# **Technical Data Sheet**

# **BV421 Mouse Anti-Human IFN-y**

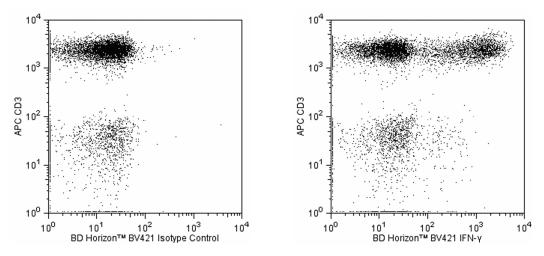
## **Product Information**

| Material Number: | 562988                                                                    |
|------------------|---------------------------------------------------------------------------|
| Alternate Name:  | IFNG; Interferon-gamma; Interferon-γ; Type II interferon; MAF             |
| Size:            | 50 tests                                                                  |
| Vol. per Test:   | 5 μl                                                                      |
| Clone:           | B27                                                                       |
| Immunogen:       | Human IFN-y Recombinant Protein                                           |
| Isotype:         | Mouse IgG1, ĸ                                                             |
| Reactivity:      | QC Testing: Human                                                         |
| Storage Buffer:  | Aqueous buffered solution containing BSA and ${\leq}0.09\%$ sodium azide. |

#### Description

The B27 monoclonal antibody specifically binds to human interferon-y (IFN-y). This is a neutralizing antibody. The use of B27 antibody for epitope mapping of human IFN-y has been described. The B27 antibody has been reported not to bind to denatured IFN-y.

The antibody was conjugated to BD Horizon<sup>TM</sup> BV421 which is part of the BD Horizon<sup>TM</sup> Brilliant Violet<sup>TM</sup> family of dyes. With an Ex Max of 407-nm and Em Max at 421-nm, BD Horizon<sup>TM</sup> BV421 can be excited by the violet laser and detected in the standard Pacific Blue<sup>TM</sup> filter set (eg, 450/50-nm filter). BD Horizon™ BV421 conjugates are very bright, often exhibiting a 10 fold improvement in brightness compared to Pacific Blue<sup>™</sup> conjugates.



Multicolor flow cytometric analysis of IFN-y expressed in stimulated human peripheral blood mononuclear cells. HiCK-1 Human Cytokine Positive Control Cells (Cat. No. 555061) were permeabilized with BD Perm/Wash™ Buffer (Cat. No. 554723). The cells were then stained with APC Mouse Anti-Human CD3 antibody (Cat. No. 561811/555335/561810) and with either BD Horizon™ BV421 Mouse IqG1, κ Isotype Control (Cat No. 562438, Left Panel) or with the BD Horizon™ BV421 Mouse Anti-Human IFN-γ antibody (Cat No. 562988, Right Panel). Two-color flow cytometric dot plots showing the expression of IFN-y (or Ig Isotype Control staining) versus CD3 were derived from gated events with the forward and side light-scatter characteristics of intact lymphocytes. Flow cytometry was performed using a BD LSR™ II Flow Cytometry 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™ BV421 under optimum conditions, and unconjugated antibody and free BD Horizon™ BV421 were removed.

#### **Application Notes**

#### Application

| Intracellular staining (flow cytometry) Routinely |        |               |                 |              | Routinely Test                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                 | ed  |
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| 877.232.8995                                      |        | 32.53.720.550 |                 | 65.6861.0633 | 55.11.5185.9995                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                |     |
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### Suggested Companion Products

| Catalog Number | Name                                         | Size      | Clone  |  |
|----------------|----------------------------------------------|-----------|--------|--|
| 554656         | Stain Buffer (FBS)                           | 500 ml    | (none) |  |
| 562438         | BV421 Mouse IgG1, k Isotype Control          | 50 µg     | X40    |  |
| 554723         | Perm/Wash Buffer                             | 100 ml    | (none) |  |
| 555061         | HiCK-1 Human Cytokine Positive Control Cells | 1.0 ml    | (none) |  |
| 561811         | APC Mouse Anti-Human CD3                     | 500 tests | UCHT1  |  |
| 555335         | APC Mouse Anti-Human CD3                     | 100 tests | UCHT1  |  |
| 561810         | APC Mouse Anti-Human CD3                     | 25 tests  | UCHT1  |  |

#### **Product Notices**

- This reagent has been pre-diluted for use at the recommended Volume per Test. We typically use  $1 \times 10^{6}$  cells in a 100-µl experimental 1. sample (a test).
- Source of all serum proteins is from USDA inspected abattoirs located in the United States. 2.
- 3. An isotype control should be used at the same concentration as the antibody of interest.
- Please refer to www.bdbiosciences.com/pharmingen/protocols for technical protocols. 4.
- Caution: Sodium azide yields highly toxic hydrazoic acid under acidic conditions. Dilute azide compounds in running water before 5. discarding to avoid accumulation of potentially explosive deposits in plumbing.
- For fluorochrome spectra and suitable instrument settings, please refer to our Multicolor Flow Cytometry web page at 6. www.bdbiosciences.com/colors.
- 7. Pacific Blue<sup>™</sup> is a trademark of Molecular Probes, Inc., Eugene, OR.
- 8 Brilliant Violet<sup>™</sup> 421 is a trademark of Sirigen.

#### References

Abrams JS, Roncarolo MG, Yssel H, Andersson U, Gleich GJ, Silver JE. Strategies of anti-cytokine monoclonal antibody development: immunoassay of IL-10 and IL-5 in clinical samples. Immunol Rev. 1992; 127:5-24. (Clone-specific: ELISA, Neutralization)

Favre C, Wijdenes J, Cabrillat H, Djossou O, Banchereau J, de Vries JE. Epitope mapping of recombinant human gamma interferon using monoclonal antibodies. Mol Immunol. 1989; 26(1):17-25. (Immunogen: Flow cytometry, Immunoprecipitation, Neutralization)

Prussin C, Metcalfe DD. Detection of intracytoplasmic cytokine using flow cytometry and directly conjugated anti-cytokine antibodies. J Immunol Methods. 1995; 188(1):117-128. (Methodology: Flow cytometry, IC/FCM Block)

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