Technical Data Sheet

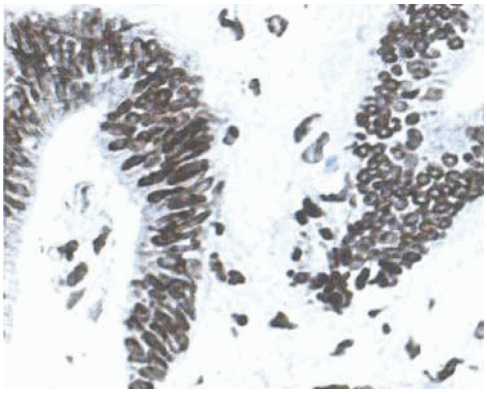
Purified Mouse Anti-Human MLH-1

Product Information

Material Number:	**550838**
Size:	1.0 ml
Concentration:	250 µg/ml
Clone:	G168-15
Immunogen:	Human recombinant MLH
Isotype:	Mouse IgG1
Reactivity:	QC Testing: Human
Storage Buffer:	Aqueous buffered solution containing BSA, goat serum, and ≤0.09% sodium azide.

Description

The repair of mismatched DNA is essential to maintaining the integrity of genetic information over time. In bacteria the DNA repair process is accomplished by the MutL, MutH, and MutS proteins. The MutS protein initially recognizes and binds to mismatched DNA. Following this, MutH, an endonuclease, and MutL form a complex with MutS and carry out an excision repair mechanism. When bacteria are deficient in one of these enzymes a mutator phenotype arises characterized by genetic instability. The important role played by DNA repair enzymes is emphasized by the fact that they are highly conserved from bacteria to yeast to mammals. In yeast the proteins are called MutS homolog 2 (MSH2), MutL homolog (MLH1), and PMS1 which is also a homolog of MutL. MSH2 is involved in the initial recognition of mismatched nucleotides during the replication mismatch repair process. It is thought that after MSH2 binds to a mismatched DNA duplex it is joined by a heterodimer of MLH1 and PMS1 which together help facilitate the later steps in mismatch repair. Biochemical studies of the human homologs of DNA mismatch repair enzymes MLH1, PMS2, and MSH2 indicate that human MSH2 protein can bind mispaired DNA, and that human MLH1 and PMS2 can exist as a heterodimer. These and other studies support the conservation of eukaryotic DNA mismatch repair mechanisms. The G168-15 antibody recognizes human MLH1 (80-85 kDa). Full-length human recombinant MLH was expressed as a fusion protein, affinity purified, and used as immunogen.



Immunohistochemistry of MLH-1 in human colon carcinoma. *Frozen sections of human colon carcinoma were reacted with the MLH-1 antibody. Cells expressing MLH-1 can be identifed by the intense brown labeling of their cell nuclei. Magnification 20X.*

Preparation and Storage

Store undiluted at 4°C.

The monoclonal antibody was purified from tissue culture supernatant or ascites by affinity chromatography.

Application Notes

Application

Western blot	Routinely Tested
Immunohistochemistry-formalin (antigen retrieval required)	Tested During Development
Immunohistochemistry-frozen	Tested During Development

Recommended Assay Procedure:

Immunohistochemistry: The MLH-1 antibody is recommended to test for immunohistochemical staining of frozen sections and formalin-fixed paraffin embedded sections. Tissues tested were human tonsil, colon carcinoma and intestine. For paraffin sections microwave oven pretreatment with BD Retrievagen A (pH 6.5) (Cat. No. 550524) is required. The staining seen is usually nuclear although some staining may be observed in the cytoplasm. The isotype control recommended for use with this antibody is purified mouse IgG1 (Cat. No. 550878). For optimal indirect immunohistochemical staining, MLH-1 antibody should be titrated (1:10 to 1:50 dilution) and visualized via a three-step staining procedure in combination with polyclonal, biotin conjugated anti-mouse Igs (multiple adsorbed) (Cat. No. 550337) as the secondary antibody and

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Streptavidin-HRP (Cat.No. 550946) together with the DAB detection system (Cat. No. 550880). Alternatively, the Anti-mouse Ig HRP Detection Kit (Cat. No. 551011) contains all the detection reagents required.

For Western blot application of the G168-15 clone reactive against MLH-1, please refer to Cat. No. 551091.

Suggested Companion Products

Catalog Number	Name	Size	Clone
550524	Retrievagen A (pH 6.0)	1000 ml	(none)
550878	Purified Mouse IgG1 κ Isotype Control	1.0 ml	MOPC-31C
550337	Biotin Goat Anti-Mouse Ig (Multiple Adsorption)	1.0 ml	Polyclonal
550946	Streptavidin HRP	50 ml	(none)
550880	DAB Substrate Kit	500 tests	(none)
551011	Anti-Mouse Ig HRP Detection Kit	200 tests	(none)
559148	Antibody Diluent for IHC	125 ml	(none)

Product Notices

1. Since applications vary, each investigator should titrate the reagent to obtain optimal results.
2. Caution: Sodium azide yields highly toxic hydrazoic acid under acidic conditions. Dilute azide compounds in running water before discarding to avoid accumulation of potentially explosive deposits in plumbing.
3. Source of all serum proteins is from USDA inspected abattoirs located in the United States.
4. This antibody has been developed for the immunohistochemistry application. However, a routine immunohistochemistry test is not performed on every lot. Researchers are encouraged to titrate the reagent for optimal performance.
5. An isotype control should be used at the same concentration as the antibody of interest.
6. Please refer to www.bdbiosciences.com/pharmingen/protocols for technical protocols.

References

Baker SM, Plug AW, Prolla TA. Involvement of mouse Mlh1 in DNA mismatch repair and meiotic crossing over. *Nat Genet.* 1996; 13(3):336-342. (Immunogen: Western blot)

Cleaver JE. It was a very good year for DNA repair. *Cell.* 1994; 76(1):1-4. (Biology)

Prolla TA, Christie DM, Liskay RM. Dual requirement in yeast DNA mismatch repair for MLH1 and PMS1, two homologs of the bacterial mutL gene. *Mol Cell Biol.* 1994; 14(1):407-415. (Biology)

Prolla TA, Pang Q, Alani E, Kolodner RD, Liskay RM. MLH1, PMS1, and MSH2 interactions during the initiation of DNA mismatch repair in yeast. *Science.* 1994; 265(5175):1091-1093. (Biology)

Su SS, Modrich P. Escherichia coli mutS-encoded protein binds to mismatched DNA base pairs. *Proc Natl Acad Sci U S A.* 1986; 83(14):5057-5061. (Biology)

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