# **Anti-Dkk3 Antibody [JE35-98]**

### **HA722396**



Product Type: Recombinant Rabbit monoclonal IgG, primary antibodies

Species reactivity: Human, Mouse, Rat

Applications: WB, IF-Cell, IHC-P, IHC-Fr, IP

Molecular Wt: Predicted band size: 38 kDa

Clone number: JE35-98

**Description:** Dickkopf-related protein 3 is a protein in the Dickkopf family that in humans is encoded by

the DKK3 gene. This gene encodes a protein that is a member of the dickkopf family. The secreted protein contains two cysteine rich regions and is involved in embryonic development through its interactions with the Wnt signaling pathway. The expression of this gene is decreased in a variety of cancer cell lines and it may function as a tumor suppressor gene. Alternative splicing results in multiple transcript variants encoding the same protein.

Immunogen: Recombinant protein within Human Dkk3 aa 151-350 / 350.

Positive control: Mouse brain tissue lysate, mouse brain treated with deglycosylation tissue lysate, human

brain tissue lysate, rat brain tissue lysate, HUVEC, human brain tissue, mouse brain tissue,

rat brain tissue.

Subcellular location: Secreted.

Database links: SwissProt: Q9UBP4 Human | Q9QUN9 Mouse

Entrez Gene: 171548 Rat

Recommended Dilutions:

WB 1:1,000 IF-Cell 1:100 IHC-P 1:1,000 IHC-Fr 1:200

IP 1-2µg/sample

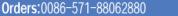
**Storage Buffer:** 1\*TBS (pH7.4), 0.05% BSA, 40% Glycerol. Preservative: 0.05% Sodium Azide.

Storage Instruction: Shipped at 4℃. Store at +4℃ short term (1-2 weeks). It is recommended to aliquot into

single-use upon delivery. Store at -20 ℃ long term.

**Purity:** Protein A affinity purified.

## Hangzhou Huaan Biotechnology Co., Ltd.



Technical: 0086-571-89986345

Service mail:support@huabio.cn



#### **Images**

 **Fig1:** Western blot analysis of Dkk3 on different lysates with Rabbit anti-Dkk3 antibody (HA722396) at 1/1,000 dilution.

Lane 1: Mouse brain tissue lysate (14 µg/Lane)

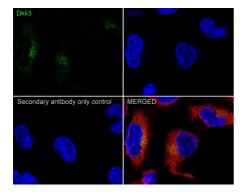
Lane 2: Mouse brain tissue lysate treated with deglycosylation (14 µg/Lane)

Lane 3: Human brain tissue lysate (20 µg/Lane) Lane 4: Rat brain tissue lysate (20 µg/Lane)

Predicted band size: 38 kDa Observed band size: 50-70 kDa

Exposure time: Lane 1-3: 10 seconds; Lane 4: 40 seconds; ECL: K1801;

4-20% SDS-PAGE gel.



**Fig2:** Immunocytochemistry analysis of HUVEC cells labeling Dkk3 with Rabbit anti-Dkk3 antibody (HA722396) at 1/100 dilution.

Cells were fixed in 4% paraformaldehyde for 20 minutes at room temperature, permeabilized with 0.1% Triton X-100 in PBS for 5 minutes at room temperature, then blocked with 1% BSA in 10% negative goat serum for 1 hour at room temperature. Cells were then incubated with Rabbit anti-Dkk3 antibody (HA722396) at 1/100 dilution in 1% BSA in PBST overnight at 4 ℃. Goat Anti-Rabbit IgG H&L (iFluor™ 488, HA1121) was used as the secondary antibody at 1/1,000 dilution. PBS instead of the primary antibody was used as the secondary antibody only control. Nuclear DNA was labelled in blue with DAPI.

Beta tubulin (M1305-2, red) was stained at 1/100 dilution overnight at  $+4^{\circ}$ C. Goat Anti-Mouse IgG H&L (iFluor 594, HA1126) was used as the secondary antibody at 1/1,000 dilution.



Fig3: Immunohistochemical analysis of paraffin-embedded human brain tissue with Rabbit anti-Dkk3 antibody (HA722396) at 1/1,000 dilution.

The section was pre-treated using heat mediated antigen retrieval with Tris-EDTA buffer (pH 9.0) for 20 minutes. The tissues were blocked in 1% BSA for 20 minutes at room temperature, washed with ddH<sub>2</sub>O and PBS, and then probed with the primary antibody (HA722396) at 1/1,000 dilution for 1 hour at room temperature. The detection was performed using an HRP conjugated compact polymer system. DAB was used as the chromogen. Tissues were counterstained with hematoxylin and mounted with DPX.

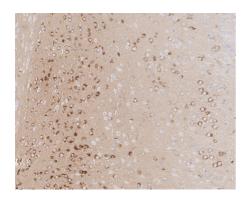
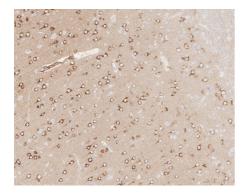


Fig4: Immunohistochemical analysis of paraffin-embedded mouse brain tissue with Rabbit anti-Dkk3 antibody (HA722396) at 1/1,000 dilution.

The section was pre-treated using heat mediated antigen retrieval with Tris-EDTA buffer (pH 9.0) for 20 minutes. The tissues were blocked in 1% BSA for 20 minutes at room temperature, washed with ddH $_2$ O and PBS, and then probed with the primary antibody (HA722396) at 1/1,000 dilution for 1 hour at room temperature. The detection was performed using an HRP conjugated compact polymer system. DAB was used as the chromogen. Tissues were counterstained with hematoxylin and mounted with DPX.



**Fig5:** Immunohistochemical analysis of paraffin-embedded rat brain tissue with Rabbit anti-Dkk3 antibody (HA722396) at 1/1,000 dilution.

The section was pre-treated using heat mediated antigen retrieval with Tris-EDTA buffer (pH 9.0) for 20 minutes. The tissues were blocked in 1% BSA for 20 minutes at room temperature, washed with ddH<sub>2</sub>O and PBS, and then probed with the primary antibody (HA722396) at 1/1,000 dilution for 1 hour at room temperature. The detection was performed using an HRP conjugated compact polymer system. DAB was used as the chromogen. Tissues were counterstained with hematoxylin and mounted with DPX.

Hangzhou Huaan Biotechnology Co., Ltd.

**//**华安生物 HUABIO www.huabio.cn Dkk3

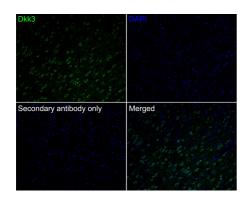
DAPI

Secondary antibody only

Merged

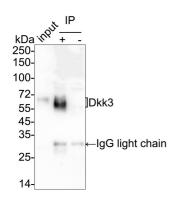
**Fig6:** Immunofluorescence analysis of frozen mouse brain tissue with Rabbit anti-Dkk3 antibody (HA722396) at 1/200 dilution.

The section was pre-treated using heat mediated antigen retrieval with sodium citrate buffer (pH 6.0) for about 2 minutes in microwave oven. The tissues were blocked in 10% negative goat serum for 1 hour at room temperature, washed with PBS, and then probed with the primary antibody (HA722396, green) at 1/200 dilution overnight at 4  $^{\circ}$ C, washed with PBS. Goat Anti-Rabbit IgG H&L (iFluor  $^{\dagger}$ M 488, HA1121) was used as the secondary antibody at 1/1,000 dilution. Nuclei were counterstained with DAPI (blue).



**Fig7:** Immunofluorescence analysis of frozen rat brain tissue with Rabbit anti-Dkk3 antibody (HA722396) at 1/200 dilution.

The section was pre-treated using heat mediated antigen retrieval with sodium citrate buffer (pH 6.0) for about 2 minutes in microwave oven. The tissues were blocked in 10% negative goat serum for 1 hour at room temperature, washed with PBS, and then probed with the primary antibody (HA722396, green) at 1/200 dilution overnight at 4  $^{\circ}$ C, washed with PBS. Goat Anti-Rabbit IgG H&L (iFluor  $^{\dagger}$  488, HA1121) was used as the secondary antibody at 1/1,000 dilution. Nuclei were counterstained with DAPI (blue).



**Fig8:** Dkk3 was immunoprecipitated from 0.2 mg mouse brain tissue lysate with HA722396 at 2  $\mu$ g/10  $\mu$ l beads. Western blot was performed from the immunoprecipitate using HA722396 at 1/1,000 dilution. Anti-Rabbit IgG for IP Nano-secondary antibody (NBI01H) at 1/5,000 dilution was used for 1 hour at room temperature.

Lane 1: Mouse brain tissue lysate (input)

Lane 2: HA722396 IP in mouse brain tissue lysate

Lane 3: Rabbit IgG instead of HA722396 in mouse brain tissue

lysate

Blocking/Dilution buffer: 5% NFDM/TBST Exposure time: 3 minutes; ECL: K1802

### Hangzhou Huaan Biotechnology Co., Ltd.

Service mail:support@huabio.cn



Note: All products are "FOR RESEARCH USE ONLY AND ARE NOT INTENDED FOR DIAGNOSTIC OR THERAPEUTIC USE".

#### **Background References**

- 1. Xu J et al. Myofiber Baf60c controls muscle regeneration by modulating Dkk3-mediated paracrine signaling. J Exp Med. 2023 Sep
- 2. Zhang LQ et al. DKK3 ameliorates neuropathic pain via inhibiting ASK-1/JNK/p-38-mediated microglia polarization and neuroinflammation. J Neuroinflammation. 2022 Jun