SEROLOGICAL ALBUMIN

DIRECTIONS FOR USE

22% and 30% Serological Albumin

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

INTENDED PURPOSE
The Serological albumin solutions are intended to be used to enhance the qualitative detection of irregular anti-erythrocytic antibodies in human plasa or serum when tested in accordance with the reccommended techniques stated in this IFU.

PRINCIPLE
When used by the recommended techniques, the reagent will not affect the first stage of haemagglutination (antibody uptake) but it will enhance the second stage (agglutination) by allowing the antibody-coated red cells to come closer together than they would in a saline medium without additives (see Limitations).

REAGENTS
Lorne 22% and 30% Serological 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. The reagents do not contain or consist of CMR substances, or endocrine disrupting substances or that could result in sensitisation or an allergic reaction by the user. 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.

STORAGE
Reagent vials should be stored at 2 - 8°C on receipt. Prolonged storage at temperatures outside this range may result in accelerated loss of reagent reactivity. This reagent has undergone transportation stability studies at 37°C and -25°C as described in document BS EN ISO 23640:2015.

SAMPLE COLLECTION AND PREPARATION
Blood samples can be collected into EDTA, citrate, CPDA anticoagulants or as a clotted sample. The samples should be tested as soon as possible following collection. If a delay in testing should occur, store the samples at 2-8°C. Samples displaying gross haemolysis or microbial contamination should not be used for testing. Blood samples showing evidence of lysis may give unreliable results. It is preferable (but not essential) to wash all blood samples with PBS or Isotonic saline before being tested.

PRECAUTIONS
1. The reagents are intended for *in vitro* diagnostic use only.
2. If a reagent vial is cracked or leaking, discard the contents immediately.
3. Do not use the reagents past the expiration date (see Vial Label).
4. Do not use the reagents if a precipitate is present.
5. Protective clothing should be worn when handling the reagents, such as disposable gloves and a laboratory coat.
6. The reagents have been filtered through a 0.2 μm capsule to reduce the bio-burden, but are not supplied sterile. 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.
7. 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.
8. The reagent contains <0.1% sodium azide. Sodium azide may be toxic if ingested and may react with lead and copper plumbing to form explosive metal azides. On disposal flush away with large volumes of water.

DISPOSAL OF REAGENT AND DEALING WITH SPILLAGES
For information on disposal of the reagents and decontamination of a spillage site see Material Safety Data Sheets, available on request.

CONTROLS AND ADVICE
1. Red cells sensitised with an *in vitro* or *in vivo* autoantibody may agglutinate spontaneously in concentrations of serological albumin as low as 6%. It is therefore essential to routinely set up control tests in which the red cells are mixed with the appropriate serological albumin solution alone.
2. The antiglobulin techniques can only be considered valid if all negative tests react positively with IgG sensitised red cells.
3. In the Recommended Techniques one volume is approximately 50μl when using the vial dropper provided.
4. Before use, let the reagent warm up to room temperature. As soon and the reagent has been used put the reagent back in storage at 2-8°C.
5. The use of the reagents and the interpretation of results must be carried out by properly trained and qualified personnel in accordance with the requirements of the country where the reagents are in use.
6. The user must determine the suitability of the reagents for use in other techniques.

REAGENTS AND MATERIALS REQUIRED
- Anti-human globulin, e.g. Lorne AHG Elite (Cat # 435010) or anti-human IgG, e.g. Lorne Anti-Human IgG (Cat # 401010).
- Coombs cell washer.
- Glass test tubes (10 x 75 mm or 12 x 75 mm).
- IgG sensitised red cells, e.g. Lorne Coombs Control Cells (Cat # 970010).
- Lorne Inert AB serum (Cat # 110010).
- PBS solution (pH 6.8–7.2) or Isotonic saline solution (pH 6.5–7.5).
- Test tube centrifuge.
- Volumetric pipettes.
- Water bath or dry heat incubator equilibrated to 37°C ± 2°C.

RECOMMENDED TECHNIQUES

A. Albumin Immediate Spin Technique
1. Prepare a 2-3% suspension of red cells in PBS or Isotonic saline.
2. Place in a labelled test tube: 2 volumes each of test serum, red cell suspension and 22% Serological Albumin.
3. Mix thoroughly and centrifuge all tubes for 20 seconds at 1000 rcf or for a suitable alternative time and force.
4. Examine supernatant for haemolysis, then gently resuspend cell button and examine macroscopically for agglutination.

B. Albumin Room Temperature Saline Phase Technique
1. Prepare a 2-3% suspension of red cells in PBS or Isotonic saline.
2. Place in a labelled test tube: 2 volumes test serum, 1 volume test cell suspension and 2 volumes 22% Serological Albumin.
3. Mix thoroughly and incubate at 18-25°C for 5-30 minutes.
4. Centrifuge all tubes for 20 seconds at 1000 rcf or for a suitable alternative time and force.
5. Examine supernatant for haemolysis, then gently resuspend cell button and examine macroscopically for agglutination.

C. Albumin 37°C Technique
1. Prepare a 2-3% suspension of red cells in PBS or Isotonic saline.
2. Place in a labelled test tube: 2 volumes test serum, 1 volume test cell suspension and 2 volumes 22% Serological Albumin.
3. Mix thoroughly and incubate at 37°C for 15-60 minutes.
4. Centrifuge all tubes for 20 seconds at 1000 rcf or for a suitable alternative time and force.
5. Examine supernatant for haemolysis, then gently resuspend cell button and examine macroscopically for agglutination.

D. Indirect Antiglobulin Technique (IAT)
1. Follow steps 1 to 3 of Albumin 37°C Technique above.
2. Wash red cells 4 times with PBS or Isotonic saline, taking care to decant saline between washes and resuspend each cell button after each wash. Completely decant saline after last wash.
3. Add 2 volumes of anti-human globulin to each dry cell button.
4. Mix thoroughly and centrifuge all tubes for 20 seconds at 1000 rcf or for a suitable alternative time and force.
5. Gently resuspend red cell button and read macroscopically for agglutination

E. Antibody Titration Technique
1. Prepare a 2-3% suspension of red cells in Lorne 22% Serological Albumin.
2. Prepare doubling dilutions of test serum in inert AB serum.
3. Add 1 volume of red cell suspension to 1 volume of each dilution.
4. Mix thoroughly and incubate at 37°C for 15-60 minutes.
5. Centrifuge all tubes for 20 seconds at 1000 rcf or for a suitable alternative time and force.
6. Gently resuspend each cell button and read macroscopically for agglutination.

INTERPRETATION OF TEST RESULTS
1. Positive: Agglutination of test red cells constitutes positive test result within accepted limitations of the test procedure.
2. Negative: No agglutination of the test red cells constitutes negative test result within accepted limitations.

STABILITY OF THE REACTIONS
1. Tube tests should be read immediately after centrifugation.
2. Washing steps should be completed without interruption and tests should be centrifuged and read immediately after addition of anti-human globulin. Delays may result in dissociation of antigen-antibody complexes, leading to false negative or weak positive reactions.
3. Caution should be exercised in the interpretation of results of tests performed at temperatures other than those recommended.

LIMITATIONS

1. Red cells with a positive DAT due to a coating of IgG cannot be typed by the indirect antiglobulin technique.
2. False positive results may occur due to the fact that agglutinins to albumin are found in a small proportion of serum samples.
3. The efficacy of albumin reagent is to be controlled throughout their use.
4. Serological Albumin will not enhance the reactivity of all blood group antibodies.
5. Serological Albumin should not be used as negative controls for potentiated IgG blood grouping reagents.
6. 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

SPECIFIC PERFORMANCE CHARACTERISTICS

1. Prior to release, each lot of Lorne Serological Albumin solution was tested using the recommended test methods listed in this IFU. The tests complied with the test requirements as stated in the current version/ issue of the 'Guidelines for the Blood Transfusion Services in the United Kingdom'
2. Prior to release, each lot of Lorne 22% and 30% Serological 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 with phenotypes that were verified by a UK transfusion centre and 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

1. The user is responsible for the performance of the reagents by any methods other than those mentioned in the Recommended Techniques.
2. Any deviations from the Recommended Techniques should be validated prior to use[5].

BIBLIOGRAPHY

1. Technical Manual, 16th Edition. American Association of Blood Banks, Bethesda, MD, 2008; Chapter 15.
2. Issitt PD. Applied Blood Group Serology, 3rd Edition. Montgomery Scientific, Miami 1985; Chapter 3
3. Guidelines for the Blood Transfusion Service in the United Kingdom, 6th Edition 2002. The Stationery Office.
4. British Committee for Standards in Haematology, Blood Transfusion Task Force. Recommendations for evaluation, validation and implementation of new techniques for blood grouping, antibody screening and cross matching. Transfusion Medicine, 1995, 5, 145-150.

AVAILABLE REAGENT SIZES

	Vial Size	Catalogue Number	Test per vial
22% Serological Albumin	10 ml	451010	100
	1000 ml	451000*	10,000
30% Serological Albumin	10 ml	452010	100
	1000 ml	452000*	10,000

*These sizes are For Further Manufacturing Use (FFMU) only and are therefore not CE marked.

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