

ORDERING INFORMATION

Catalog Number: MAB3367

Clone: 341504

Lot Number: XOG01

Size: 100 μg

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

Storage: -20° C

Reconstitution: sterile PBS

Specificity: human/mouse Wnt-8b

Immunogen: E. coli-derived rmWnt-8b

Ig class: rat IgG_{2A}

Recommended Application: Immunohistochemistry

Other Application: Direct ELISA

Monoclonal Anti-human/mouse Wnt-8b Antibody

Background

Mouse Wnt-8b is a 40 - 50 kDa secreted glycoprotein member of the Wnt family of proteins. Mature mouse Wnt-8b is 329 aa in length and contains 24 cysteines plus three potential N-linked glycosylation sites. Little is known about Wnt-8b. It appears to play a role in forebrain development. The mature mouse protein is 100%, 98% and 97% identical to mature rat, canine and human Wnt-8b, respectively.

Preparation

This antibody was produced from a hybridoma resulting from the fusion of a mouse myeloma with B cells obtained from a rat immunized with purified, *E. coli*-derived, recombinant mouse Wingless related integration site family member 8b (rmWnt-8b; Accession # AAD31816; aa 22 - 52 and 195 - 269, connected by a Gly-Ser linker). The IgG fraction of the tissue culture supernatant was purified by Protein G affinity chromatography.

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 $500 \mu g/mL$.

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

This antibody was selected for its ability to detect mouse Wnt-8b in direct ELISAs. In this format, this antibody shows no cross-reactivity with rmWnt-1, -2b, -3a, -4, -5a, -5b, -8a, -9b, -10b, rhWnt-2, -7a, -7b, or -9a. Immunohistochemical staining of the human neural cell line, NTera2, demonstrates that this antibody also recognizes human Wnt-8b.

Applications

Immunohistochemistry - This antibody was used at a concentration of 10 µg/mL to detect Wnt-8b on mouse fetal brain tissue sections. Sections were fixed with PBS containing 4% paraformaldehyde for 20 minutes at room temperature and blocked with PBS containing 10% normal donkey serum, 0.1% Triton[®] X-100, and 1% BSA for 45 minutes at room temperature. After blocking, cells were incubated with diluted primary antibody overnight at 4° C followed by Rhodamine Red[™]-coupled anti-rat IgG at room temperature in the dark for one hour. Between each step, cells were washed with PBS containing 0.1% BSA.

Direct ELISA - This antibody can be used at 0.5 - 1.0 μ g/mL with the appropriate secondary reagents to detect human or mouse Wnt-8b. The detection limit for rmWnt-8b is approximately 5 ng/well.

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

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