

AM26533AF-N

Monoclonal Antibody to Mesothelin - Azide Free

Alternate names:	CAK1 antigen, MPF, MSLN, Pre-pro-megakaryocyte-potentiating factor
Quantity:	0.1 mg
Concentration:	1.0 mg/ml
Background:	Mesothelin (MSLN) is a GPI-linked cell surface glycoprotein expressed in the mesothelial lining of the body cavities and in mullerian duct epithelium related cancer cells (ovarian, pancreatic cancer, mesothelioma). Both human and mouse mesothelin bind to ovarian cancer antigen CA125/MUC16 with high affinity, mediating cell attachment in vitro , suggesting that mesothelin may facilitate ovarian cancer metastasis.
Uniprot ID:	Q61468
NCBI:	NP_061345.1
GeneID:	56047
Host / Isotype:	Rat / IgG2a
Recommended Isotype Controls:	SM26A
Clone:	B35
Immunogen:	Murine hemangioblast-like cell line LO
Format:	State: Liquid purified Ig fraction Purification: Protein G Agarose Chromatography Buffer System: PBS containing 50% Glycerol, pH 7.2. No preservative is contained.
Applications:	Flow Cytometry: 10 µg/ml (final concentration). For more details see Protocol below. Other applications not tested. Optimal dilutions are dependent on conditions and should be determined by the user.
Specificity:	This antibody reacts with Mesothelin.
Species Reactivity:	Tested: Human (Jurkat), Mouse.
Add. Information:	This product was originally produced by MBL International.
Storage:	Upon receipt, store (in aliquots) at -20 °C. Avoid repeated freezing and thawing. Shelf life: One year from despatch.
General Readings:	1. Gubbels, J. A., et al., Mol. Cancer 5 , 50-64 (2006). 2. Ho, M., et al., Clin. Cancer Res. 11 , 3814-3820(2005). 3. Rump, A., et al., J. Biol. Chem. 279 , 9190-9198(2004). 4. Nakayama, K., et al., J. Biol. Chem . 274 , 24766-24772 (1999). 5. Kojima, T., et al., J. Biol. Chem. 270 , 21984-21990(1995).

Protocols:

Flow cytometric analysis for floating cells

We usually use Fisher tubes or equivalents as reaction tubes for all steps described below.

- 1) Wash the cells 3 times with washing buffer [PBS containing 2% fetal calf serum (FCS) and 0.1% NaN₃].
- 2) Resuspend the cells with washing buffer (5x10⁶ cells/mL).
- 3) Add 50 µL of the cell suspension into each tube, and centrifuge at 500 x g for 1 minute at room temperature (20~25°C). Remove supernatant by careful aspiration.
- 4) Add 10 µL of normal goat serum containing 1 mg/mL normal human IgG and 0.1% NaN₃ to the cell pellet after tapping. Mix well and incubate for 5 minutes at room temperature.
- 5) Add 30 µL of the primary antibody at the concentration as suggested in the APPLICATIONS diluted in the washing buffer. Mix well and incubate for 30 minutes at room temperature.
- 6) Add 1 mL of the washing buffer followed by centrifugation at 500 x g for 1 minute at room temperature. Remove supernatant by careful aspiration.
- 7) Add 30 µL of 1:100 FITC conjugated anti-rat IgG diluted with the washing buffer. Mix well and incubate for 15 minutes at room temperature.
- 8) Add 1 mL of the washing buffer followed by centrifugation at 500 x g for 1 minute at room temperature. Remove supernatant by careful aspiration.
- 9) Resuspend the cells with 500 µL of the washing buffer and analyze by a flow cytometer.

(Positive control for Flow cytometry; LO)

Pictures:

Flow cytometric analysis of mouse Mesothelin expression on LO (left) and Jurkat (right). Open histogram indicates the reaction of isotypic control to the cells. Shaded histograms indicate the reaction of AM26533AF-N to the cells.

