

## Technical Data Sheet

## Purified NA/LE Hamster Anti-Mouse CD80

## Product Information

Material Number:	553765
Alternate Name:	B7-1
Size:	0.5 mg
Concentration:	1.0 mg/ml
Clone:	16-10A1
Immunogen:	Mouse CD80 (B7) Transfected Cell Line
Isotype:	Armenian Hamster IgG2, $\kappa$
Reactivity:	QC Testing: Mouse Reported: Dog
Storage Buffer:	No azide/low endotoxin: Aqueous buffered solution containing no preservative, 0.2 $\mu$ m sterile filtered. Endotoxin level is $\leq 0.01$ EU/ $\mu$ g ( $\leq 0.001$ ng/ $\mu$ g) of protein as determined by the LAL assay.

## Description

The 16-10A1 antibody reacts with CD80 (B7-1). This member of the Ig superfamily, along with CD86 (B7-2), participates in T-cell co-stimulation via interactions with CD28 and CD152 (CTLA-4). CD80 has been reported to be constitutively expressed on dendritic cells, monocytes, and peritoneal macrophages; and it is inducible on B cells by various means, including activation by LPS, IL-4, and the cross-linking of surface Ig. Expression of CD80 has been reported to be greatly enhanced on splenic B cells following activation by LPS, with peak expression occurring between 48 and 72 hours. It has been reported that the activation of purified B cells with LPS can induce CD80 expression in as few as 18 hours. The 16-10A1 antibody has been reported to block binding of CTLA-4 Ig to CD80 and to block T-cell activation by Con A-elicited peritoneal exudate cells and CD80-transfected cell lines. However, 16-10A1 antibody alone is not able to block T-cell activation by antigen-presenting cells. CD86 (B7-2) is an alternate ligand for CD28 and CD152 (CTLA-4). Preliminary reports indicate that the 16-10A1 mAb may block the binding of rat anti-CD80 mAb clone 1G10 (Cat. No. 553368). In addition, it has been reported that the 16-10A1 antibody may cross-react with an activation antigen expressed on IFN- $\gamma$ -activated alveolar macrophages of the dog.

## 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
Blocking	Reported
Immunoprecipitation	Reported
Immunohistochemistry-frozen	Reported

## Recommended Assay Procedure:

**Flow Cytometry:** For flow cytometry of leukocytes, it is recommended that Mouse BD Fc Block™ (Cat. No. 553141) be used. When using Mouse BD Fc Block™ (clone 2.4G2), it is important that the second-step anti-hamster IgG antibody does not cross-react with the 2.4G2 mAb (rat IgG2b,  $\kappa$ ); we have found that the anti-hamster IgG cocktail (Cat. No. 554010) to be effective. Since this antigen is expressed at low density, it may be desirable to amplify staining by using a biotinylated second-step reagent followed by a "bright" third-step reagent, such as PE streptavidin (Cat. No. 554061).

## BD Biosciences

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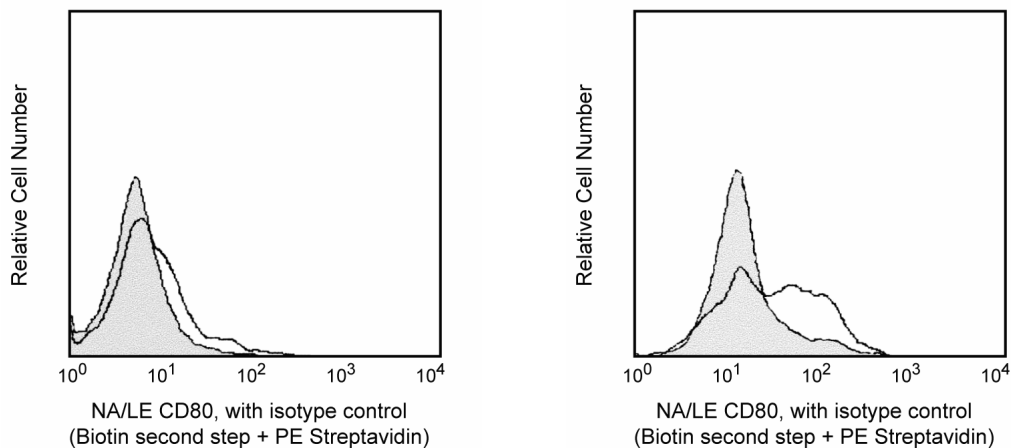
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**Expression of membrane CD80 by mouse splenocytes.** Freshly isolated (left panel) or 72-hour LPS-stimulated BALB/c splenocytes (right panel) were pretreated with Mouse BD Fc Block™ (anti-mouse CD16/CD32 mAb 2.4G2, Cat. No. 553141) and stained with either purified 16-10A1 mAb (open histograms) or purified hamster IgG isotype control mAb (shaded histograms). Staining was detected with biotinylated mouse anti-hamster IgG (Cat. No. 554010) followed by PE streptavidin (Cat. No. 554061). Flow cytometry was performed on a FACScan™ (BD Biosciences, San Jose, CA).

## Suggested Companion Products

Catalog Number	Name	Size	Clone
559339	Purified NA/LE Hamster IgG2, κ Isotype Control	0.5 mg	B81-3
554010	Biotin Mouse Anti-Armenian and Syrian Hamster IgG Cocktail	0.5 mg	(none)
554061	PE Streptavidin	0.5 mg	(none)
553141	Purified Rat Anti-Mouse CD16/CD32 (Mouse BD Fc Block™)	0.1 mg	2.4G2

## Product Notices

- Since applications vary, each investigator should titrate the reagent to obtain optimal results.
- Although hamster immunoglobulin isotypes have not been well defined, BD Biosciences Pharmingen has grouped Armenian and Syrian hamster IgG monoclonal antibodies according to their reactivity with a panel of mouse anti-hamster IgG mAbs. A table of the hamster IgG groups, Reactivity of Mouse Anti-Hamster Ig mAbs, may be viewed at [http://www.bdbiosciences.com/documents/hamster\\_chart\\_11x17.pdf](http://www.bdbiosciences.com/documents/hamster_chart_11x17.pdf).
- Please refer to [www.bdbiosciences.com/pharmingen/protocols](http://www.bdbiosciences.com/pharmingen/protocols) for technical protocols.

## References

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Herold KC, Vezys V, Koons A, Lenschow D, Thompson C, Bluestone JA. CD28/B7 costimulation regulates autoimmune diabetes induced with multiple low doses of streptozotocin. *J Immunol*. 1997; 158(2):984-991. (Biology: Immunohistochemistry, In vivo exacerbation)

Razi-Wolf Z, Freeman GJ, Galvin F, Benacerraf B, Nadler L, Reiser H. Expression and function of the murine B7 antigen, the major costimulatory molecule expressed by peritoneal exudate cells. *Proc Natl Acad Sci U S A*. 1992; 89(9):4210-4214. (Immunogen: Blocking, Immunoprecipitation)

Sojka DK, Donepudi M, Bluestone JA, Mokyr MB. Melphalan and other anticancer modalities up-regulate B7-1 gene expression in tumor cells. *J Immunol*. 2000; 164(12):6230-6236. (Biology)