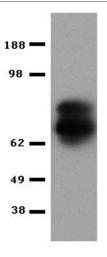


Anti-Mouse Perforin Purified

Catalog Number: 14-9392 RUO: For Research Use Only



Immunoblotting of CTLL-2 lysates using Anti-Mouse Perforin Purified at 2 μ g/ml and revealed with Anti-Rat HRP.

Product Information

Contents: Anti-Mouse Perforin Purified

REF Catalog Number: 14-9392 Clone: eBioOMAK-D Concentration: 0.5 mg/ml Host/Isotype: Rat IgG2a, κ Formulation: aqueous buffer, 0.09% sodium azide, may contain

carrier protein/stabilizer

Temperature Limitation: Store at 2-8°C.

LOT Batch Code: Refer to Vial

Use By: Refer to Vial

Se By: Refer to Vial
Caution, contains Azide

Description

The eBioOMAK-D antibody reacts with mouse perforin (pore-forming protein, pfp, Prf). Perforin is one of the cytolytic mediators present in the cytoplasmic granules of cytotoxic T lymphocytes (CTL) and natural killer cells (NK). Perforin is involved in the killing function by CTLs and NKs and has an important role in the immune response against tumors and virus infections.

By immunobloting, eBioOMAK-D recognizes a ~70kDa band in lysates of CTLL-2 mouse cytotoxic cell line and in lysates of IL-2 stimulated but not unstimulated mouse splenocytes. By multi-color intracellular flow cytometric analysis, eBioOMAK-D staining is increased upon stimulation (IL-2 or anti-CD3/28). Intracellular flow staining results showing upregulation of protein expression have been confirmed by immunoblotting. Furthermore, stimulated Perforin Knock-out (developed by Walsh) splenocytes do not stain with eBioOMAK-D nor is any protein detectable by western blotting with eBioOMAK-D as well as other anti-mouse perforin antibodies. Please note that the Kagi perforin knock-out mice may synthesize a truncated form of the protein which may be recognized by eBioOMAK-D.

In IL-2 stimulated mouse splenocytes, NK cells (as determined by CD49b staining) contain perforin while CD8 cells contain little to none and can vary with culture conditions. This has been confirmed by staining and western blotting the two populations using both OMAK-D and P1-8 antibodies. In contrast stimulation of splenocytes with anti-CD3/CD28 antibodies does result in an increase of perforin on both NK cells and CD8 cells.

eBioOMAK-D is also crossreactive to human perforin and co-stains CD56 positive cells in PBMC.

Expression of perforin and Granzyme B do not always correlate (as discussed above in the CD8 population of IL-2 stimulated splenocytes). Granzyme B typically is expressed earlier and at higher levels. Expression of Granzyme B is dramatically increased (more than 10,00 fold based on mRNA estimates and significantly at the protein level based on western blotting and flow analysis) compared to a minimal increase (10-100 fold) in perforin mRNA and protein with IL-2 stimulation.

For intracellular staining and flow cytometric analysis with direct conjugates of anti-mouse perforin, it is highly recommended to use the Foxp3 buffer system (cat. 00-5523). Other buffers may yield varying results. For more information, please contact technical support at tech@ebioscience.com.

Applications Reported

This eBioOMAK-D antibody has been reported for use in immunoblotting (WB).

Applications Tested

This eBioOMAK-D antibody has been tested by immunblotting at 2ug/ml.

References

Fehniger TA, Cai SF, Cao X, Bredemeyer AJ, Presti RM, French AR, Ley TJ. Acquisition of Murine NK Cell Cytotoxicity Requires the Translation of a Pre-existing Pool of Granzyme B and Perforin mRNAs Immunity 2007 May (Epub) (eBioOMAK-D, IC, PubMed)

Liu CC, Walsh CM, Young JD. Perforin: structure and function. Immunol Today. 1999. 16(4):194-201.

Opferman JT, Ober BT, Ashton-Rickardt PG. Linear differentiation of cytotoxic effectors into memory T lymphocytes. Science. 1999. 283 (5408):1745-8.

Slifka MK, Rodriguez F, Whitton JL. Rapid on/off cycling of cytokine production by virus-specific CD8+ T cells. Nature. 1999. 401(6748):76-9.

Walsh CM, Matloubian M, Liu CC, Ueda R, Kurahara CG, Christensen JL, Huang MT, Young JD, Ahmed R, Clark WR. Immune function in mice lacking the perforin gene. Proc Natl Acad Sci U S A. 1994. 91(23):10854-8.

Related Products

00-5523 Foxp3 Staining Buffer Set

00-8222 IC Fixation Buffer

11-9994 Anti-Human Perforin FITC (dG9 (delta G9))

12-9994 Anti-Human Perforin PE (dG9 (delta G9))

14-4321 Rat IgG2a K Isotype Control Purified

14-8822 Anti-Mouse Granzyme B Purified (16G6)

88-8823 Fixation & Permeabilization Buffers

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