Technical Data Sheet

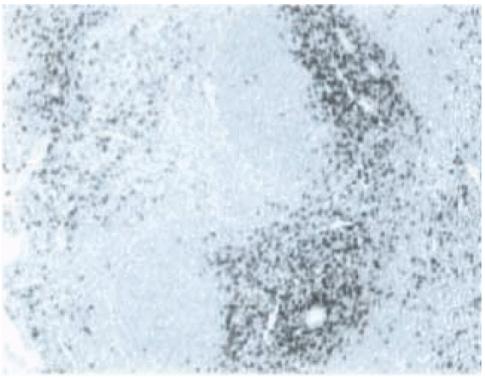
Purified Mouse Anti-Rat CD8a

Product Information

Material Number:	**550298**
Alternate Name:	Cd8a; CD8α; CD8 alpha; OX-8 membrane antigen
Size:	1.0 ml
Concentration:	15.625 µg/ml
Clone:	OX-8
Immunogen:	High-molecular-weight rat thymocyte glycoproteins
Isotype:	Mouse (BALB/c) IgG1, κ
Reactivity:	QC Testing: Rat
Storage Buffer:	Aqueous buffered solution containing BSA, goat serum, and ≤0.09% sodium azide.

Description

The OX-8 antibody reacts with the hinge-like membrane-proximal domain of the 32 kDa α chain of the CD8 differentiation antigen. A truncated CD8 α' isoform has not been detected in the rat. The CD8 α and β chains (CD8a and CD8b, respectively) form a heterodimer on the surface of most thymocytes and a subpopulation of mature T lymphocytes (i.e., MHC class I-restricted T cells, including most T suppressor/cytotoxic cells). Intestinal intrapithelial lymphocytes, many CD8+ T cells of athymic rats, many activated CD4+ T cells, and most NK cells express CD8a without CD8b. It has been suggested that the expression of the CD8a/CD8b heterodimer is restricted to thymus-derived T lymphocytes. OX-8 antibody does not react with resting CD4+ T helper cells. CD8 is an antigen coreceptor on the T-cell surface which interacts with MHC class I molecules on antigen-presenting cells. It participates in T-cell activation through its association with the T-cell receptor complex and protein tyrosine kinase Ick. Macrophages have also been reported to express CD8 α and β chains, which are involved in signal transduction. Soluble OX-8 mAb partially blocks in vitro MLR and CTL activity.



Immunohistochemical staining of Rat T lymphocytes. *The paraffin-embedded section of normal rat spleen was stained with Purified Mouse Anti-Rat CD8a (Cat. No. 550298). CD8+ lymphocytes around the central arterioles of the white pulp are identified by the brown staining.*

Preparation and Storage

Store undiluted at 4°C.

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

Application Notes

Application

Flow cytometry	Routinely Tested
Immunohistochemistry-frozen	Tested During Development
Immunohistochemistry-formalin (antigen retrieval required)	Tested During Development
Immunohistochemistry-zinc-fixed	Tested During Development
Immunohistochemistry-paraffin	Tested During Development
Immunoprecipitation	Reported
Immunoaffinity Chromatography	Reported
Western blot	Reported
Blocking	Reported

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Recommended Assay Procedure:

Immunohistochemistry: The OX-8 antibody is recommended to test for immunohistochemical staining of acetone-fixed frozen sections and paraffin sections. For paraffin sections no pretreatment is required. Tissues tested were rat spleen and thymus. The antibody stains the CD8 subset of T lymphocytes. The isotype control recommended for use with this antibody is purified mouse IgG1 (Cat. No. 550878). For optimal indirect immunohistochemical staining, the OX-8 antibody should be titrated (1:10 to 1:50 dilution) and visualized via a three-step staining procedure in combination with biotinylated polyclonal anti-mouse Ig (multiple adsorbed) (Cat. No. 550337) as the secondary antibody and Streptavidin-HRP (Cat. No. 550946) together with the DAB detection system (Cat. No. 550880). More conveniently, the anti-mouse Ig HRP detection kit (Cat. No. 551011) that contains the biotinylated secondary antibody, antibody diluent, streptavidin-HRP and DAB substrate can be used for staining. For more protocol information please visit **http://www.bdbiosciences.com/resources/cellbiology/index.jsp**

Suggested Companion Products

Catalog Number	Name	Size	Clone
550337	Biotin Goat Anti-Mouse Ig (Multiple Adsorption)	1.0 ml	Polyclonal
550880	DAB Substrate Kit	500 tests	(none)
550946	Streptavidin HRP	50 ml	(none)
551011	Anti-Mouse Ig HRP Detection Kit	200 tests	(none)
550878	Purified Mouse IgG1 κ Isotype Control	1.0 ml	MOPC-31C
559148	Antibody Diluent for IHC	125 ml	(none)

Product Notices

1. Since applications vary, each investigator should titrate the reagent to obtain optimal results.
2. Source of all serum proteins is from USDA inspected abattoirs located in the United States.
3. This antibody has been developed for the immunohistochemistry application. However, a routine immunohistochemistry test is not performed on every lot. Researchers are encouraged to titrate the reagent for optimal performance.
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.
5. 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 affect the results of functional studies, we recommend the NA/LE (No Azide/Low Endotoxin) antibody format, if available, for in vitro and in vivo use.
6. An isotype control should be used at the same concentration as the antibody of interest.
7. Please refer to www.bdbiosciences.com/pharmingen/protocols for technical protocols.

References

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Brideau RJ, Carter PB, McMaster WR, Mason DW, Williams AF. Two subsets of rat T lymphocytes defined with monoclonal antibodies.. *Eur J Immunol.* 1980; 10:609-615. (Immunogen: Flow cytometry)

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Mitnacht R, Bischof A, Torres-Nagel N, Hunig T. Opposite CD4/CD8 lineage decisions of CD4+8+ mouse and rat thymocytes to equivalent triggering signals: correlation with thymic expression of a truncated CD8 alpha chain in mice but not rats. *J Immunol.* 1998; 160(2):700-707. (Clone-specific: Immunoprecipitation, Western blot)

Stitz L, Sobbe M, Bilzer T. Preventive effects of early anti-CD4 or anti-CD8 treatment on Borna disease in rats. *J Virol.* 1992; 66(6):3316-3323. (Clone-specific: Blocking)

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Wallgren AC, Karlsson-Parra A, Korsgren O. The main infiltrating cell in xenograft rejection is a CD4+ macrophage and not a T lymphocyte. *Transplantation* . 1995; 60(6):594-601. (Clone-specific: Immunohistochemistry)

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