



## Instructions For Use

### Anti-A, Anti-B & Anti-A+B (Agglutination Method)

Cat no.	item	size
4101 101	Anti A	10 mL
4102 101	Anti B	10 mL

#### INTENDED USE

Anti-A, Anti-B and Anti-AB Reagent is intended for the in vitro Qualitative diagnostic determination of Blood groups A, B and AB in Human Blood.

#### Background

Monoclonal antibodies are derived from hybridoma cell lines, created by fusing mouse antibody producing B lymphocytes with mouse myeloma cells. Each hybridoma cell line produces homogenous antibodies of only one immunoglobulin class, which are identical in their chemical structure and immunological activity.

Human red Blood cell antigens can be divided into four groups A, B, AB and O depending on the presence or absence of the corresponding antigens on the red blood cells. Approximately 41% of the Caucasian population have the A antigen, 9% have the B antigen, 4% have both A and B antigens, while remaining have neither A nor B antigen

#### Assay Principle

Human red blood cells possessing A and/or B antigen will agglutinate in the presence of antibody directed towards the antigen.

Agglutination of red blood cells with Anti-A, Anti-B, Anti-AB reagents is a positive test result and indicates the presence of the corresponding antigen.

Absence of agglutination of red blood cells with Anti-A, Anti-B, Anti-AB reagents is a negative test result and indicates the absence of the corresponding antigen.

#### Note

- In-vitro diagnostic reagent for laboratory and professional use only. Not for medicinal use.
- The reagent contains sodium azide 0.1% as preservative. Avoid contact with skin and mucosa. On disposal flush with large quantities of water.
- Extreme turbidity may indicate microbial contamination or denaturation of protein due to thermal damage. Such reagent should be discarded.
- blood grouping reagents are not from human sources, hence contamination due to HBsAg and HIV is practically excluded.

#### Reagents

Anti-A, Anti-B and Anti-AB are ready-to-use reagents prepared from supernatants of mouse hybridoma cell cultures. These antibodies of immunoglobulin class IgM are a mixture of several monoclonal antibodies of the same specificity but having the capability of recognizing different epitopes of the human red blood cell antigens A and B. Each batch of reagent undergoes quality control at various stages of manufacture for its specificity, avidity and performance.

#### Reagent Storage and Stability

- Store the reagent at 2-8 °C. Don't freeze.
- The shelf life of reagent is as per the expiry date mentioned on the reagent vial label

#### Precautions and Warnings

Do not ingest or inhale. In case of contact with eyes or skin; rinse immediately with plenty of soap and water. In case of severe injuries; seek medical advice immediately.

Symbols in Product Labeling			
	Authorized Representative		Expiration date
	For in-vitro diagnostic use		CAUTION, consult instructions for use
	Catalogue number		Manufactured by
	Lot number		Temperature Limit
	Consult instructions for use		

#### Specimen Collection and Storage

No special preparation of the patient is required prior to sample collection by approved techniques. Samples should be stored at 2-8 °C if not tested immediately. Do not use haemolysed samples. Anticoagulated blood using various anticoagulants should be tested within the below mentioned time period:

- EDTA or heparin : 2 days
- Sodium citrate or Sodium oxalate : 14 days
- ACD or CPD : 28 days

Clotted whole blood should be tested within 14 days

#### Additional Material Required For Slide And Tube Tests

Glass slides (50x75 mm), Test tubes (12x75 mm), Pasteur pipettes, isotonic saline, Centrifuge, timer, mixing sticks.

#### Procedure

- 1-Bring reagent and samples to room temperature before testing.
- 2-Slide Test Place one drop of Anti-A or Anti-B or Anti-AB reagent on a clean glass slide.
- 3-To each reagent drop, add one small drop (50 µl) of whole blood.
- 4-Mix well with a mixing stick uniformly over an area of approximately 2.5 cm<sup>2</sup>.
- 5-Rock the slide gently, back and forth. Observe for agglutination macroscopically at two minutes.
- 6-Tube Test Prepare a 2-3% suspension of the red cells to be tested in Isotonic saline.
- 7-Place one drop of Anti-A, Anti-B, Anti-AB into correspondingly labeled test tubes.
- 8-Pipette into each of the test tubes, one drop ( 50µl) of the test red cell suspension and mix well.
- 9-Centrifuge for **1 minute** at 1000 rpm (125 g) or **20 seconds** at 3400 rpm (1000 g) or incubate at room temperature for **20-30 minutes**.
- 10-Gently resuspend the cell button, observing for agglutination macroscopically.

#### Interpretation Of Results (Slide and Tube tests)

Agglutination is a positive test result and indicates the presence of A and/or B antigen. Do not interpret peripheral drying or fibrin strands as agglutination. No agglutination is a negative test results and indicates the absence of A and/or B antigen.

#### References

- 1.Kohler C. & Milstein C. (1975), Continuous cultures of fused cells secreting antibody of predefined specificity, Nature, 256, 495-497.
- 2.Lee H.H., Rouger P, Germain C., Muller A. & Salmon C. (1983), The production and standardisation of monoclonal antibodies as AB blood group typing reagents.
- 3.Symposium of International Association of Biological Standardisation on Monoclonal antibodies.
- 4.Human Blood Groups by Geoff Daniels, 1st Ed., Blackwell Science, Oxford 1995.
- 5.HMSO, Guidelines for the Blood Transfusion Services, 2nd Ed., 1994.