Technical Data Sheet

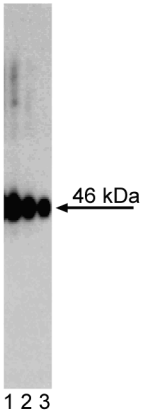
Purified Mouse anti-Oct3/4 (Human Isoform A)

Product Information

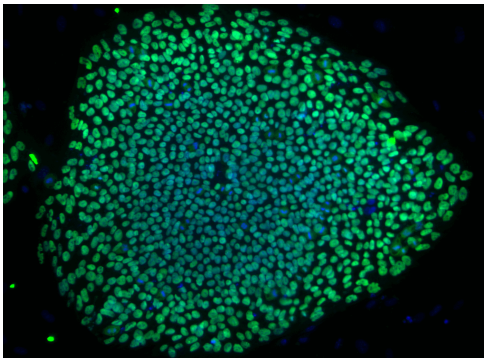
Material Number:	**561555**
Alternate Name:	Oct3/4A, Oct-3A, OTF-3, NF-A3, OTF4, POU5F1, MGC22487
Entrez Gene ID:	5460, 18999
Size:	0.1 mg
Concentration:	0.5 mg/ml
Clone:	O50-808
Immunogen:	Human Oct3/4 Isoform A Recombinant Protein
Isotype:	Mouse (BALB/c) IgG1, κ
Reactivity:	QC Tested: Human Tested in Development: Mouse
Target MW:	46 kDa
Storage Buffer:	Aqueous buffered solution containing ≤0.09% sodium azide.

Description

Development of a multicellular organism from a single fertilized egg 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 (ES) cell lines and embryonal carcinomas (EC). The human POU5F1 gene can encode various splice variants, two of which are Oct3/4A and Oct3/4B. Both isoforms share identical POU DNA-binding and C-terminal domains but differ in their N-terminal domain. The N-terminal domain of Oct3/4B is inhibitory to the DNA binding domain and therefore cannot stimulate transcription of Oct3/4-dependent genes. Oct3/4B can be detected in both pluripotent and some differentiated cell types in both the nucleus and cytoplasm, but its function is unclear. There is not an equivalent to Oct3/4B in mouse. Oct3/4A is expressed in the nucleus and has been demonstrated to orchestrate the transcription of Oct3/4-dependent genes. It has been demonstrated that the expression of Oct3/4 isoforms can vary greatly in different cell types, and discrimination of these is crucial for assessing Oct3/4 expression and function. The O50-808 monoclonal antibody recognizes human Oct3/4 Isoform A and mouse Oct3/4.



Western Blot analysis of Oct3/4 Isoform A in human embryonic stem (ES) cells. *Lysate from H9 human ES cells (WiCell,Madison, WI) grown in mTESR™1 medium (StemCell Technologies) on BD Matrigel™ hESC-qualified Matrix (Cat. No. 354277) (15 µg/lane) was probed with Purified Mouse anti-Oct3/4 (Human Isoform A) monoclonal antibody at 0.125 (lane 1), 0.06 (lane 2), and 0.03 (lane 3) µg/ml. Oct3/4A is identified as a band of 46 kDa.*



Immunofluorescent staining of Oct3/4 Isoform A in human embryonic stem (ES) cells. *H9 human ES cells (WiCell, Madison, WI) passage 33 grown on irradiated mouse fibroblasts were fixed with BD Cytofix™ fixation buffer (Cat. No. 554655), permeabilized, and stained with Purified Mouse anti-Oct3/4 (Human Isoform A) monoclonal antibody (pseudo-colored green) at 0.06 µg/mL. The second-step reagent was AlexaFluor® 488 goat anti-mouse Ig (Life Technologies), and cell nuclei were counter stained with Hoechst 33342 (pseudo-colored blue). The images were captured on a BD Pathway™ 435 Cell Analyzer and merged using BD Attovision™ Software. The cells were permeabilized with BD™ Phosflow Perm Buffer III (Cat. No. 558050); 1x BD Perm/Wash™ Buffer (Cat No. 554723) is also suitable for permeabilization.*

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Preparation and Storage

The monoclonal antibody was purified from tissue culture supernatant or ascites by affinity chromatography.
Store undiluted at 4°C.

Application Notes

Application

Western blot	Routinely Tested
Bioimaging	Tested During Development
Intracellular staining (flow cytometry)	Tested During Development
Immunofluorescence	Tested During Development

Suggested Companion Products

Catalog Number	Name	Size	Clone
354277	BD Matrigel™ hESC-qualified Matrix, 5 ml vial	NA	(none)
353219	BD Falcon™ 96-well Imaging Plate	NA	(none)
554002	HRP Goat Anti-Mouse Ig	1.0 ml	(none)
558050	Perm Buffer III	125 ml	(none)
554723	Perm/Wash Buffer	100 ml	(none)
554655	Fixation Buffer	100 ml	(none)

Product Notices

1. Since applications vary, each investigator should titrate the reagent to obtain optimal results.
2. Please refer to www.bdbiosciences.com/pharmingen/protocols for technical protocols.
3. 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.
4. Sodium azide is a reversible inhibitor of oxidative metabolism; therefore, antibody preparations containing this preservative agent must not be used in cell cultures nor injected into animals. Sodium azide may be removed by washing stained cells or plate-bound antibody or dialyzing soluble antibody in sodium azide-free buffer. Since endotoxin may also affect the results of functional studies, we recommend the NA/LE (No Azide/Low Endotoxin) antibody format, if available, for in vitro and in vivo use.

References

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