

Technical Data Sheet

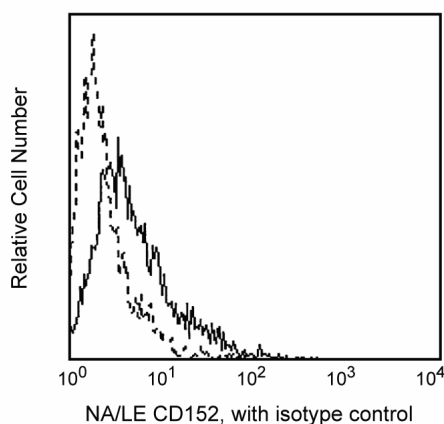
Purified NA/LE Mouse Anti-Human CD152

Product Information

Material Number:	555850
Alternate Name:	CTLA-4; AILIM; Cytotoxic T-lymphocyte protein 4
Size:	0.5 mg
Concentration:	1.0 mg/ml
Clone:	BNI3
Isotype:	Mouse (BALB/c) IgG2a, κ
Reactivity:	QC Testing: Human
Storage Buffer:	No azide/low endotoxin: Aqueous buffered solution containing no preservative, 0.2 μ m sterile filtered. Endotoxin level is ≤ 0.01 EU/ μ g (≤ 0.001 ng/ μ g) of protein as determined by the LAL assay.

Description

The BNI3 monoclonal antibody specifically binds to the human cytolytic T lymphocyte-associated antigen, CTLA-4. CTLA-4 is transiently expressed on activated CD28+ T cells and binds to CD80 and CD86 present on antigen presenting cells (APC) with high avidity. This interaction appears to deliver a negative regulatory signal to the T cell. There are recent reports that indicate that CTLA-4 is also expressed on B cells when cultured with activated T cells, suggesting a possible role of CTLA-4 in the regulation of B-cell response. Immobilized BNI3.1 antibody enhances T-cell proliferation induced by antibody-mediated crosslinking of CD3 and CD28. Recent studies have shown that CD152 can be expressed by regulatory T (Treg) cells. After cellular fixation and permeabilization, the BNI3 antibody can stain intracellular CD152 expressed in T cells including Treg cells. Clone BNI3.1 was studied in the VI Leukocyte Typing Workshop.



Profile of concanavalin-A-stimulated 3-day peripheral blood mononuclear cells analyzed by flow cytometry.

Preparation and Storage

Store undiluted at 4°C.

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

This preparation contains no preservatives, thus it should be handled under aseptic conditions.

Application Notes

Application

Flow cytometry	Routinely Tested
Functional assay	Tested During Development

Recommended Assay Procedure:

For flow cytometric applications, a three step labeling procedure is recommended for amplifying signal. Suggested protocol for 3-step staining using concanavalin-A-stimulated peripheral blood mononuclear cells method:

1. Incubate 100 μ l concanavalin-A-stimulated 3-day peripheral blood mononuclear cells (1×10^6) with primary (unconjugated) antibody for 20-30 minutes at room temperature.
2. Add 2 mls of 1X lysing buffer (10X Pharm Lyse™ Lysing Buffer, Cat. No. 555899) and incubate for 10-15 minutes. Centrifuge and aspirate.

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3. Wash once with PBS/0.1% sodium azide/1% heat-activated fetal bovine serum (PBS-FBS). Centrifuge and aspirate.
4. Add biotinylated goat anti-mouse Ig (Cat. No. 553999) and incubate for 20-30 minutes at room temperature.
5. Wash once with PBS-FBS. Centrifuge and aspirate.
6. Add PE-Streptavidin (Cat. No. 554061) and incubate for 20-30 minutes in the dark at room temperature.
7. Wash once with PBS-FBS. Centrifuge and aspirate. Resuspend in 0.5 ml of PBS-FBS and analyze by flow cytometry.

Suggested Companion Products

Catalog Number	Name	Size	Clone
554061	PE Streptavidin	0.5 mg	(none)
554645	Purified NA/LE Mouse IgG2a, κ Isotype Control	0.5 mg	G155-178
553999	Biotin Goat Anti-Mouse Ig (Multiple Adsorption)	0.5 mg	Polyclonal
555899	Lysing Buffer	100 ml	(none)

Product Notices

1. Since applications vary, each investigator should titrate the reagent to obtain optimal results.
2. An isotype control should be used at the same concentration as the antibody of interest.
3. Please refer to www.bdbiosciences.com/pharming/en/protocols for technical protocols.

References

Barclay NA, Brown MH, Birkeland ML, et al, ed. *The Leukocyte Antigen FactsBook*. San Diego, CA: Academic Press; 1997. (Biology)

Kuiper HM, Brouwer M, Linsley PS, van Lier RA. Activated T cells can induce high levels of CTLA-4 expression on B cells. *J Immunol*. 1995; 155(4):1776-1783. (Biology)

Lindsten T, Lee KP, Harris ES, et al. Characterization of CTLA-4 structure and expression on human T cells. *J Immunol*. 1993; 151(7):3489-3499. (Biology)

Morton PA, Fu XT, Stewart JA, et al. Differential effects of CTLA-4 substitutions on the binding of human CD80 (B7-1) and CD86 (B7-2). *J Immunol*. 1996; 156(3):1047-1054. (Biology)

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