Technical Data Sheet

Alexa Fluor® 647 Mouse anti-Mouse CD64 a and b Alloantigens

Product Information

Material Number: 558539 Alternate Name: FcgRI Size $0.1 \, \text{mg}$ 0.2 mg/ml Concentration: X54-5/7.1 Clone:

Immunogen: Mouse CD64 a Alloantigen Isotype: Mouse (NOD/Lt) IgG1, κ

Reactivity: Confirmed postivie strains: BALB/c, C57BL/6

> Reported positive strains: 129, A, AKR, ALR, BUB, C3H, C57BL/10, C57BLKS, C57BR, C58, CBA, CE, DBA/2, HRS, MRL, NON, NZB, NZO,

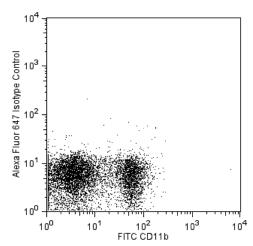
NZW, PL, SJL, ST, SWR

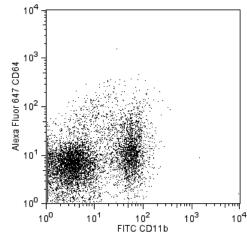
reported negative strains: ABH, NOD

Storage Buffer: Aqueous buffered solution containing protein stabilizer and ≤0.09% sodium

Description

The monoclonal alloantibody X54-5/7.1 reacts with FcyRI (CD64) encoded by the more common Fcgr1a and Fcgr1b alleles. The alloantigens generated by the Fcgr1a and Fcgr1b alleles, have been confirmed positive in mouse strains BALB/c and C57BL/6 and reported positive in strains 129, A, AKR, ALR, BUB, C3H, C57BL/10, C57BLKS, C57BR, C58, CBA, CE, DBA/2, HRS, MRL, NON, NZB, NZO, NZW, PL, SJL, ST, SWR. The a and b alloantigens have been reported negative in mouse strains ABH, NOD. CD64, a key receptor in the development of immune responses, has a dual role as a low affinity receptor for IgG3 and high affinity receptor for IgG2a linking innate and adaptive immunities. CD64 mediates endocytosis, phagocytosis, antibody-dependent cellular toxicity, cytokine release and superoxide generation. CD64 is expressed largely on macrophages and dendritic cells. For more information regarding clone X54-5/7.1 and the alloantigens it recognizes, please refer to the reference by Tan et al listed below.





Flow cytometric analysis of Alexa Fluor® 647 Anti-Mouse CD64 recognizing a and b Alloantigens on mouse bone marrow cells. Isolated murine bone marrow cells were preincubated with Mouse BD Fc Block™ purified Anti-Mouse CD16/CD32 mAb 2.4G2 (Cat. No. 553141/553142). The cells were then stained with FITC Anti-CD11b (clone M1/70, Cat. No. 553310) and either Alexa Fluor® 647 Anti-CD64 (clone X54-5/7.1, Cat. No. 558539, right panel) or a Alexa Fluor® 647 Mouse IgG1 isotype control (Cat. No. 557732, left panel). Flow cytometry was performed on a BD FACSCalibur™ System and the dot plots were derived from the gated events based on light scattering characteristics of viable bone marrow cells

Preparation and Storage

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

The antibody was conjugated to Alexa Fluor® 647 under optimum conditions, and unreacted Alexa Fluor® 647 was removed. The monoclonal antibody was purified from tissue culture supernatant or ascites by affinity chromatography.

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Application Notes

Application

Flow cytometry Routinely Tested	
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Recommended Assay Procedure:

For flow cytometry of cell suspensions from peripheral lymphoid tissues, it is recommended that the cells be pre-incubated with Mouse BD Fc BlockTM purified anti-mouse CD16/CD32 mAb 2.4G2 (Cat. No.553141/553142).

Suggested Companion Products

Catalog Number	Name	Size	Clone	
557732	Alexa Fluor® 647 Mouse IgG1 κ Isotype Control	100 tests	MOPC-21	
553141	Purified Rat Anti-Mouse CD16/CD32 (Mouse BD Fc Block TM)	0.1 mg	2.4G2	
553142	Purified Rat Anti-Mouse CD16/CD32 (Mouse BD Fc Block™)	0.5 mg	2.4G2	

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. 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.
- Alexa Fluor® 647 fluorochrome emission is collected at the same instrument settings as for allophycocyanin (APC).
- 5. 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.
- 6. Alexa Fluor® is a registered trademark of Molecular Probes, Inc., Eugene, OR.
- 7. An isotype control should be used at the same concentration as the antibody of interest.

References

Kishimoto T, von dem Borne AEG, Goyert SM,et al., ed. *Leucocyte Typing VI: White Cell Differentiation Antigens*. London: Garland Publishing; 1997. (Biology) Tan PS, Gavin AL, Barnes N, et al. Unique monoclonal antibodies define expression of Fc gamma RI on macrophages and mast cell lines and demonstrate heterogeneity among subcutaneous and other dendritic cells. *J Immunol.* 2003; 170(5):2549-2556. (Biology)

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