



Mouse anti-MLH1

Cat. No.: MSK045-05 (0.5 ml Concentrate)

Instructions for use

Intended use

This antibody is designed for the specific localisation of MLH1 in formalin-fixed, paraffin-embedded tissue sections. Anti-MLH1 antibody is intended for in vitro diagnostic use.

Specifications

Specificity:	Anti-human MLH1
Clone:	G168-15
Isotype:	Mouse IgG1 kappa
Species reactivity:	Human +, others not tested

Summary and Description

MLH1 (mutL homologue 1) belongs to the so-called mismatch repair proteins (MMR proteins) like MLH3, MSH2, MSH3, MSH4, MSH5, MSH6, PMS1, and PMS2.

Defects in the genes coding for these proteins lead to microsatellite instability (MSI) and considerable higher mutation rates. Hereditary nonpolyposis colorectal cancers (HNPCC) often show germline mutations in the *mmr* protein associated genes. These mutations result in decreased or abnormal protein production. The majority of HNPCC is characterised by damages in the MLH1 and MSH2 encoding genes, sometimes also in damages in MSH6 and PMS2 encoding genes.

Reagent provided

Mouse monoclonal antibody in buffer with carrier protein and preservative for stabilisation in the following formats:

Concentrate: 0.5 ml (Cat. No. MSK045-05) Immunoglobulin concentration: 62.5 µg/ml

Dilution of primary antibody

Dilution of Zytomed Systems' concentrated antibody depends on the detection system used. The final working dilution must always be determined by the user.

For dilution of this antibody we recommend the special Antibody Diluent B (Cat. No. ZUC051) leading to significantly stronger signals in immunohistochemistry.

The validation of staining protocol should be done by an experienced specialist. For Zytomed Systems' recommendations see chapter 'Staining procedure'.

Storage and handling

The antibody should be stored at 2-8°C without further dilution.

Dilutions of the concentrated antibody should be done with a suitable antibody dilution buffer (e.g. ZUC051 from Zytomed Systems). The diluted antibody should be stored at 2-8°C after use. Stability of this working solution depends on various parameters and has to be confirmed by appropriate controls.

The antibody provided is suitable for use until the expiry date indicated on the label, if stored at 2-8°C. Do not use product after the expiry date. Positive and negative controls should be run simultaneously with all specimens. If unexpected staining is observed which cannot be explained by variations in laboratory procedures and a problem with the antibody is suspected, contact Zytomed Systems' technical support or your local distributor.

Precautions

Use through qualified personnel only.

Wear protective clothing to avoid contact of reagents and specimens with eye, skin and mucous membranes. If reagents or specimens come in contact with sensitive area, wash with large amounts of water.

Microbial contamination of the reagent must be avoided, since otherwise non-specific staining may occur.

Sodium azide (NaN₃), used for stabilisation, is not considered hazardous material in the concentration used. Reaction of sodium azide with lead or copper in drainage pipes can result in the formation of highly explosive metallic azides.

Sodium azide should be discarded in a large volume of running water to avoid formation of deposits. Material safety data sheets (MSDS) are available upon request.

Staining procedure

Refer to the following table for conditions specifically recommended for this antibody. Also refer to detection system data sheets for guidance on specific staining protocols or other requirements.

Parameters	Zytomed Systems recommendations
*Pre-treatment	Heat Induced Epitope Retrieval (for example in Citrate Buffer pH 6.0)
*Control tissue	Colon carcinoma
*Working dilution	1:25-1:50 (for concentrates)
*Incubation time	30 - 60 minutes

Quality control

The recommended positive control tissue for this antibody is colon carcinoma. We recommend carrying out a positive and a negative control with every staining run. Please refer to the instructions of the detection system for guidance on general quality control procedures.

Troubleshooting

If you observe unusual staining or other deviations from the expected results please read these instructions carefully, refer to the instructions of the detection system for relevant information or contact your local distributor.

Expected results

This antibody stains positive in nuclei of MLH1 expressing cells in formalin-fixed, paraffin-embedded tissue sections. When staining colon carcinomas for MLH1 expression adjacent normal colon mucosa can be used as internal positive control.

Interpretation of the staining results is solely the responsibility of the user. Any experimental result should be confirmed by a medically established diagnostic procedure.

Limitations of the Procedure

Immunohistochemistry is a complex technique involving both histological and immunological detection methods. Tissue processing and handling prior to immunostaining, for example variations in fixation and embedding or the inherent nature of the tissue can cause inconsistent results (Nadji and Morales, 1983). Endogenous peroxidase, pseudoperoxidase activity in erythrocytes or biotin may cause non-specific staining depending on the detection system used. Tissues containing Hepatitis B Surface Antigen (HBsAg) may give false positive results with HRP (horse radish peroxidase) detection systems (Omata *et al*, 1980). Inadequate counterstaining and mounting can influence the interpretation of the results.

Zytomed Systems warrants that the product will meet all requirements described from its shipping date until the expiry date is reached, if the product is stored and utilised as recommended. No additional guarantees can be given. Under no circumstances shall Zytomed System be liable for any damages arising out of the use of the reagent provided.

Performance characteristics

Zytomed Systems has conducted studies to evaluate the performance of the antibody for use with a standard detection system. The product has been found to be sensitive and specific to the antigen of interest with minimal or no cross-reactivity.

Bibliography

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Explanations of the symbols on the product label

Symbols are used in accordance with ISO 15223-1. Further symbols on the product label might be:



GSH07: Warning / Attention

RUO

For Research Use Only