# biotechne

# Human ABCG2 Antibody

Monoclonal Mouse IgG2B Clone # 5D3 Catalog Number: MAB995

**R**Dsystems

DESCRIPTION	
Species Reactivity	Human
Specificity	Detects human ABCG2 in flow cytometry and immunocytochemistry.
Source	Monoclonal Mouse IgG <sub>2B</sub> Clone # 5D3
Purification	Protein A or G purified from hybridoma culture supernatant
Immunogen	3T3 cells transduced with human ABCG2
Formulation	Lyophilized from a 0.2 µm filtered solution in PBS with Trehalose.

# APPLICATIONS

Please Note: Optimal dilutions should be determined by each laboratory for each application. General Protocols are available in the Technical Information section on our website.				
	Recommended Concentration	Sample		
Flow Cytometry	0.25 µg/10 <sup>6</sup> cells	Human ABCG2 transfected CHO-S cells; RPMI 8226 human multiple myeloma cell line		
Immunocytochemistry	8-25 μg/mL	See Below		
CyTOF-ready	Ready to be labeled using established co conjugation.	Ready to be labeled using established conjugation methods. No BSA or other carrier proteins that could interfere with conjugation.		

# DATA

## Immunocytochemistry



ABCG2 in JAR Human Cell Line. ATP-Binding Cassette Transporter G2 (ABCG2) was detected in immersion fixed JAR human choriocarcinoma cell line using Mouse Anti-Human ABCG2 Monoclonal Antibody (Catalog # MAB995) at 10 µg/mL for 3 hours at room temperature. Cells were stained using the NorthernLights™ 493-conjugated Anti-Mouse IgG Secondary Antibody (green; Catalog # NL009) and counterstained with DAPI (blue). View our protocol for Fluorescent ICC Staining of Cells on Coverslips.

### Immunocytochemistry



ABCG2 in Human ABCG2 transfected CHO Chinese Hamster Cell Line. ABCG2 was detected in immersion fixed Human ABCG2 transfected CHO Chinese hamster ovary cell line using Mouse Anti-Human ABCG2 Monoclonal Antibody (Catalog # MAB995) at 10 µg/mL for 3 hours at room temperature. Cells were stained using the NorthernLights<sup>™</sup> 557-conjugated Anti-Mouse IgG Secondary Antibody (red; Catalog # NL007) and counterstained with DAPI (blue). View our protocol for Fluorescent ICC Staining of Cells on Coverslips.



8226 cells by Flow Cytometry RPMI 8226 cells were stained with Mouse Anti-Human ABCG2 Monoclonal Antibody (Catalog # MAB995, filled histogram) or isotype control antibody (Catalog # MAB0041, open histogram)

Detection of ABCG2 in RPMI

followed by Phycoerythrinconjugated Anti-Mouse IgG Secondary Antibody (Catalog # F0102B). View our protocol for Staining Membrane-associated

Proteins.

Immunocytochemistry



#### ABCG2 in A549 Human Cell Line. ABCG2 was detected in

immersion fixed A549 human lung carcinoma cell line using Mouse Anti-Human ABCG2 Monoclonal Antibody (Catalog # MAB995) at 8 µg/mL for 3 hours at room temperature. Cells were stained using the NorthernLights™ 557conjugated Anti-Mouse IgG Secondary Antibody (red; Catalog # NL007) and counterstained with DAPI (blue). Specific staining was localized to plasma membrane. View our protocol for Fluorescent ICC Staining of Non-adherent Cells.

PREPARATION AND STORAGE		
Reconstitution	Reconstitute at 0.5 mg/mL in sterile PBS.	
Shipping	The product is shipped at ambient temperature. Upon receipt, store it immediately at the temperature recommended below.	
Stability & Storage	<ul> <li>Use a manual defrost freezer and avoid repeated freeze-thaw cycles.</li> <li>12 months from date of receipt, -20 to -70 °C as supplied.</li> <li>1 month, 2 to 8 °C under sterile conditions after reconstitution.</li> <li>6 months, -20 to -70 °C under sterile conditions after reconstitution.</li> </ul>	

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# BACKGROUND

**R**Dsystems

Hematopoietic stem cells are known to express a membrane transporter molecule, known as P-glycoprotein (Pgp), that is encoded by the multidrug resistance gene 1 (MDR1) (1, 2). Expression of Pgp appears to confer a proliferative advantage to stem cells through its anti-apoptotic effects (3, 4). An additional transporter molecule known as ABCG2 (ATP-binding cassette gene 2) or Bcrp1 (Breast cancer resistance protein 1), first identified in a breast cancer cell line (5), is expressed on stem cells (6). ABCG2 belongs to a family of molecules that span the cell membrane six times and can exist as either homo or hetero dimers linked by a short intracellular flexible linker region that plays an important role in the efflux of a wide range of substrates (7, 8). Although these transporter molecules have initially been thought to play a role in drug resistance, they have been found to have utility in better characterizing primitive stem cells. For example, the "side-population" of hematopoietic stem cells, characterized by their inability to retain high levels of the intracellular staining dyes Hoechst 33342 and Rhodamine 123, has been found to express high levels of ABCG2. Of interest is the observation that ABCG2 function has been linked to the efflux of the Hoechst dye (6). Furthermore, there is now evidence that this monoclonal can be used as a cell surface marker to identify hematopoietic stem cells within the bone marrow fraction of lineage negative cells (6). The expression of ABCG2 appears greatest on CD34<sup>-</sup> cells and is downregulated with the acquisition of CD34 on the cell surface (6).

### References:

- 1. Chaudhary, P.M. and I.B. Roninson (1991) Cell 66:85.
- 2. Sorrentino, B.P. et al. (1995) Blood 86:491.
- 3. Pallis, M. and N. Russell (2000) Blood 95:2897.
- 4. Johnstone, R.W. et al. (1999) Blood 93:1075.
- 5. Doyle, L.A. et al. (1998) Proc. Natl. Acad. Sci. USA 95:15665.
- 6. Zhou, S. et al. (2001) Nat. Medicine 7:1028.
- 7. Hrycyna, C.A. et al. (1998) Biochem. 37:13660.
- 8. Bunting, K.D. (2002) Stem Cells 20:11.