

### Analyte Specific Reagent.

Analytical and performance characteristics are not established.

#### SPECIFICITY

The CD80 antigen (B7, B7-1, or BB1) is a highly glycosylated single-chain transmembrane protein, structurally similar to CD86 (B7-2 or B70), with a molecular weight of 60 kDa, under non reducing conditions (1, 2). Its extracellular region is composed of two Ig-like domains. CD80 shares with CD86 the same co-receptors on T cells, CD28 and CD152 (CTLA-4) (3).

CD80 and CD86 have a critical role in one costimulatory pathway involved in the prevention of antigen-specific T-cell tolerance (anergy), mediated by ligation of CD28 on T cells by its ligands, CD80 and CD86 on antigen-presenting cells (4). Interactions between CD28 on T-cells and CD80 (or CD86) on activated B cells result in enhanced T-cell activation (1, 5). CD152 (CTLA-4) binds CD80 and CD86 with a higher affinity and probably functions as a negative regulator for T-cell activation (6, 7). The MAB104 monoclonal antibody (mAb) reacts with *in vitro* activated B lymphocytes, some B cell lines, and weakly with a small proportion of non-activated B cells (8). This antibody also reacts with activated T cells but not with peripheral monocytes and T cells (3, 8, 9).

The MAB104 mAb was assigned to the CD80 cluster of differentiation at the 6th International Workshop on Human Leucocyte Differentiation Antigens in Kobe, Japan, in 1996 (3).

#### REAGENT

IOTest CD80-PE Conjugated Antibody

PN IM1976U – 2 mL Liquid – 20 µL / test\*.

#### Clone

MAB104

#### Isotype

IgG1, mouse

#### Immunogen

Jijoye cells (Human Burkitt Lymphoma cell line)

#### Hybridoma

NS1 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.

#### Fluorescence

PE (orange-red)  
Excites at 486 – 580 nm  
Emits at 568 – 590 nm

#### REAGENT CONTENTS

This reagent is provided in phosphate-buffered saline, with 0.1% sodium azide (NaN<sub>3</sub>) as preservative, and 2.0 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. Do not use antibody beyond the expiration date on the label.
3. Samples and all material coming in contact with them should be handled as if capable of transmitting infection and disposed of with proper precautions.
4. Never pipet by mouth and avoid contact of samples with skin and mucous membranes
5. Minimize exposure of reagent to 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. Minimize exposure to light.

#### EVIDENCE OF DETERIORATION

Any change in the physical appearance of this PE-labeled reagent (clear colorless to pinkish 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 monoclonal antibody may be used directly from the vial. Bring reagent to 18 – 25°C prior to use.

#### SELECTED RESEARCH REFERENCES

1. Boussiotis, V.A., Freeman, G.J., Gribben, J.G., Daley, J., Gray, G., Nadler, L.M., "Activated human B lymphocytes express three CTLA-4 counter receptors that costimulate T-cell activation", 1993, Proc. Natl. Acad. Sci. USA, 90, 11059-11063.
2. Engel, P., Gribben, J.G., Freeman, G.J., Zhou, L.J., Nozawa, Y., Abe, M., Nadler, L.M., Wakasa, H., Tedder, T.F., "The B7-2 (B70) costimulatory molecule expressed by monocytes and activated B lymphocytes is the CD86 differentiation antigen", 1994, Blood, 84, 1402-1407.
3. Jones, M., Mason, D.Y., "CD80 Workshop Panel report", 1997, Leucocyte Typing VI, White Cell Differentiation Antigens. Kishimoto, T., et al., Eds., Garland Publishing, Inc., 186-187.

4. Gause, W.C., Halvorson, M.J., Lu, P., Greenwald, R., Linsley, P., Urban, J.F., Finkelman, F.D., "The function of costimulatory molecules and the development of IL-4 producing T cells", 1997, Immunol. Today, 18, 115-120.
5. June, C.H., Ledbetter, J.A., Linsley, P.S., Thompson, C.B., "Role of the CD28 receptor in T-cell activation", 1990, Immunol. Today, 11, 211-216
6. Linsley, P.S., Nadler, S.G., Bajorath, J., Peach, R., Leung, H.T., Rogers, J., Bradshaw, J., Stebbins, M., Leytze, G., Brady, W., Malacko, A.R., Marquardt, H., Shaw, S.-Y., "Binding stoichiometry of the cytotoxic T lymphocyte-associated molecule-4 (CTLA-4)", 1995, J. Biol. Chem., 270, 15417-15424.
7. Linsley, P.S., Golstein, P., "Lymphocyte activation: T-cell regulation by CTLA-4", 1996, Curr. Biology, 6, 398-400.
8. Vallé, A., Garrone, P., Yssel, H., Bonnefoy, J.Y., Freedman, Freeman, G., "MAB104, a new monoclonal antibody, recognizes the B7 antigen that is expressed on activated B cells and HTLV-1-transformed T cells", 1990, Immunology, 69, 531-535.
9. Vallé, A., Aubry, J.P., Durand, I., Banchereau, J., "IL-4 and IL-2 upregulate the expression of antigen B7, the B cell counterstructure to T cell CD28: an amplification mechanism for T-B cell interactions", 1991, Int. Immunol., 3, 3, 229-235.

#### PRODUCT AVAILABILITY

IOTest CD80-PE Conjugated Antibody

PN IM1976U – 2 mL Liquid – 20 µL / test\*.

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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(\*) : 20 µL is the quantity of product sufficient to stain

5 x 10<sup>5</sup> cells in a standard immunofluorescence assay