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Datasheet for ABIN1305233

Anti-Mouse CD45R/B220 Magnetic Particles

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Overview

Quantity:	10 mL
Target:	CD45 (PTPRC)
Reactivity:	Mouse
Host:	Rat
Clonality:	Monoclonal
Conjugate:	Magnetic Particles
Application:	Separation (Sep)

Product Details

Brand:	BD IMag™

Target Details

Target:	CD45 (PTPRC)
Alternative Name:	CD45R/B220 (PTPRC Products)
Background:	BD IMag™ anti-mouse CD45R/B220 Particles - DM are magnetic nanoparticles that have monoclonal antibody conjugated to their surfaces. These particles are optimized for the positive selection or depletion of CD45R/B220-bearing leukocytes using the BD IMagnet™. CD45R/B220 is reportedly expressed on B lymphocytes at all stages from pro-B through mature and activated B cell. It is also has been reported to be found on abnormal T cells involved in the lymphadenopathy of lpr/lpr and gld/gld mutant mice, on lytically active subsets of lymphokine-activated killer cells (NK cells and non-MHC-restricted CTL), on apoptotic T lymphocytes of mice injected with bacterial superantigen, on a population of NK-cell precursors in the bone

marrow, and on B-lymphocyte, T-lymphocyte, and macrophage progenitors in fetal liver. The CD45R/B220 antigen is reportedly not on hematopoietic stem cells, plasma cells, resting T lymphocytes, or MHC-restricted CTL. Leukocytes are labeled with BD IMag™ anti-mouse CD45R/B220 Particles - DM according to the Magnetic Labeling Protocol. This labeled cell suspension is then placed within the magnetic field of the BD IMagnet™ (Cat. No. 552311). Labeled cells migrate toward the magnet (positive fraction), leaving the unlabeled cells in suspension so they can be drawn off (negative fraction). The tube is then removed from the magnetic field for resuspension of the positive fraction. The separation is repeated twice to increase the purity of the positive fraction. The magnetic separation steps are diagrammed in the Separation Flow Chart. After the positive fraction is washed, the small size of the magnetic particles allows the positive fraction to be further evaluated in downstream applications such as flow cytometry.

Application Details

Protocol:

1. Prepare a single-cell suspension from the lymphoid tissue of interest according to standard laboratory procedures. Remove clumps of cells and/or debris by passing the suspension through a 70-µm nylon cell strainer.
2. Dilute BD IMag™ Buffer (10X) (Cat. No. 552362) 1:10 with sterile distilled water or prepare 1X BD IMag™ buffer by supplementing Phosphate Buffered Saline with 0.5% BSA, 2mM EDTA, and 0.09% sodium azide. Place on ice. Although our experience indicates that use of Mouse BD Fc Block™ purified anti-mouse CD16/CD32 mAb 2.4G2 (Cat. No. 553141) is not required for optimal cell separation, some laboratories may want to use it in their studies. If adding Mouse BD Fc Block™, proceed to Step 3. If not adding Mouse BD Fc Block™, proceed to Step 4.
3. Add Mouse BD Fc Block™ at 0.25 µg/10e6 cells, and incubate on ice for 15 minutes.
4. Wash cells with at least an equal volume of 1X BD IMag™ buffer, and carefully aspirate all the supernatant.
5. Vortex the BD IMag™ anti-mouse CD45R/B220 Particles - DM thoroughly, and add 50 µl of particles for every 10e7 total cells.
6. MIX THOROUGHLY. Refrigerate at 6°C - 12°C for 30 minutes.
7. Bring the BD IMag-particle labeling volume up to 1-8 x 10e7 cells/ml with 1X BD IMag™ buffer, and immediately place the tube on the BD IMagnet™. Incubate at room temperature for 6-8 minutes.
8. With the tube on the BD IMagnet™, carefully aspirate off the supernatant. This supernatant contains the negative fraction.
9. Remove the tube from the BD IMagnet™, and add 1X BD IMag™ buffer to the same volume as

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Application Details

	in Step 7. Gently resuspend cells by pipetting briefly, and return the tube to the BD IMagnet™ for another 2-4 minutes. 10. With the tube on the BD IMagnet™, carefully aspirate off the supernatant and discard. 11. Repeat steps 9 and 10. 12. After the final wash step, resuspend the positive fraction in an appropriate buffer and at an appropriate concentration for further analysis. NOTE: Avoid nonspecific labeling by working quickly and adhering to recommended incubation times.
Restrictions:	For Research Use only

Handling

Format:	Liquid
Buffer:	Aqueous buffered solution containing BSA and ≤0.09 % sodium azide.
Preservative:	Sodium azide
Precaution of Use:	This product contains Sodium azide: a POISONOUS AND HAZARDOUS SUBSTANCE which should be handled by trained staff only.
Storage:	4 °C
Storage Comment:	Store undiluted at 4° C. Page 1 of 3551513 Rev. 1

Publications

Product cited in:

Kobata, Takasaki, Asahara, Hong, Masuko-Hongo, Kato, Hirose, Shirai, Kayagaki, Yagita, Okumura, Nishioka: "Apoptosis with FasL+ cell infiltration in the periphery and thymus of corrected autoimmune mice." in: **Immunology**, Vol. 92, Issue 2, pp. 206-13, (1998) (PubMed).

Sagara, Sugaya, Tokoro, Tanaka, Takano, Kodama, Nakauchi, Takahama: "B220 expression by T lymphoid progenitor cells in mouse fetal liver." in: **Journal of immunology (Baltimore, Md. : 1950)**, Vol. 158, Issue 2, pp. 666-76, (1997) (PubMed).

Renno, Hahne, Tschopp, MacDonald: "Peripheral T cells undergoing superantigen-induced apoptosis in vivo express B220 and upregulate Fas and Fas ligand." in: **The Journal of experimental medicine**, Vol. 183, Issue 2, pp. 431-7, (1996) (PubMed).

Rolink, ten Boekel, Melchers, Fearon, Krop, Andersson: "A subpopulation of B220+ cells in

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murine bone marrow does not express CD19 and contains natural killer cell progenitors." in: **The Journal of experimental medicine**, Vol. 183, Issue 1, pp. 187-94, (1996) (PubMed).

Laouar, Ezine: "In vivo CD4+ lymph node T cells from lpr mice generate CD4-CD8-B220+TCR-beta low cells." in: **Journal of immunology (Baltimore, Md. : 1950)**, Vol. 153, Issue 9, pp. 3948-55, (1994) (PubMed).

There are more publications referencing this product on: Product page

Images

Flow Cytometry

Image 1.