



Cat. No.: BMS033 (16 ml Ready-to-use)

Instructions for use

Intended use

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

Specifications

Specificity: Human MLH1
Clone: G168-15
Isotype: Mouse IgG1 κ

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 antibodies in buffer with carrier protein and preservative for stabilisation in the formats:

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Dilution of primary antibody

None

Storage and handling

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

If necessary, dilutions of the antibody should be done in 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 cocktail 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. ProClin300 is used for stabilisation. Material safety data sheets (MSDS) are available upon request.

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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.

<u>Parameters</u> <u>Zytomed Systems recommendations</u>

*Pre-treatment: Heat Induced Epitope Retrieval (for example in Citrate Buffer pH 6.0)

*Control tissue colon carcinoma

*Working dilution None

*Incubation time 30 – 60 minutes

Quality control

The recommended positive control tissues 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 against MLH1 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. Further details about the expression pattern of MLH1 can be found in the chapter 'Summary and Description'.

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:

Rüschoff J et al. Pathologe 25:178-192, 2004 Umar A et al. J Natl Cancer Inst 96:261-268, 2004 Marra G, Schär P. Biochem J 338:1-13, 1999 Omata M et al. Am J Clin Pathol 73: 626-632, 1980 Peiro G, et al. Mod Pathol. 2001 Aug;14(8):777-83 Young J et al. Am J Pathol 159:2107-2116, 2001 Kim JC et al. Clin Cancer Res 10:6159-6168, 2004 Nadji M, Morales AR. Ann N Y Acad Sci 420:134-138, 1983 Vilkin, A, et al. Hum Pathol 2015;46:1705–11 Djordjevic B, Broaddus RR Surg Pathol Clin 2016;9:289-99



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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

For Research Use Only

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