Anti-A, Anti-B, Anti-AB and Anti-D

MONOCLONAL ABO BLOOD GROUPING REAGENTS
(Tube, Slide and Microplate Tests)

REF: **Anti-A500100** **Anti-B501100** **Anti-D502100** **Anti-AB503100**

Principle :

The test is based on the agglutination principle. Agglutination occurs when red cells possessing the antigen are tested against the corresponding antibody.

Summary:

The BioMed ABO grouping reagents are prepared using the in-vitro culture supernatants of hybridized immunoglobulin-secreting mouse cell. Lines the antibodies are diluted in a phosphate buffer containing sodium chloride, EDTA and bovine albumin resulting in a reagent which is optimized for tube, slide or microplate testing. Anti-A is colored with a blue dye, Anti-B is colored with a yellow dye while Anti-AB is not colored,

Anti -D has been prepared from a blend of both **IgG and IgM anti-Ds. The IgG anti-D** agglutinates with the D and low grade weak D (D) Phenotypes by the indirect antiglobulin test (IAT). The IgM anti-D agglutinates with D positive red cells which would include the majority of D variants Value (with the exception of D, and a high proportion of weak D or D phenotypes. The antibodies are diluted in phosphate buffer containing sodium chloride, Bovine albumin and macromolecular potentiators resulting In reagent optimization for tube, slide and microplate testing, the reagents contain 0.1% sodium azide as a preservative and are supplied ready-to-use.

Samples :

Blood samples with or without anticoagulant may be used. Ideally samples should be tested as soon as possible. However samples may be Stored between 2°C and 8°C provided the following criteria applies: EDTA and heparinised samples should be tested within 48 hours

- Clotted samples within 14 days.
- ACD, CPD or CPDA-1 up to date of expiry or within 35 days of bleeding

SPECIMEN COLLECTION :

Blood samples with or without anticoagulant may be used. Ideally samples should be tested as soon as possible; however samples which have been stored at 2°C – 8°C for up to two days may be tested provided there is no evidence of haemolysis

TEST PROCEDURES:

TUBE TECHNIQUE:

1- Prepare a 2-3% suspension of red cells in isotonic buffered saline (pH6.9)
2- Into a glass tube dispense 100µl of the BioMed ABO Grouping reagent and the 2-3% cell suspension.
3- Mix well and incubate at room temperature (18°C – 25°C) for 5 – 15 minutes.
4- Centrifuge at 1000g for 15 seconds.
5- Gently resuspend the cell button and examine macroscopically for signs of agglutination.
6-Record the results.

SLIDE TECHNIQUE:

1. Dispense onto a glass slide 1 drop of the BioMed ABO Grouping reagent and 1 drop Of 35 – 45% cells suspended in their own or group compatible plasma or serum.
2. Using a clean stirrer, mix and spread the reagent.
3. Tilt the slide back and forth, observing for signs of agglutination.
4. After no more than two minutes samples not showing agglutination are interpreted as negative.
5. Record the results.

MICROTITRE PLATE TECHNIQUE

1. Prepare a 2-3% suspension of red cells in isotonic buffered saline (pH6.9).
2. Dispense into the appropriate well of a U-well micro titration plate 40µl of the BioMed ABO Grouping reagent and 40µl of the 2-3% cell suspension.
3. Mix well; ensuring no cross-well contamination occurs.
4. Incubate at room temperature (18°C – 25°C) for 5 to 15 minutes.
5. Centrifuge the micro plate at 140 rcf for 1 minute.
6. Tilt the plate at an angle of between 60° and 90° for up to three minutes and observe for streaming. Negative reactions allow the cells to flow downwards in a uniform stream. Positive reactions remain as distinct buttons, either on the bottom of the well or occasionally sliding down the side. The cell buttons can be resuspended using carefully controlled agitation on a micro plate shaker, then examined for agglutination either visually or by using an automatic plate reader.
7. Record results.

STABILITY OF REACTIONS:

*Tube and micro plate results should be read immediately following Centrifugation and the results interpreted and recorded without delay.
*Slide Tests should be interpreted within two minutes to minimize the Incidence of false positive results which can be caused by the drying of the reagents.

QUALITY CONTROL :

* To confirm the performance of the BioMed ABO Blood Grouping reagents it is recommended that the appropriate Antigen-positive and Antigen-negative cells be tested with each run of tests performed.
* Reverse grouping is recommended as confirmation of grouping interpretations on patients older than 6 months of age.

EXPECTED VALUES :

Reagent			Cell Suspension				ABO Group
Anti-A	Anti-B	Anti-A,B	A1	A2	B	O	
+	–	+	–	–	+	–	A
–	+	+	+	+	–	–	B
+	+	+	–	–	–	–	AB
–	–	–	+	+	+	–	O

If the results obtained with the serum do not correlate with the red cell test, further investigation is required.

LIMITATIONS:

1. False Positive or False Negative results may occur due to contamination of test materials, improper cell concentration,
Centrifugation, incubation time or temperature, or any deviation from the recommended test procedure.
2. Weaker reactions may be observed with stored rather than with fresh blood.
3. ABO antigens are not fully developed at birth – therefore weaker reactions may occur with cord or neonatal red cells.
4. Cord samples contaminated with Wharton's jelly may give false positive results.
5. Blood samples of weak A and B sub groups may give rise to false negative or weak reactions.

STABILITY DATA

*Kit components are stable to expiry if stored at 2oC to 8oC.
*Do not freeze or expose to elevated temperatures.

NOTE:

*Marked turbidity may indicate reagent deterioration or contamination.
*As there is no known test method which can guarantee that products derived from Human or animal sources are free from infectious agents, the handling and disposal of these reagents should therefore be done with care.

PRESENTATION:

Product Code:	A-10	Anti-A	10 x 10 ml Pack
	B-10	Anti-B	10 x 10 ml Pack
	AB-10	Anti-AB	10 x 10 ml Pack
	D -10	Anti-D	10 x 10 ml Pack
Product Code:	A-3	Anti-A	3 x 10 ml Pack
	B-3	Anti-B	3 x 10 ml Pack
	AB-3	Anti-AB	3 x 10 ml Pack
	D -3	Anti-D	3 x 10 ml Pack

BIIBLIOGRAPHY:

Kholer G., Milstein C. (1975) Continuous culture of fused cells secreting antibody of predefined specificity. Nature, 256, 495 – 497.
Race R.R., Sanger R. Blood Groups in Man, 6thed. Oxford: Blackwell Scientific, 1975: 323-49.
Issitt P.D. Applied Blood Group Serology 3rd Ed. Miami: Montgomery Scientific, 1985: 127-153
Mollison P.L. Blood Transfusion in Clinical Medicine, 8th ed. Oxford: Blackwell Scientific, 1987: Ch 8.
Voak D., et al, (1982). Monoclonal anti-A and anti-B development as cost effective reagents. Med. Lab. Sci., 39, 109-122.

	Consult Instructions for Use
	Caution, Consult accompanying
IVD	In Vitro Diagnostic Medical Device
8°C 2°C	Temperature Limitation
	Manufacturer
EC REP	Authorized Representative in the European Community
REF	Catalogue Number
LOT	Batch Code
	Use by

EGY- CHEM for lab technology
Badr City, Industrial Area Piece 170
250 Faddan In East of Elrubaki, EGYPT.
Office Tel: +202 26236727 / +202 26236598
Factory Tel: + 202 23108170 / +202 23108171
Fax: +202 26240986
www.egy-chem.com