## **Technical Data Sheet**

# **BV421 Mouse Anti-Human CD66b**

#### **Product Information**

Material Number: 562940

Alternate Name: CEACAM8; CGM6; NCA-95

 Size:
 100 tests

 Vol. per Test:
 5 μl

 Clone:
 G10F5

 Immunogen:
 Human Granulocytes

 Isotype:
 Mouse (BALB/c) IgM, κ

 Reactivity:
 QC Testing: Human

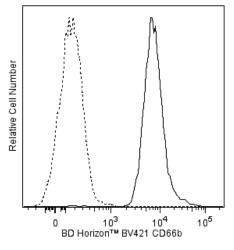
 Workshop:
 V 5T-127, MA020

Storage Buffer: Aqueous buffered solution containing BSA and ≤0.09% sodium azide.

### Description

The G10F5 monoclonal antibody specifically binds to CD66b, also known as Carcinoembryonic antigen-related cell adhesion molecule 8 (CEACAM8). CD66b is a glycosylphosphatidylinositol (GPI) linked protein with a molecular weight of 100 kDa expressed on granulocytes. This molecule was previously clustered as CD67 in the Fourth Human Leucocyte Differentiation Antigen (HLDA) Workshop and renamed CD66b in the Fifth HLDA Workshop. CD66b is a member of the carcinoembryonic antigen (CEA)-like glycoprotein family present on granulocytes and referred to as non-specific crossreacting antigens (NCA). Granulocyte activation induced with soluble stimulators (calcium ionophore, phorbol myristate acetate, N-formylmethionyl- leucyl-phenylalanine) results in release and increased expression of NCA. Findings suggest that these molecules may play a role in phagocytosis, chemotaxis and adherence.

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<sup>TM</sup> BV421 conjugates are very bright, often exhibiting a 10 fold improvement in brightness compared to Pacific Blue<sup>TM</sup> conjugates.



Flow cytometric analysis of CD66b expression on human peripheral blood granulocytes. Whole blood was stained with either BD Horizon™ BV421 Mouse Anti-Human CD66b antibody (Cat. No. 562940; solid line histogram) or with BD Horizon™ BV421 Mouse IgM, κ Isotype Control (Cat. No. 562704; dashed line histogram). The erythrocytes were lysed with BD Pharm Lyse™ Lysing Buffer (Cat. No. 555899). The fluorescence histograms were derived from events with the forward and side light-scatter characteristics of viable granulocytes. Flow cytometry 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<sup>TM</sup> BV421 under optimum conditions, and unconjugated antibody and free BD Horizon<sup>TM</sup> BV421 were removed.

### **Application Notes**

Application

Flow cytometry Routinely Tested

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#### **Suggested Companion Products**

Catalog Number	Name	Size	Clone
554656	Stain Buffer (FBS)	500 ml	(none)
562704	BV421 Mouse IgM, κ Isotype Control	50 μg	G155-228
555899	Lysing Buffer	100 ml	(none)

#### **Product Notices**

- This reagent has been pre-diluted for use at the recommended Volume per Test. We typically use 1 × 10<sup>6</sup> cells in a 100-µl experimental sample (a test).
- 2. Source of all serum proteins is from USDA inspected abattoirs located in the United States.
- 3. An isotype control should be used at the same concentration as the antibody of interest.
- 4. Please refer to www.bdbiosciences.com/pharmingen/protocols for technical protocols.
- 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.
- For fluorochrome spectra and suitable instrument settings, please refer to our Multicolor Flow Cytometry web page at www.bdbiosciences.com/colors.
- 7. Pacific Blue™ is a trademark of Molecular Probes, Inc., Eugene, OR.
- 8. Brilliant Violet™ 421 is a trademark of Sirigen.

#### References

Hemler ME, Kassner P, Bodorova J. CD66 and CD67 cluster workshop report. In: Schlossman SF, Boumsell L, Gilks W, et al, ed. Leukocyte Typing V: White Cell Differentiation Antigens. New York: Oxford University Press; 1995:889-899. (Clone-specific: Immunohistochemistry, Immunoprecipitation)

Knapp W, Dörken B, Gilks WR, et al, ed. *Leucocyte Typing IV*. New York, NY: Oxford University Press; 1989:1-1182. (Clone-specific)
Kuijpers TW, van der Schoot CE, Hoogerwerf M, Roos D. Cross-linking of the carcinoembryonic antigen-like glycoproteins CD66 and CD67 induces neutrophil

aggregation. *J Immunol*. 1993; 151(9):4934-4940. (Biology)
Kuroki M, Matsuo Y, Kinugasa T, Matsuoka Y. Augmented expression and release of nonspecific cross-reacting antigens (NCAs), members of the CEA family, by human neutrophils during cell activation. *J Leukoc Biol*. 1992; 52(5):551-557. (Biology)

Lund-Johansen F, Olweus J, Horejsi V, et al. Activation of human phagocytes through carbohydrate antigens (CD15, sialyl-CD15, CDw17, and CDw65). *J Immunol.* 1992; 148(10):3221-3229. (Clone-specific: Blocking, Flow cytometry)

Schlossman S, Boumell L, et al, ed. Leucocyte Typing V. New York: Oxford University Press; 1995. (Clone-specific)

Thompson JS, Brown SA, Rhoades JL, Burch J, Oberle EM. G10F5 (Workshop no. 310) reacts with a Pronase resistant epitope whose tissue distribution differs from CD15 monoclonal antibodies. In: McMichael AJ, ed. *Leucocyte Typing III: White Cell Differentiation Antigens*. Oxford: Oxford University Press; 1987:713-714. (Immunogen)

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