

LORNE LABORATORIES LTD.

SEROLOGICAL ENHANCEMENT MEDIUM

DIRECTIONS FOR USE

LISS Ready for use: For Potentiating Serological Techniques.

SUMMARY

Reducing the ionic strength of a test system increases the rate of red blood cell antigen-antibody binding. Low and Messeter in 1974 showed that the use of a low ionic strength solution enhances the rate of antibody uptake in first stage of agglutination, allowing incubation times to be shortened.

PRINCIPLE

When used by the recommended techniques, the solution will reduce the ionicstrength of a test system, increase the rate of red blood cell antigen-antibody binding and permits a substantial reduction in incubation time and an increases in the test sensitivity with many antibody specificities (see Limitations).

REAGENT

Lorne LISS ready for use is a low ionic strength solution containing glycine, sodium chloride and phosphate buffer. The reagent is supplied at the 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 Label.

STORAGE

Reagent vials should be stored at 10 - 30°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.

PRECAUTIONS

- The reagent is intended for in vitro diagnostic use only. 1.
- If vial is cracked or leaking, discard the contents immediately. 2.
- 3. Do not use the reagent past the expiration date (see Vial Label).
- Do not use the reagent if a precipitate is present. 4.
- 5. Protective clothing should be worn when handling the reagent, such as disposable gloves and a laboratory coat.
- The reagent has been filtered through a 0.2 μ m 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 6. indicate reagent deterioration or contamination.
- The reagent contains < 0.1% sodium azide. Sodium azide may be toxic if 7. ingested and may react with lead and copper plumbing to form explosive metal azides. On disposal flush away with large volumes of water.
- 8. Contact with LISS together with bleach causes accelerated corrosion of base metals such as copper and iron. This should be borne in mind when considering the use of bleach for decontaminating pluming or apparatus with metal parts, which have also been in contact with LISS

DISPOSAL OF REAGENT AND DEALING WITH SPILLAGES

For information on disposal of the reagent and decontamination of a spillage site see Material Safety Data Sheets, available on request.

CONTROLS AND ADVICE

- It is recommended Lorne Precise Weak Anti-D and appropriate red cells 1. (ideally R1r and rr) be tested in parallel with each batch of tests. Tests must be considered invalid if controls do not show the expected results.
- The antiglobulin technique can only be considered valid if all negative tests 2. react positively with IgG sensitised red cells
- 3 The LISS solution, red cell suspensions and test sera should be at room temperature prior to use to avoid encountering unwanted positive reactions due to "cold" antibodies.
- In the Recommended Techniques one drop is approximately 50 µl when 4. using the vial dropper provided
- The use of the reagent and the interpretation of results must be carried out 5. by properly trained and qualified personnel in accordance with the requirements of the country where the reagents are in use.
- The user must determine the suitability of the reagent for use in other 6 techniques

REAGENTS AND MATERIALS REQUIRED

- Anti-human globulin i.e. Lorne AHG Elite (Cat # 435010 or 415010) or antihuman IgG i.e. Lorne Anti-Human IgG (Cat # 401010 or 402010).
- Coombs cells washer.
- Glass test tubes (10 x 75 mm or 12 x 75 mm).
- IgG sensitised red cells i.e. Lorne Coombs Control Cells (Cat # 970010).
- Lorne Precise Weak Anti-D (Cat # 209005). PBS solution (pH 6.8-7.2) or Isotonic saline solution (pH 6.5-7.5). •
- Positive (ideally R1r) and negative (rr) control red cells.
- Volumetric pipettes.
- Water bath or dry heat incubator equilibrated to $37^{\circ}C \pm 2^{\circ}C$.

RECOMMENDED TECHNIQUE

- Wash red cells at least twice in PBS or Isotonic saline and then wash once 1. in Lorne LISS "Ready For Use".
- Resuspend red cells to 1.5-2.0% in LISS "Ready For Use". 2.
- 3. Equal volumes of LISS suspended red cells and serum should be mixed thoroughly for LISS procedures, e.g. 2 volumes of 1.5-2% cell suspension and 2 volumes of serum.

LIMITATIONS

- The suspension of red cells in LISS is associated with an accelerated deterioration in the expression of $Fy^a,\,Fy^b,\,s$ and S antigens and therefore 1. red cells suspended in LISS should be discarded within 24 hours of their preparation. Adherence to 1:1 volumetric ratio of cell suspension to serum and thorough
- 2. For optimum sensitivity, LISS IAT should be incubated for a minimum of 15
- 3. minutes at 37°C.
- 4. In order to avoid non-specific uptake of autologous complement red cells should be washed at least twice in NISS before they are finally washed and resuspended in LISS
- Not all antigen-antibody reactions are enhanced by LISS techniques. 5.

SPECIFIC PERFORMANCE CHARACTERISTICS

- Prior to release, each lot of Lorne LISS "Ready For Use" has been shown 1. to enhance many antigen-antibody reactions when used by the Recommended Techniques.
- 2. The solution complies with the recommendations contained in the latest issue of the Guidelines for the UK Blood Transfusion Services.

DISCLAIMER

- The user is responsible for the performance of the reagent by any methods 1. other than those mentioned in the Recommended Techniques.
- Any deviations from the **Recommended Techniques** should be validated prior to use¹⁰. 2

BIBLIOGRAPHY

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- 2. Moore C., Mollison P.L. Use of low ionic strength saline medium in manual tests for antibody detection. Transfusion 1976; 16. 291-296.
- Wicker B., Wallas C.H. A comparison of low ionic strength saline medium 3. with routine methods for antibody detection. Transfusion 1976; 16. 469-472.
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- 5. techniques in blood bank management. Med. Lab. Sci. 1980; 37. 119-125.
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- 7. Voak D., Downie M., Haigh T.J., Cook N. Improved antiglobulin tests to detect difficult antibodies: detection of Anti-Kell by LISS. Med. Lab. Sci. 1982; **39**. 363-370.
- Phillips P.K., Bebbington C. The pH, conductivity and osmolality of low 8. ionic strength solutions used within the U.K. for the antiglobulin test. Transfusion Medicine 1991; 1. 155-158.
- Guidelines for the Blood Transfusion Service in the United Kingdom. 9. H.M.S.O. Current Edition.
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AVAILABLE REAGENT SIZES

Vial Size	Catalogue Number	
4x250 ml	470250	
20x250 ml	470020	
1x2500 ml	470025*	

*This size is For Further Manufacturing Use (FFMU) only and are therefore not CE marked.

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TABLE OF SYMBOLS

LOT	Batch Number	IVD	<i>in-vitro</i> Diagnostic
REF	Catalogue Reference		Store At
	Expiry Date		Manufacturer
≡i	Read Pack Insert		