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

Anti-IDH2-R172K (Human) mAb

CODE No. D328-3

CLONALITY Monoclonal
CLONE KMab-1
ISOTYPE Rat IgG2b κ
QUANTITY 100 µL, 1 mg/mL

SOURCE Purified IgG from hybridoma supernatant
FORMULATION 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 µg/mL

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) Parsons, D. W., *et al.*, *Science* **321**, 1807-1812 (2008)

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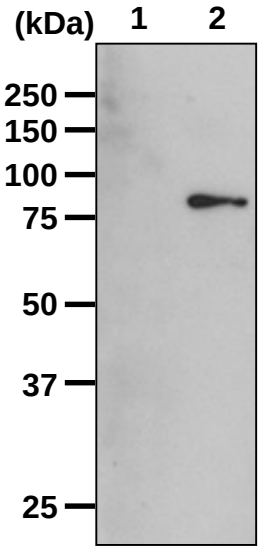
SDS-PAGE & Western blotting

1) Mix 5 μg samples in 10 μL of Laemmli's sample buffer.
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^2 for 1 hr. in a semi-dry transfer system (Transfer Buffer: 25 mM Tris, 190 mM glycine, 20% methanol). 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).
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).
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 1 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-R172K
Lane 1: IDH2 (Wild type)
Lane 2: IDH2-R172K
Immunoblotted with D328-3