cells possess the antigen, it is unlikely that the corresponding antibody is present unless the autologous control, in addition to reagent panel cells, is agglutinated.
    - 2) If no conclusion of antibody identification can be made, a sample should be submitted to Red Cross Lab for further study. Upon antibody identification, any donor units for crossmatch should be

- screened for the corresponding antigen with available antisera, and crossmatched to confirm compatibility.
- 3) Add on Ab ID in Sunquest Blood Order Processing by clicking on the "Add a Test" keyboard in the patient testing section and hit the "M" key. Once the test has been added, result with the proper code from the Antigen/Antibody codes list (See UPPK BB-0140.01).
  - 4) Add on the auto control performed with the antibody panel in Sunquest BOP by clicking on "a" key on the "Add a Test" keyboard. Fill in with the proper reactions and interpretation for the auto control.
  - 5) If the patient's auto control is positive, perform a DAT with the Anti-IgG, -C3d Polyspecific Coombs reagent (;DBS in the "Add a Test" box in BOP). Remember to QC the Polyspecific Coombs reagent if not already performed for the day. If the DAT is positive with the Polyspecific Coombs reagent, then perform a DAT with the Monospecific Coombs (Anti-IgG) reagent (use the "Q" key on the "Add a Test" keyboard) to determine if the DAT is positive from IgG or Compliment.
  - 6) If antigen typing of units is performed, add on the antigen typing test, after appropriate units have been allocated, by using the "Z" key on the "Add a Unit Test" keyboard. Once the test has been added to all appropriate units, result with the proper code from the Anigen/Antibody code list (See UPPK BB-0140.01).
  - 7) If an antibody titer is requested, send to Unity Point Health-Methodist Lab.

## VII. LIMITATIONS

- A. Negative reactions and subsequent positive reactions with IgG coated red blood cells indicate that the serum contains no detectable antibodies against one of the antigens present on the Reagent Red Blood Cells.
- B. Because some antibodies show dosage effect, the antigen density on the Reagent Red Blood Cells needs to be considered when evaluating the test results (homozygous or heterozygous hereditary disposition). A heterozygous expression of the antigen may result in non-detection of weak antibodies depending on the test method used.
- C. In very rare cases HLA-antigens within the product may lead to false positive reactions.
- D. The reactivity of the product may decrease during the dating period and therefore should not be used after the expiration date. The rate of decrease in reactivity is partially dependent on individual donor characteristics that are neither controlled nor predicted by the manufacturer.
- E. Negative reactions will be obtained if the sample contains antibodies present in concentrations too low to be detected by the test method employed. No test method is capable of detecting all red cell antibodies.



**VIII. REFERENCES**

- A. Bio-Rad Medical Diagnostics GmbH, Dreieich, Germany, Reagent Red Blood Cells, Biotestcell® 8, 11, and 11 Plus, 186186/19, Rev. 08/2016.
- B. AABB Technical Manual, Nineteenth Edition, Bethesda, Maryland 20817, Rev. 2017.

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<b>REVISION HISTORY (began tracking 2011)</b>			
Rev	Description of Change	Author	Effective Date
01/24/18	3 days and 7 days for Outpatient Surgical	Jenny Turner	01/24/18
10/22/18	Reagent Kits Bio-testcell	Jenny Turner	10/22/18
12/09/18	Added Sunquest resulting information.	Jenny Turner	12/09/18
09/10/19	Added resulting info in SQ for Auto Control and DAT with AB panel.	Jenny Turner	09/10/19

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