Technical Data Sheet

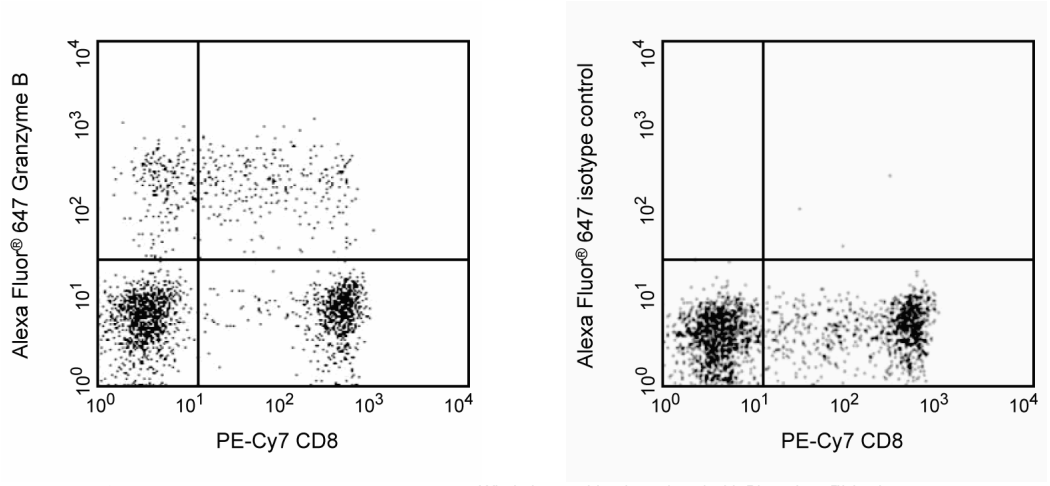
Alexa Fluor® 647 Mouse Anti-Human Granzyme B

Product Information

Material Number:	**561999**
Alternate Name:	GZMB; Granzyme-2; CCPI; CGL1; CSPB; CTLA1; CTSGL1; GRB; HLP; SECT
Size:	25 µg
Concentration:	0.2 mg/ml
Clone:	GB11
Immunogen:	Human Granzyme B
Isotype:	Mouse IgG1, κ
Reactivity:	QC: Human
Storage Buffer:	Aqueous buffered solution containing ≤0.09% sodium azide.

Description

The GB11 antibody specifically reacts with human granzyme B, a serine protease of approximately 32 kDa. Granzyme B is stored in the granules of cytotoxic T lymphocytes and NK cells along with the pore-forming protein perforin. In the classic model of target cell lysis, perforins create holes in the target cell membrane allowing entrance of granzymes. Granzyme B has been shown to act on specific substrates including caspase-3, -7, -9, and -10 which in turn give rise to enzymes that mediate apoptosis. Granzyme B may also be involved in the hydrolysis of extracellular matrix components. Detectable levels of granzyme B have been detected in sera from healthy volunteers. The immunogen used to generate the GB11 hybridoma was human granzyme B isolated from an NK cell line.



***Expression of granzyme B by peripheral blood lymphocytes**. Whole human blood was lysed with PharmLyse™ Lysing buffer (Cat No. 555899) prior to staining with GB11. Whole lysed human blood was subsequently fixed, permeabilized and stained with PE-Cy™7 Mouse anti-Human CD8 (Cat. No. 557746) and either Alexa Fluor® 647 Mouse anti-Human Granzyme B (left panel) or Alexa Fluor® 647 Mouse IgG1 κ Isotype Control (Cat. No. 557732; right panel) by using Pharmingen's staining protocol. To demonstrate specificity of staining, the binding of Alexa Fluor® 647-GB11 was blocked by preincubation of the fixed/permeabilized cells with an excess of unlabelled GB11 antibody (10 µg/10^6 cells, data not shown) prior to staining. The dot plots in the figure were derived from gating on cells with the forward- and side- scatter characteristics of lymphocytes.*

Preparation and Storage

Store undiluted at 4°C and protected from prolonged exposure to light. Do not freeze.

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

The antibody was conjugated to Alexa Fluor® 647 under optimum conditions, and unreacted Alexa Fluor® 647 was removed.

Application Notes

Application

Intracellular staining (flow cytometry)	Routinely Tested

BD Biosciences

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Suggested Companion Products

Catalog Number	Name	Size	Clone
555899	Lysing Buffer	100 ml	(none)
557746	PE-Cy™7 Mouse Anti-Human CD8	100 tests	RPA-T8
557732	Alexa Fluor® 647 Mouse IgG1 κ Isotype Control	100 tests	MOPC-21

Product Notices

1. Since applications vary, each investigator should titrate the reagent to obtain optimal results.
2. Please refer to www.bdbiosciences.com/pharmingen/protocols for technical protocols.
3. For fluorochrome spectra and suitable instrument settings, please refer to our Fluorochrome Web Page at www.bdbiosciences.com/colors.
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. Alexa Fluor® 647 fluorochrome emission is collected at the same instrument settings as for allophycocyanin (APC).
6. The Alexa Fluor®, Pacific Blue™, and Cascade Blue® dye antibody conjugates in this product are sold under license from Molecular Probes, Inc. for research use only, excluding use in combination with microarrays, or as analyte specific reagents. The Alexa Fluor® dyes (except for Alexa Fluor® 430), Pacific Blue™ dye, and Cascade Blue® dye are covered by pending and issued patents.
7. Alexa Fluor® is a registered trademark of Molecular Probes, Inc., Eugene, OR.
8. An isotype control should be used at the same concentration as the antibody of interest.

References

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