Technical Data Sheet

BV605 Mouse Anti-Human TRA-1-60 Antigen

Product Information

Material Number: 563187 Alternate Name: TRA-1-60(R) 50 Tests Size Vol. per Test: 5 μl TRA-1-60 Clone:

Human Embryonal Carcinoma Cell Line Immunogen:

Isotype: Mouse (BALB/c) IgM, κ Reactivity: QC Testing: Human

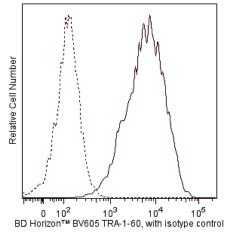
Storage Buffer: Aqueous buffered solution containing BSA and ≤0.09% sodium azide.

Description

The TRA-1-60 monoclonal antibody reacts with the neuraminidase-resistant form of a pluripotent-stem-cell-specific epitope on a high-molecular-weight transmembrane glycoprotein. The TRA-1-60 antigen is a sialylated epitope on the same keratan sulfate core molecule, podocalyxin, as 4 other distinct antigens on tumor-derived cell lines, TRA-1-81, GCTM2, K4, and K21. The expression of TRA-1-60 antigen is stage-specific and can be used to characterize embryonic cells and monitor their differentiation. The antigen is found on teratocarcinoma (embryonal carcinoma or EC), embryonic inner cell mass (but not morula or trophoblast), and embryonic stem (ES) cells. TRA-1-60 antigen is released into the serum of patients bearing testicular tumors containing EC cells. As human EC and ES cells undergo differentiation, expression of TRA-1-60 antigen is lost. Expression of TRA-1-60 antigen has also been observed on a rhesus monkey ES cell line (Thomson et al, 1995).

This antibody is conjugated to BD Horizon BV605 which is part of the BD Horizon BrilliantTM Violet family of dyes. With an Ex Max of 407-nm and Em Max of 602-nm, BD Horizon BV605 can be excited by a violet laser and detected with a standard 610/20-nm filter set. BD Horizon BV605 is a tandem fluorochrome of BD Horizon BV421 and an acceptor dye with an Em max at 605-nm. Due to the excitation of the acceptor dye by the green (532 nm) and yellow-green (561 nm) lasers, there will be significant spillover into the PE and BD Horizon PE-CF594 detectors off the green or yellow-green lasers. BD Horizon BV605 conjugates are very bright, often exhibiting brightness equivalent to PE conjugates and can be used as a third color off of the violet laser.

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).



Flow cytometric analysis of TRA-1-60 expression of human embryonic stem (ES) cells. H9 human ES cells (WiCell, Madison, WI) passage 48 grown in mTESR™1 media (StemCell Technologies) were harvested with Accutase™ (Cat. No. 561527) and stained with either BD Horizon™ BV605 Mouse anti-TRA-1-60 (Cat. No. 563187; solid line) or BD Horizon™ BV605 Mouse IgM, κ isotype control (Cat. No. 563517; dashed line). Flow cytometry was performed on a BD LSR Fortessa™ 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™ BV605 under optimum conditions, and unconjugated antibody and free BD Horizon™ BV605 were removed.

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Application Notes

Application

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Flow cytometry	Routinely Tested

Suggested Companion Products

Catalog Number	Name	Size	Clone	
561527	Accutase TM Cell Detachment Solution	100 mL	(none)	
563517	BV605 Mouse IgM, κ Isotype Control	50 μg	G155-228	
554656	Stain Buffer (FBS)	500 mL	(none)	
563794	Brilliant Stain Buffer	5 mL	(none)	

Product Notices

- 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
- Source of all serum proteins is from USDA inspected abattoirs located in the United States.
- 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.
- 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.
- 6. For fluorochrome spectra and suitable instrument settings, please refer to our Multicolor Flow Cytometry web page at www.bdbiosciences.com/colors.
- Please observe the following precautions: Absorption of visible light can significantly alter the energy transfer occurring in any tandem fluorochrome conjugate; therefore, we recommend that special precautions be taken (such as wrapping vials, tubes, or racks in aluminum foil) to prevent exposure of conjugated reagents, including cells stained with those reagents, to room illumination.
- Although every effort is made to minimize the lot-to-lot variation in the efficiency of the fluorochrome energy transfer, differences in the residual emission from BD HorizonTM BV421 may be observed. Therefore, we recommend that individual compensation controls be performed for every BD Horizon™ BV605 conjugate.
- mTESRTM1 is a trademark of StemCell Technologies.
- Accutase is a registered trademark of Innovative Cell Technologies, Inc.
- CFTM is a trademark of Biotium, Inc.

References

Andrews PW, Banting G, Damanov I, Arnaud D, Avner P. Three monoclonal antibodies defining distinct differentiation antigens associated with different high molecular weight polypeptides on the surface of human embryonal carcinoma cells. Hybridoma. 1984; 3(4):347-361. (Immunogen)

Badcock G, Pigott C, Goepel J, Andrews PW. The human embryonal carcinoma marker antigen TRA-1-60 is a sialylated keratan sulfate proteoglycan. Cancer Res. 1999; 59:4715-4719. (Clone-specific)

Draper JS, Pigott C, Thomson JA, Andrews PW. Surface antigens of human embryonic stem cells: changes upon differentiation in culture. J Anat. 2002; 200:249-258. (Clone-specific)

Henderson JK, Draper JS, Baillie HS, et al. Preimplantation human embryos and embryonic stem cells show comparable expression of stage-specific embryonic antigens. Stem Cells. 2002; 20:329-337. (Clone-specific)

Schopperle WM, DeWolf WC. The TRA-1-60 and TRA-1-81 human pluripotent stem cell markers are expressed on podocalyxin in embryonal carcinoma. Stem Cells, 2007; 25:723-730, (Clone-specific)

Thomson JA, Itskovitz-Eldor J, Shapiro SS, et al. Embryonic stem cell lines derived from human blastocysts. Science, 1998; 282;1145-1147. (Clone-specific) Thomson JA, Kalishman J, Golos TG, et al. Isolation of a primate embryonic stem cell line. Proc Natl Acad Sci U S A. 1995; 92:7844-7848. (Clone-specific)

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