

CSF1R | Validation File

TARGET CSF1R

CLONE NAME FER216

DESCRIPTION mouse monoclonal

ANTIGEN USED ecCSF1R-Fc-6His recombinant protein (84kDa-extracellular portion)

ISOTYPE IgG1

SPECIES REACTIVITY human

LOCALIZATION membrane

POSITIVE CONTROL Tonsil

STORAGE BUFFER Tissue culture supernatant: 0.02% sodium azide

STORAGE Aliquot and store at 4C. Do not freeze



Recommended



Inconclusive



Not Recommended



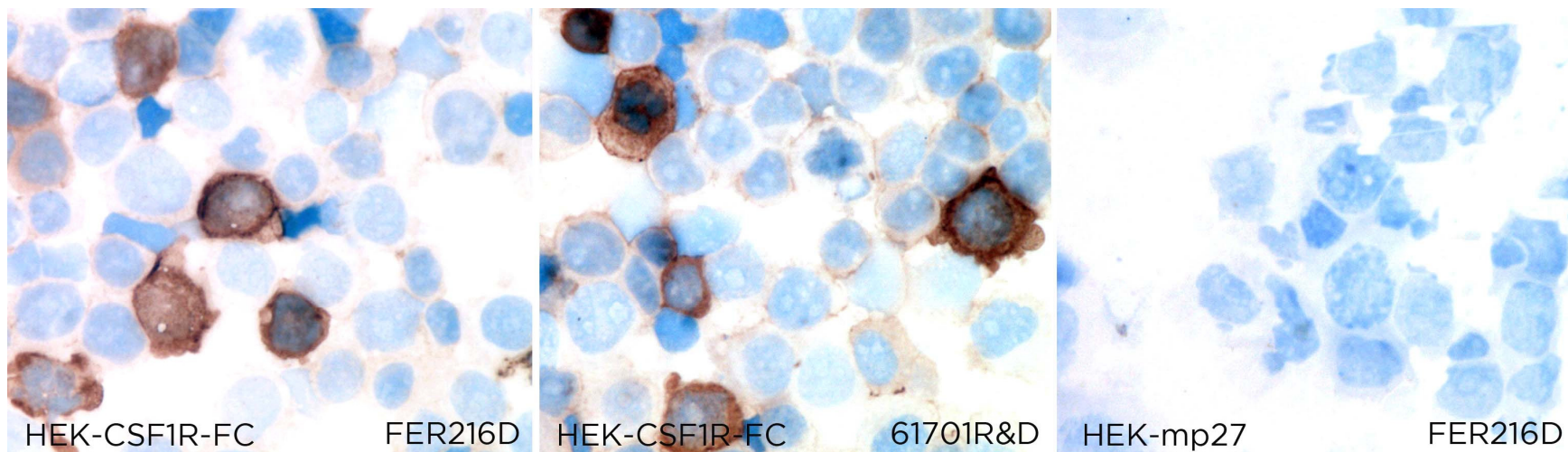
Not Tested

APPLICATIONS

● | ICC | *Immunocytochemistry*

FER216 mAb is able to detect human CSF1R protein in immunocytochemistry

To confirm that FER216 mAb recognizes human CSF1R protein, immunocytochemistry on frozen cytospin preparations of Fc-tagged human CSF1R expressed in HEK293T cell line was performed. Labeling with the anti-CSF1R antibody (R&D SYSTEMS Clone 61701) confirmed the efficiency of transfection. HEK-mp27 was used as negative control.



● | WB | **Western Blotting**

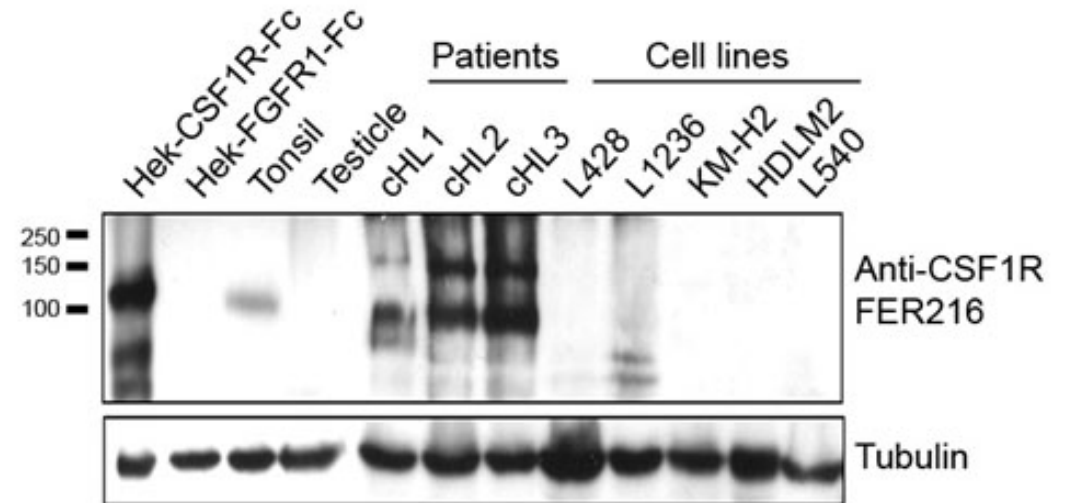
FER216 mAb is able to detect human CSF1R protein by WB.

DILUTION undiluted supernatant

Predicted molecular weight: **107kDa**
Observed molecular weight: **107kDa**

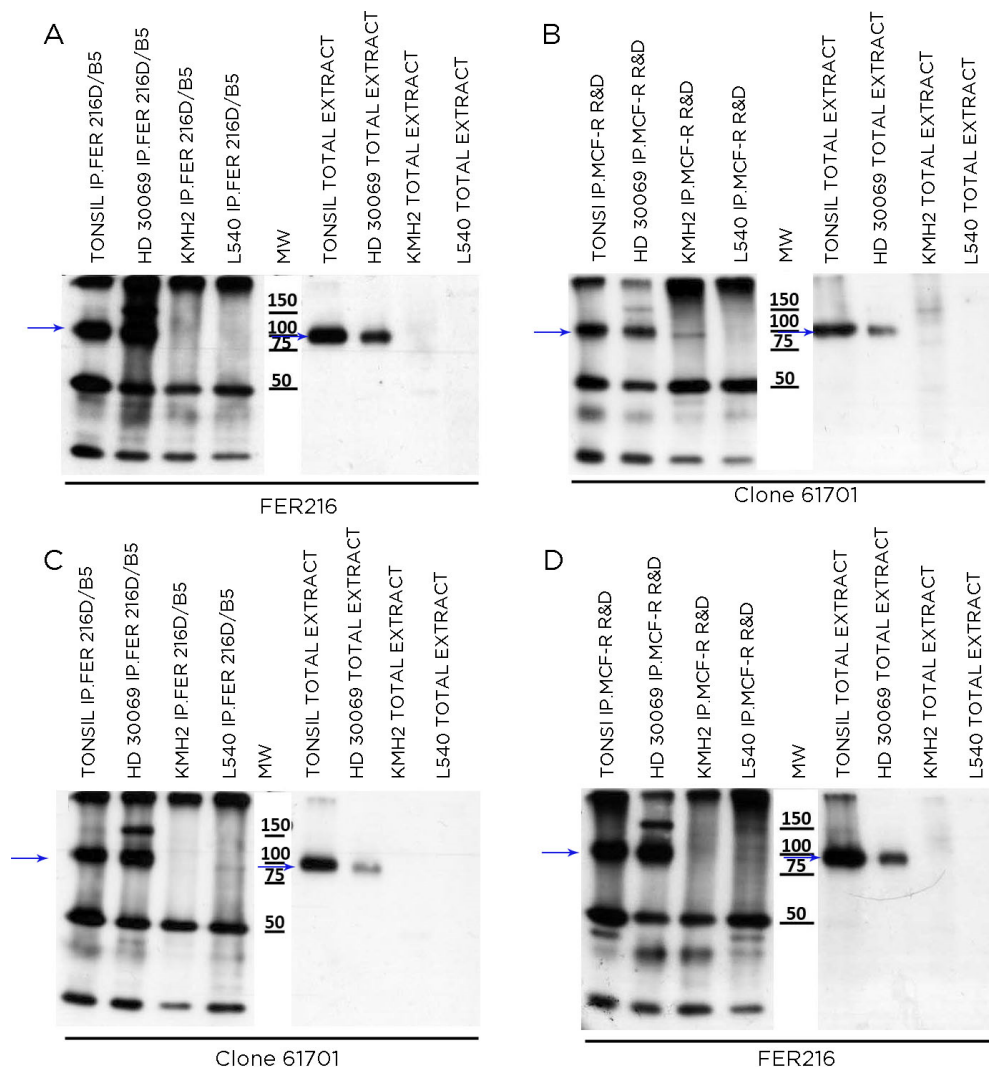
LANES

Lane 1 Hek-CSF1R-Fc	(20ug) (+)
Lane 2 Hek-FGFR1-Fc	(20ug) (-)
Lane 3 Tonsil	(100ug) (+)
Lane 4 Testicle	(100ug) (-)
Lane 5 HL Case 1	(100ug) (+)
Lane 6 HL Case 2	(100ug) (+)
Lane 7 HL Case 3	(100ug) (+)
Lane 8 L428 cell line	(100ug) (-)
Lane 9 L1236 cell line	(100ug) (-)
Lane10 KMH2 cell line	(100ug) (-)
Lane 11 HDLM2 cell line	(100ug) (-)
Lane 12 L540 cell line	(100ug) (-)



| IP | **Immunoprecipitation**

FER216 mAb can be used to detect CSF1R protein by IP.



Result:

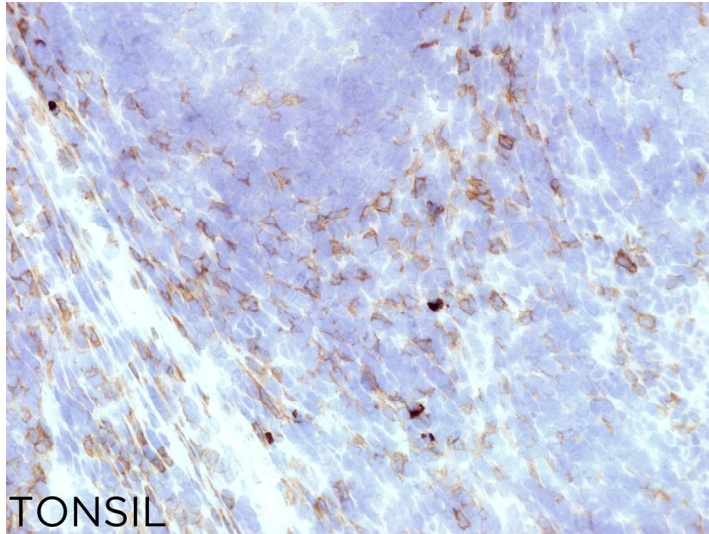
- Immunoprecipitation of protein extracts from human tonsil, one HD case, KMH2, and L540 with FER216 mAb (30ul/IP) followed by western blotting with FER216 (1:5 dilution).
- Immunoprecipitation of protein extracts from human tonsil, HD case, KMH2, and L540 with R&D Systems antibody (clone 61701-1ul/IP) followed by western blotting with the same antibody (1:50 dilution).
- Immunoprecipitation of protein extracts from human tonsil, one HD case, KMH2, and L540 with FER216 mAb (30ul/IP) followed by western blotting with R&D Systems antibody (1:50 dilution).
- Immunoprecipitation of protein extracts from human tonsil, one HD case, KMH2, and L540 with R&D Systems antibody (clone 61701-1ul/IP) followed by western blotting with FER216 antibody (1:5 dilution).

● | IHC-F | ***Immunohistochemistry (frozen)***

FER216 mAb can be used to detect CSF1R protein in human frozen tissues.

TISSUE SAMPLE Human tonsil

DILUTION neat (supernatant)



● | IHC-P | **Immunohistochemistry (paraffin)**

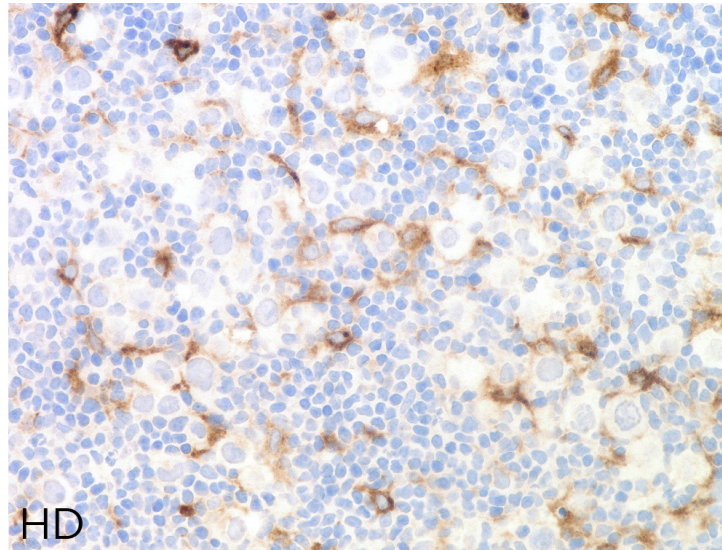
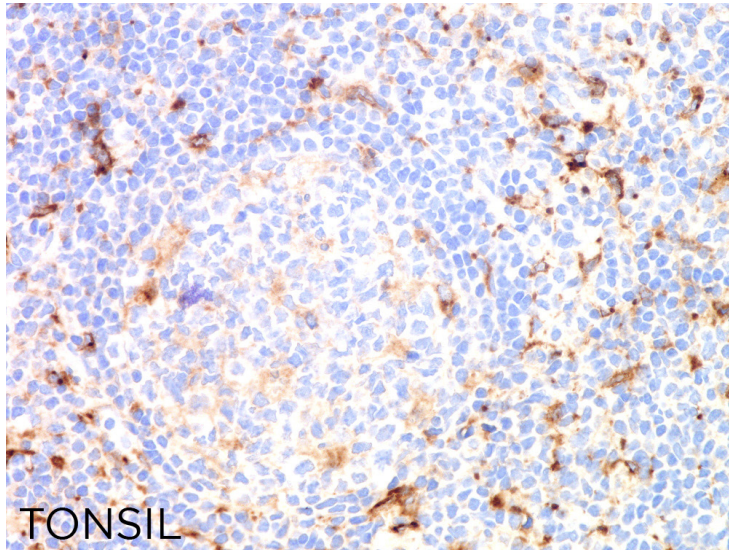
FER216 mAb can be used to detect CSF1R protein in human paraffin tissues

TISSUE SAMPLE tonsil

DILUTION 1:10 (supernatant)

ANTIGEN RETRIEVAL 20 minutes ER2 (Tris-EDTA)

DETECTION SYSTEM Novolink kit (BondMax Leica)



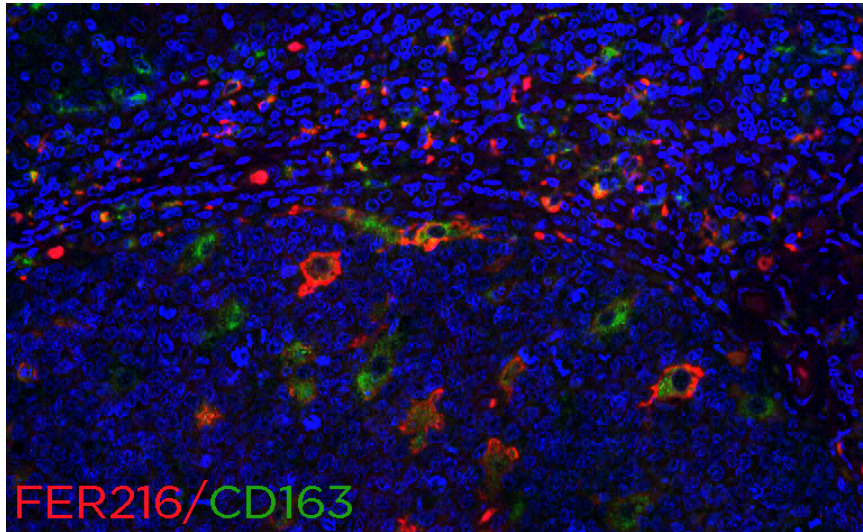
● | IF | **Immunofluorescence (paraffin)**

FER216 mAb can be used to detect CSF1R protein by immunofluorescence

TISSUE SAMPLE Human tonsil

DILUTION 1:10 supernatant

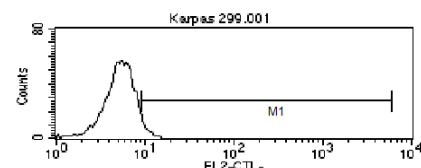
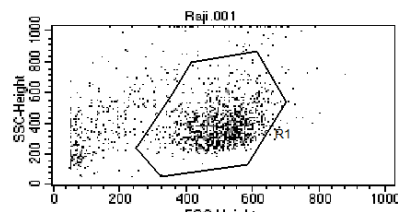
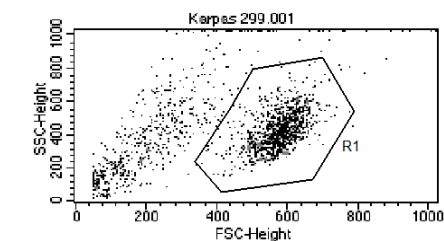
ANT. RETRIEVAL 20 minutes ER2 (Tris-EDTA)



FER216 mAb can be used to detect CSF1R protein by flow cytometry.

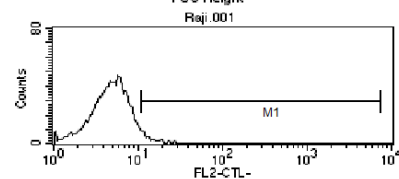
CELLS 500.000 cell/tube of Karpas 299 and Raji human cell lines

DILUTION 1/200, 1/400 and 1/800



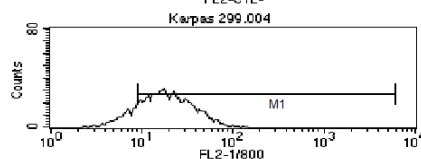
Gate: G1
Total Events: 7956
Gated Events: 5079
X Parameter: FL2-H FL2-CTL- (Log)

Marker	Left	Right	Events	% Gated	% Total	Mean
All	1	9910	5079	100.00	63.84	5.49
M1	9	6043	183	3.60	2.30	11.29



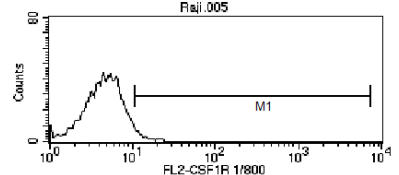
Gate: G1
Total Events: 7488
Gated Events: 5331
X Parameter: FL2-H FL2-CTL-

Marker	Left	Right	Events	% Gated	% Total	Mean
All	1	9910	5331	100.00	71.19	5.22
M1	11	7432	162	3.04	2.16	15.03



Gate: G1
Total Events: 8536
Gated Events: 5083
X Parameter: FL2-H FL2-1/800 (Lo)

Marker	Left	Right	Events	% Gated	% Total	Mean
All	1	9910	5083	100.00	59.55	21.63
M1	9	6043	4166	81.96	48.81	24.94



Gate: G1
Total Events: 7837
Gated Events: 5369
X Parameter: FL2-H FL2-CSF1R

Marker	Left	Right	Events	% Gated	% Total	Mean
All	1	9910	5369	100.00	68.51	5.01
M1	11	7432	140	2.61	1.79	14.49

SOLD BY: Millipore

REFERENCES

Martín-Moreno AM, Roncador G, Maestre L, Mata E, Jiménez S, Martínez-Torrecedrada JL, Reyes-García AI, Rubio C, Tomás JF, Estévez M, Pulford K, Piris MA, García JF. CSF1R Protein Expression in Reactive Lymphoid Tissues and Lymphoma: Its Relevance in Classical Hodgkin Lymphoma. PLoS One 2015 Jun 12;10(6).