



10 X CONCENTRATED LOW IONIC STRENGTH SOLUTION (LISS) DIRECTIONS FOR USE

LISS Concentrate: 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 ionic strength of a test system, increase the rate of red blood cell antigen-antibody binding and permits a substantial reduction in incubation time and an increase in the test sensitivity with many antibody specificities (see **Limitations**).

REAGENT

Lorne LISS Concentrate is a solution of glycine, phosphate buffer and 0.3 M sodium chloride. The solution is supplied at a stronger concentration than needed for serological use. It must be diluted 10 times in deionised water before being used with all recommended techniques mentioned. For lot reference number and expiry date see **Bottle 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

1. The reagent is intended for *in vitro* diagnostic use only.
2. If bottle is cracked or leaking, discard the contents immediately.
3. Do not use the reagent past the expiration date (see **Vial Label**).
4. Do not use the reagent if a precipitate is present.
5. Protective clothing should be worn when handling the reagent, such as disposable gloves and a laboratory coat.
6. The reagent has been filtered through a 0.2 µm 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.
7. 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.
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 plumbing 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

1. It is recommended Lorne Precise Weak Anti-D and appropriate red cells (ideally R_{1r} and rr) be tested in parallel with each batch of tests. Tests must be considered invalid if controls do not show the expected results.
2. The antiglobulin technique can only be considered valid if all negative tests 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.
4. In the **Recommended Techniques** one drop is approximately 50 µl when using the vial dropper provided
5. The use of the reagent 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 reagent for use in other techniques.

REAGENTS AND MATERIALS REQUIRED

- Anti-human globulin i.e. Lorne AHG Elite (Cat # 435010 or 415010) or anti-human IgG i.e. Lorne Anti-Human IgG (Cat # 401010 or 401010).
- Conductivity meter.
- 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).
- Osmolality reader.
- pH meter.
- PBS solution (pH 6.8-7.2) or Isotonic saline solution (pH 6.5-7.5).
- Distilled or deionised water.
- Positive (ideally R_{1r}) and negative (rr) control red cells.
- Volumetric pipettes.
- Water bath or dry heat incubator equilibrated to 37°C ± 2°C.

DILUTION OF LISS CONCENTRATE

1. Check container of LISS Concentrate for deposits of solutes which if present should be thoroughly re-dissolved before dilution of concentrate.
2. Accurately dilute 1 volume of Lorne LISS Concentrate with 9 volumes of good quality distilled or deionised water. The diluted solution should be measured and be within the following parameters:
 - pH: 6.7 ± 0.2 at 22°C ± 1°C.
 - Conductivity: 3.7 ± 0.3 mS/cm at 22°C ± 1°C.
 - Osmolality: 295 ± 10 mOsm/Kg
3. LISS "Ready for Use" is stable at 18-25°C for 4 weeks, provided that contamination is avoided.
4. If stored at 2-8°C, LISS "Ready for Use" should be brought to room temperature prior to use.
5. Discard solution if it is turbid.

RECOMMENDED TECHNIQUE

1. Wash red cells at least twice in PBS or Isotonic saline and then wash once in Lorne LISS "Ready For Use".
2. Resuspend red cells to 1.5-2.0% in LISS "Ready For Use".
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

1. 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 red cells suspended in LISS should be discarded within 24 hours of their preparation.
2. Adherence to 1:1 volumetric ratio of cell suspension to serum and thorough mixing is essential to the integrity of the low ionic test system.
3. For optimum sensitivity, LISS IAT should be incubated for a minimum of 15 minutes at 37°C.
4. In order to avoid non-specific uptake of autologous complement red cells should be washed at least twice in LISS before they are finally washed and resuspended in LISS.
5. Not all antigen-antibody reactions are enhanced by LISS techniques.

SPECIFIC PERFORMANCE CHARACTERISTICS

1. Prior to release, each lot of Lorne LISS Concentrate has been shown 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

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

BIBLIOGRAPHY

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8. Phillips P.K., Bebbington C. The pH, conductivity and osmolality of low ionic strength solutions used within the U.K. for the antiglobulin test. *Transfusion Medicine* 1991; **1**. 155-158.
9. Guidelines for the Blood Transfusion Service in the United Kingdom. H.M.S.O. Current Edition.
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
AVAILABLE REAGENT SIZES

Vial Size	Catalogue Number
500 ml	460500
2500 ml	460025

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TABLE OF SYMBOLS

	Batch Number		<i>In-vitro</i> Diagnostic
	Catalogue Reference		Store At
	Expiry Date		Manufacturer
	Read Pack Insert		