ACCUCARE™

Bovine Serum Albumin (BSA) 22% and 30%

Accucare BSA, a high protein solution is designed for in vitro diagnostic use to detect incomplete antibody sensitization phenomena. It has been found that there are antibodies which have failed to agglutinate red cells in normal saline but do so in a high protein solution. Incomplete anti RBC antibodies can be used in conjunction with 22% and 30% BSA to detect the respective antigen either microscopically or macroscopically. For In Vitro Diagnostic Test use only.

ORDER INFORMATION

REF	Pack Size
BSA 10	1 X 10mL
BSA 100	10 X 10 mL
BSA 1000	1 X 1000mL

CLINICAL SIGNIFICANCE

Serological albumin was first recognised as a potentiator of certain antigen-antibody interactions in 1945 by Diamond. Since then, methods employing Bovine Serum albumin have been widely used for the detection or quantitation of antibodies. Bovine Serum albumin has also been shown to enhance the sensitivity of the indirect antiglobulin test for some antibody specificities.

METHOD

Hemagglutination technique.

PRINCIPLE

Incomplete antibodies (IgG) do not agglutinate red blood cells in saline medium but will cause firm agglutination when mixed or suspended in 30% BSA. No other protein has been found to be as effective as BSA while giving negative reactions free from pseudoagglutination.

REAGENT

Accucare 22% and 30% Bovine Serum Albumin are prepared from a mixture of bovine serum albumin, and buffered saline. No artificial avidity enhancers or high molecular weight agglutination potentiators are added to any BSA preparation. None of the BSA reagents do contain sodium caprylate. Each BSA reagent is supplied at optimal dilution for use with all the recommended techniques stated below without the need for further dilution or addition. For lot reference number and expiry date see Vial Labels.

REAGENT PREPARATION

The reagent supplied is ready to use. Protect from Bright Light.

REAGENT STORAGE AND STABILITY

This product will be well-preserved within utility limit till the expiry date, if stored at temperature between +2°C and +8°C.

Caution: Do not freeze.

WARNING AND PRECAUTIONS

- For in vitro diagnostic use.
- Do not use components beyond the expiration date.
- If the reagent vial is cracked or leaking, discard the contents immediately.
- Do not use the reagents if a precipitate is present.
- Exercise the normal precautions required for handling all laboratory reagents.
- The reagents have been filtered through a 0.2µm capsule to reduce the bio-burden. Once a vial has been opened the contents should remain viable up until the expiry date as long as there is no marked turbidity, which can indicate reagent deterioration or contamination.
- The reagent contains preservative. Do not swallow. Avoid contact with skin and mucous membranes.
- The BSA has been obtained from a closed herd in the female line since 1980, in which no animal has been clinically suspected of having Bovine Spongiform Encephalopathy (BSE), and which has not been fed rations containing ruminant derived protein during that period.
- Consider Blood specimen as potentially infectious, handle and dispose it as per national applicable guideline.
- For information on disposal of the reagent and decontamination of a spillage site see **Material Safety Data Sheets**, available on request.

WASTE MANAGEMENT

Please refer to local legal requirements.

MATERIALS REQUIRED BUT NOT PROVIDED

- Test tubes (8X50mm),
- Pipettes,
- Centrifuge
- (0.9% NaCl) saline.
- General laboratory equipment

SAMPLE COLLECTION AND PRESERVATION

- Blood should be drawn by an aseptic technique with an anticoagulant. The specimen should be tested as soon as possible after collection.
- Use properly stored or fresh anti coagulated blood. Fresh 10% RBC-Saline suspension or Rh positive and Rh negative RBC and fresh samples of suspected incomplete antiseras.

PREPARATION OF 10% RBC-SALINE SUSPENSION

1. Add approximately 5 volumes of isotonic saline to the anti coagulated whole blood.
2. Mix gently and centrifuge for 2 minutes at 1500 rpm.
3. Remove the supernatant and wash the sedimented RBCs three more times with normal saline, as above.
4. After final wash, take 100µl of sedimented RBC and dilute it to 1.0 ml with normal saline. Mix thoroughly before use.

ASSAY PROCEDURE

DETECTION OF Rh D ANTIGEN

1. Take two known incomplete anti Rh D serums.
2. Dispense one volume of each incomplete serum in to the two rows of precipitin tubes respectively.
3. Add one volume each of unknown Rh D cell suspension (10%) to the 1st and 2nd row of precipitin tubes. Add known or confirmed Rh 'D' positive and Rh 'D' negative cells also in one set of tubes as control.
4. Mix the tubes thoroughly and incubate at 37°C for 1 ½ hours.
5. Add drop of ACCUCARE BSA through the wall of the tube and allow it to run down to the bottom without disturbing the cell sediment.
6. Incubate for 30 minutes at 37°C
7. Observe the results either microscopically or macroscopically

DETECTION OF ANTI Rh D ANTIBODIES

1. Take one volume of known standard 10% RhD negative cells in precipitin tube.
2. Mix one volume of unknown test antiserum to one tube each if the above standard cell suspension.
3. Mix thoroughly and incubate for 1 ½ hours at 37°C.
4. Add one volume of 22% ACCUCARE BSA through the sides of the tubes to run down to the bottom of the tube.
5. Care should be taken not to disturb the RBC while adding

BSA.

6. Incubate the tubes for another 30 minutes at 37°C
7. Read the results microscopically or macroscopically.

Stability of the reactions

1. Washing steps should be completed without interruption and tests centrifuged and read immediately after addition of the reagent.
2. Delays may result in dissociation of antigen-antibody complexes, causing false negative or weak positive results. Caution should be exercised in the interpretation of results of tests performed at temperatures other than those recommended.

INTERPRETATION

ACCUCARE BSA displaces serum-saline mixture by forming a layer of aggregation immediately above the cell button. It will be seen that the agglutinates formed are not easily broken down, when the tubes are gently shaken. Positive results can be seen as above, but all negative results should be checked microscopically for weak reactions, which are occasionally encountered for the presence of 'D' antigens. Similarly weak incomplete anti Rh D antibodies also can give weak results. By this way Anti Rh D anti bodies can be detected as well as can be typed for its specificity and strength.

PERFORMANCE CHARACTERISTICS

1. The reagents have been characterised by the procedures mentioned in the Recommended Techniques.
2. Prior to release, each lot of Accucare 22% and 30% Bovine Serum Albumin have been shown to enhance agglutination of Rh and other antibodies when used according to Recommended Techniques.
3. Each lot is tested to assure specificity in an antibody-free system with red cells known to possess the most frequently inherited blood group antigens.
4. The Quality Control of the reagents was performed using red cells that had been washed with PBS or Isotonic saline prior to use.
5. The reagents comply with the recommendations contained in the latest issue of the Guidelines for the UK Blood Transfusion Services.

DISCLAIMER

- Each facility should verify the optimum spin time for the specific centrifuge in use.
- Manual techniques are to be performed according to the manufacturer's instructions.
- Each deviation from these instructions is the sole responsibility of the user.
- Used tests must be discarded as hazardous material. Manage waste according to local, state and national regulations.

LIMITATION OF THE PROCEDURE

- Red cells with a positive DAT due to a coating of IgG cannot be typed by the indirect antiglobulin technique.
- False positive results may occur due to the fact that agglutinins to albumin are found in a small proportion of serum samples.
- The efficacy of albumin reagent is to be controlled throughout their use.
- Bovine Serum Albumin will not enhance the reactivity of all blood group antibodies.
- Bovine Serum Albumin should not be used as negative controls for potentiated IgG blood grouping reagents.
- False positive or false negative results may occur due to:
  - Contamination of test materials
  - Improper cell concentration
  - incubation time or temperature
  - Improper or excessive centrifugation
  - Improper storage of test materials or omission of reagent
  - Introduction of human serum/gamma globulins into test

BIBLIOGRAPHY

1. Doumas, B.T Watson, W.A. and Biggs, H.G.: Clinica Cheimca Acta 31:87, 1971.
2. Korngold, I.: Prog. Clin. Pathol. 1:398, 1966

GLOSSARY OF SYMBOL

	Consult Instruction for Use	LOT	Lot Number
REF	Catalog Number		Date of Manufacturing
	Store between		Use By or Expiration Date
	Manufacturer	IVD	For *in vitro* Diagnostic use only
	Keep away from sunlight	CONT	Content of the kit

LAB-CARE DIAGNOSTICS (INDIA) PVT. LTD.
C1 Type, Shed No.: 3225, Chemical Zone,
GIDC Sarigam – 396155, Dist. Valsad, Gujarat, India.
Tel.: +91 22 2554 2109 /1558
Email: accucarediagnostics.com; Website: www.labcarediagnostics.com