For Research Use Only. Not for use in diagnostic procedures.



Anti-Mutated IDH1/2 (Human) mAb

CODE No.	D332-3
CLONALITY	Monoclonal
CLONE	MsMab-2
ISOTYPE	Rat IgG2a κ
QUANTITY	100 μL, 1 mg/mL
SOURCE REACTIVITY	Purified IgG from hybridoma supernatant This clone strongly reacts with IDH1-R132L and IDH2-R172M. Please see the reference 3) and 4) for more details.
FORMULATION	PBS containing 50% glycerol. No preservative is contained.
STORAGE	This antibody solution is stable for one year from the date of purchase when stored at -20°C.

APPLICATION-CONFIRMED

<u>Western blotting</u> 5 µg/mL for chemiluminescence detection system

APPLICATIONS-REPORTED

ImmunohistochemistryReference 1) and 2)ELISAReference 4)

SPECIES CROSS REACTIVITY on WB

Species	Human	Mouse	Rat	Hamster
Sample Recombinant protein		Not tested	Not tested	Not tested
Reactivity	+			

Entrez Gene ID 3417, 3418 (Human)

REFERENCES

1) Hayashi, A., et al., Pathol. Int. 66, 578-582 (2016) [IHC]

2) Moriya, K., et al., Cancer Sci. 105, 359-362 (2015) [IHC]

3) Liu, X., et al., Cancer Med. 2, 803-814 (2013) [WB]

4) Ogasawara, S., et al., Monoclon. Antib. Immunodiagn. Immunother: 32, 377-381 (2013) [WB, ELISA]

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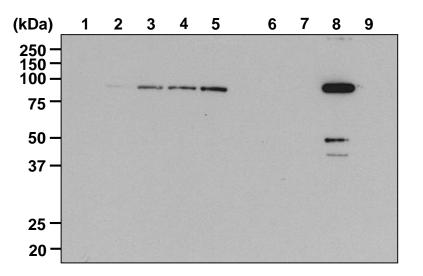
RELATED PRODUCTS

- D332-3 Anti-Mutated IDH1/2 (Human) mAb (MsMab-2)
- D309-3 Anti-IDH1 mAb (RMab-3)
- D336-3 Anti-IDH1 (Human) mAb (RcMab-1)
- D299-3 Anti-IDH1-R132H (Human) mAb (HMab-1)
- D300-3 Anti-IDH1-R132S (Human) mAb (SMab-1)
- D331-3 Anti-IDH1-R132G (Human) mAb (GMab-r1)
- D311-3 Anti-IDH2 mAb (RMab-22)
- D330-3 Anti-IDH2 mAb (KrMab-3)
- D328-3 Anti-IDH2-R172K (Human) mAb (KMab-1)
- D337-3 Anti-IDH2-R172M (Human) mAb (MMab-1)
- D338-3 Anti-IDH2-R172W (Human) mAb (WMab-1)

SDS-PAGE & Western blotting

- 1) The recombinant protein is dissolved in Laemmli's sample buffer at $10 \,\mu\text{g/mL}$.
- 2) Boil the samples for 2 min. and centrifuge. Load 10 µL of the sample per lane in a 1-mm-thick SDS-polyacrylamide gel (12.5% acrylamide) for electrophoresis.
- 3) Blot the protein to a polyvinylidene difluoride (PVDF) membrane at 1 mA/cm² for 1 hr. in a semi-dry transfer system (Transfer Buffer: 25 mM Tris, 190 mM glycine, 20% MeOH). See the manufacturer's manual for precise transfer procedure.
- 4) To reduce nonspecific binding, soak the membrane in 10% skimmed milk (in PBS, pH 7.2) for 1 hr. at room temperature.
- 5) Incubate the membrane with primary antibody diluted with 1% skimmed milk (in PBS, pH 7.2) as suggested in the **APPLICATIONS** for 1 hr. at room temperature. (The concentration of antibody will depend on the conditions.)
- 6) Wash the membrane with PBS-T [0.05% Tween-20 in PBS] (5 min. x 3 times).
- 7) Incubate the membrane with HRP-conjugated anti-rat IgG antibody diluted with 1% skimmed milk (in PBS, pH 7.2) for 1 hr. at room temperature.
- 8) Wash the membrane with PBS-T (5 min. x 3 times).
- 9) Wipe excess buffer on the membrane, and then incubate it with appropriate chemiluminescence reagent for 1 min. Remove extra reagent from the membrane by dabbing with paper towel, and seal it in plastic wrap.
- 10) Expose to an X-ray film in a dark room for 10 min. Develop the film as usual. The condition for exposure and development may vary.

(Positive controls for Western blotting; Recombinant human mutated IDH1 and IDH2)



Western blot analysis of human IDH1 and IDH2 proteins

Lane 1: IDH1 (Wild type) Lane 2: IDH1-R132H Lane 3: IDH1-R132S Lane 4: IDH1-R132G Lane 5: IDH1-R132L Lane 6: IDH2 (Wild type) Lane 7: IDH2-R172K Lane 8: IDH2-R172M Lane 9: IDH2-R172W

Immunoblotted with Anti-Mutated IDH1/2 (Human) mAb (D332-3)

The samples were kindly provided by Dr. Yukinari Kato. (Department of Regional Innovation, Tohoku University Graduate School of Medicine)