

MONOCLONAL ANTIBODY

Anti-Osteopontin (SPP1) (Human) mAb

Code No.	Clone	Subclass	Quantity	Concentration
W052-3	4A7D	Mouse IgG2a κ	100 μ L	1 mg/mL

BACKGROUND: Osteopontin, also known as SPP1, is a secreted phosphoglycoprotein that belongs to the SIBLING family. Osteopontin binds to hydroxyapatite and is involved in anchoring of osteoclasts to the mineral matrix of bones. It also acts as a cytokine that mediates type I immune responses through enhancing the production of interferon-gamma and interleukin-12 while reducing production of interleukin-10. Osteopontin has been identified as a biomarker for ovarian cancer, prostate cancer, and non-small cell lung cancer.

SOURCE: This antibody was purified from hybridoma culture supernatant by Protein A affinity column chromatography.

IMMUNOGEN: Human Osteopontin (SPP1) expressed Ba/F3 transfectants generated from SST-REX (signal sequence trap by retrovirus-mediated expression screening).

FORMULATION: 100 μ g IgG in 100 μ L volume of PBS containing 50% glycerol, pH 7.2. No preservative is contained.

STORAGE: This antibody solution is stable for one year from the date of purchase when stored at -20°C .

REACTIVITY: This antibody reacts with human Osteopontin (SPP1) on Flow cytometry.

APPLICATIONS:

- Flow cytometry: 1-10 μ g/mL
- Western blotting: Not tested
- Immunoprecipitation: Not tested
- Immunohistochemistry: Not tested
- Immunocytochemistry: Not tested

Detailed procedure is provided in the following **PROTOCOL**.

INTENDED USE:

For Research Use Only. Not for use in diagnostic procedures.

Entrez Gene ID:

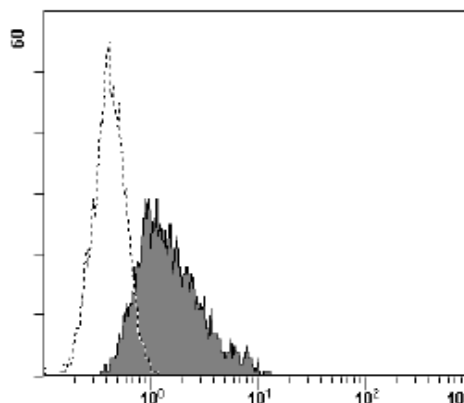
6696 (Human)

REFERENCES:

- 1) Kim, J. H., *et al.*, *JAMA* **287**, 1671-1679 (2002)
- 2) Ashkar, S., *et al.*, *Science* **287**, 860-864 (2000)
- 3) Kojima, T and Kitamura, T., *Nat. Biotechnol.* **17**, 487-490 (1999)
- 4) Reinholt, F. P., *et al.*, *PNAS* **87**, 4473-4475 (1990)

SPECIES CROSS REACTIVITY:

Species	Human	Mouse	Rat	Hamster
Cells	Transfectant	Not tested	Not tested	Not tested
Reactivity on FCM	+			



Flow cytometric analysis of human Osteopontin (SPP1) expression on Ba/F3 transfectant. Open histograms indicate the reaction of isotypic control to the cells. Shaded histograms indicate the reaction of W052-3 to the cells.

PROTOCOL:

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 0.05% NaN_3].
- 2) Resuspend the cells with washing buffer (2.5×10^6 cells/mL).
- 3) Add 200 μ L of cell suspension into each tube. And centrifuge at 500 x g for 1 minute at room temperature ($20\sim 25^{\circ}\text{C}$). Remove supernatant by careful decantation.
- 4) Add 20 μ L of Clear Back (human Fc receptor blocking reagent, MBL; code no. MTG-001) to the cell pellet after tapping. Mix well and incubate for 5 minutes at room

temperature.

- 5) Add 50 μ L of the primary antibody at the concentration as suggest 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 decantation.
- 7) Add 50 μ L of 1:200 anti-mouse IgG-PE (Beckman Coulter; code no. IM0855) diluted with the washing buffer. Mix well and incubate for 30 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 decantation.
- 9) Resuspend the cells with 500 μ L of the washing buffer and analyze by a flow cytometer.

(Positive control for Flow cytometry; transfectant)

RELATED PRODUCTS:

CY-P1035	Anti-Osteopontin pAb
K0032-3	Anti-Osteoprotegerin (OPG) mAb
D140-3	Anti-CD44 (Human) mAb
D140-4	Anti-CD44 (Human) mAb-FITC
D140-5	Anti-CD44 (Human) mAb-PE
D050-3	Anti-CD29 (Integrin β 1) (Human) mAb
D050-5	Anti-CD29 (Integrin β 1) (Human) mAb-PE
W005-3	Anti-BTN2A1 (Human) mAb
W008-3	Anti-Carboxypeptidase D (Human) mAb
W010-3	Anti-CCDC107 (Human) mAb
W011-3	Anti-Dystroglycan (Human) mAb
W017-3	Anti-EphA2 (Human) mAb
W029-3	Anti-IGFBP1 (Human) mAb
W031-3	Anti-IGFBP6 (Human) mAb
W039-3	Anti-MANSC1 (Human) mAb
W041-3	Anti-Neuroplastin (Human) mAb
W046-3	Anti-CD201 (EPCR) (Human) mAb
W049-3	Anti-QSOX1 (Human) mAb
W050-3	Anti-RECK (Human) mAb
W052-3	Anti-Osteopontin (SPP1) (Human) mAb
W072-3	Anti-CD358 (DR6) (Human) mAb
W074-3	Anti-CRELD1 (Human) mAb
W077-3	Anti-GRK5 (Human) mAb
W080-3	Anti-ADAMTS1 (Human) mAb
W086-3	Anti-LYPD3 (C4.4A) (Human) mAb
W089-3	Anti-C11orf24 (Human) mAb
W109-3	Anti-TMED2 (Human) mAb
W111-3	Anti-DLL4 (Human) mAb
W117-3	Anti-TINAGL1 (Human) mAb
W124-3	Anti-GPR56 (Human) mAb
W125-3	Anti-GPR56 (Human) mAb
W128-3	Anti-CD318 (CDCP1) (Human) mAb
W147-3	Anti-TYRO3 (Human) mAb
W158-3	Anti-HEXA (Human) mAb
W164-3	Anti-RHBDD3 (Human) mAb
W172-3	Anti-CD172a (SIRP α) (Human) mAb
W181-3	Anti-Apolipoprotein D (Human) mAb
W194-3	Anti-FAM171A1 (Human) mAb
W253-3	Anti-Glypican 1 (Human) mAb
W321-3	Anti-FGFRL1 (Human) mAb
W357-3	Anti-CD105 (Endoglin) (Human) mAb
W358-3	Anti-CD300A (Human) mAb
W359-3	Anti-CD300C (Human) mAb
M076-3	Mouse IgG2a (isotype control)
MTG-001	Clear Back (Human Fc receptor blocking reagent)