

## Technical Data Sheet

## Purified Hamster Anti-Mouse CD80

## Product Information

<b>Material Number:</b>	553766
<b>Alternate Name:</b>	B7-1
<b>Size:</b>	0.5 mg
<b>Concentration:</b>	0.5 mg/ml
<b>Clone:</b>	16-10A1
<b>Immunogen:</b>	Mouse CD80 (B7) Transfected Cell Line
<b>Isotype:</b>	Armenian Hamster IgG2, $\kappa$
<b>Reactivity:</b>	QC Testing: Mouse Reported: Dog
<b>Storage Buffer:</b>	Aqueous buffered solution containing $\leq 0.09\%$ sodium azide.

## 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.

This antibody is routinely tested by flow cytometric analysis. Other applications were tested at BD Biosciences Pharmingen during antibody development only or reported in the literature.

## Preparation and Storage

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

Store undiluted at 4° C.

## Application Notes

## Application

Flow cytometry	Routinely Tested
Immunohistochemistry-frozen	Reported
Immunoprecipitation	Reported
Blocking	Reported
In vivo exacerbation	Reported

## Recommended Assay Procedure:

**Precautions for 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 streptavidin-PE (Cat. No. 554061).

**Caution:** Sodium azide is a reversible inhibitor of oxidative metabolism; therefore, antibody preparations containing this preservative agent must not be used in cell cultures nor injected into animals. Sodium azide may be removed by washing stained cells or plate-bound antibody or dialyzing soluble antibody in sodium azide-free buffer. Since endotoxin may also effect the results of functional studies, we recommend the NA/LE™ (No Azide/Low Endotoxin) antibody format for in vitro and in vivo use.

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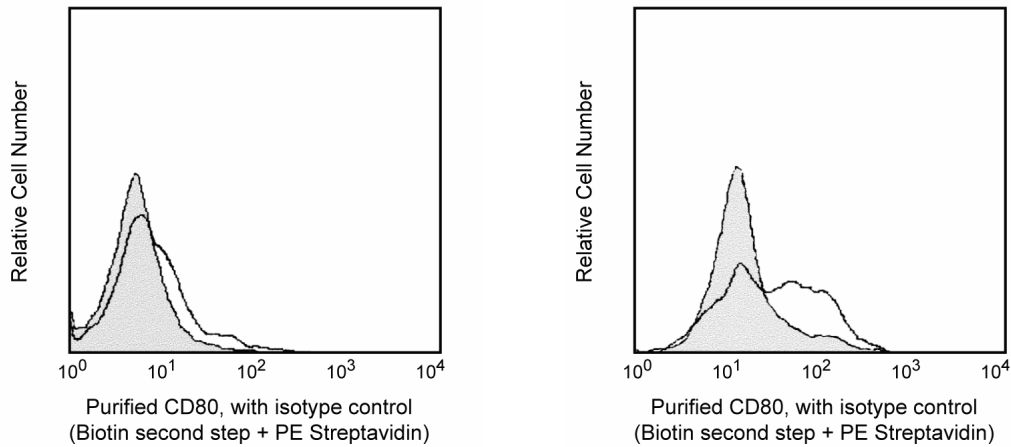
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**Expression of membrane CD80 by mouse splenocytes.** Freshly isolated (left panel) or 72-hour LPS-stimulated BALB/c splenocytes (right panel) were pretreated with Mouse BD Fc Block™ (anti-mouse CD16/CD32 mAb 2.4G2, Cat. No. 553141) and stained with 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 streptavidin-PE (Cat. No. 554061). Flow cytometry was performed on a FACScan™ (BD Biosciences, San Jose, CA).

### Suggested Companion Products

Catalog Number	Name	Size	Clone
554010	Biotin Mouse Anti-Armenian and Syrian Hamster IgG Cocktail	0.5 mg	(none)
554061	Streptavidin PE	0.5 mg	(none)
559277	Purified Hamster IgG2 κ Isotype Control	0.5 mg	B81-3
553141	Purified Rat Anti-Mouse CD16/CD32 (Mouse BD Fc Block™)	0.1 mg	2.4G2
553765	Purified NA/LE Hamster Anti-Mouse CD80	0.5 mg	16-10A1

### Product Notices

1. Since applications vary, each investigator should titrate the reagent to obtain optimal results.
2. Please refer to [www.bdbiosciences.com/pharminggen/protocols](http://www.bdbiosciences.com/pharminggen/protocols) for technical protocols.
3. 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/pharminggen/hamster\\_chart\\_11x17.pdf](http://www.bdbiosciences.com/pharminggen/hamster_chart_11x17.pdf).
4. Caution: Sodium azide yields highly toxic hydrazoic acid under acidic conditions. Dilute azide compounds in running water before discarding to avoid accumulation of potentially explosive deposits in plumbing.

### References

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