Technical Data Sheet

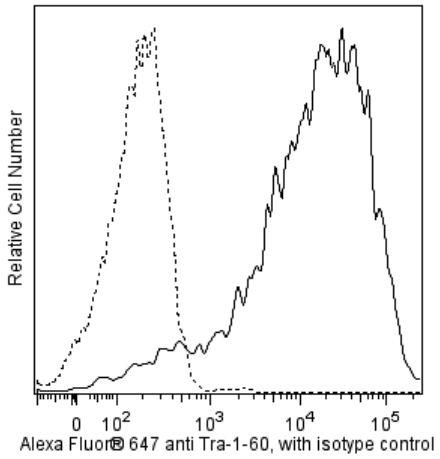
Alexa Fluor® 647 Mouse anti-Human TRA-1-60 Antigen

Product Information

Material Number:	**560850**
Alternate Name:	TRA-1-60(R)
Size:	100 tests
Vol. per Test:	5 µl
Clone:	TRA-1-60
Immunogen:	Human Embryonal Carcinoma Cell Line
Isotype:	Mouse (BALB/c) IgM, κ
Reactivity:	QC Testing: Human
Storage Buffer:	Aqueous buffered solution containing BSA, protein stabilizer, 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).



Flow cytometric analysis of human ES cell line. *H9 human embryonic stem (ES) cells (WiCell, Madison, WI) were stained with either Alexa Fluor® 647 Mouse Anti-Human TRA-1-60 (solid line) or Alexa Fluor® 647 Mouse IgM, κ Isotype Control (Catalog No. 560806, dashed line), incubated in the dark for 20 minutes at room temperature and analyzed by flow cytometry on a BD™ LSR II 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 to Alexa Fluor® 647 under optimum conditions, and unreacted Alexa Fluor® 647 was removed.

Application Notes

Application

Flow cytometry	Routinely Tested

Recommended Assay Procedure:

Bioimaging: MN 560850 has been optimized for flow cytometry. For Bioimaging, investigators are encouraged to use MN 560122.

Suggested Companion Products

Catalog Number	Name	Size	Clone
554656	Stain Buffer (FBS)	500 ml	(none)
560806	Alexa Fluor® 647 Mouse IgM, κ Isotype Control	0.1 mg	G155-228
560122	Alexa Fluor® 647 Mouse anti-Human TRA-1-60 Antigen	100 tests	TRA-1-60

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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 × 10^6 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. Alexa Fluor® 647 fluorochrome emission is collected at the same instrument settings as for allophycocyanin (APC).
4. Alexa Fluor® is a registered trademark of Molecular Probes, Inc., Eugene, OR.
5. The Alexa Fluor®, Pacific Blue™, and Cascade Blue® dye antibody conjugates in this product are sold under license from Molecular Probes, Inc. for research use only, excluding use in combination with microarrays, or as analyte specific reagents. The Alexa Fluor® dyes (except for Alexa Fluor® 430), Pacific Blue™ dye, and Cascade Blue® dye are covered by pending and issued patents.
6. 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.
7. Source of all serum proteins is from USDA inspected abattoirs located in the United States.
8. For fluorochrome spectra and suitable instrument settings, please refer to our Multicolor Flow Cytometry web page at www.bdbiosciences.com/colors.
9. Please refer to www.bdbiosciences.com/pharmingen/protocols for technical protocols.

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: Immunofluorescence, Immunoprecipitation, Radioimmunoassay)

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: Immunoprecipitation, Western blot)

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: Flow cytometry)

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: Flow cytometry, Immunofluorescence)

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: Flow cytometry)

Thomson JA, Itskovitz-Eldor J, Shapiro SS, et al. Embryonic stem cell lines derived from human blastocysts. *Science.* 1998; 282:1145-1147. (Clone-specific: Immunocytochemistry (cytospins))

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: Immunocytochemistry (cytospins))

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