

# Anti-PGBD5 Antibody [7-F8-5]

## M1012-1



<b>Product Type:</b>	Mouse monoclonal IgG1, primary antibodies
<b>Species reactivity:</b>	Human, Mouse, Rat
<b>Applications:</b>	WB, IF-Cell, IHC-P
<b>Molecular Wt:</b>	Predicted band size: 58 kDa
<b>Clone number:</b>	7-F8-5

<b>Description:</b>	PiggyBac Transposable Element Derived 5 is an enzyme that in humans is encoded by the PGBD5 gene. PGBD5 is a DNA transposase related to the ancient PiggyBac transposase first identified in the cabbage looper moth, <i>Trichoplusia ni</i> . The gene is believed to have been domesticated over 500 million years ago in the common ancestor of cephalochordates and vertebrates. The putative catalytic triad of the protein composed of three aspartic acid residues is conserved among PGBD5-like genes through evolution, and is distinct from other PiggyBac-like genes. PGBD5 has been shown to be able to transpose DNA in a sequence-specific, cut-and-paste fashion. PGBD5 has also been proposed to mediate site-specific DNA rearrangements in human tumors. In mature mice brain tissue PGBD5 is found primarily in regions of the olfactory bulb, hippocampus, and cerebellum. In embryonic mice brain tissue PGBD5 is found not only in the medial pallium and prepontine isthmus, which are embryonic brain areas that give rise to the development of the hippocampus and cerebellum but also in areas in the embryonic brain that give rise to the hypothalamus and medulla. PGBD5 is expressed in the majority of human pediatric solid tumors. It's upregulated in sporadic Creutzfeldt-Jakob disease. PGBD5 is associated with frontotemporal dementia, where it gets most expressed in neurons, followed by oligodendrocytes, mature astrocytes, fetal astrocytes, endothelial cells and then microglia/macrophages.
<b>Immunogen:</b>	Synthetic peptide with Human PGBD5 aa 61-110 / 524.
<b>Positive control:</b>	Human brain tissue lysate, Mouse brain tissue lysate, SH-SY5Y, Neuro-2a, C6, human lung squamous cell carcinoma tissue, mouse hippocampus tissue, mouse cerebellum tissue, rat cerebellum tissue.
<b>Subcellular location:</b>	Nucleus
<b>Database links:</b>	SwissProt: Q8N414 Human   D3YZI9 Mouse Entrez Gene: 292098 Rat
<b>Recommended Dilutions:</b>	
<b>WB</b>	1:500-1:2,000
<b>IF-Cell</b>	1:100-1:1,000
<b>IHC-P</b>	1:200-1:400
<b>Storage Buffer:</b>	1*PBS (pH7.4), 0.2% BSA, 40% Glycerol. Preservative: 0.05% Sodium Azide.
<b>Storage Instruction:</b>	Shipped at 4°C. Store at +4°C short term (1-2 weeks). It is recommended to aliquot into single-use upon delivery. Store at -20°C long term.
<b>Purity:</b>	Protein A affinity purified.

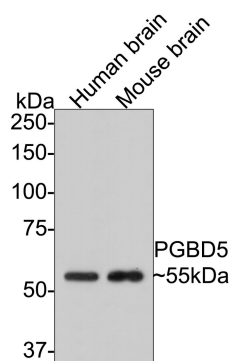
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**Fig1:** Western blot analysis of PGBD5 on different lysates with Mouse anti-PGBD5 antibody (M1012-1) at 1/500 dilution.

Lane 1: Human brain tissue lysate

Lane 2: Mouse brain tissue lysate

Lysates/proteins at 20 µg/Lane.

Predicted band size: 58 kDa

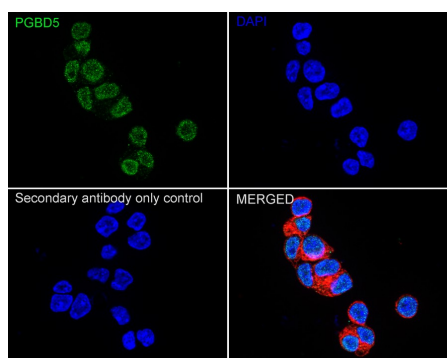
Observed band size: 55 kDa

Exposure time: 2 minutes;

8% SDS-PAGE gel.

Proteins were transferred to a PVDF membrane and blocked with 5% NFDM/TBST for 1 hour at room temperature. The primary antibody (M1012-1) at 1/500 dilution was used in 5% NFDM/TBST at room temperature for 2 hours. Goat Anti-Mouse IgG - HRP Secondary Antibody (HA1006) at 1:150,000 dilution was used for 1 hour at room temperature.

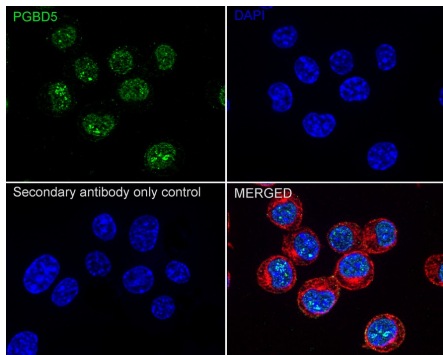
**Fig2:** Immunocytochemistry analysis of SH-SY5Y cells labeling PGBD5 with Mouse anti-PGBD5 antibody (M1012-1) at 1/100 dilution.



Cells were fixed in 4% paraformaldehyde for 15 minutes at room temperature, permeabilized with 0.1% Triton X-100 in PBS for 15 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 Mouse anti-PGBD5 antibody (M1012-1) at 1/100 dilution in 1% BSA in PBST overnight at 4 °C. Goat Anti-Mouse IgG H&L (iFluor™ 488, HA1125) 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 (ET1602-4, red) was stained at 1/100 dilution overnight at +4°C. Goat Anti-Rabbit IgG H&L (iFluor™ 594, HA1122) was used as the secondary antibody at 1/1,000 dilution.

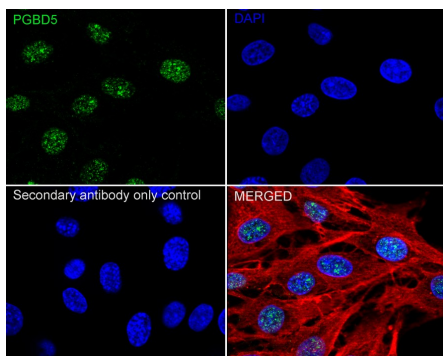
**Fig3:** Immunocytochemistry analysis of Neuro-2a cells labeling PGBD5 with Mouse anti-PGBD5 antibody (M1012-1) at 1/100 dilution.



Cells were fixed in 4% paraformaldehyde for 15 minutes at room temperature, permeabilized with 0.1% Triton X-100 in PBS for 15 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 Mouse anti-PGBD5 antibody (M1012-1) at 1/100 dilution in 1% BSA in PBST overnight at 4 °C. Goat Anti-Mouse IgG H&L (iFluor™ 488, HA1125) 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.

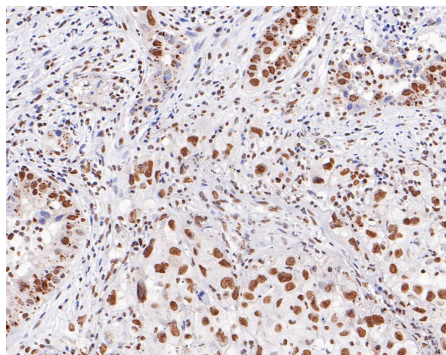
beta Tubulin (ET1602-4, red) was stained at 1/100 dilution overnight at +4°C. Goat Anti-Rabbit IgG H&L (iFluor™ 594, HA1122) was used as the secondary antibody at 1/1,000 dilution.

**Fig4:** Immunocytochemistry analysis of C6 cells labeling PGBD5 with Mouse anti-PGBD5 antibody (M1012-1) at 1/1,000 dilution.



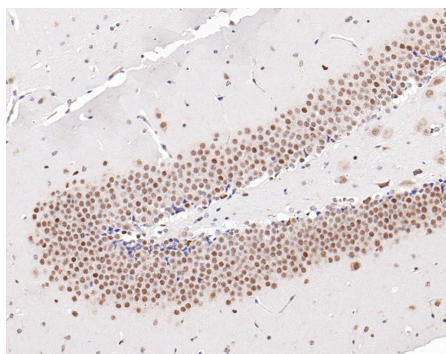
Cells were fixed in 4% paraformaldehyde for 15 minutes at room temperature, permeabilized with 0.1% Triton X-100 in PBS for 15 minutes at room temperature, then blocked with 1% BSA in 10% negative goat serum for 1 hour at room temperature. Cells were then incubated with Mouse anti-PGBD5 antibody (M1012-1) at 1/1,000 dilution in 1% BSA in PBST overnight at 4 °C. Goat Anti-Mouse IgG H&L (iFluor™ 488, HA1125) 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 (ET1602-4, red) was stained at 1/100 dilution overnight at +4°C. Goat Anti-Rabbit IgG H&L (iFluor™ 594, HA1122) was used as the secondary antibody at 1/1,000 dilution.



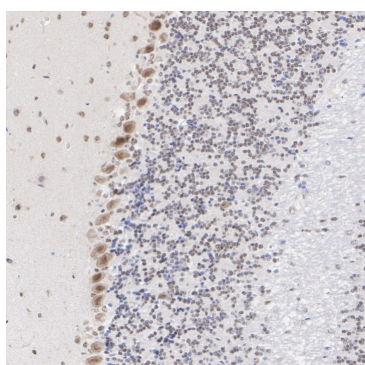
**Fig5:** Immunohistochemical analysis of paraffin-embedded human lung squamous cell carcinoma tissue with Mouse anti-PGBD5 antibody (M1012-1) at 1/400 dilution.

The section was pre-treated using heat mediated antigen retrieval with sodium citrate buffer (pH 6.0) (high pressure) for 2 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 (M1012-1) at 1/400 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.



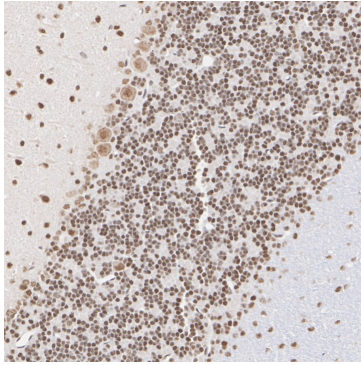
**Fig6:** Immunohistochemical analysis of paraffin-embedded mouse hippocampus tissue with Mouse anti-PGBD5 antibody (M1012-1) at 1/400 dilution.

The section was pre-treated using heat mediated antigen retrieval with sodium citrate buffer (pH 6.0) (high pressure) for 2 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 (M1012-1) at 1/400 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.



**Fig7:** Immunohistochemical analysis of paraffin-embedded mouse cerebellum tissue with Mouse anti-PGBD5 antibody (M1012-1) at 1/200 dilution.

The section was pre-treated using heat mediated antigen retrieval with sodium citrate buffer (pH 6.0) (high pressure) for 2 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 (M1012-1) at 1/200 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.



**Fig8:** Immunohistochemical analysis of paraffin-embedded rat cerebellum tissue with Mouse anti-PGBD5 antibody (M1012-1) at 1/200 dilution.

The section was pre-treated using heat mediated antigen retrieval with sodium citrate buffer (pH 6.0) (high pressure) for 2 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 (M1012-1) at 1/200 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.

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

### Background References

1. "The status, quality, and expansion of the NIH full-length cDNA project: the Mammalian Gene Collection (MGC)." The MGC Project Team *Genome Res.* 14:2121-2127(2004)
2. "The DNA sequence and biological annotation of human chromosome 1." Gregory S.G., Barlow K.F., McLay K.E., Kaul R., Swarbreck D., Dunham A., Scott C.E., Howe K.L., Woodfine K., Spencer C.C.A., Jones M.C., Gillson C., Searle S., Zhou Y., Kokocinski F., McDonald L., Evans R., Phillips K. Bentley D.R. *Nature* 441:315-321(2006).

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