

BM4007**Monoclonal Antibody to Macrophage F4/80 antigen - Purified**

Alternate names:	Cell surface glycoprotein EMR1, EMR1 hormone receptor, Emr1, Gpf480
Quantity:	0.2 mg
Concentration:	0.4 mg/ml (after reconstitution)
Background:	<p>F4/80 antigen is a 160 kDa cell surface glycoprotein that is a member of the EGF TM7 family of proteins which shares 68% overall amino acid identity with human EGF module containing mucin like hormone receptor 1 (EMR1). Expression of F4/80 is heterogeneous and is reported to vary during macrophage maturation and activation. The F4/80 antigen is expressed on a wide range of mature tissue macrophages including Kupffer cells, Langerhans, microglia, macrophages located in the gut lamina propria, peritoneal cavity, lung, thymus, bone marrow stroma and macrophages in the red pulp of the spleen. F4/80 expression has also been reported on a subpopulation of dendritic cells but is absent from macrophages located in T cell areas of the spleen and lymph node. The ligands and biological functions of the F4/80 antigen have not yet been determined but recent studies suggest a role for F4/80 in the generation of efferent CD8+ve regulatory T cells.</p>
Uniprot ID:	Q61549
NCBI:	NP_034260
GeneID:	13733
Host / Isotype:	Rat / IgG2a
Recommended Isotype Controls:	SM15P, SM15PX
Clone:	BM8
Immunogen:	<p>Cultured macrophages. The F4/80 antigen is a 125 kD extracellular membrane protein sensitive to 2-Mercaptoethanol.</p>
Format:	<p>State: Lyophilized purified IgG fraction Purification: Immunoaffinity Chromatography Buffer System: Phosphate buffered saline pH 7.2 (PBS) Preservatives: 0.05% (v/v) Kathon CG Stabilizers: 5 mg/ml BSA Reconstitution: Restore by adding 0.5 ml distilled water</p>
Applications:	<p>Immunohistochemistry on Frozen Sections: 0.4 µg/ml (1/1000). Immunohistochemistry on Paraffin Sections: 4 µg/ml (1/100). Proteinase K pretreatment for antigen retrieval is recommended. Positive Control: Mouse spleen. Has been described to work in FACS. Other applications not tested. Optimal dilutions are dependent on conditions and should be determined by the user.</p>

Specificity:

This antibody is useful for the detection of major subpopulations of resident tissue macrophages. The antigen expression increases upon maturation of macrophage precursors in bone marrow and blood as well as in ontogeny.

This clone *BM8* is the only macrophage marker that is able to distinguish non-destructive from destructive inflammation processes in the pancreas and has been shown to be a unique histological marker of the progression from peri-insulinitis to β -cell destruction and diabetes in a mouse diabetes model.

Antigen Distribution on Tissue and Isolated Sections: The antigen is detected on tissue fixed macrophages in all organs tested so far (spleen, lymph nodes, thymus, liver, skin). It is also present on Langerhans cells in the skin and Kupffer cells in the liver. In complete Freund's adjuvant induced granulomas the antigen is expressed by inflammatory macrophages, but is absent from epitheloid cells.

The antigen is expressed *in vitro* on over 80% of M-CSF stimulated bone marrow derived macrophages, after a few days of culture. It is absent from granulocytes, lymphocytes and thrombocytes.

Species: Mouse. This antibody stains also Human heart macrophages. Other species not tested.

Storage:

Store lyophilized at 2-8°C for 6 months or at -20°C long term.

After reconstitution store the antibody undiluted at 2-8°C for one month or (in aliquots) at -20°C long term.

Avoid repeated freezing and thawing.

Shelf life: one year from despatch.

Product Citations:**Purchased from Acris:**

1. Kakehashi H, Nishioku T, Tsukuba T, Kadowaki T, Nakamura S, Yamamoto K. Differential regulation of the nature and functions of dendritic cells and macrophages by cathepsin E. *J Immunol.* 2007 Nov 1;179(9):5728-37. PubMed PMID: 17947645.
2. Kimura H, Miyashita H, Suzuki Y, Kobayashi M, Watanabe K, Sonoda H, et al. Distinctive localization and opposed roles of vasohibin-1 and vasohibin-2 in the regulation of angiogenesis. *Blood.* 2009 May 7;113(19):4810-8. doi: 10.1182/blood-2008-07-170316. Epub 2009 Feb 9. PubMed PMID: 19204325.
3. Torzewski M, Shaw PX, Han KR, Shortal B, Lackner KJ, Witztum JL, et al. Reduced *in vivo* aortic uptake of radiolabeled oxidation-specific antibodies reflects changes in plaque composition consistent with plaque stabilization. *Arterioscler Thromb Vasc Biol.* 2004 Dec;24(12):2307-12. Epub 2004 Nov 4. PubMed PMID: 15528482.
4. Stangl S, Gehrmann M, Riegger J, Kuhs K, Riederer I, Sievert W, et al. Targeting membrane heat-shock protein 70 (Hsp70) on tumors by cmHsp70.1 antibody. *Proc Natl Acad Sci U S A.* 2011 Jan 11;108(2):733-8. doi: 10.1073/pnas.1016065108. Epub 2010 Dec 27. PubMed PMID: 21187371.
5. Rydman, E. Inflammatory effects of nanosized titanium dioxide and carbon nanotube pulmonary exposure. Thesis 2016. <https://helda.helsinki.fi/handle/10138/166835>.
6. ISSN 2412-9690 Actualscience 2016, 35-36, Tom 2, № 10

General Readings:

1. Malorny U, Michels E, Sorg C. A monoclonal antibody against an antigen present on mouse macrophages and absent from monocytes. *Cell Tissue Res.* 1986;243(2):421-8. PubMed PMID: 3948241.

2. Kraal G, Rep M, Janse M. Macrophages in T and B cell compartments and other tissue macrophages recognized by monoclonal antibody MOMA-2. An immunohistochemical study. Scand J Immunol. 1987 Dec;26(6):653-61. PubMed PMID: 3321409.
3. Leenen PJ, de Bruijn MF, Voerman JS, Campbell PA, van Ewijk W. Markers of mouse macrophage development detected by monoclonal antibodies. J Immunol Methods. 1994 Sep 14;174(1-2):5-19. PubMed PMID: 8083537.
4. Smit MJ, Duursma AM, Koudstaal J, Hardonk MJ, Bouma JM. Infection of mice with lactate dehydrogenase-elevating virus destroys the subpopulation of Kupffer cells involved in receptor-mediated endocytosis of lactate dehydrogenase and other enzymes. Hepatology. 1990 Nov;12(5):1192-9. PubMed PMID: 2172137.
5. Rosmalen JG, Martin T, Dobbs C, Voerman JS, Drexhage HA, Haskins K, et al. Subsets of macrophages and dendritic cells in nonobese diabetic mouse pancreatic inflammatory infiltrates: correlation with the development of diabetes. Lab Invest. 2000 Jan;80(1):23-30. PubMed PMID: 10652999.
6. Schaller E, Macfarlane AJ, Rupec RA, Gordon S, McKnight AJ, Pfeffer K. Inactivation of the F4/80 glycoprotein in the mouse germ line. Mol Cell Biol. 2002 Nov;22(22):8035-43. PubMed PMID: 12391169.

Protocols:**Protocol with frozen, ice-cold acetone-fixed sections:**

The whole procedure is performed at room temperature

1. Wash in PBS
2. Block endogenous peroxidase
3. Wash in PBS
4. Block with 10% normal goat serum in PBS for 30min. in a humid chamber
5. Incubate with primary antibody (dilution see datasheet) for 1h in a humid chamber
6. Wash in PBS
7. Incubate with secondary antibody (peroxidase-conjugated goat anti rat IgG (H+L) minimal-cross reaction to mouse) for 1h in a humid chamber
8. Wash in PBS
9. Incubate with AEC substrate (3-amino-9-ethylcarbazol) for 12min.
10. Wash in PBS
11. Counterstain with Mayer's hemalum.

Protocol with formalin-fixed, paraffin-embedded sections:

The whole procedure is performed at room temperature

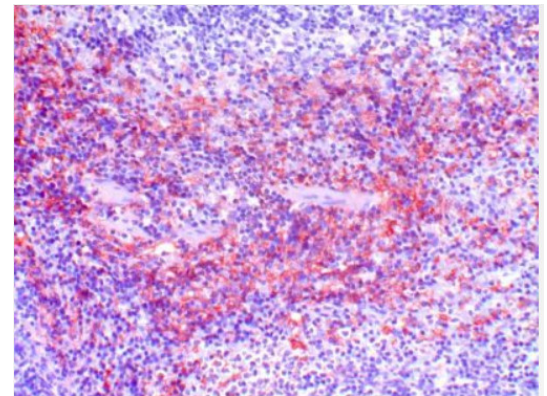
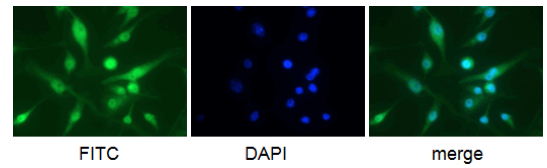
1. Deparaffinize and rehydrate tissue section
2. Incubate the tissue section with proteinase K for 7min.
3. Wash in distilled water
4. Block endogenous peroxidase
5. Wash in PBS
6. Block with 10% normal goat serum in PBS for 30min. in a humid chamber
7. Incubate with primary antibody (dilution see datasheet) for 1h in a humid chamber
8. Wash in PBS
9. Incubate with secondary antibody (peroxidase-conjugated goat anti rat IgG (H+L) minimal-cross reaction to mouse) for 1h in a humid chamber
10. Wash in PBS

11. Incubate with AEC substrate (3-amino-9-ethylcarbazol) for 12min.
12. Wash in PBS
13. Counterstain with Mayer's hemalum.

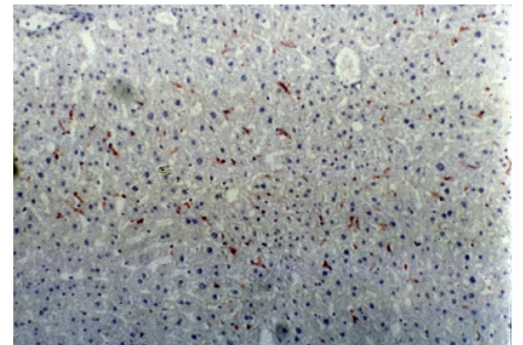
Pictures:

Mouse anti-Macrophage F4/80 antigen antibody Cat.-No BM4007F (5 µg/ml) on Raw 204.4 cells. Cells were fixed in 1% PFA, permeabilized in 0.25% Triton X 100 in PBS, blocked in 1% BSA in PBS.

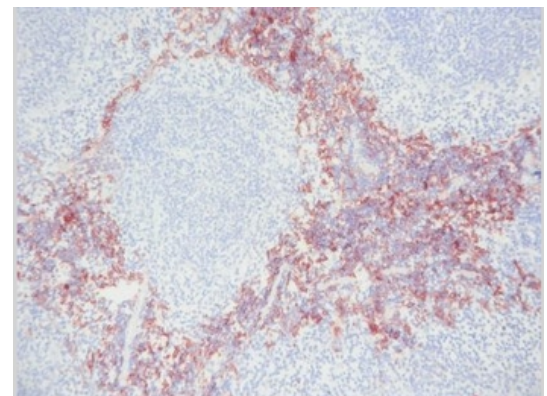
BM4007 F4/80 antibody staining of Mouse Spleen Paraffin.



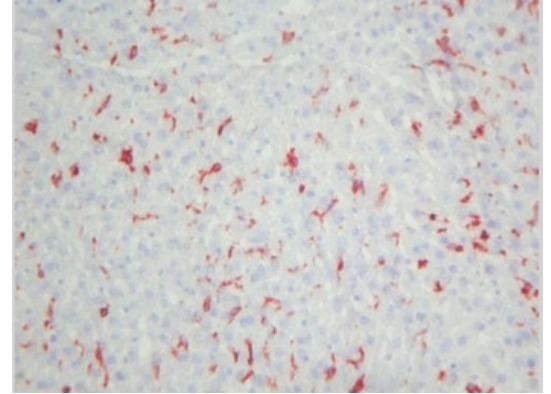
BM4007 F4/80 antibody staining of Mouse Liver Section.



BM4007 F4/80 antibody staining of Mouse Spleen Frozen.



BM4007 F4/80 Antigen antibody staining of Mouse Liver Frozen Section.



Mouse anti-Macrophage F4/80 antigen antibody Cat.-No BM4007F on thioglycollate elicited mouse peritoneal macrophages. Purple: BM4007F; Green: Isotype control SM15F; Percentages reflect % positive after subtraction of negative control, using M1 marker.

