

ORDERING INFORMATION

Catalog Number: MAB2417

Clone: 257219

Lot Number: UVD01

Size: 100 µg (sufficient for 200 mL of Blotting solution)

Formulation: 0.2 µm filtered solution in PBS with 5% trehalose

Storage: -20° C

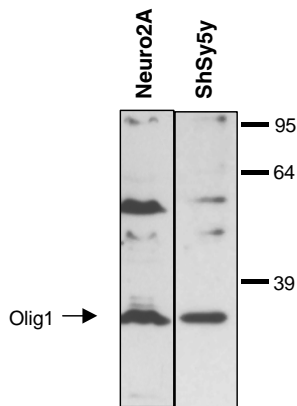
Specificity: human and mouse Olig1

Immunogen: *E. coli*-derived rhOlig1

Ig class: mouse IgG_{2b}

Recommended Applications:
Western blot
Immunohistochemistry

Other Application:
Direct ELISA



Detection of Olig1 with MAB2417.

Lysates from mouse Neuro2A and human ShSy5y cells were resolved by SDS-PAGE, transferred to Immobilon-P membrane, and immunoblotted with 0.5 µg/mL anti-Olig1 as described in *Protocols for Immunoblotting*. A one minute exposure to film is shown.

Preparation

This antibody was produced from a hybridoma resulting from the fusion of a mouse myeloma with B cells obtained from a mouse immunized with purified, *E. coli*-derived, recombinant human Oligodendrocyte transcription factor-1 (rhOlig1; Accession # XM_170977). The IgG fraction of the tissue culture supernatant was purified by Protein G affinity chromatography. Olig1 and Olig2, which have been implicated in oligodendrogenesis, are expressed in the region of the ventral ventricular zone of late embryonic spinal cord where oligodendrocyte progenitors appear. Olig3 is transiently expressed in different types of progenitors of embryonic central nervous system and then disappears in the course of development. In adult, expression of Olig3 is primarily detected in neural tissues.

Formulation

Lyophilized from a 0.2 µm filtered solution in phosphate-buffered saline (PBS) with 5% trehalose.

Reconstitution

Reconstitute with sterile PBS. If 0.2 mL of PBS is used, the antibody concentration will be 500 µg/mL.

Storage

Lyophilized samples are stable for twelve months from date of receipt when stored at -20° C to -70° C. Upon reconstitution, the antibody can be stored at 2° - 8° C for 1 month without detectable loss of activity. Reconstituted antibody can also be aliquotted and stored frozen at -20° C to -70° C in a manual defrost freezer for six months without detectable loss of activity. **Avoid repeated freeze-thaw cycles.**

Specificity

The antibody was selected for its ability to detect endogenous human and mouse Olig1 in direct ELISAs and western blots. In direct ELISAs, this antibody does not cross-react with rhOlig2 or rhOlig3.

Applications

Western Blot - An antibody concentration of 0.5 µg/mL is recommended.

Direct ELISA - This antibody can be used at 0.5 - 1.0 µg/mL with the appropriate secondary reagents to detect human Olig1. The detection limit for rhOlig1 is approximately 0.5 ng/well.

Optimal dilutions should be determined by each laboratory for each application.

Protocols for Immunoblotting:

Blotting Buffer

25 mM Tris, pH 7.5
0.15 M NaCl
0.05% Tween 20

Blocking Solution

5% nonfat dry milk
in Blotting Buffer
pH to 7.5

1. Transfer the electrophoresed proteins onto a PVDF membrane and incubate the membrane for 1 hour at room temperature in Blocking Solution.
2. Incubate the membrane for 2 hours at room temperature or overnight at 2 - 8° C in Blocking Solution containing 0.5 µg/mL anti-human/mouse Olig1.
3. Wash the membrane at room temperature for 60 minutes with 3 or more changes of Blotting Buffer. Changing the membrane containers often reduces background.
4. Incubate the membrane at room temperature for 1 hour in Blocking Solution containing a 1:2000 dilution of goat anti-mouse IgG-HRP (R&D Systems, Catalog # HAF007).
5. Wash the membrane for 60 minutes with 3 or more changes of Blotting Buffer.
6. Detect with WesternGlo™ Chemiluminescent Detection Substrate (R&D Systems #AR004) or equivalent.

Cell lysates for western blotting: To prepare total cell lysates, solubilize cells in 2X SDS gel sample buffer (20 mM dithiothreitol, 6% SDS, 0.25 M Tris, pH 6.8, 10% glycerol, and bromophenyl blue) and sonicate with a probe sonicator using 3 - 4 bursts of 5 - 10 seconds each. Heat extracts in a boiling water bath for 5 minutes and load onto polyacrylamide gels. Samples may be diluted with 1X SDS sample buffer to the desired concentration

Immunohistochemistry - This antibody was used at a concentration of 8-25 µg/mL with appropriate secondary reagents to detect Olig1 in paraffin-embedded human astrocytoma and glioma tissue sections. For chromogenic detection of labeling, the use of R&D Systems Cell and Tissue Staining Kits (CTS Series) is recommended.

For immunohistochemistry images, please refer to our website at <http://www.RnDSystems.com/go/ihc>.