Technical Data Sheet

**BV786 Mouse Anti-Human CD152**

**Product Information**

- **Material Number:** 563931
- **Alternate Name:** CTLA-4; AILIM; Cytotoxic T-lymphocyte protein 4
- **Size:** 50 Tests
- **Vol. per Test:** 5 µl
- **Clone:** BNI3
- **Immunogen:** Human CTLA4 Recombinant Protein
- **Isotype:** Mouse (BALB/c) IgG2a, κ
- **Reactivity:** QC Testing: Human
  Tested in Development: Rhesus, Cynomolgus, Baboon
- **Storage Buffer:** Aqueous buffered solution containing BSA and ≤0.09% sodium azide.

**Description**

The BNI3 monoclonal antibody specifically binds to the human cytolytic T lymphocyte-associated antigen (CTLA-4), also known as CD152. CTLA-4 is transiently expressed on activated CD28+ T cells and binds to CD80 and CD86 present on antigen presenting cells (APC) with high avidity. This interaction appears to deliver a negative regulatory signal to the T cell. Recent reports indicate that CTLA-4 is also expressed on B cells when cultured with activated T cells, suggesting a role for CTLA-4 in the regulation of B-cell response. Immobilized BNI3 antibody enhances T-cell proliferation induced by antibody-mediated crosslinking of CD3 and CD28. Recent studies have shown that CD152 can be expressed by regulatory T (Treg) cells. After cellular fixation and permeabilization, the BNI3 antibody can stain intracellular CD152 expressed in T cells including Treg cells. Clone BNI3 was studied in the VI Leukocyte Typing Workshop.

Clone BNI3 also cross-reacts with a subset of peripheral blood lymphocytes of baboon, and both rhesus and cynomolgus macaque monkeys, following Concanavalin A (Con A) treatment. The distribution of BNI3+ cells following activation is similar to that observed with peripheral blood lymphocytes from normal human donors.

The antibody was conjugated to BD Horizon BV786 which is part of the BD Horizon Brilliant™ Violet family of dyes. This dye is a tandem fluorochrome of BD Horizon BV421 with an Ex Max of 405-nm and an acceptor dye with an Em Max at 786-nm. BD Horizon BV786 can be excited by the violet laser and detected in a filter used to detect Cy™7-like dyes (e.g., 780/60-nm filter).

**Flow cytometric analysis of CD152 expression on activated human peripheral blood mononuclear cells.**

Human peripheral blood mononuclear cells were stimulated with Concanavalin A (Sigma C-5275) for 3 days and then stained with either BD Horizon™ BV786 Mouse IgG2a, κ Isotype Control (Cat. No. 563732; dashed line histogram) or BD Horizon™ BV786 Mouse Anti-Human CD152 antibody (Cat. No. 563931; solid line histogram). The fluorescence histograms were derived from gated events with the forward and side light-scatter characteristics of viable activated cells. Flow cytometric analysis was performed using a BD™ LSR II Flow Cytometer System.
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™ BV786 under optimum conditions, and unconjugated antibody and free BD Horizon™ BV786 were removed.

Application Notes

Application

| Flow cytometry | Routinely Tested |

Recommended Assay Procedure:
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).

Suggested Companion Products

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<th>Name</th>
<th>Size</th>
<th>Clone</th>
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<tbody>
<tr>
<td>554656</td>
<td>Stain Buffer (FBS)</td>
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<tr>
<td>554657</td>
<td>Stain Buffer (BSA)</td>
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<td>563732</td>
<td>BV786 Mouse IgG2a, κ Isotype Control</td>
<td>50 µg</td>
<td>G155-178</td>
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<tr>
<td>563794</td>
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<td>566349</td>
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<tr>
<td>566385</td>
<td>Brilliant Stain Buffer Plus</td>
<td>1000 Tests</td>
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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 × 10⁶ cells in a 100-µl experimental sample (a test).
2. An isotype control should be used at the same concentration as the antibody of interest.
3. Source of all serum proteins is from USDA inspected abattoirs located in the United States.
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. For fluorochrome spectra and suitable instrument settings, please refer to our Multicolor Flow Cytometry web page at www.bdbiosciences.com/colors.
6. Cy is a trademark of GE Healthcare.
7. BD Horizon Brilliant Violet 786 is covered by one or more of the following US patents: 8,110,673; 8,158,444; 8,227,187; 8,455,613; 8,575,303; 8,354,239.
8. 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.
9. Species testing during development may have been performed with a different format of the same clone. Selected applications have been tested for cross-reactivity.

References


Morton PA, Fu XT, Stewart JA, et al. Differential effects of CTLA-4 substitutions on the binding of human CD80 (B7-1) and CD86 (B7-2). J Immunol. 1996; 156(3):1047-1054. (Biology)