Technical Data Sheet

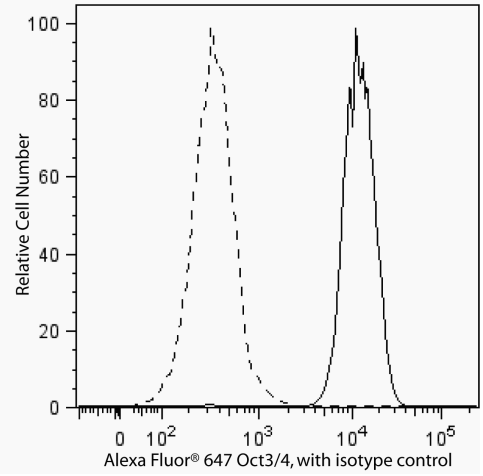
Alexa Fluor® 647 Mouse anti-Oct3/4

Product Information

Material Number:	**560329**
Alternate Name:	Oct3, OTF3, Oct4, OTF4, POU5F1
Size:	50 tests
Vol. per Test:	20 µl
Clone:	40/Oct-3
Immunogen:	Mouse Oct3 aa. 252-372 Recombinant Protein
Isotype:	Mouse IgG1, κ
Reactivity:	QC tested: Human Tested in Development: Mouse
Storage Buffer:	Aqueous buffered solution containing BSA, protein stabilizer, and ≤0.09% sodium azide.

Description

Development of a multicellular organism from a single fertilized cell is regulated by the coordinated activity of DNA transcription factors. Oct3/4, a member of the POU family of transcription factors, functions in pluripotent cells of early embryonic stem cell (ES) lines and embryonal carcinomas (EC). Other members of the POU family include Oct1, Oct2, Pit-1, and unc-86. The POU domain, a 150-amino acid region that determines binding specificity, is conserved among these proteins and consists of 3 subdomains: POU-specific A and B subdomains and a homeobox-like subdomain. Oct3/4 is expressed in undifferentiated cells, but is lost as cells are induced to differentiate. Oct3/4 is not expressed in adult tissues. The interaction of Oct3/4 with SOX2, another embryonic transcription factor, produces an active complex that regulates expression of genes such as Nanog, UTF1, and FGF4. Although Oct3/4 is specifically phosphorylated on serine residues, this modification is not required for DNA binding, but may affect its transactivation potential. Thus, Oct3/4 is a transcription factor that plays an important role in determining early steps of embryogenesis and differentiation.



Flow cytometric analysis of Alexa Fluor® 647 anti-Oct3/4 in H9 cells. *H9 human embryonic stem (ES) cells (WiCell, Madison, WI) were grown on BD Matrigel™ hESC-qualified matrix (Cat. No. 354277). Cells were dissociated into a single cell suspension and fixed (BD Cytofix™ buffer, Cat. No. 554655), permeabilized with BD Phosflow™ Perm/Wash Buffer I (Cat. No. 557885), and then stained with either Alexa Fluor® 647 Mouse anti-Oct3/4 (solid line) or Alexa Fluor® 647 Mouse IgG1, κ Isotype Control (Cat. No. 557783, dashed line). Flow cytometry was performed on a BD FACSCanto™ II flow cytometry system. This antibody also works in BD™ Phosflow Perm Buffer II and III.*

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

Intracellular staining (flow cytometry)	Routinely Tested

BD Biosciences

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877.232.8995	888.259.0187	32.53.720.550	0120.8555.90	65.6861.0633	55.11.5185.9995

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Suggested Companion Products

Catalog Number	Name	Size	Clone
554655	Fixation Buffer	100 ml	(none)
557885	Perm/Wash Buffer I	125 ml	(none)
557783	Alexa Fluor® 647 Mouse IgG1 κ Isotype control	50 tests	MOPC-21

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. Please refer to www.bdbiosciences.com/pharmingen/protocols for technical protocols.
3. For fluorochrome spectra and suitable instrument settings, please refer to our Fluorochrome Web Page at www.bdbiosciences.com/colors.
4. An isotype control should be used at the same concentration as the antibody of interest.
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. Alexa Fluor® 647 fluorochrome emission is collected at the same instrument settings as for allophycocyanin (APC).
7. 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.
8. Source of all serum proteins is from USDA inspected abattoirs located in the United States.
9. Alexa Fluor® is a registered trademark of Molecular Probes, Inc., Eugene, OR.

References

Nishimoto M, Fukushima A, Okuda A, Muramatsu M. The gene for the embryonic stem cell coactivator UTF1 carries a regulatory element which selectively interacts with a complex composed of Oct-3/4 and Sox-2. *Mol Cell Biol.* 1999; 19(8):5453-5465. (Biology)

Okamoto K, Okazawa H, Okuda A, Sakai M, Muramatsu M, Hamada H. A novel octamer binding transcription factor is differentially expressed in mouse embryonic cells. *Cell.* 1990; 60(3):461-472. (Biology)

Pan G, Thomson JA. Nanog and transcriptional networks in embryonic stem cell pluripotency. *Cell Res.* 2007; 17:42-49. (Biology)

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Vigano MA, Staudt LM. Transcriptional activation by Oct-3: evidence for a specific role of the POU-specific domain in mediating functional interaction with Oct-1. *Nucleic Acids Res.* 1996; 24(11):2112-2118. (Biology)

Yuan H, Corbi N, Basilico C, Dailey L. Developmental-specific activity of the FGF-4 enhancer requires the synergistic action of Sox2 and Oct-3. *Genes Dev.* 1995; 9(21):2635-2645. (Biology)