

Mouse monoclonal antibody against MLH1 (Clone ISM063)

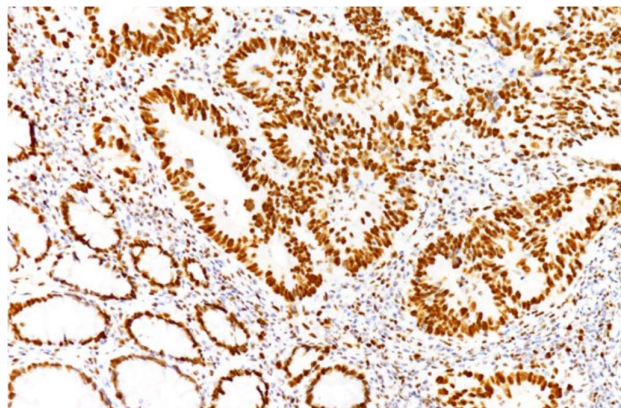


Figure 1 Human colorectal adenocarcinoma stained with anti-MLH1 antibody.

Product identification

ISC088-R3	3 ml ready-to-use (RTU)
ISC088-R7	7 ml ready-to-use (RTU)
ISC088-C02	0.2 ml concentrated
ISC088-C05	0.5 ml concentrated

Summary and explanation

The DNA-mismatch repair (MMR) pathway corrects base substitution and insertion-deletion errors. MMR dysfunction leads to the cellular acquisition of both point mutations and the instability of long, repetitive DNA sequences (ie, microsatellite instability [MSI]). Inherited MMR dysfunction is associated with increased rates of carcinogenesis. The MMR machinery has an additional function of sensing DNA lesions, triggering cell-cycle checkpoints and apoptosis. MutL protein homolog 1 (MLH1) is a pivotal member of the MMR pathway. The human MLH1 is a 756 amino-acids protein that can be roughly divided into two domains: the N-terminal domain (NTD) and the C-terminal domain (CTD).

Principle of the procedure

The stated primary antibody is suitable for immunohistochemical staining of FFPE tissue sections based on specific antigen-antibody reaction. Using a detection system linked to HRP or alkaline phosphatase the antigen visualization is performed via specific binding of the primary antibody. Secondary antibody is binding to the primary antibody, and the enzyme complex labels this complex. The enzymatic activation of the chromogen results in a visible reaction product at the antigen site. Each step is incubated for a precise time and temperature and requires interposed washing steps. The specimen may then be counterstained. Results are interpreted using a light microscope.

Materials provided

Primary antibody	MLH1
Host	Mouse
Subclass	IgG
Clone	ISM063
Species Cross-reactivity	Human. Others-not known
Applications	Immunohistochemistry
Epitope Retrieval	Heat-induced epitope retrieval
Ready-to-use antibody	Prediluted antibody in antibody diluent buffer
Recommended working dilution range	1:50 to 1:100

Product label shows the specific lot number.

Prediluted antibody is ready-to-use and optimized for staining. No further dilution, reconstitution, mixing, or titration is needed.

Antibody concentrate is optimized for dilution within dilution range using InSituChem® Antibody Diluent for IHC (Cat. No. D005-50). Indicated dilution range should be considered as recommendation and depends on different facts (tissue, fixation, incubation conditions, etc.). Optimum dilution to be determined in user's own system.

Materials required but not provided

The following materials may be required for staining but are not provided with the primary antibody.

- Positive and negative controls
- Microscope slides (positively charged) and cover slips
- Water bath
- Humidified chamber
- Staining jars
- Stopwatch
- Xylene or xylene substitute
- Ethanol
- Deionized or distilled water
- Antigen retrieval reagent, e.g. Antigen Enhancer (HIER buffer), Cat. No. D004-500
- Detection system, e.g. IHC Complete Detection system (Goat anti mouse/rabbit HRP, DAB staining), Cat. No. D001-15
- Wash buffer: e.g. IHC Wash Buffer, Cat. No. D003-500
- Tap water/bluing reagent (e.g. ammonia water)
- Light microscope

Storage and handling

Store at 2 - 8 °C.

When stored correctly antibody is stable to the expiration date indicated on the vial. Do not use after expiration date.

To ensure proper reagent delivery and stability of the antibody, replace the dispenser cap after every use and immediately place the bottle cool in an upright position.

Staining procedure

1. Cut 3-4 μm section of formalin-fixed paraffin-embedded tissue and place on positively charged slides.
2. Dry at 65°C for 2 hours.
3. Deparaffinize, rehydrate, and epitope retrieve. Upon completion, rinse with 3 changes of distilled or deionized water.
4. If using HRP detection system, place slides in peroxide block for 10 minutes; rinse. If using AP detection system, omit this step.
5. Apply the antibody and incubate for 60 minutes; rinse.
6. Apply the InSituVison™ Polymer Rabbit/Mouse Detection System for 30 minutes; rinse.
7. Apply ample amount of DAB or AEC chromogen and incubate; rinse.
8. Dehydrate and coverslip.

Interpretation of results

The immunostaining procedure causes a colored reaction product to precipitate at the antigen sites localized by the primary antibody.

Cellular localization: Cell nucleus.

A qualified pathologist experienced in immunohistochemistry procedures must evaluate positive and negative tissue controls before interpreting patient specimens.

Positive staining intensity should be assessed within the context of any background staining of the negative reagent control.

Note: A negative result means that the antigen in question was not detected, but not that the antigen is not present in the cells/tissues tested. An antibody panel may be used to support the results in some circumstances. Additionally, the morphology of each tissue sample should be examined utilizing a hematoxylin and eosin stained section. A qualified pathologist must interpret the patient's morphologic findings and pertinent clinical data.

Warnings and precautions

1. Application only by qualified and trained personnel.
2. There are no estimated health risks, if the product is used as directed. MSDS is available on request.
3. Product contains sodium azide as preservative. Pure sodium azide is toxic. The concentration of sodium azide in this reagent is < 0.1 % and is not classified hazardous. See MSDS.
4. As with any product derived from biological sources, proper handling procedures should be used.
5. Do not use reagents after expiration date.
6. Take reasonable precautions when handling reagents. Use protective clothing and gloves.

7. All hazardous materials should be disposed according to guidelines for hazardous waste disposal. Materials of human or animal origin should be handled as biohazardous materials and disposed of with proper precautions.
8. Avoid microbial contamination of reagents as it may cause incorrect results.

Literature

[1] Lawrence, Y.R., Moughan, J., Magliocco, A.M., Klimowicz, A.C., Regine, W.F., Mowat, R.B., DiPetrillo, T.A., Small, W., Jr, Simko, J.P., Golan, T., Winter, K.A., Guha, C., Crane, C.H. and Dicker, A.P. (2018), Expression of the DNA repair gene MLH1 correlates with survival in patients who have resected pancreatic cancer and have received adjuvant chemoradiation: NRG Oncology RTOG Study 9704. Cancer, 124: 491-498.




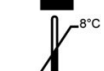



[2] Pingping Jia, Weihang Chai,
The MLH1 ATPase domain is needed for suppressing aberrant formation of interstitial telomeric sequences, DNA Repair, 65, 2018, 20-25.

Date of publication or revision

2024-06-10

Change(s) made: -

Explanation of symbols

	Catalog number
	Batch code
	Use by
	Temperature limitation
	Do not use if package damaged
	Consult instructions for use
	Caution