



Anti- human MGMT Mouse Monoclonal Primary Antibody

Clone: UMAB56

IVD

REF CE00017

CATALOG NUMBER

C0017MA10-MA 1.0 mL

ENGLISH

Intended use

Anti-human MGMT (Clone: UMAB56) Mouse Monoclonal Primary Antibody is intended for detection of O⁶-methylguanine DNA methyltransferase expression in frozen or formalin fixed human tissues and cells. The clinical interpretation of any positive staining or its absence should be complemented by morphological and histological studies with proper controls. Evaluations should be made within the context of the patient's clinical history and other diagnostic tests by a qualified pathologist. The antibody is intended for *in vitro* diagnostic (IVD) use.

Background

Expressed by the MGMT gene, O⁶-methylguanine DNA methyltransferase repairs DNA by demethylating O⁶-methylguanine to guanosine. The O⁶-methylguanine causes errors during DNA replication and transcription by base-pairs to thymine rather than cytidine. MGMT methylation is associated with the loss of its protein expression. MGMT expression is variable in cancer tissues.

Alternative names: O⁶-alkylguanine DNA alkyltransferase, AGT, AGAT

Reagent provided

Anti-human MGMT Mouse Monoclonal Primary Antibody (Clone: UMAB56) is provided in liquid form in 20mM Sodium phosphate, 150mM Sodium chloride, 0.2% BSA, 0.09% Sodium azide, pH 7.4. The isotype of the antibody is IgG1. The protein concentration is approximately 0.6 +/- 0.05 mg/mL.

For immunohistochemistry, the primary antibody may be used at a working dilution of 1:100 – 1:200 for formalin-fixed, paraffin-embedded human tissue. It can be dependent upon the detection system used. These are guidelines only, and optimal dilutions should be determined by the individual laboratory.

Immunogen

Full length recombinant protein of human MGMT (NP_002403) produced in E. coli.

Specificity

The specificity of the anti-human MGMT Mouse Monoclonal Primary Antibody was established on human breast cancer, normal human brain and heart tissues. The anti- MGMT presented no staining on normal human brain and heart, and positive staining on human breast cancer tissue using immunohistochemical (IHC) test methods.

Materials Required but Not Supplied

Antibody diluent, HIER solution, Antibody detection kits, Chromogen, Staining reagents, negative and positive tissue control slides are not included.

Precautions

1. For use by trained professionals only.
2. This product contains sodium azide (NaN_3), a chemical highly toxic in pure form. At product concentrations, though not classified as hazardous, NaN_3 may react with lead and copper plumbing to form highly explosive build-ups of metal azides. Upon disposal, flush with large volumes of water to prevent metal azide build-up in plumbing.
3. Wear appropriate Personal Protective Equipment to avoid contact with eyes and skin.
4. Unused reagents should be disposed of according to local, State, and Federal regulations.

Storage

Store at 2-8°C. Do not use the product past the expiration date indicated on the label. If reagents are stored under any other conditions, the end user must verify the acceptability of those conditions. There are no obvious signs to indicate instability of this product therefore, positive and negative controls should be run simultaneously with patient specimens.

Specimen Preparation

Paraffin Sections

Anti-human MGMT Mouse Monoclonal Primary Antibody can be used on paraffin-embedded tissue sections at a working dilution of 1:100 to 1:200. We recommend 20 minutes of pretreatment with Heat Induced Epitope Retrieval (HIER) using Accel HIER buffer from GBI Labs (B22C-100). The dilutions are estimates; the actual staining results may vary due to reagents and detection protocols used. Validation of antibody performance and final protocol are the responsibility of the end user.

Staining procedure

Manual Staining Procedure

1. Deparaffinize slides.
2. Submerge slides in 3% peroxidase quenching solution for ~10 minutes and rinse with distilled water 2 times, 2 minutes each.
3. Heat Induced Epitope Retrieval is required for this antibody. We recommend incubating the slides in Accel pH 8.5 HIER buffer [GBI Labs B22C-100] at 95-100 °C for 20 minutes or using the pressure cooker at high pressure for 3 minutes.
4. Wash with PBS buffer 3 times, 2 minutes each before staining. Apply serum blocking solution.[Optional]
5. Apply primary antibody and incubate for 30-60 minutes at room temperature. After incubation wash with PBS-T 3 times, 2 minutes each.
6. Apply secondary antibody and incubate according to the data sheet of the detection system. Wash with PBS-T 3 times, 2 minutes each.
7. Apply enzyme conjugate and incubate according to data sheet of detection system. Wash with PBS-T 3 times, 2 minutes each.
8. Apply chromogen and incubate 5-10 minutes and rinse with distilled water.

Staining interpretation

The cellular staining pattern for Anti-human MGMT Mouse Monoclonal Primary Antibody is selectively nuclear and cytosolic.

Performance Characteristics

Normal human brain and heart tissues were shown to be negative for this antibody.

Anti-human MGMT Mouse Monoclonal Primary Antibody (Clone: UMAB56) produced nucleus staining selectively when screened on human breast cancer.

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