Technical Data Sheet

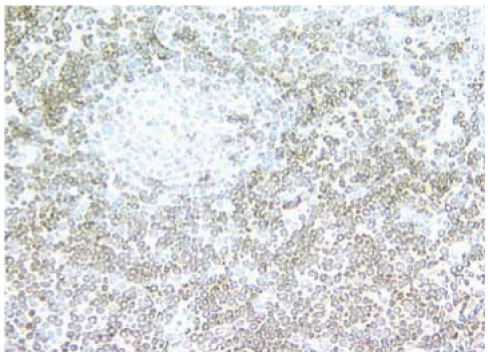
Purified Mouse Anti-Human CD3

Product Information

Material Number:	**550367**
Size:	1.0 ml
Concentration:	125 µg/ml
Clone:	HIT3a
Isotype:	Mouse IgG2a, κ
Reactivity:	QC Testing: Human
Workshop:	V 5T-CD03.05
Storage Buffer:	Aqueous buffered solution containing BSA, goat serum, and ≤0.09% sodium azide.

Description

Reacts with the human ε-chain, a 20 kDa subunit of CD3/T cell antigen receptor complex found on 70-80% of normal human peripheral blood lymphocytes and 60-85% of thymocytes. Studies from the HLDA Workshop show this antibody to be mitogenic when used in conjuction with pokeweed mitogen. CD3 plays a role in signal transduction during antigen recognition. HIT3a antibody does not stain intracellular CD3 unlike the other CD3 clone, UCHT1.



Immunohistochemical staining of T lymphocytes. *Frozen sections of normal human tonsil was reacted with the CD3 antibody. T lymphocytes can be identified by the intense brown labeling of their cell membranes. Amplification 20X.*

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-paraffin	Not Recommended

Recommended Assay Procedure:

Immunohistochemistry: The clone HIT3a specific for human CD3 is recommended to test for immunohistochemical staining of acetone-fixed frozen sections. Tissues tested were human spleen and tonsil. The antibody stains peripheral T lymphocytes and some thymocytes. The isotype control recommended for use with this antibody is purified mouse IgG2a (Cat. No. 550339). For optimal indirect immunohistochemical staining, the HIT3a antibody should be titrated (1:10 to 1:50 dilution) and visualized via a three-step staining procedure in combination with biotin anti-mouse IgG2a (Cat. No. 550332) as the secondary antibody and Streptavidin-HRP (Cat. No. 550946) together with the DAB detection system (Cat. No. 550880). A detailed protocol of the immunohistochemical procedure is available on our website at www.bdbiosciences.com/pharmingen/protocols. The clone HIT3a is not recommended for formalin-fixed paraffin embedded sections.

Suggested Companion Products

Catalog Number	Name	Size	Clone
550339	Purified Mouse IgG2a κ Isotype Control	1.0 ml	C1.18.4
550332	Biotin Rat Anti-Mouse IgG2a	1.0 ml	R19-15
559148	Antibody Diluent for IHC	125 ml	(none)
550880	DAB Substrate Kit	500 tests	(none)

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550946	Streptavidin HRP	50 ml	(none)

Product Notices

1. Since applications vary, each investigator should titrate the reagent to obtain optimal results.
2. 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.
3. Source of all serum proteins is from USDA inspected abattoirs located in the United States.
4. An isotype control should be used at the same concentration as the antibody of interest.
5. 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.
6. Please refer to www.bdbiosciences.com/pharmingen/protocols for technical protocols.

References

Beverley PC, Callard RE. Distinctive functional characteristics of human "T" lymphocytes defined by E rosetting or a monoclonal anti-T cell antibody. *Eur J Immunol.* 1981; 11(4):329-334. (Biology)

Knapp W, Dörken B, Gilks WR, et al, ed. *Leucocyte Typing IV.* New York, NY: Oxford University Press; 1989:1-1182. (Biology)

McMichael AJ, Beverly PCL, Gilks W, et al, ed. *Leukocyte Typing III: White Cell Differentiation Antigens.* New York: Oxford University Press; 1987. (Biology)

Schlossman SF, Boumsell L, Gilks W, et al, ed. *Leukocyte Typing V: White Cell Differentiation Antigens.* New York: Oxford University Press; 1995. (Clone-specific)

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