

## Technical Data Sheet

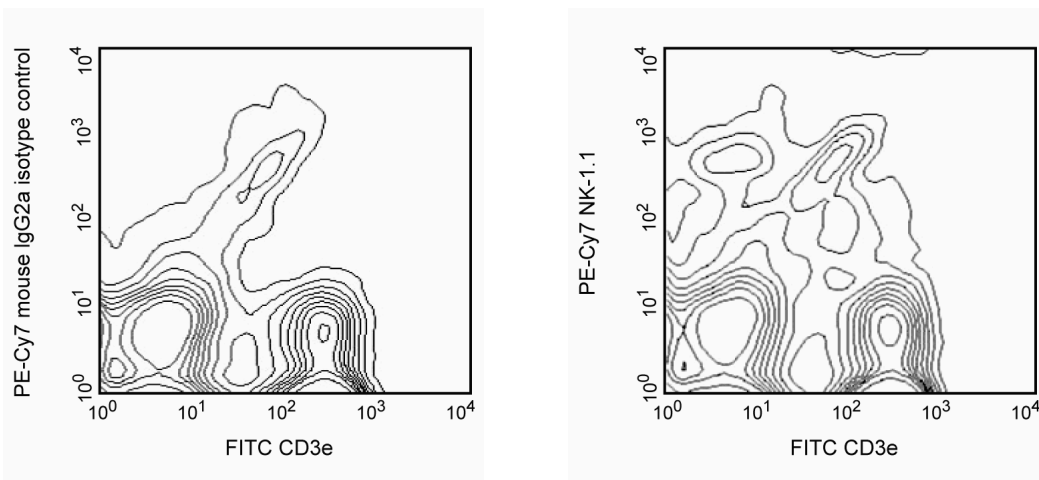
## PE-Cy™7 Mouse Anti-Mouse NK-1.1

## Product Information

<b>Material Number:</b>	<b>562062</b>
<b>Alternate Name:</b>	NKR-P1B and NKR-P1C
<b>Size:</b>	25 µg
<b>Concentration:</b>	0.2 mg/ml
<b>Clone:</b>	PK136
<b>Immunogen:</b>	Mouse NK-1+ Spleen and Bone Marrow Cells
<b>Isotype:</b>	Mouse (C3H x BALB/c) IgG2a, κ
<b>Reactivity:</b>	QC Testing: Mouse
<b>Storage Buffer:</b>	Aqueous buffered solution containing ≤0.09% sodium azide.

## Description

In the mouse, at least three members of the *Klrb* (Killer cell lectin-like receptor, subfamily *b*; formerly *NKR-P1*) gene family have been identified (*Klrb1a/NKR-P1A*, *Klrb1b/NKR-P1B*, and *Klrb1c/NKR-P1C*); but in the human gene family, a single homologue has been designated *KLRB1*, *NKR-P1A*, or *CD161*. The KLRB1/NKR-P1 family of proteins are type-II-transmembrane C-type lectin receptors. KLRB1C/NKR-P1C activates NK-cell cytotoxicity, while KLRB1B/NKR-P1B functions as an inhibitory receptor. KLRB1B/NKR-P1B protein has intracellular Immunoreceptor Tyrosine-based Inhibitory Motif (ITIM), while KLRB1C/NKR-P1C lacks ITIM and activates via association with Fc Receptor γ chain. Strikingly, KLRB1B/NKR-P1B and KLRB1C/NKR-P1C share 96% amino acid sequence identity in their extracellular C-type lectin domains. The PK136 antibody reacts with the NK-1.1 surface antigen encoded by the *Klrb1c/NKR-P1C* gene expressed on natural killer (NK) cells in selected strains of mice (eg, C57BL, FVB/N, NZB, but not A, AKR, BALB/c, CBA/J, C3H, C57BR, C58, DBA/1, DBA/2, NOD, SJL, 129) and the antigen encoded by the *Klrb1b/NKR-P1B* gene expressed only on Swiss NIH and SJL mice, but not on C57BL/6. Expression of KLRB1C/NKR-P1C protein is correlated with the ability to lyse tumor cells in vitro and to mediate rejection of bone marrow allografts. The NK-1.1 marker is useful in defining NK cells; however, the antigen is also expressed on a rare, specialized population of T lymphocytes (NK-T cells) and some cultured monocytes. Plate-bound PK136 mAb, in combination with low concentrations of IL-2, induces proliferation of a subset of NK cells.



**Two-color analysis of NK-1.1 expression on splenocytes.** C57BL/6 splenocytes were stained with FITC Hamster anti-Mouse CD3e mAb (Cat. No. 553061/553062) and either PE-Cy™7 Mouse IgG2a, κ Isotype Control (Cat. No. 552868, left panel) or PE-Cy™7 Mouse anti-Mouse NK-1.1 (right panel). NK-1.1+ CD3e- NK cells and NK-1.1[dim] CD3e+ NK-T cells are detected. Please note that the dead leukocytes were not excluded in this experiment, and the typical diagonal dead-cell population is partially obscured in the right panel. Flow cytometry was performed on a BD FACSCalibur™ Flow Cytometry System.

## Preparation and Storage

Store undiluted at 4°C and protected from prolonged exposure to light. Do not freeze.

The monoclonal antibody was purified from tissue culture supernatant or ascites by affinity chromatography.

The antibody was conjugated with PE-Cy7 under optimum conditions, and unconjugated antibody and free PE-Cy7 were removed.

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## Application Notes

### Application

Flow cytometry

Routinely Tested

### Suggested Companion Products

Catalog Number	Name	Size	Clone
553061	FITC Hamster Anti-Mouse CD3e	0.1 mg	145-2C11
552868	PE-Cy7 <sup>TM</sup> Mouse IgG2a, κ Isotype Control	0.1 mg	G155-178
554656	Stain Buffer (FBS)	500 ml	(none)
553142	Purified Rat Anti-Mouse CD16/CD32 (Mouse BD Fc Block <sup>TM</sup> )	0.5 mg	2.4G2

### Product Notices

1. Since applications vary, each investigator should titrate the reagent to obtain optimal results.
2. Please refer to [www.bdbiosciences.com/pharmingen/protocols](http://www.bdbiosciences.com/pharmingen/protocols) for technical protocols.
3. PE-Cy7 is a tandem fluorochrome composed of R-phycoerythrin (PE), which is excited by 488-nm light and serves as an energy donor, coupled to the cyanine dye Cy7, which acts as an energy acceptor and fluoresces maximally at 780 nm. PE-Cy7 tandem fluorochrome emission is collected in a detector for fluorescence wavelengths of 750 nm and higher. Although every effort is made to minimize the lot-to-lot variation in the efficiency of the fluorochrome energy transfer, differences in the residual emission from PE may be observed. Therefore, we recommend that individual compensation controls be performed for every PE-Cy7 conjugate. PE-Cy7 is optimized for use with a single argon ion laser emitting 488-nm light, and there is no significant overlap between PE-Cy7 and FITC emission spectra. When using dual-laser cytometers, which may directly excite both PE and Cy7, we recommend the use of cross-beam compensation during data acquisition or software compensation during data analysis.
4. Cy is a trademark of Amersham Biosciences Limited. This conjugated product is sold under license to the following patents: US Patent Nos. 5,486,616; 5,569,587; 5,569,766; 5,627,027.
5. This product is subject to proprietary rights of Amersham Biosciences Corp. and Carnegie Mellon University and made and sold under license from Amersham Biosciences Corp. This product is licensed for sale only for research. It is not licensed for any other use. If you require a commercial license to use this product and do not have one return this material, unopened to BD Biosciences, 10975 Torreyana Rd, San Diego, CA 92121 and any money paid for the material will be refunded.
6. Please observe the following precautions: Absorption of visible light can significantly alter the energy transfer occurring in any tandem fluorochrome conjugate; therefore, we recommend that special precautions be taken (such as wrapping vials, tubes, or racks in aluminum foil) to prevent exposure of conjugated reagents, including cells stained with those reagents, to room illumination.
7. Warning: Some APC-Cy7 and PE-Cy7 conjugates show changes in their emission spectrum with prolonged exposure to formaldehyde. If you are unable to analyze fixed samples within four hours, we recommend that you use BD<sup>TM</sup> Stabilizing Fixative (Cat. No. 338036).
8. For fluorochrome spectra and suitable instrument settings, please refer to our Fluorochrome Web Page at [www.bdbiosciences.com/colors](http://www.bdbiosciences.com/colors).
9. Caution: Sodium azide yields highly toxic hydrazoic acid under acidic conditions. Dilute azide compounds in running water before discarding to avoid accumulation of potentially explosive deposits in plumbing.
10. An isotype control should be used at the same concentration as the antibody of interest.

### References

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Carlyle JR, Martin A, Mehra A, Attisano L, Tsui FW, Zuniga-Pflucker JC. Mouse NKR-P1B, a novel NK1.1 antigen with inhibitory function. *J Immunol*. 1999; 162(10):5917-5923. (Clone-specific: Immunoprecipitation)

Koo GC, Peppard JR. Establishment of monoclonal anti-NK-1.1 antibody. *Hybridoma*. 1984; 3(3):301-303. (Immunogen: Cytotoxicity, Flow cytometry)

Kung SK, Su RC, Shannon J, Miller RG. The NKR-P1B gene product is an inhibitory receptor on SJL/J NK cells. *J Immunol*. 1999; 162(10):5876-5887. (Clone-specific: Blocking)

Reichlin A, Yokoyama WM. Natural killer cell proliferation induced by anti-NK1.1 and IL-2. *Immunol Cell Biol*. 1998; 76(2):143-152. (Clone-specific: Induction)

Sentman CL, Kumar V, Koo G, Bennett M. Effector cell expression of NK1.1, a murine natural killer cell-specific molecule, and ability of mice to reject bone marrow allografts. *J Immunol*. 1989; 142(6):1847-1853. (Clone-specific: Depletion)

Yu YY, Kumar V, Bennett M. Murine natural killer cells and marrow graft rejection. *Annu Rev Immunol*. 1992; 10:189-213. (Biology)