

Technical Data Sheet

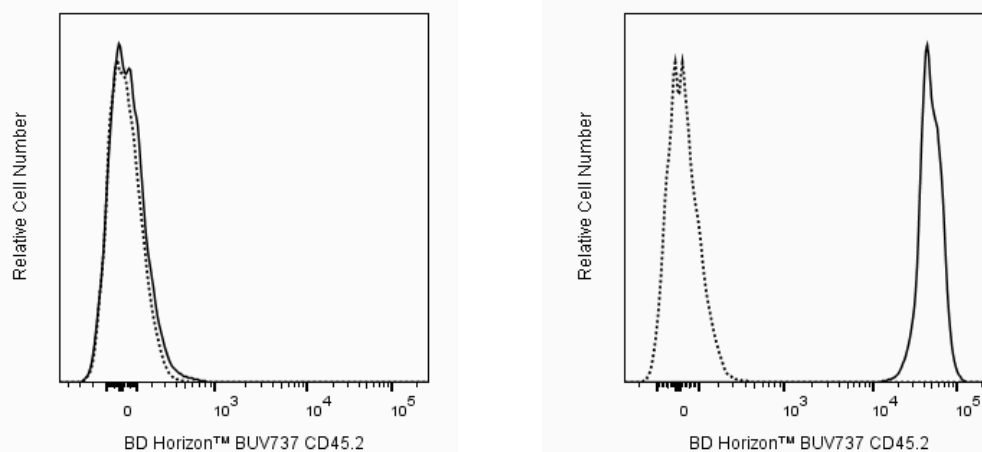
BUV737 Mouse Anti-Mouse CD45.2**Product Information**

Material Number:	612779
Alternate Name:	Ly-5.2; T200; LCA; Leukocyte common antigen; Ptprc
Size:	25 µg
Concentration:	0.2 mg/ml
Clone:	104
Immunogen:	B10.S mouse thymocytes and splenocytes
Isotype:	Mouse (SJL) IgG2a, κ
Reactivity:	QC Testing: Mouse
Storage Buffer:	Aqueous buffered solution containing ≤0.09% sodium azide.

Description

The 104 monoclonal antibody recognizes the CD45 (Leukocyte Common Antigen) present on all leucocytes of most mouse strains (eg, A, AKR, BALB/c, CBA/Ca, CBA/J, C3H/He, C57BL, C57BR, C57L, C58, DBA/1, DBA/2, NZB, SWR, 129). This alloantigen was originally named Ly-5.1, and this was the designation at the time that the antibody was characterized. The designation was later changed from Ly-5.1 to Ly-5.2 to conform with the convention that the .2 alloantigen designations be assigned to the C57BL/6 strain. mAb 104 has been reported not to react with leucocytes of the mouse strains expressing the CD45.1 alloantigen (eg, RIII, SJL/J, STS/A, and DA). CD45 is a member of the Protein Tyrosine Phosphatase (PTP) family: its intracellular (COOH-terminal) region contains two PTP catalytic domains, and the extracellular region is highly variable due to alternative splicing of exons 4, 5, and 6 (designated A, B, and C, respectively), plus differing levels of glycosylation. The CD45 isoforms detected in the mouse are cell type-, maturation-, and activation state-specific. The CD45 isoforms play complex roles in T-cell and B-cell antigen receptor signal transduction. The 104 antibody has been reported to inhibit some responses of B cells, from mice expressing the CD45.2 alloantigen, to certain antigens and LPS. In addition, reduction of serum IgG levels and amelioration of autoimmune renal pathology were reported in mAb 104-treated systemic lupus erythematosus-prone mice.

The antibody was conjugated to BD Horizon BUV737 which is part of the BD Horizon Brilliant™ Ultraviolet family of dyes. This dye is a tandem fluoro-chrome with an Ex Max near 350 nm and an Em Max near 737 nm. BD Horizon Brilliant BUV737 can be excited by the ultraviolet laser (355 nm) and detected with a 740/35 nm filter. Due to the excitation of the acceptor dye by the red laser line, there may be significant spillover into red laser detectors with filters in the 700-720 nm range.



Flow cytometric analysis of CD45.2 expression on C57BL/6 mouse splenocytes. Splenic leucocytes from an SJL mouse (Left Panel) or a C57BL/6 mouse (Right Panel) were preincubated with Purified Rat Anti-Mouse CD16/CD32 antibody (Mouse BD Fc Block™) (Cat. No. 553141/553142). The cells were then stained with either BD Horizon™ BUV737 Mouse IgG2a, κ Isotype Control (Cat. No. 612765; dashed line histograms) or BD Horizon BUV737 Mouse Anti-Mouse CD45.2 antibody (Cat. No. 612778/612779; solid line histograms) at 1 µg/test. BD Via-Probe™ Cell Viability 7-AAD Solution (Cat. No. 555815/555816) was added to cells right before analysis. The fluorescence histograms showing CD45.2 expression (or Ig Isotype control staining) were derived from gated events with the forward and side light-scatter characteristics of viable (7-AAD-negative) leucocytes. Flow cytometry and data analysis were performed using a BD LSRFortessa™ X-20 Cell Analyzer System and FlowJo™ software. Data shown on this Technical Data Sheet are not lot specific.

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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 with BD Horizon BUV737 under optimum conditions, and unconjugated antibody and free BD Horizon BUV737 were removed.

Application Notes

Application

Flow cytometry

Routinely Tested

Recommended Assay Procedure:

BD™ CompBeads can be used as surrogates to assess fluorescence spillover (Compensation). When fluorochrome conjugated antibodies are bound to BD CompBeads, they have spectral properties very similar to cells. However, for some fluorochromes there can be small differences in spectral emissions compared to cells, resulting in spillover values that differ when compared to biological controls. It is strongly recommended that when using a reagent for the first time, users compare the spillover on cells and BD CompBead to ensure that BD CompBeads are appropriate for your specific cellular application.

For optimal and reproducible results, BD Horizon Brilliant Stain Buffer should be used anytime two or more BD Horizon Brilliant dyes are used in the same experiment. Fluorescent dye interactions may cause staining artifacts which may affect data interpretation. The BD Horizon Brilliant Stain Buffer was designed to minimize these interactions. More information can be found in the Technical Data Sheet of the BD Horizon Brilliant Stain Buffer (Cat. No. 563794/566349) or the BD Horizon Brilliant Stain Buffer Plus (Cat. No. 566385).

Note: When using high concentrations of antibody, background binding of this dye to erythroid cell subsets (mature erythrocytes and precursors) has been observed. For researchers studying these cell populations, or in cases where light scatter gating does not adequately exclude these cells from the analysis, this background may be an important factor to consider when selecting reagents for panel(s).

Suggested Companion Products

Catalog Number	Name	Size	Clone
563794	Brilliant Stain Buffer	100 Tests	(none)
566349	Brilliant Stain Buffer	1000 Tests	(none)
566385	Brilliant Stain Buffer Plus	1000 Tests	(none)
612778	BUV737 Mouse Anti-Mouse CD45.2	50 µg	104
612765	BUV737 Mouse IgG2a, κ Isotype Control	50 µg	G155-178
553141	Purified Rat Anti-Mouse CD16/CD32 (Mouse BD Fc Block™)	0.1 mg	2.4G2
553142	Purified Rat Anti-Mouse CD16/CD32 (Mouse BD Fc Block™)	0.5 mg	2.4G2
555815	Cell Viability Solution	500 Tests	(none)
555816	Cell Viability Solution	100 Tests	(none)
555899	Lysing Buffer	100 mL	(none)
554656	Stain Buffer (FBS)	500 mL	(none)
554657	Stain Buffer (BSA)	500 mL	(none)

Product Notices

1. Since applications vary, each investigator should titrate the reagent to obtain optimal results.
2. An isotype control should be used at the same concentration as the antibody of interest.
3. 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.
4. For fluorochrome spectra and suitable instrument settings, please refer to our Multicolor Flow Cytometry web page at www.bdbiosciences.com/colors.
5. BD Horizon Brilliant Ultraviolet 737 is covered by one or more of the following US patents: 8,110,673; 8,158,444; 8,227,187; 8,575,303; 8,354,239.
6. BD Horizon Brilliant Stain Buffer is covered by one or more of the following US patents: 8,110,673; 8,158,444; 8,575,303; 8,354,239.
7. Please refer to www.bdbiosciences.com/pharming/protocols for technical protocols.

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