Anti-Human MICB Antibody [PSH10-74] - BSA and Azide free (Detector)

HA723239

Product Type:	Recombinant Rabbit monoclonal IgG, primary antibodies
Species reactivity:	Human
Applications:	ELISA(Det)
Clone number:	PSH10-74

Description: Widely expressed membrane-bound protein which acts as a ligand to stimulate an activating receptor KLRK1/NKG2D, expressed on the surface of essentially all human natural killer (NK), gammadelta T and CD8+ alphabeta T-cells. Up-regulated in stressed conditions, such as viral and bacterial infections or DNA damage response, serves as signal of cellular stress, and engagement of KLRK1/NKG2D by MICA triggers NK-cells resulting in a range of immune effector functions, such as cytotoxicity and cytokine production. Widely expressed with the exception of the central nervous system where it is absent. Expressed in many, but not all, epithelial tumors of lung, breast, kidney, ovary, prostate and colon. In hepatocellular carcinomas, expressed in tumor cells but not in surrounding non-cancerous tissue. An inflammatory disease with autoimmune features and a complex genetic component. It primarily affects the joints and is characterized by inflammatory changes in the synovial membranes and articular structures, widespread fibrinoid degeneration of the collagen fibers in mesenchymal tissues, and by atrophy and rarefaction of bony structures.

Immunogen: Recombinant protein within Human MICB aa 23-309 (HA210907).

Positive control: Recombinant Human MICB protein (HA210907).

Subcellular location: Cell membrane.

Database links: SwissProt: Q29980 Human

Recommended Dilutions:

ELISA(Det) Use at an assay dependent concentration. Can be paired for Sandwich ELISA with Rabbit monoclonal [PSH10-73] to Human MICB antibody (Capture) (HA723238) and Recombinant Human MICB protein (HA210907) as the standard. The reference range value is 39.1-5,000 pg/ml.

Storage Buffer: PBS (pH7.4).

Storage Instruction: Store at +4℃ after thawing. Aliquot store at -20℃. Avoid repeated freeze / thaw cycles.

Purity: Protein A affinity purified.

Images

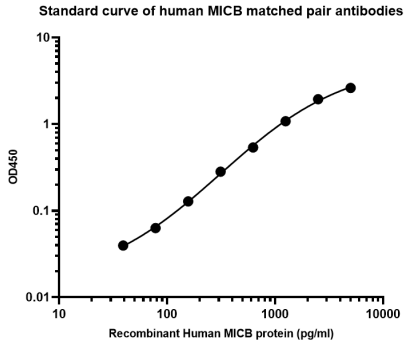

Fig1: Sandwich ELISA analysis of Human MICB matched pair antibodies

Elisa assay was performed by coating wells of a 96-well plate with 50 µl per well of capture antibody (HA723238) diluted in carbonate/bicarbonate buffer, at a concentration of 5 µg/mL overnight at 4℃. Wells of the plate were washed, blocked with 150 µl 0.05% tween-20 1% BSA blocking buffer, and incubated with serial diluted Recombinant Human MICB protein (HA210907) starting from 5,000 pg/ml to 0 pg/ml and detect antibody (HA723239, Biotin, 0.2 µg/ml) for 1 hour at 30℃ with shaking. Then the plate was washed and incubated with 50 µl per well of SA-HRP for 0.5 hour at 30℃ with shaking. Detection was performed using an Ultra TMB Substrate for 10 minutes at room temperature in the dark. The reaction was stopped with sulfuric acid and absorbances were read on a spectrophotometer at 450 nm.

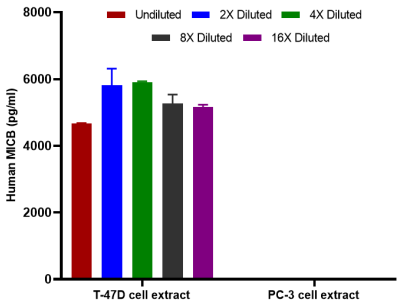

Fig2: Interpolated concentrations of native MICB in T-47D and PC-3 extract samples based on a 1,000 µg/ml extract load.

The concentrations of MICB were measured in duplicates, interpolated from the MICB standard curve and corrected for sample dilution. Undiluted samples are T-47D extract 100% and PC-3 extract 100%. The interpolated dilution factor corrected values are plotted (mean +/- SD, n=2). The mean MICB concentration was determined to be 5,368 pg/ml in T-47D extract and undetectable in PC-3 extract.

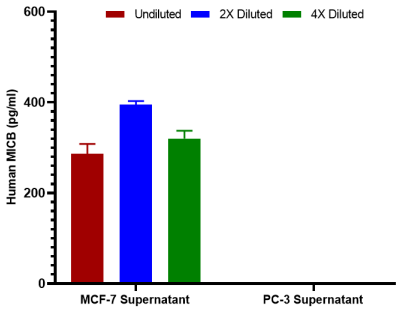

Fig3: Interpolated concentrations of native MICB in MCF-7 and PC-3 cell culture supernatant.

The concentrations of MICB were measured in duplicates, interpolated from the MICB standard curve and corrected for sample dilution. Undiluted samples are MCF-7 cell culture supernatant 100% and PC-3 cell culture supernatant 100%. The interpolated dilution factor corrected values are plotted (mean +/- SD, n=2). The mean MICB concentration was determined to be 334.07 pg/ml in MCF-7 cell culture supernatant and undetectable in PC-3 cell culture supernatant.

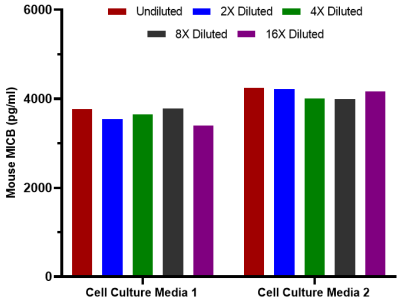

Fig4: Interpolated concentrations of spiked MICB in human cell culture media samples.

The concentrations of MICB were interpolated from the MICB standard curves and corrected for sample dilution. Undiluted samples are as follows: cell culture media 50%.

Background References

1. Steinle A., Li P., Morris D.L., Groh V., Lanier L.L., Strong R.K., Spies T. Interactions of human NKG2D with its ligands MICA, MICB, and homologs of the mouse RAE-1 protein family. Immunogenetics 53:279-287 (2001)
2. Sutherland C.L., Chalupny N.J., Schooley K., VandenBos T., Kubin M., Cosman D. UL16-binding proteins, novel MHC class I-related proteins, bind to NKG2D and activate multiple signaling pathways in primary NK cells. J. Immunol. 168:671-679 (2002)