Technical Data Sheet

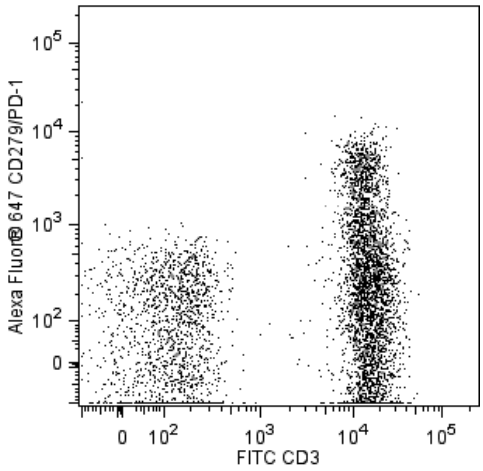
Alexa Fluor® 647 Mouse anti-Human CD279 (PD-1)

Product Information

Material Number:	**560838**
Alternate Name:	hPD-1; PD1; PDCD1; Programmed cell death 1; SLEB2
Size:	100 tests
Vol. per Test:	5 µl
Clone:	EH12.1
Immunogen:	Human PD-1 Recombinant Protein
Isotype:	Mouse IgG1, κ
Reactivity:	QC testing: Human
Storage Buffer:	Aqueous buffered solution containing BSA and ≤0.09% sodium azide.

Description

The EH12.1 monoclonal antibody specifically binds to CD279. CD279 is an immunoregulatory receptor that is expressed on activated T cells, B cells and myeloid cells and contains an immunoreceptor tyrosine-based inhibitory motif (ITIM) in the cytoplasmic region. Mice deficient in CD279 show a breakdown of peripheral tolerance and manifest multiple autoimmune symptoms. PD-L1 and PD-L2 are ligands of CD279 and are members of the B7 gene family. Interaction of CD279:PD-Ligands results in inhibition of T cell proliferation and cytokine secretion. Reports suggest that the B7/CTLA-4 pathway functions primarily to attenuate, limit, and/or terminate naïve T-cell activation in secondary lymphoid organs. The PD-ligand:CD279 pathway, on the other hand, may primarily attenuate, limit, and/or terminate T-, B-, and myeloid cell activation/effector function at sites of inflammation in the periphery.



Flow cytometric analysis of CD279 (PD-1) expression on human peripheral blood lymphocytes. *Whole blood was stained with Alexa Fluor® 647 Mouse anti-Human CD279 (PD-1) (clone EH12.1, Cat. No. 560838) and FITC Mouse Anti-Human CD3 (clone UCHT1, Cat. No. 555332). The erythrocytes were lysed with BD PharmLyse™ Lysing Buffer (Cat. No. 555899). A two-color flow cytometric dot plot showing the correlated expression patterns of CD3 versus CD279 (PD-1) was derived from gated events with the forward and side light-scatter characteristics of viable lymphocytes. Flow cytometry was performed using a BD LSR™ II flow cytometry system.*

Preparation and Storage

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.

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

Application Notes

Application

Flow cytometry	Routinely Tested

Suggested Companion Products

Catalog Number	Name	Size	Clone
555332	FITC Mouse Anti-Human CD3	100 tests	UCHT1
555899	Lysing Buffer	100 ml	(none)
557714	Alexa Fluor® 647 Mouse IgG1 κ Isotype Control	100 tests	MOPC-21
554656	Stain Buffer (FBS)	500 ml	(none)

BD Biosciences

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Product Notices

1. This reagent has been pre-diluted for use at the recommended Volume per Test. We typically use 1 × 10^6 cells in a 100-µl experimental sample (a test).
2. Please refer to www.bdbiosciences.com/pharmingen/protocols for technical protocols.
3. Since applications vary, each investigator should titrate the reagent to obtain optimal results.
4. Alexa Fluor® 647 fluorochrome emission is collected at the same instrument settings as for allophycocyanin (APC).
5. For fluorochrome spectra and suitable instrument settings, please refer to our Fluorochrome Web Page at www.bdbiosciences.com/colors.
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. 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.
8. Alexa Fluor® is a registered trademark of Molecular Probes, Inc., Eugene, OR.
9. Source of all serum proteins is from USDA inspected abattoirs located in the United States.

References

Bennett F, Luxenberg D, Ling V, et al. Program death-1 engagement upon TCR activation has distinct effects on costimulation and cytokine-driven proliferation: attenuation of ICOS, IL-4, and IL-21, but not CD28, IL-7, and IL-15 responses. *J Immunol.* 2003; 170(2):711-718. (Biology)

Carter L, Fouser LA, Jussif J, et al. PD-1:PD-L inhibitory pathway affects both CD4(+) and CD8(+) T cells and is overcome by IL-2. *Eur J Immunol.* 2002; 32:634-643. (Biology)

Dorfman DM, Brown JA, Shahsafaei A, Freeman GJ. Programmed death-1 (PD-1) is a marker of germinal center-associated T cells and angioimmunoblastic T-cell lymphoma. *Am J Surg Pathol.* 2006; 30:802-810. (Clone-specific: Immunohistochemistry)

Freeman GJ, Long AJ, Iwai Y, et al. Engagement of PD-1 immunoinhibitory receptor by a novel B7 family member leads to negative regulation of lymphocyte activation. *J Exp Med.* 2000; 192:1027-1034. (Biology)

Latchman Y, Wood CR, Chernova T, et al. PD-L2 is a second ligand for PD-1 and inhibits T cell activation. *Nat Immunol.* 2001; 2(3):261-268. (Biology)

Nishimura H, Minato N, Nakano T, Honjo T. Immunological studies on PD-1 deficient mice: implication of PD-1 as a negative regulator for B cell responses. *Int Immunol.* 1998; 10(10):1563-1572. (Biology)