

# Anti-IDH2-R172W (Human) mAb

**CODE No.** D338-3

**CLONALITY** Monoclonal  
**CLONE** WMab-1  
**ISOTYPE** Rat IgG2b  $\kappa$   
**QUANTITY** 100  $\mu$ L, 1 mg/mL

**SOURCE** Purified IgG from hybridoma supernatant  
**FORMURATION** PBS containing 50% Glycerol (pH 7.2). 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

Western blotting 1  $\mu$ g/mL for chemiluminescence detection system

## APPLICATIONS-REPORTED

Immunohistochemistry Reference 1)  
Immunocytochemistry Reference 1)

## SPECIES CROSS REACTIVITY on WB

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

**Entrez Gene ID** 3418 (Human)

**REFERENCES** 1) Kato, Y. and Kaneko, M. K., *Biochem Biophys Res Commun.* In press (2013) [WB, IHC, IC]  
2) Parsons, D. W., *et al.*, *Science* **321**, 1807-1812 (2008)

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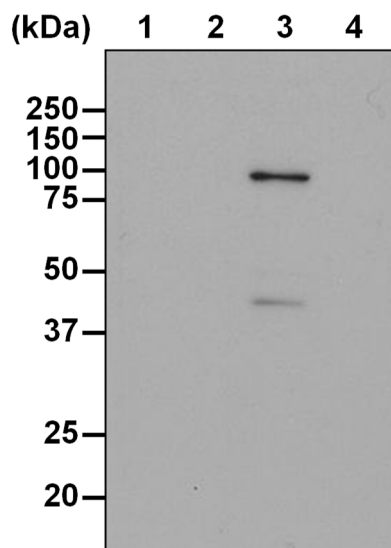
**RELATED PRODUCTS**

D338-3	Anti-IDH2-R172W (Human) mAb (WMab-1)
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)
D336-3	Anti-IDH1 (Human) mAb (RcMab-1)
D309-3	Anti-IDH1 mAb (RMab-3)
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)

### **SDS-PAGE & Western blotting**

- 1) The recombinant protein is dissolved in Laemmli's sample buffer at 10 µg/mL.
- 2) Boil the samples for 3 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/cm<sup>2</sup> 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 **APPLICATION** 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 the 1:10,000 anti-IgG (Rat) pAb-HRP (MBL; code no. IM-0825) 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 3 min. Develop the film as usual. The condition for exposure and development may vary.

(Positive control for Western blotting; recombinant protein)



#### ***Western blot analysis of IDH2-R172W***

- Lane 1: IDH2 (Wild type)
- Lane 2: IDH2-R172K
- Lane 3: IDH2-R172W
- Lane 4: IDH2-R172M

Immunoblotted with Anti-IDH2-R172W mAb (D338-3)