IOTest Anti-OSCAR-PE

PN A24987 – 100 tests – Liquid – 20 µL/test – Clone 11.1CN5

For Research Use Only. Not for use in diagnostic procedures.

SPECIFICITY

OSCAR, an abbreviation of osteoclast-associated receptor, is a new member of the leukocyte receptor complex (LRC) (1). Human OSCAR is a cell-surface, monomeric, N-glycosylated protein of approximately 45 kDa. The amino acid sequence shows homology with that of members of the KIR family. The protein exhibits two C2-type immunoglobulin extracellular domains, a transmembrane section that contains a positively charged arginine residue, and a short intracytoplasmic tail with no recognizable signaling motif. The gene coding for OSCAR has been mapped to chromosome 19q13 (2).

Unlike murine OSCAR that is expressed only by osteoclasts, human OSCAR is widely expressed in cells of the myeloid lineage, and in particular, at all stages of myeloid dendritic cell maturation. *In vitro* derived macrophages, granulocytes, and monocyte-derived dendritic cells also express OSCAR. In contrast, T, B, and NK cells do not express OSCAR.

OSCAR associates with the ITAM-adapter FcRγ and not with other signaling molecules, due to the presence of the charged residue in the transmembrane domain. Ligation of OSCAR by specific antibodies promotes dendritic cell (DC) survival by an ERK- and PI3K-dependent pathway. That survival is linked to expression of the Bcl-2 and Bcl-x_L anti-apoptotic molecules. Ligation of OSCAR also leads to maturation of DC with secretion of chemokines recruiting Th_2 and regulatory cells, but no significant effect on naïve T cell proliferation is noted. The two ITAM-bearing adapter chains DAP12 and FcRγ for TREM2 (3) and OSCAR, respectively, are involved in DC differentiation and function, but a clear difference may be noted with FcRγ for OSCAR in the involvement of ERK in the survival, and in the capacity to induce cytokine secretion (4).

REAGENT

IOTest Anti-OSCAR-PE Conjugated antibody
PN A24987 - 100 tests - Liquid - 20 µL/test

Clone	11.1CN5
Isotype	IgG1, Mouse
Immunogen	Human OSCAR extracellular domain fused to human Fcγ fragment
Hybridoma	X63 x balb/c
Source	Ascites fluid or supernatant of in vitro cultured hybridoma cells.
Purification	Affinity chromatography
Conjugation	R Phycoerythrin (PE)
Molar Ratio	PE / Ig : 0.5 - 1.5
Fluorescence	Excites at 488 nm Emits at 575 nm

REAGENT CONTENTS

This antibody is provided in phosphate-buffered saline, containing 0.1% sodium azide and 2 mg/mL bovine serum albumin.

APPLICATION

Flow cytometry.

STATEMENTS OF WARNING

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 considered potentially infectious 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 antibody beyond the expiration date on the label.
5. Do not expose reagents to strong light during storage or incubation.
6. Avoid microbial contamination of reagents or incorrect results might occur.
7. Use good laboratory practices when handling this reagent.

STORAGE CONDITIONS AND STABILITY

This reagent is stable up to the expiration date when stored at 2 – 8°C. Do not freeze.

REAGENT PREPARATION

No reconstitution is necessary. This monoclonal antibody may be used directly from the vial. Bring reagent to 18 – 25°C prior to use.

PROCEDURE

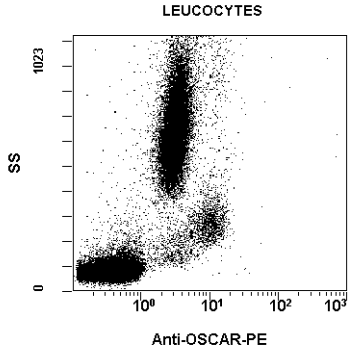
This reagent is designed for Flow Cytometry.
Assay volume: 20 µL per 5 x 10^5 cells in one test, or per 100 µL whole blood.
It is recommended to establish the right range of antibody dilutions to be used for the experiment.

EXAMPLE DATA

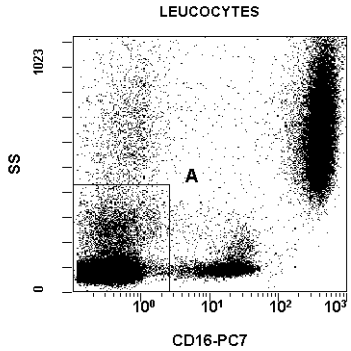
Histograms below are obtained from a normal whole blood sample stained with CD33-FITC (PN IM1135), Anti-OSCAR-PE (PN A24987), CD14-ECD (PN IM2707), CD85k (ILT3)-PC5 (PN IM3579) and CD16-PC7 (PN 6607418). Red blood cells are lysed with VersaLyse (PN IM3648).

Acquisition is with the BECKMAN COULTER CYTOMICS FC 500 flow cytometer. Analysis is with the CYTOMICS CXP analysis software.

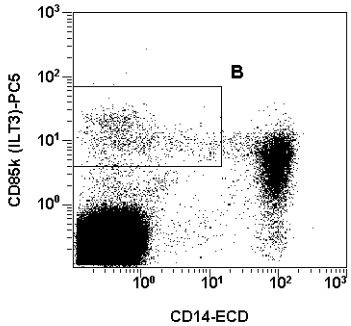
Histogram 1 shows Anti-OSCAR-PE *versus* side scatter (SS).



Histogram 2 shows CD16-PC7 *versus* SS. A gate region A is drawn around $CD16^-$ events.



Histogram 3, gated on the A region, shows CD14-ECD *versus* CD85k (ILMT3)-PC5.
A gate region B, set around $CD14^-$, $CD85k^+$ events, encompasses all $CD16^-$ $CD14^-$ $CD85k^+$ events that represent dendritic cells.



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IG-720–RUO_L_AH

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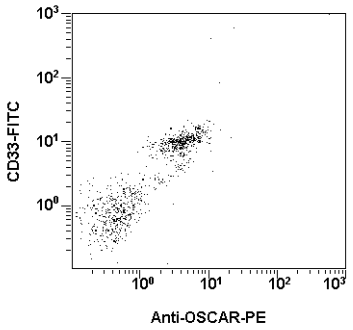
Histogram 4, gated on the A and B regions, shows lack of expression of OSCAR on plasmacytoid DC ($CD33^{-}$) while expression on myeloid DC ($CD33^{+}$) cells.



SELECTED RESEARCH REFERENCES

1. Kim, N., Takami, M., Rho, J., Josien, R., Choi, Y., “A novel member of the leukocyte receptor complex regulates osteoclast differentiation", 2002, J. Exp. Med., 195, 2, 201-209.
2. Merck, E., Gaillard, C., Gorman, D., Montero-Julian, F., Durand, I., Zurawski, S., Menetrier-Caux, C., Carra, G., Lebecque, S., Trinchieri, G., Bates, E., “OSCAR is an FcRγ-associated receptor that is expressed by myeloid cells and is involved in antigen presentation and activation of human dendritic cells", 2004, Blood, 104, 5, 1386-1395.
3. Paloneva, J., Mandelin, J., Kiialainen, A., Böhling, T., Prudlo, J., Hakola, P., Haltia, M., Konttinen, Y., Peltonen, L., “DAP12/TREM2 deficiency results in impaired osteoclast differentiation and osteoporotic features", 2003, J. Exp. Med., 198, 4, 669-675.
4. Merck, E., de Saint-Vis, B., Scuiller, M., Gaillard, C., Caux, C., Trinchieri, G., Bates, E., "Fc receptor gamma-chain activation via hOSCAR induces survival and maturation of dendritic cells and modulates Toll-like receptor responses", 2005, Blood, prepublished online.

TRADEMARKS

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