

## INSTRUCTIONS FOR USE

# Anti-M monoclonal (IgG)



## Klon: M-11H2

FOR IN VITRO DIAGNOSTIC USE

### **1. Description of product**

Anti-M (clone **M-11H2**) is made from monoclonal mouse IgG antibodies. Anti-M monoclonal (IgG) aids for the detection of the corresponding antigens on erythrocytes and is suitable for the test tube procedure.  $\text{NaN}_3$  (< 0.1%) is added to the test reagent as a preservative.

### **2. Principle of the test**

The testing method indicated is based on the principle of hemagglutination. Incubation of test red cells with the test reagent will result in a specific antigen-antibody reaction if the corresponding antigen is present on the test red cells. Visible detection of this reaction is demonstrated by agglutination of the cells. No agglutination indicates a negative test result, and within the accepted limitations of the test procedure, indicates the absence of the corresponding antigen.

### **3. Storage and stability**

Store the test reagent at 2...8°C. Do not freeze! Allow the reagent to come to room temperature before use and store again at 2...8°C after use.

Once Anti-M has been opened the first time, the test reagent may be used up to the expiration date indicated on the label if the specified storage conditions are observed.

Do not use the reagent past the expiration date indicated on the label.

### **4. Preparation of samples**

The blood samples should be collected according to the customary medical procedure. Blood samples with and without anticoagulants (EDTA, citrate) are suitable for testing. Do not use hemolytic samples! Testing should take place without delay whenever possible. If this is not possible, store blood samples at 2...8°C.

If erythrocytes are stored for too long before testing, the red blood cell antigens may change, which can lead to weakened reactions (see 9. Important Notes/Limitations of the Method).

### **5. Additional materials required**

0.9% NaCl solution (isotonic saline)

Test tubes (75 x 12 mm)

Single-use Pasteur pipettes

Centrifuge

## **6. Test procedure (Tube test)**

1. Wash the red cells to be examined at least once in cold isotonic saline and then prepare a 2 – 3% suspension of test red cells in isotonic saline.
2. Mix 1 drop of monoclonal test reagent and 2 drops of the test red cell suspension in a labeled test tube and incubate at room temperature for 5 minutes.  
For titer determination an incubation time of 10 minutes are recommended.
3. Centrifuge 1 minute at 180 - 270 x g (~1000 rpm) or at an alternative rpm with an appropriate time adjustment.
4. Resuspend the cells by gently shaking the tube and examine macroscopically for agglutination.

**Comments:** Do not examine the test microscopically.  
Erythrocytes that are positive with regard to the M antigen, and erythrocytes that are negative with regard to the M antigen, as well as a negative control for monoclonal test reagents and an autocontrol to test for autoagglutination must also be tested as controls. The determination of antigens should be carried out with at least 2 different test reagents. When using two monoclonal test reagents, two different clones should be used, if possible.

## **7. Interpretation of the results**

Agglutination of test red cells with the test reagent indicates the presence of the M antigen (within the accepted limitations of the test procedure).

No agglutination of test red cells with the test reagent indicates the absence of the M antigen (within the accepted limitations of the test procedure).

If no agglutination occurs with the test red cells known to be positive for the antigen or if agglutination occurs with the test red cells known to be negative for the antigen or with the negative control for monoclonal test reagents or with the auto-control the test results should not be interpreted. If different test results occur with two different test reagents, repeat the determination of the antigen with another test method and/or an other test reagent.

Pay attention to the limitations of procedure and important directions (s. 9. Important notes/Limitations of the method).

## **8. Stability of reactions**

All test results must be interpreted immediately once the test is performed.

## **9. Important notes/Limitations of the method**

1. The test reagent is designed for in vitro diagnostic use only and should be used only by properly trained, qualified personnel.
2. By treatment with enzymes the M antigen on erythrocytes may be destroyed. Therefore the test reagent should not be used for the testing of enzyme-treated erythrocytes.
3. Hemolytic samples should not be used.
4. The strength of positive reactions depends on the age of the used blood.
5. Light cloudiness does not influence the reactivity of the product.
6. In rare cases, spontaneous and non-specific agglutinations may occur with red cells coated with immunoglobulins in vivo. In such instances similar phenomena would most likely occur in blood grouping tests of other blood group systems as well. Therefore, as a control, a negative control for monoclonal test reagents and an autologous patient serum should always be tested as well. If the control tests also show a positive reaction, the result of the blood type determination cannot be interpreted.
7. Suspensions of unwashed red cells in plasma or serum promote false positive reactions. The use of well washed red cells can reduce the occurrence of such false positive reactions.

8. Reading the results of the test too late, agitating the red cell sediment too strongly, and other deviations from the indicated testing procedure can lead to weaker or false negative results.
9. False negative results or unexpected weak reactions may be caused by storing the red cells for too long and/or under inappropriate conditions and/or by a cell concentration that is too low.
10. False negative or false positive results can result from inappropriate techniques, incorrect centrifugation or incubation, dirty tubes, incorrect pH of solutions and/or contaminated materials and samples.
11. A microbial or chemical contamination of the test reagent must be absolutely avoided because this shortens the shelf life of the product and can lead to false results.
12. No single centrifugation speed or time can be recommended for all types of available centrifuges or test applications. Centrifuges should be calibrated individually to determine the optimal time and speed required to produce a clear supernatant and a clearly delineated red cell button that can be easily resuspended.
13. Deviation from the recommended Instructions for Use may result in less than optimal product performance. User-defined deviations such as modifications of test procedures, serum dilution for use in automat or cards, freezing of serum on microtiter plates etc. may require validation by the user.
14. Whether transfusions or transplantations have taken place should always be taken into consideration when interpreting the results. Any history of transfusions and/or transplantations, as well as the patient's medication history, should be taken into consideration when interpreting results.

#### **10. Warnings and instructions for disposal**

All materials of biological origin used for the test, especially the red cells to be tested, should be regarded as potentially infectious. Therefore, appropriate safety precautions are recommended when handling biological materials (do not pipette using the mouth; wear protective gloves when performing the test; disinfect hands after testing). Biological material should be inactivated before disposal (e.g. in an autoclave). Disposables should be autoclaved or incinerated after use.

Spillage of potentially infectious materials should be removed immediately with absorbent paper tissue and the contaminated areas swabbed with a suitable standard disinfectant or 70% alcohol. Material used to clean spills, including gloves, should be inactivated before disposal (e.g. in an autoclave).

The test reagent contains  $\text{NaN}_3$  as a preservative. The reagent contains < 0.1%  $\text{NaN}_3$  which is not considered to be a harmful concentration. Nevertheless avoid contact with the skin and mucous membranes. The copper and lead used in some plumbing systems can react with azides to form explosive salts. The quantities of azide used in this reagent are small; nevertheless when disposing of azide-containing materials, they should be flushed away with a large volume of water.









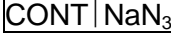

Disposal of all specimen and test materials should be in accordance with state and local law.

**11. Package sizes**                      See price list.

#### **12. Bibliography**

Applied Blood Group Serology, PD Issitt and DJ Anstee  
4<sup>th</sup> Edition, Montgomery Scientific, Durham SC, 1998

Technical manual of the American Association of Blood Banks, 15<sup>th</sup> ed., 2005

Explanation of symbols used on Labelling	
	For in vitro diagnostic use
	Storage temperature
	Batch code
	Use by
REF	Catalogue number
	Consult instructions for use
	Monoclonal IgG
	Clone
	Origin: murine
	Contains Natriumazide
	Titer

Instructions for use	Issue: February 2010
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