BOVINE SERUM ALBUMIN (22%-30%)

Diagnostic reagent for the in-vitro usage to enhance the sensitivity of the indirect antiglobulin test for some antibodyspecificities.

REF: V/BAA01.010	200 tests	REF: V/BAA05.010	1000 tests

CLINICAL SIGNIFICANCE [1]

Bovine serum albumin (BSA) is a globular protein (~66,000 Da molecular weight) that is used in numerous biochemical applications due to its stability and lack of interference within biological reactions.
Incomplete antibodies have the ability to combine with their specific antigens in the first stage of agglutination but will not produce visible agglutination without the use of special techniques. Addition of bovine albumin to the cell suspension enable some of these antibodies to complete the second stage of agglutination. Albumin has been shown to enhance the sensitivity of the indirect antiglobulin test for some antibody specificities.

METHOD PRINCIPLE [2]

Because BSA is a small, stable, moderately non-reactive protein, it is often used as a blocker in immunohistochemistry. This binding of BSA to nonspecific binding sites increases the chance that the antibodies will bind only to the antigens of interest. The 22% and 30% Bovine albumin reagents are prepared from bovine serum albumin. No artificial avidity enhancers or high molecular weight agglutination potentiators are added to Bovine albumin reagents. These reagents do not contain sodium caprylate and should be used as supplied by the methods described; their suitability for use in other techniques must be determined by the user.

REAGENT COMPOSITION

Reagent : Bovine Albumin	
Bovine Serum Albumin	22% or 30%
Sodium Azide	0.1%

PRECAUTIONS AND WARNINGS

Reagent to be handled by entitled and professionally educated person. Do not ingest or inhale as reagent (R) contains sodium azide which is classified as dangerous substance for environment.

Good Laboratories practices using appropriate precautions should be followed in:

- Wearing personnel protective equipment (overall, gloves, glasses,..).
- Do not pipette by mouth.
- In case of contact with eyes or skin; rinse immediately with plenty of soap and water. In case of severe injuries; seek medical advice immediately.
- Respect country requirement for waste disposal.
  S56: dispose of this material and its container at hazardous or special waste collection point.
  S57: use appropriate container to avoid environmental contamination.
  S61: avoid release in environment.

For further information, refer to the **Lab.Vie** Bovine Serum Albumin reagentmaterial safety data sheet.

REAGENT PREPARATION, STORAGE AND STABILITY

Lab.Vie Bovine Albumin reagents are supplied ready-to-use and stable up to the expiry date labeled on the bottles when properly stored refrigerated at 2–8°C.

Deterioration
The **Lab.Vie** Bovine Albumin reagent is normally clear, do not use if it is turbid.

SPECIMEN COLLECTION AND PRESERVATION

Serum or plasma
Fresh serum obtained from a fully clotted specimen should be used in compatibility or antibody identification procedures. Red cells obtained from samples with or without anticoagulants can be used in antigen detection tests. Fresh Serum or plasma should be used for test performing as soon as possible, If testing is delayed store samples at 2 – 8 °C. Serum or Plasma can be separated from the cells and frozen

EQUIPMENT REQUIRED NOT PROVIDED

- Sterile Syringe
- Analytical tubes and automatic pipet
- Centrifuge and spectrophotometer

ASSAY PROCEDURE

Many assay procedures can be described according the wide techniques varieties for the Bovine Albumin reagent usages.

Technique (1): Albumin Replacement Method
1- Prepare a 2 - 3% suspension of red cells in isotonic buffered saline (pH 6.9).
2- Place in a glass test tube : 1 Volume of serum or plasma and 1 Volume of 2-3% cell suspension.
3- Mix well and incubate at 37 °C for 45 - 90 minutes.
4- With a fine pipette remove the supernatant saline-serum mixture, leaving the button of red cells.
5- Add one volume of the 22% Bovine Albumin. Taking care not to disturb the cell button.
6- Without mixing re-incubate at 37 °C for 15-30 minutes.
7- Examine for agglutination. Reactions may be examined with an optical aid, or microscopically and record the results.

Technique (2): Albumin Displacement Method
1- Follow steps 1-3 of albumin replacement method.
2- Upwardly displace the supernatant saline-serum mixture by carefully allowing one volume of Bovine albumin 30% to run down the inside wall of the test tube.
3- Follow steps 6 and 7 of the Albumin Replacement Method

Technique (3): Albumin Mix Method
1- Prepare a 2 - 3% suspension of red cells in isotonic buffered saline (pH 6.9).
2- Place in a glass test tube: 1 Volume of serum or plasma, 1 Volume of 2-3% cell suspension and 2 Volumes of Bovine Albumin 22%.
3- Mix well and incubate at 37 °C for 15 - 60 minutes.
4- Centrifuge at 900 - 1000 rcf for 30 seconds.
5- Gently resuspend the cell button, examine for agglutination and record the result

Technique (4): Indirect Antiglobulin Test

1- Follow steps 1-3 of albumin mix method.
2- Wash the cells 3-4 times in isotonic buffered saline, decanting the saline completely after each wash.
3- Add two volumes of Bovine Albumin to the dry cell button.
4- Mix gently and centrifuge at 900 -1000 rpm (100 RCF) for 15 seconds. Gently resuspend the cell button and examine for agglutination.
5- Record the result
6- Confirm validity of all negative reactions by using IgG sensitized red cells.

Technique (5): Antibody Titration Procedure

1- Prepare dilution of test serum in either normal group AB serum or 6% bovine albumin (the late can be prepared by mixing 1 part 30% bovine albumin with 4 parts isotonic buffered Saline).
2- Prepare a 2% suspension of appropriate washed red cells in 22% or 30% bovine albumin.
3- Add 1 volume of 2% cell suspension to 1 volume of each serum dilution, mix well and incubate at 37 °C for 15 - 60 minutes.
4- Centrifuge at 900 -1000 rpm (100 RCF) for 30 seconds.
5- Gently resuspend the cell button, Examine for agglutination and record the result.
6- An antiglobulin test may be performed on those cells showing weak or negative results.

STABILITY OF THE REACTIONS

1. Read all tube and slide tests immediately.
2. Avoid delay in completing of washing steps, as it may result in dissociation of antigen antibody complexes.
3. Slide tests should be interpreted within two minutes to ensure specificity and to avoid the false negative or weak positive reactions.

LIMITATIONS

Bovine albumin will not enhance the reactivity of all blood group antibodies. Bovine albumin solutions should not be used as negative controls for potentiated IgG blood grouping reagents. False positive or false negative results may occur due to improper technique or contaminated test materials.

QUALITY CONTROL

To ensure adequate quality control, it is recommended to carefully read the instructions for good performance and storage of reagent. For any questions or problems please contact the **Lab.Vie** technical support.

PERFORMANCE CHARACTERISTICS

Lab.Vie Bovine serum Albumin reagent 22% and 30% have been shown to enhance agglutination of Rh and other antibodies when used according to insert methodologies. Each lot is tested to assure specificity in an antibody- free system with red cells known to process the most frequently inherited blood group antigens

REFERENCES

1. Majorek KA, Porebski PJ, Dayal A, Zimmerman MD, Jablonska K, Stewart AJ, Chruszcz M, Minor W (October 2012). "Structural and immunologic characterization of bovine, horse, and rabbit serum albumins". Molecular Immunology. 52 (3–4): 174–82.
2. Tips for Reducing ELISA Background". Biocompare. Compare Networks. October 8, 2012

SYMBOLS IN PRODUCT LABELLING

IVD	For in-vitro diagnostic use	Σ	Number of <n> test in the pack
LOT	Batch Code/Lot number		Caution
REF	Catalogue Number		Do not use if package is damaged
	Temperature Limitation		Consult Instruction for use
	Expiration Date		
	Manufactured by		