Blood Grouping Reagent

Anti-M (MNS1)

Seraclone® Murine Monoclonal
(BS57)

FOR IN-VITRO DIAGNOSTIC USE
For Tube Testing
MEETS FDA POTENCY REQUIREMENTS
U.S. License Number: 1845
Rx only

PACKAGE SIZE
[REF] 808410100 [VOL] 2 mL Seraclone® Anti-M (MNS1)

INTENDED USE
For the determination of the M (MNS1) antigen of red blood cells using the tube test.

SUMMARY
Antibodies to the M antigen are usually of the IgM class and appear as saline reactive or cold agglutinins. However, in rare cases an IgG anti-M may cause hemolytic disease of the fetus and newborn (HDFN) and hemolytic transfusion reactions (HTR)[1]. The complex system of the MNS system consists of over 40 antigens carried on two glycophorin molecules. M, N, S, ¿, and U antigens are the most important antigens of the MNS system with regard to transfusion medicine.

The frequencies of the common phenotypes are shown in the table.

Phenotypes and Frequencies in the MNS System[1]

Phenotype	Whites	Blacks
M+N-	30	25
M+N+	49	49
M-N+	21	26
S+s-	10	6
S+s+	42	24
S-s+	48	68
S-s-	0	2

Bio-Rad Anti-M, Anti-N, Anti-S, and Anti-¿ Blood Group Reagents are used to test for the presence or absence of the M, N, S, and ¿ antigens.
They are used principally in the resolution of antibody problems or in family studies.

PRINCIPLES OF THE TEST
The test principle is hemagglutination. The antibodies in Anti-M (MNS1), bind to the corresponding antigens on red blood cells and cause an antigen-antibody reaction visible as red blood cell agglutination.

REAGENT
[IVD]
OBSERVABLE INDICATIONS.
Do not use if markedly turbid.
Do not use damaged vials.

As the reactive components Seraclone® Anti-M contains murine monoclonal antibodies of the immunoglobulin class IgM. They are derived from hybridoma cell lines which are created by fusing mouse antibody producing B lymphocytes with mouse myeloma cells and demonstrate consistent specificity and reproducibility characteristic for monoclonal antibodies.

The antibodies are diluted in an isotonic saline solution containing bovine albumin and macromolecular potentiators.
Seraclone® Anti-M (MNS1) clone BS57 (IgM)

Preservative: 0.1% Sodium azide.

PRECAUTIONS
- For In-vitro diagnostic use.
- Store at 2 to 8°C.
- Do not use beyond the expiration date.
- Do not use if turbid.
- Use the reagent as it is supplied, do **NOT** acidify.
- Handle and dispose of reagents as potentially infectious.
- Caution: Do not pipette by mouth. The absence of murine viruses has not been determined.
- Caution: This Product Contains Natural Rubber Latex Which May Cause Allergic Reactions.
- Warning: Contains sodium azide (NaN_3), which may react with lead or copper plumbing to form explosive azides. If discarded in the sink, flush with large amounts of water to prevent the build-up of explosive metal azides.
- The bovine albumin used for the production of this reagent is sourced from donor animals of U.S. origin that have been inspected by U.S. Veterinary Service inspectors to be disease free.
- **Do Not** incubate at 2 to 8°C.
- Consult **downloads.bio-rad.com** to download the valid version of this instruction for use.

SPECIMEN COLLECTION
Fresh samples of clotted, EDTA or citrate anticoagulated whole blood collected following general blood sampling guidelines are acceptable. The specimen should be tested as soon as possible after collection. If testing is delayed, EDTA and clotted specimens should be stored at 2 to 8°C, citrated specimens (donor segments) at 1 to 6°C.
Note: Blood specimens exhibiting gross hemolysis or contamination should not be used.
Clotted samples or those collected in EDTA may be tested within ten days from collection. Donor blood stored in citrate anticoagulant may be tested until the expiration date of the donor unit.

MATERIALS
Materials provided
- Seraclone® Anti-M

Materials required but not provided
- Pipettes
- Isotonic saline or Phosphate Buffered Saline (PBS; pH 7.2 +/-0.1)
- Glass tubes 10 x 75mm or 12 x 75mm
- Interval timer
- Markers
- Agglutination viewer (optional).

TEST PROCEDURE
Tube test
1. Prepare a 3 to 5% suspension of red blood cells to be tested in saline.
2. Place one drop reagent into an appropriately labelled tube.
3. Add one drop (approx. 40 to 50 µL) of red blood cell suspension into the tube and mix.
4. Incubate at room temperature (15 to 30°C) for 30 to 60 minutes **DO NOT CENTRIFUGE**.
5. Gently dislodge red blood cell button and observe for macroscopic agglutination. Negative reactions may be examined with an agglutination viewer, however, microscopic examination is not recommended.
6. Record results.

STABILITY OF REACTION
Following incubation, all tube tests should be read immediately and results interpreted without delay. Time delays may cause a dissociation of the antigen-antibody complexes resulting to false negative or more often weak positive reactions.

QUALITY CONTROL
The reactivity of all blood typing reagents should be confirmed by testing with known positive and negative red blood cells on each day of use.

To confirm the reactivity or specificity of Bio-Rad Monoclonal Anti-M, Blood Grouping Reagent, it should be tested with antigen-positive (preferably from heterozygous individuals) and antigen-negative red blood cells, respectively.

Each reagent is satisfactory for use if it reacts only with antigen-positive red blood cells.

INTERPRETATION OF RESULTS
Agglutination of the red blood cells is a positive result and indicates the presence of the corresponding antigen. No agglutination is a negative result and indicates the absence of the corresponding antigen.

Antigen Frequency (%)

	Caucasians	Blacks
M	78%	74%
N	72%	75%

An agglutination viewer may facilitate the reading of tube tests (as recommended by the AABB Technical Manual)[1].

LIMITATIONS
- **DO NOT CENTRIFUGE.** False positive interpretation may occur when the test is centrifuged.
- Samples with cold agglutinins or rouleaux formation may show false positive results in testing with monoclonal antibodies. Results on these samples must be interpreted with caution. False positive results or reaction suspected to be due to cold agglutinins should be resolved according to in-house procedures.
- Due to the nature of monoclonal antibodies, i.e. recognition of a single epitope, discrepant results (strengths of reaction) may be found in comparison with human polyclonal test sera. This may be due to antigen variants.
- In case of ambiguous results it is recommended to wash red blood cells at least 2 times.
- Grossly icteric blood samples, blood samples with abnormally high concentrations of protein or blood samples from patients who have received plasma expanders of high molecular weight may give false positive results.
- Some conditions that may cause false positive results are:
  - Contamination of sample or reagents
  - Autoantibodies
  - Improper storage or preparation of red blood cells
  - Cross reaction with patient's medication (e.g. antibiotics)
  - Mixed field reaction due to patients pre-transfusion history

SPECIFIC PERFORMANCE CHARACTERISTICS
Testing is performed in accordance with FDA recommended methods.
The final release testing is performed according to the product specific SOPs. As part of the release process each lot of Bio-Rad blood group reagent is tested according to the package insert method against a panel of antigen positive red blood cells (heterozygous antigen expression and if possible weakened antigen

expression) to insure suitable reactivity. The products meet FDA potency requirements. The specificity testing for the presence of contaminating antibodies is performed according to the product specific SOPs.
For the product performance it is necessary to adhere to the recommended method in the instructions for use.

The performance of the Bio-Rad Anti-M was confirmed against a FDA approved reference reagent in a Multi Center Field Trial.

For Technical Support or further product information, contact Bio-Rad Laboratories, Inc., at 800-224-6723.

NOTE
Manual techniques are to be performed according to the manufacturer's instructions. Each deviation from these instructions is the sole responsibility of the user.
Used tests must be discarded as hazardous material. Manage waste according to local, state and national regulations.

GLOSSARY OF SYMBOLS

Symbol	Definition	Symbol	Definition
[LOT]	Batch Code	[IVD]	*In vitro* diagnostic medical device
!	Consult the instructions for use for important cautionary information such as warnings and precautions	I	Consult instructions for use
M	Manufacturer	e	Use by YYYY-MM-DD
s	Contains sufficient quantity for <n> tests	[REF]	Catalog number
t	Temperature limitation	[VOL]	Volume

BIBLIOGRAPHY
1. John D. Roback, MD et al. Technical Manual 17th Edition, Bethesda, MA: AABB, 2011.
2. Marion E. Reid, Christine Lomas-Francis, The Blood Group Antigen FactsBook, New York, NY: Academic Press, 2004.

Key: Underline = Addition of changes ◄ = Deletion of text