



ATLAS BOVINE ALBUMIN SOLUTION, 22% & 30%

Bovine Albumin Solution for the completion of second stage agglutination of incomplete antibodies and the enhancement of the sensitivity of indirect antiglobulin tests

INTRODUCTION & PRINCIPLES

ATLAS 22% and 30% Bovine Albumin Solutions are prepared from bovine serum albumin without the addition of artificial avidity enhancers or high molecular weight agglutination potentiators. No sodium caprylate was added, although Sodium Azide 0.1% was added to preserve the solution. ATLAS Bovine Albumin solutions are subjected during preparations to high temperature and low pH conditions for extended periods sufficient to completely inactivate BSE-like agents.

ATLAS Bovine Albumin solutions are added to cell suspension to enhance the ability of incomplete antibodies, that can combine with their specific antigens only in the first stage of agglutination, to complete the second stage of agglutination.

PRECAUTIONS

- 1.The reagent have to be stored at 2-8°C.
- 2.This reagent is derived from animal sources, thus, appropriate care must be taken in the use and disposal of this reagent as there are no known test methods that can guarantee absence of infectious agents.
- 3.Don't Freeze the reagents.

4. Do not use the reagent if it is marked with turbidity as this may indicate reagent deterioration or contamination.
- 5.The reagent contains 0.1% Sodium Azide as preservative.
6. The reagent should be used as supplied and in accordance to the procedure mentioned below. Do not use beyond expiration date.

PREPARING THE SPECIMEN

For antibody identification procedures, only fresh serum obtained from fully clotted specimen should be used. For antigen detection procedures, red cells can be obtained from samples drawn with/without anticoagulants. The test should be performed as soon as possible to avoid false results. If testing is delayed samples should be stored at 2° to 8°C and tested within two days provided there is no evidence of haemolysis or it should be stored frozen. In this case red cells should be removed from the serum.

ALBUMIN MIX PROCEDURE

- 1.Prepare a 2-3% suspension of red cells sample in isotonic buffered saline (pH6.9).
- 2.Place 2 volumes of serum or plasma in a glass test tube.
- 3.Add 1 volume of the 2-3% red cell suspension.
- 4.Add 2 volumes of ATLAS Bovine Albumin Solution and mix the content thoroughly.
- 5.Incubate for 15-60 minutes at 37°C.
- 6.Centrifuge for 30 seconds at 900-1000 RCF.
- 7.Gently re-suspend the cells button and observe immediately for agglutination with an optical aid or microscopically.

ALBUMIN REPLACEMENT PROCEDURE

- 1.Prepare a 2-3% suspension of red cells sample in isotonic buffered saline (pH6.9).
- 2.Place 1 volume of serum or plasma in a glass test tube.
- 3.Add 1 volume of the 2-3% red cell suspension and mix thoroughly.
- 4.Incubate for 45-90 minutes at 37°C.
- 5.Remove the supernatant saline-serum mixture using a fine pipette without disturbing the button of red cells.
- 6.Add 1 volume of ATLAS Bovine Albumin Solution 22%. Care must be taken so as not to disturb the cell button. Do not mix.
- 7.Reincubate for 15-30 minutes at 37°C.

- 8.Observe immediately for agglutination with an optical aid or microscopically.

ALBUMIN DISPLACEMENT PROCEDURE

- 1.Prepare a 2-3% suspension of red cells sample in isotonic buffered saline (pH6.9).
- 2.Place 1 volume of serum or plasma in a glass test tube.
- 3.Add 1 volume of the 2-3% red cell suspension and mix thoroughly.
- 4.Incubate for 45-90 minutes at 37°C.
- 5.Allow one volume of ATLAS Bovine Albumin solution 30% to run down the inside wall of the test tube so as to upwardly displace the supernatant serum-saline mixture. Do not mix.
- 6.Reincubate for 15-30 minutes at 37°C.
- 7.Observe immediately for agglutination with an optical aid or microscopically.

INDIRECT ANTIGLOBULIN PROCEDURE

- 1.Prepare a 2-3% suspension of red cells sample in isotonic buffered saline (pH6.9).
- 2.Place 2 volumes of serum or plasma in a glass test tube.
- 3.Add 1 volume of the 2-3% red cell suspension.
- 4.Add 2 volumes of ATLAS Bovine Albumin Solution and mix the content thoroughly.
- 5.Incubate for 15-60 minutes at 37°C.
- 6.Using large volumes of Isotonic buffer saline (pH 6.9), wash the red cells at least four times. Care must be taken when discarding the saline completely after each wash.
- 7.Add two volumes of ATLAS Premium Anti-Human Globulin reagent to the dry cell button in the tube.
- 8.Gently Mix the tube contents and centrifuge for 15 seconds at 900-1000 RCF.
- 9.Gently resuspend the cells button and observe immediately for agglutination with an optical aid or microscopically.

ANTI-BODY TITRATION PROCEDURE

- 1.Prepare doubling dilutions of test serum in either normal group AB serum or 6% bovine albumin (6% bovine albumin can be prepared by mixing 4 parts of isotonic buffered saline with 1 part of ATLAS Bovine Albumin 30% solution).

2. Prepare a 2% cell suspension of appropriate washed red cells in ATLAS Bovine Albumin 22% or 30% solution.
3. Mix 1 volume of each serum dilution with 1 volume of 2% cell suspension.
4. Mix thoroughly and incubate for 15-60 minutes at 37°C.
5. Centrifuge for 30 seconds at 900-1000 RCF.
6. Gently resuspend each cell button and observe immediately for agglutination with an optical aid or microscopically. Weak positive or negative results may be further checked by performing an agglutination test.

READING THE RESULT

Presence of agglutination indicate positive reaction.

Lack of agglutination indicate negative reaction.

All results should be read immediately after performing the tests according to the above instructions. False negative or weak positive results may appear, if reading was delayed, as this may result in antigen-antibody dissociation.

All negative or weak positive results in the indirect anti-human globulin test must be confirmed by using IgG sensitised red cells (e.g. Coombs Control Cells).

PROCEDURE LIMITATION

1. False positive/ negative results may occur from contamination from test materials, improper storage of test materials, improper washing, improper incubation time or temperature, delayed reading of results, or any deviation from the recommended test procedures.
2. Improper centrifugation may also cause false results. The mentioned centrifugation times should be regarded as suggestions only. An appropriate centrifugation time should be determined by that which produces the strongest reaction of anti-body with antigen-positive cells, yet allows easy resuspension of antigen-negative red cells.
3. It is essential to routinely set up control tests in which the test red cells are mixed with the appropriate albumin solution alone and to establish the efficacy of the albumin solution throughout its use.
4. Bovine albumin solutions should not be used as negative controls for potentiated IgG blood grouping reagents.
5. The use of bovine albumin will not enhance the reactivity of all blood group antibodies.

QUALITY CONTROL

ATLAS Bovine Albumin Solutions 22% and 30% are checked prior to release for their reactivity to assure specificity in antibody-free system with red cells known to possess the most frequently inherited blood group antigens. ATLAS Bovine Albumin Solutions have been shown to enhance agglutination of Rh and other antibodies when used according to the mentioned procedures.

REFERENCES

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PPI024A01
Revision B (12.06.2004)