



LECTIN BLOOD GROUPING REAGENTS  
DIRECTIONS FOR USE

**Anti-N Lectin: For Tube and Bio-Rad-ID Techniques.**

**SUMMARY**

The N antigen is part of the MNSs system and was reported in 1927. Anti-N generally reacts at room temperature and so is rarely implicated in Haemolytic Transfusion Reactions and Haemolytic Disease of the Newborn.

Anti-M	Anti-N	Phenotype	Caucasians %	Afro-Americans %
+	0	M+N-	28	26
+	+	M+N+	50	44
0	+	M-N+	22	30

**PRINCIPLE**

The reagent will cause agglutination (clumping) of red cells, that carry the N antigen, after centrifugation. No agglutination generally indicates the absence of the N antigen (see **Limitations**).

**REAGENT**

Lorne Anti-N Lectin blood grouping reagent is prepared from an extract of *Vicia* seeds or leaves, diluted with a sodium chloride solution containing bovine albumin. The reagent is supplied at optimal dilution for use with all 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 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 reagent is 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 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 reagents, 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. No known tests can guarantee that products derived from human or animal sources are free from infectious agents. Care must be taken in the use and disposal of each vial and its contents.

**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 a positive control (ideally heterozygous cells) and a negative control be tested in parallel with each batch of tests. Tests must be considered invalid if controls do not show expected results.
2. In the **Recommended Techniques** one volume is approximately 50µl when using the vial dropper provided.
3. Use of reagent and interpretation of results must be carried out by properly trained and qualified personnel in accordance with the requirements of the country where the reagent is in use. The user must determine suitability of the reagent for use in other techniques.

**REAGENTS AND MATERIALS REQUIRED**

- Bio-Rad ID-Cards (NaCl, Enzyme test and Cold Agglutinins).
- Bio-Rad ID-Centrifuge.
- Bio-Rad ID-CellStab or ID-Diluent 2.
- Bio-Rad ID-Incubator equilibrated at 37°C ± 2°C.
- Glass test tubes (10 x 75 mm or 12 x 75 mm).
- PBS solution (pH 6.8-7.2) or Isotonic saline solution (pH 6.5-7.5).
- Positive (ideally M+N+) and negative (M+M+) control red cells.
- Test tube centrifuge.
- Volumetric pipettes.

- Water bath or dry heat incubator equilibrated to 37°C ± 2°C.

**RECOMMENDED TECHNIQUES**

**A. Tube Technique**

1. Prepare a 2-3% suspension of red cells in PBS or Isotonic saline.
2. Place in a labelled test tube: 1 volume of Lorne reagent and 1 volume of red cell suspension.
3. Mix thoroughly and then incubate at 37°C for 15 minutes.
4. Centrifuge all the 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

**B. Bio-Rad ID Micro Typing Technique**

1. Prepare a 0.8% suspension of red cells in ID-CellStab or ID-Diluent 2.
2. Remove aluminium foil from as many microtubes on a Bio-Rad NaCl, Enzyme test and Cold Agglutinins gel card as needed.
3. Place in appropriate microtube: 50 µl of red cell suspension and 25µl of Lorne reagent.
4. Incubate the ID-Card(s) for 15 minutes at 37°C.
5. Centrifuge ID-Card(s) in a Bio-Rad ID centrifuge.
6. Read macroscopically for agglutination.

**INTERPRETATION OF TEST RESULTS**

1. **Positive:** Agglutination of red cells constitutes a positive test result and within the accepted limitations of the test procedure, indicates the presence of the N antigen on the red cells.
2. **Negative:** No agglutination of the red cells constitutes a negative result and within the accepted limitations of the test procedure, indicates the absence of the N antigen on the red cells.

**STABILITY OF THE REACTIONS**

1. Tube tests must be read immediately after centrifugation. Delays may cause dissociation of antigen-antibody complexes leading to false negative or weak positive reactions.
2. Caution should be exercised in the interpretation of results of tests performed at temperatures other than those recommended.

**LIMITATIONS**

1. Stored blood may give weaker reactions than fresh blood.
2. False positive or false negative results may also occur due to:
  - Contamination of test materials
  - Improper storage, cell concentration, incubation time or temperature
  - Improper or excessive centrifugation
  - Deviation from the recommended techniques

**SPECIFIC PERFORMANCE CHARACTERISTICS**

1. The reagent has been characterised by all the procedures mentioned in the **Recommended Technique**.
2. Prior to release, each lot of Lorne Anti-N Lectin reagent is tested by the **Recommended Technique** against a panel of antigen-positive red cells to ensure suitable reactivity.
3. The Quality Control of the reagent was performed using red cells that had been washed twice with PBS or Isotonic saline prior to use.
4. The reagent 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 method other than those mentioned in the **Recommended Techniques**.
2. Any deviations from the **Recommended Techniques** should be validated prior to use<sup>6</sup>.

**BIBLIOGRAPHY**

1. Widman FK. Technical Manual, 9<sup>th</sup> Edition. American Association of Blood Banks, Arlington, VA, 1985; Chapter 8
2. Race RR, Sanger R. Blood Groups in Man, 6<sup>th</sup> Edition. Blackwell Scientific, Oxford 1975; Chapter 2
3. Mollison PL. Blood Transfusion in Clinical Medicine, 8<sup>th</sup> Edition. Blackwell Scientific, Oxford 1987; Chapter 7
4. Issitt PD. Applied Blood Group Serology, 3<sup>rd</sup> Edition. Montgomery Scientific, Miami 1985; Chapter 6
5. Guidelines for the Blood Transfusion Service in the United Kingdom. H.M.S.O. Current Edition.

6. 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
2 ml	312002
1000 ml	312000*

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

For the availability of other sizes, please contact:

**Lorne Laboratories Limited**  
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 Danehill  
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**TABLE OF SYMBOLS**

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