Anti-MSR1 / CD204 Antibody [PSH29-03]

HA724831

Product Type:	Recombinant Rabbit monoclonal IgG, primary antibodies
Species reactivity:	Mouse
Applications:	WB, IF-Cell, IHC-P, FC, IP
Molecular Wt:	Predicted band size: 50 kDa
Clone number:	PSH29-03

Description: Macrophage scavenger receptor 1, also known as MSR1. MSR1 has also been designated CD204 (cluster of differentiation 204). This gene encodes the class A macrophage scavenger receptors, which include three different types (1, 2, 3) generated by alternative splicing of this gene. These receptors or isoforms are trimeric integral membrane glycoproteins and have been implicated in many macrophage-associated physiological and pathological processes including atherosclerosis, Alzheimer's disease, and host defense. They were thought to be expressed macrophage-specific, but recently shown to be present on different dendritic cells classes, too. The isoforms type 1 and type 2 are functional receptors and are able to mediate the endocytosis of modified low density lipoproteins (LDLs). The isoform type 3 does not internalize modified LDL (acetyl-LDL) despite having the domain shown to mediate this function in the types 1 and 2 isoforms. It has an altered intracellular processing and is trapped within the endoplasmic reticulum, making it unable to perform endocytosis. The isoform type 3 can inhibit the function of isoforms type 1 and type 2 when co-expressed, indicating a dominant negative effect and suggesting a mechanism for regulation of scavenger receptor activity in macrophages.

Immunogen:	Recombinant protein within mouse MRS1 aa 79-458.
Positive control:	RAW264.7 (Mouse monocytic macrophage leukemia cell) cell lysate, RAW264.7 treated with 50nM PMA and 5μg/mL Lipopolysaccharide for 24 hours cell lysate, J774A.1 (Mouse monocyte macrophage) cell lysate.
Subcellular location:	Membrane.
Database links:	SwissProt: P30204 Mouse
Recommended Dilutions:	
WB	1:5,000
IF-Cell	1:500-1:5,000
IHC-P	1:200-1:1,000
FC	1:1,000
IP	1-2μg/sample
Storage Buffer:	1*PBS (pH7.4), 0.1% BSA, 40% Glycerol, 0.2% Proclean 950.
Storage Instruction:	Shipped at 4℃. Store at +4℃ short term (1-2 weeks). Store at -20℃ long term.
Purity:	Protein A affinity purified.

Images

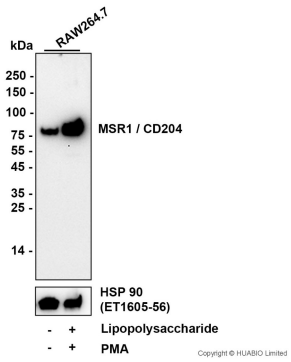


Fig1: Western blot analysis of MSR1 / CD204 on different lysates with Rabbit anti-MSR1 / CD204 antibody (HA724831) at 1/5,000 dilution.

Lane 1: RAW264.7 (Mouse monocytic macrophage leukemia cell) cell lysate
Lane 2: RAW264.7 treated with 50nM PMA and 5μg/mL Lipopolysaccharide for 24 hours cell lysate

Lysates/proteins at 20 μg/Lane.
Exposure time: 3 minutes; ECL: K1801

Blocking: 5% NFDM/TBST, 1 hour at room temperature
Primary antibody: HA724831, 1/5,000 in primary antibody dilution buffer (K1803), overnight at 4 °C
Secondary antibody: Goat anti-Rabbit IgG-HRP (HA1001), 1/50,000 in 5% NFDM/TBST, 1 hour at room temperature

Predicted band size: 50 kDa
Observed band size: 70-90 kDa

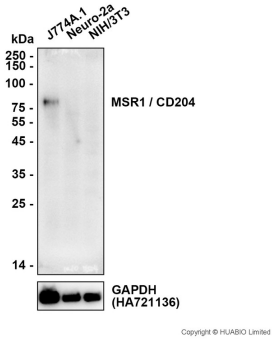


Fig2: Western blot analysis of MSR1 / CD204 on different lysates with Rabbit anti-MSR1 / CD204 antibody (HA724831) at 1/5,000 dilution.

Lane 1: J774A.1 (Mouse monocyte macrophage) cell lysate
Lane 2: Neuro-2a (Mouse brain neuroblastoma cell) cell lysate
Lane 3: NIH/3T3 (Mouse fibroblast) cell lysate

Lysates/proteins at 30 μg/Lane.
Exposure time: 3 minutes; ECL: K1802

Negative control: Neuro-2a, NIH/3T3 (PMID: 35937296).

Blocking: 5% NFDM/TBST, 1 hour at room temperature
Primary antibody: HA724831, 1/5,000 in primary antibody dilution buffer (K1803), overnight at 4 °C
Secondary antibody: Goat anti-Rabbit IgG-HRP (HA1001), 1/50,000 in 5% NFDM/TBST, 1 hour at room temperature

Predicted band size: 50 kDa
Observed band size: 70-90 kDa

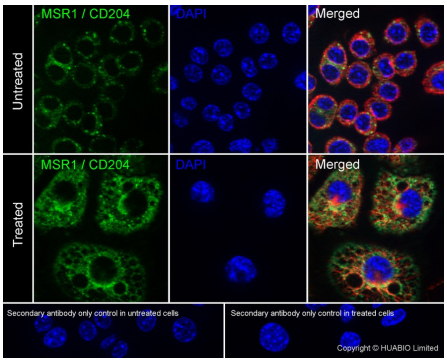


Fig3: Application: Immunocytochemistry (IF-cell)

Species: Mouse
Sample: RAW264.7 (Mouse monocytic macrophage leukemia cell) treated with or without 50 nM PMA and 5 μg/mL LPS for 24 hours

Fixation: 4% Paraformaldehyde, 15 minutes at room temperature.
Permeabilization: 0.1% Triton X-100, 15 minutes at room temperature.
Blocking: 1% BSA + 10% normal goat serum, 1 hour at room temperature.
Antibody dilution buffer: 1% BSA in PBST.

Primary antibody: HA724831, 1/5,000, overnight at 4°C.
Secondary antibody: Goat Anti-Rabbit IgG (iFluor™ 488, HA1121), 45 minutes at room temperature.
Counterstain: Beta tubulin (HA601187, Red), 1/100, overnight at 4℃. The nuclear counterstain was DAPI (Blue).

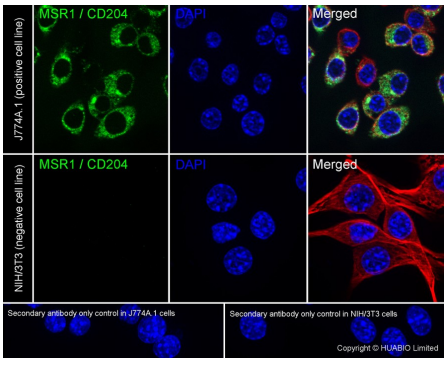


Fig4: Application: Immunocytochemistry (IF-cell)

Species: Mouse
Sample: J774A.1 (Mouse monocyte macrophage) / NIH/3T3 (Mouse fibroblast)

Fixation: 4% Paraformaldehyde, 15 minutes at room temperature.
Permeabilization: 0.1% Triton X-100, 15 minutes at room temperature.
Blocking: 1% BSA + 10% normal goat serum, 1 hour at room temperature.
Antibody dilution buffer: 1% BSA in PBST.

Primary antibody: HA724831, 1/500, overnight at 4℃.
Secondary antibody: Goat Anti-Rabbit IgG (iFluor™ 488, HA1121), 45 minutes at room temperature.
Counterstain: Beta tubulin (HA601187, Red), 1/100, overnight at 4℃. The nuclear counterstain was DAPI (Blue).

Negative control: NIH/3T3 (PMID: 35937296).

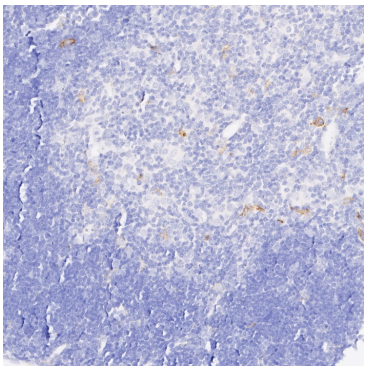


Fig5: Application: Immunohistochemistry (IHC-P)

Species: Mouse
Tissue: Thymus
Sample: Paraffin-embedded section

Antigen retrieval: Heat-mediated, Tris-EDTA buffer (pH 9.0), 20 minutes at 95°C.

Wash buffer: 1× TBST
Endogenous peroxidase blocking: 3% H_2O_2, 10 minutes at room temperature.
Blocking: 1% BSA + 10% normal goat serum, 10 minutes at room temperature.
Primary antibody: HA724831, 1/200, 1 hour at room temperature.
Secondary antibody: HA1119, 20 minutes at room temperature.

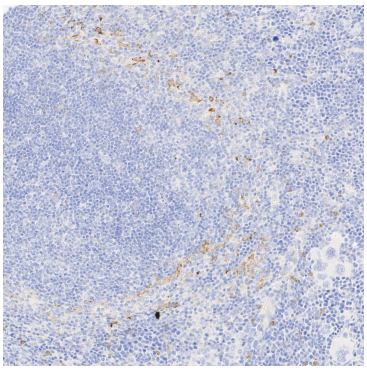


Fig6: Application: Immunohistochemistry (IHC-P)

Species: Mouse
Tissue: Spleen
Sample: Paraffin-embedded section

Antigen retrieval: Heat-mediated, Tris-EDTA buffer (pH 9.0), 20 minutes at 95°C.

Wash buffer: 1× TBST
Endogenous peroxidase blocking: 3% H_2O_2, 10 minutes at room temperature.
Blocking: 1% BSA + 10% normal goat serum, 10 minutes at room temperature.
Primary antibody: HA724831, 1/200, 1 hour at room temperature.
Secondary antibody: HA1119, 20 minutes at room temperature.

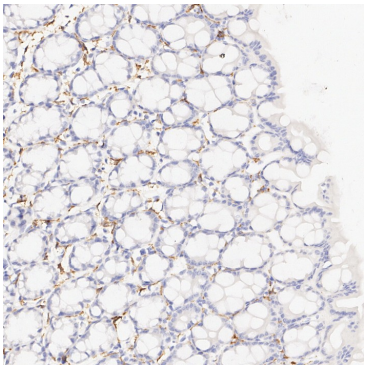


Fig7: Application: Immunohistochemistry (IHC-P)

Species: Mouse
Tissue: Colon
Sample: Paraffin-embedded section

Antigen retrieval: Heat-mediated, Tris-EDTA buffer (pH 9.0), 20 minutes at 95°C.

Wash buffer: 1× TBST
Endogenous peroxidase blocking: 3% H_2O_2, 10 minutes at room temperature.
Blocking: 1% BSA + 10% normal goat serum, 10 minutes at room temperature.
Primary antibody: HA724831, 1/200, 1 hour at room temperature.
Secondary antibody: HA1119, 20 minutes at room temperature.

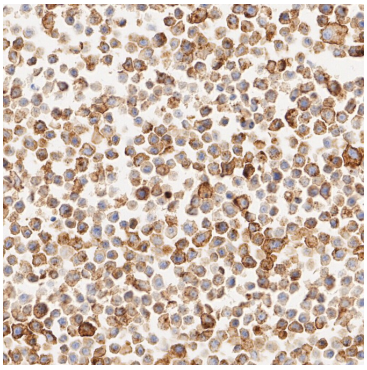


Fig8: Application: Immunohistochemistry (IHC-P)

Species: Mouse
Tissue: RAW264.7 (Mouse monocytic macrophage leukemia cell)
Sample: Paraffin-embedded section

Antigen retrieval: Heat-mediated, Tris-EDTA buffer (pH 9.0), 20 minutes at 95°C.

Wash buffer: 1× TBST
Endogenous peroxidase blocking: 3% H_2O_2, 10 minutes at room temperature.
Blocking: 1% BSA + 10% normal goat serum, 10 minutes at room temperature.
Primary antibody: HA724831, 1/1,000, 1 hour at room temperature.
Secondary antibody: HA1119, 20 minutes at room temperature.

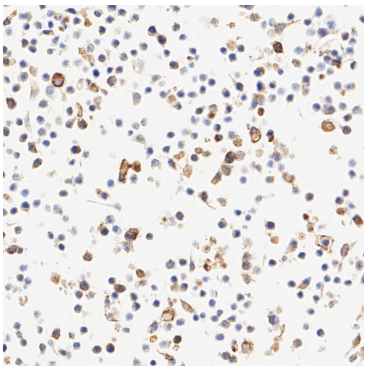


Fig9: Application: Immunohistochemistry (IHC-P)

Species: Mouse
Tissue: J774A.1 (Mouse monocyte macrophage)
Sample: Paraffin-embedded section

Antigen retrieval: Heat-mediated, Tris-EDTA buffer (pH 9.0), 20 minutes at 95°C.

Wash buffer: 1× TBST
Endogenous peroxidase blocking: 3% H_2O_2, 10 minutes at room temperature.
Blocking: 1% BSA + 10% normal goat serum, 10 minutes at room temperature.
Primary antibody: HA724831, 1/1,000, 1 hour at room temperature.
Secondary antibody: HA1119, 20 minutes at room temperature.

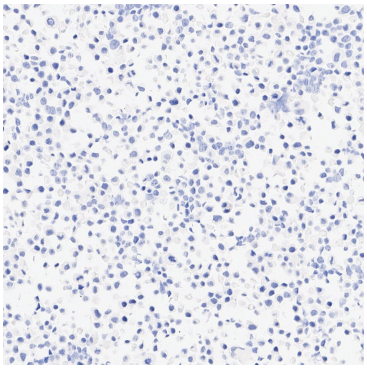


Fig10: Application: Immunohistochemistry (IHC-P)

Species: Mouse
Tissue: Neuro-2a (Mouse brain neuroblastoma cell)
Sample: Paraffin-embedded section

Negative control: Neuro-2a (PMID: 35937296).

Antigen retrieval: Heat-mediated, Tris-EDTA buffer (pH 9.0), 20 minutes at 95°C.

Wash buffer: 1× TBST
Endogenous peroxidase blocking: 3% H_2O_2, 10 minutes at room temperature.
Blocking: 1% BSA + 10% normal goat serum, 10 minutes at room temperature.
Primary antibody: HA724831, 1/1,000, 1 hour at room temperature.
Secondary antibody: HA1119, 20 minutes at room temperature.

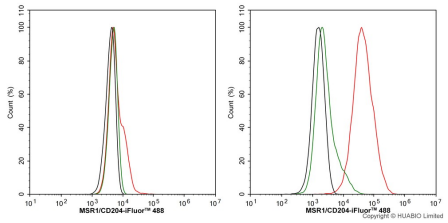


Fig11: Application: Flow Cytometry

Species: Mouse
Sample: NIH/3T3 (Mouse fibroblasts) (Left, negative) / J774A.1 (Mouse monocyte macrophage)

Blocking: 1% BSA + 10% normal goat serum, 15 minutes at room temperature.
Antibody dilution buffer: 1x PBS.

Primary antibody: HA724831(1/1,000, Red) compared with Rabbit IgG Isotype Control (HA722127, Green), 15 minutes at room temperature.
Secondary antibody: Goat Anti-Rabbit IgG (iFluor™ 488, HA1121), 15 minutes at room temperature.

Negative control: NIH/3T3 (PMID: 35937296).

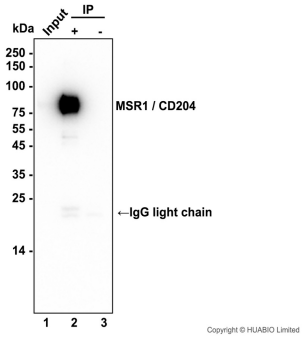


Fig12: Immunoprecipitation (IP)

MSR1 / CD204 was immunoprecipitated in 0.2 mg RAW264.7 (Mouse monocytic macrophage leukemia cell) treated with 50nM PMA and 5μg/mL LPS for 24 hours cell lysate with HA724831 at 2 μg/10 μl beads. Western blot was performed from the immunoprecipitate using HA724831 at 1/5,000 dilution. Anti-Rabbit IgG for IP, AlpSdAbs® VHH(HRP) (025-100-005) at 1/5,000 dilution was used for 1 hour at room temperature.

Lane 1: RAW264.7 treated with 50nM PMA and 5μg/mL LPS for 24 hours cell lysate (input)
Lane 2: HA724831 IP in RAW264.7 treated with 50nM PMA and 5μg/mL LPS for 24 hours cell lysate
Lane 3: Rabbit IgG instead of HA724831 in RAW264.7 treated with 50nM PMA and 5μg/mL LPS for 24 hours cell lysate

Exposure time: 2 seconds
Blocking: 5% NFDM/TBST, 1 hour at room temperature

Primary dilution: HA724831, 1/5,000 in primary antibody dilution buffer (K1803), 2 hours at room temperature

Predicted band size: 50 kDa
Observed band size: 70-90 kDa

Note: All products are "FOR RESEARCH USE ONLY AND ARE NOT INTENDED FOR DIAGNOSTIC OR THERAPEUTIC USE".

Background References

1. Sheng W et al. Role of macrophage scavenger receptor MSR1 in the progression of non-alcoholic steatohepatitis. Front Immunol. 2022 Dec
2. Gudgeon J et al. The role of macrophage scavenger receptor 1 (MSR1) in inflammatory disorders and cancer. Front Immunol. 2022 Oct