IOTest® CD158b1/b2,j-PE

Formerly designated as CD158b-PE – PN IM2278U – Liquid 1 mL – 20 µL / test* – Clone GL183

Analyte Specific Reagent.

Analytical and performance characteristics are not established.

SPECIFICITY

NK cells are innate immune effectors: they can exert natural cytotoxicity and secrete cytokines and chemokines in the absence of sensitization (1). NK cells also mediate antibody-dependent cellular cytotoxicity (ADCC) via FcγRIII (CD16). Using multiple receptors, NK cells can select (with Natural Killer Receptors: NKRs) and engage (with NKG2D (2), and Natural Cytotoxicity Receptors: NCRs) a variety of target cells for their lysis. While the presence of HLA-Class I antigens on target cells can inhibit NK cells cytotoxicity, the absence of HLA-Class I antigens does not, by itself, activate NK cells (3): this is the basis of the "missing self" concept (3) that refers to the negative regulation provided by inhibitory receptors when they sense adequate HLA-Class I molecules on target cells (3). The complex mosaic expression of activating or inhibiting receptors on the different NK cell subsets adds more possibilities to the regulation of NK cell activity. The KIR (Killer-cell Immunoglobulin-like Receptor) acronym is used to designate certain inhibitory or activating receptors of HLA-Class I molecules. These receptors are expressed on NK cells and on a subset of T cells. Reports on the KIR and CD nomenclatures have been published (4, 5).

According to this nomenclature, the CD158b1 molecule also known as KIR2DL2 (KIR, 2 extracellular Ig like domain, long cytoplasmic tail, 2) and the CD158b2 molecule also known as KIR2DL3 (KIR, 2 extracellular Ig like domain, long cytoplasmic tail, 3) identify the p58.2 and p58.3 receptors respectively, and the CD158j molecule also known as KIR2DS2 (KIR2D, short cytoplasmic tail, 2) identifies the p50.2 receptor. p58.2/p58.3 and p50.2 receptors specifically recognize HLA molecules of the Cw3 supertype, including Cw3, Cw1, Cw7 and Cw8 alleles within the HLA-Cw series (6, 7). Brother-receptors p58.1 and p50.1 are recognized by monoclonal antibodies (mAbs) belonging to CD158a and CD158h, respectively.

CD158b1/b2 and CD158j molecules are expressed by human NK cell subsets and by human T lymphocyte subsets, the latest being, in most instances, $CD8^+CD4^-$ TCRalpha/beta$^+$ (8, 9).

The GL183 mAb reacts with the extracytoplasmic identical region of p58.2, p58.3 and p50.2 molecules, thus it can be classified as CD158b1/b2,j (10 – 13). It was assigned to the CD158b overall cluster of differentiation at the 6th International Workshop on Human Leucocyte Differentiation Antigens (HLDA) in Kobe, Japan, in 1996 (14).

REAGENT

IOTest CD158b1/b2,j-PE Conjugated Antibody

PN IM2278U – Liquid 1 mL – 20 µL / test*.

Clone	GL183
Isotype	IgG1, mouse
Immunogen	Human NK clone E57
Hybridoma	P3V1 x Balb/c
Source	Ascites fluid
Purification	Ion exchange or affinity chromatography
Conjugation	R-phycoerythrin (PE) is conjugated at 0.5 – 1.5 moles of PE per mole of Ig.

Excitation wavelength: 488 nm

Maximum emission wavelength: 575 nm

Main emission color: Orange-red

REAGENT CONTENTS

This reagent is provided in phosphate-buffered saline containing 0.1% sodium azide (NaN_3) as preservative, and 2 mg/mL bovine serum albumin (BSA).

STATEMENT OF WARNINGS

1. This reagent contains 0.1% sodium azide. Sodium azide under acid conditions yields hydrazoic acid, an extremely toxic compound. Azide compounds should be flushed with running water while being discarded. These precautions are recommended to avoid deposits in metal piping in which explosive conditions can develop. If skin or eye contact occurs, wash excessively with water.
2. Specimens, samples and all material coming in contact with them should be handled as if capable of transmitting infection and disposed of with proper precautions.
3. Never pipet by mouth and avoid contact of samples with skin and mucous membranes.
4. Do not use reagent beyond the expiration date on the vial label.
5. Minimize exposure of reagent to light during storage or incubation.
6. Avoid microbial contamination of reagent or erroneous results may occur.
7. Use good laboratory practices when handling this reagent.

STORAGE CONDITIONS AND STABILITY

This reagent is stable up to the expiration date printed on the vial label when stored at 2 –8°C in the dark. Do not freeze. Minimize exposure to light.

EVIDENCE OF DETERIORATION

Any change in the physical appearance of this PE-labeled reagent (clear, colorless to pink liquid) or any major variation in values obtained for control samples may indicate deterioration and the reagent should not be used.

REAGENT PREPARATION

No preparation is necessary. This reagent is used directly from the vial. Bring reagent to 18 – 25°C prior to use.

SELECTED RESEARCH REFERENCES

1. Moretta, A., Bottino, C., Vitale, M., Pende, D., Cantoni, C., Mingari, M.C., Biassoni, R., Moretta, L., "Activating receptors and coreceptors involved in human natural killer cell-mediated cytolysis", 2001, Annu. Rev. Immunol., 19, 197-223.
2. Houchins, J.P., Yabe, T., McSherry, C., and Bach, F.H., "DNA sequence analysis of NKG2, a family of related cDNA clones encoding type II integral membrane proteins on human natural killer cells", 1991, J. Exp. Med., 173, 1017-1020.
3. Watzl, C., "The NKG2D receptor and its ligands - recognition beyond the "missing self"?", 2003, Microbes and Infection, 5, 31-37.
4. Marsh, S.G.E., Parham, P., Dupont, B., Geraghty, D.E., Trowsdale, J., Middleton, D., Vilches, C., Carrington, M, Witt, C., Guethlein, L.A., Shilling, H., Garcia, C.A., Hsu, K.C., and Wain, H., "Killer-cell immunoglobulin-like receptor (KIR) nomenclature report, 2002", 2003, Immunogenetics, 55, 220-226.
5. André, P., Biassoni, R., Colonna, M., Cosman, D., Lanier, L.L., Long, O., Lopez-Botet, M., Moretta, A., Moretta, L., Parham, P., Trowsdale, J., Vivier, E., Wagtmann, N., and Wilson, M.J., "New nomenclature for MHC receptors", 2001, Nature Immunol., 2, 661.
6. Moretta, A., Vitale, M., Bottino, C., Orengo, A.M., Morelli, L., Augugliaro, R., Barbaresi, M., Ciccone, E., Moretta, L., "p58 molecules as putative receptors for major histocompatibility complex (MHC) class I molecules in human Natural Killer (NK) cells. Anti-p58 antibodies reconstitute lysis of MHC class I-protected cells in NK clones displaying different specificities", 1993, J. Exp. Med., 178, 597-604.
7. Ciccone, E., Pende, D., Viale, O, Than, A., Di Donato, C., Orengo, A.M., Biassoni, R., Verdiani, S., Amoroso, A.,

*** 20 µL is the quantity of product sufficient to stain 5×10^5 cells in a standard immunofluorescence assay**

BECKMAN COULTER

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Moretta, A., Moretta, L., "Involvement of HLA class I alleles in Natural Killer (NK) cell-specific functions: Expression of HLA-Cw3 confers selective protection from lysis by alloreactive NK clones displaying a defined specificity (specificity 2)", 1992, J. Exp. Med., 176, 963-971.

8. Ferrini, S., Cambiaggi, A., Meazza, R., Sforzini, S., Marciano, S., Mingari, M.C., Moretta, L., "T cell clones expressing the natural killer cell-related p58 receptor molecule display heterogeneity in phenotypic properties and p58 function", 1994, Eur. J. Immunol., 24, 2294-2298.
9. Mingari, M.C., Schiavetti, F., Ponte, M., Vitale, C., Maggi, E., Romagnani, S., Demarest, J., Pantaleo, G., Fauci, A.S., Moretta, L., "Human $CD8^+$ T lymphocyte subsets that express HLA class I-specific inhibitory receptors represent oligoclonally or monoclonally expanded cell populations", 1996, Immunology, 93, 12433-12438.
10. Moretta, A., Tambussi, G., Bottino, C., Tripodi, G., Merli, A., Ciccone, E., Pantaleo, G., Moretta, L., "A novel surface antigen expressed by a subset of human $CD3^-CD16^+$ Natural Killer cells", 1990, J. Exp. Med., 171, 695-714.
11. Wagtmann, N., Biassoni, R., Cantoni, C., Verdiani, S., Malnati, M.S., Vitale, M., Bottino, C., Moretta, L., Moretta, A., Long, E.O., "Molecular clones of the p58 NK cell receptor reveal immunoglobulin-related molecules with diversity in both the extra- and intracellular domains", 1995, Immunity, 2, 439-449.
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14. Moretta, L., Poggi, A., Nanni, L., Bottino, C., Melioli, G., Biassoni, R., Moretta, A., "Natural killer cell antigens: section report", 1997, Leucocyte Typing VI, White Cell Differentiation Antigens. Kishimoto, T., et al., Eds., Garland Publishing, Inc., 265-267.

PRODUCT AVAILABILITY

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PE is licensed under patent 4,520,110

For additional information in the USA, call 800-526-7694.
Outside the USA, contact your local Beckman Coulter representative

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TRADEMARKS

Manufactured by:
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*** 20 µL is the quantity of product sufficient to stain 5×10^5 cells in a standard immunofluorescence assay**