Oligodendrocyte Marker Antibody Sampler Kit

HAK21061

Contains Product	Quantity	Applications	Species reactivity	MW(kDa)
Olig2 [ET1604-29]	20μl	WB,IHC-P,IHC-Fr,IF-Tissue,mIHC,IF-Cell	H,M,R	32 kDa
Myelin Basic Protein [ET1702-15]	20μl	WB,IHC-P,IF-Tissue,IHC-Fr,mIHC	H,M,R	33 kDa
CNPase [ET1702-46]	20μl	WB,IHC-P,IF-Tissue	H,M,R	48 kDa
Myelin PLP [HA722964]	20μl	WB,IHC-P,IF-Tissue,IHC-Fr	H,M,R	30 kDa
MAG [HA721818]	20μl	WB,IHC-P,IF-Tissue,IP,IHC-Fr	H,M,R	69 kDa
MOG [ET1705-16]	20μl	WB,IHC-P,IF-Tissue,IHC-Fr	H,M,R	28 kDa
NG2 [ET1703-16]	20μl	WB,IHC-P	H,M,R	251 kDa
SOX10 [HA721240]	20μl	WB,IHC-P,IF-Cell,IF-Tissue,FC,IHC-Fr,IP	H,M,R	50 kDa
HRP-Goat Anti-Rabbit IgG (H+L) [HA1001]	100μl	WB,ELISA,IHC-P	Rab	

Description: The Oligodendrocyte Marker Antibody Sampler Kit provides an economical means of detecting proteins identified as oligodendrocyte markers by immunofluorescence and western blot. This kit includes enough primary antibodies to perform at least two western blot experiments per primary antibody.

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

Storage Instruction: Store at +4℃ after thawing. Aliquot store at -20℃. Avoid repeated freeze / thaw cycles.

Background Oligodendrocytes are the myelinating glial cells of the central nervous system (CNS). Myelin basic protein (MBP) is an abundant CNS myelin membrane protein that plays an important role in nerve myelination. CNPase (2', 3'-cyclic nucleotide 3'-phosphodiesterase) is an enzyme highly expressed in oligodendrocytes. Myelin proteolipid protein (PLP1) corresponds to the majority of myelin proteins in the CNS, providing support to axons and modulating axonal growth. Myelin-associated glycoprotein (MAG) is localized in oligodendroglial membranes of myelin sheaths. Chondroitin sulfate proteoglycan 4 (CSP4, NG2) is a type I membrane glycoprotein expressed by oligodendroglial precursor cells (OPCs). SRY-box 10 (Sox10) is a high-mobility group transcription factor expressed throughout oligodendrocyte development required for myelin gene expression. Myelin-oligodendrocyte glycoprotein (MOG) is a type I membrane bound glycoprotein of the immunoglobulin superfamily that is enriched in the outer lamella of the myelin sheath. Olig2 is a basic helix–loop–helix (bHLH) transcription factor necessary for oligodendrocyte and motor neuron differentiation and development in the CNS.

Database links: UniProt ID: Q13516, Q9EQW6, 22121, P02686, P04370, P02688, P09543, P16330, P13233, P60201, P60202, P60203, P20916, P20917, P07722, Q16653, Q61885, Q63345, Q6UVK1, Q8VHY0, Q00657, P56693, Q04888, O55170

Images

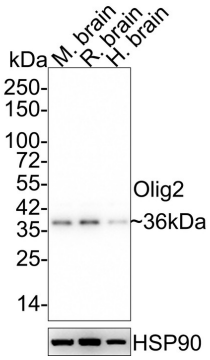

Fig1: Western blot analysis of Olig2 on different lysates with Rabbit anti-Olig2 antibody (ET1604-29) at 1/5,000 dilution.

Lane 1: Mouse brain tissue lysate (20 µg/Lane)
Lane 2: Rat brain tissue lysate (20 µg/Lane)
Lane 3: Human brain tissue lysate (20 µg/Lane)

Predicted band size: 32 kDa
Observed band size: 36 kDa
Exposure time: 5 minutes 30 seconds;
4-20% 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 (ET1604-29) at 1/5,000 dilution was used in 5% NFDM/TBST at 4℃ overnight. Goat Anti-Rabbit IgG - HRP Secondary Antibody (HA1001) at 1/50,000 dilution was used for 1 hour at room temperature.

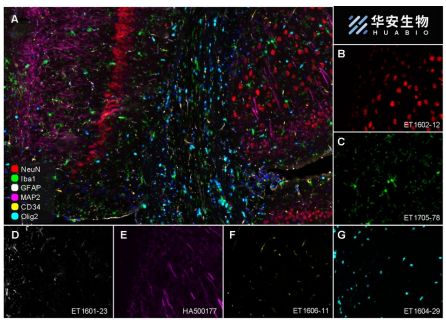

Fig2: Fluorescence multiplex immunohistochemical analysis of mouse brain (Formalin/PFA-fixed paraffin-embedded sections). Panel A: the merged image of anti-NeuN (ET1602-12, red), anti-Iba1 (ET1705-78, green), anti-GFAP (ET1601-23, gray), anti-Olig2 (ET1604-29, cyan), anti-MAP2 (HA500177, magenta) and anti-CD34 (ET1606-11, yellow) on mouse brain. HRP Conjugated UltraPolymer Goat Polyclonal Antibody HA1119/HA1120 was used as a secondary antibody. The immunostaining was performed with the Sequential Immuno-staining Kit (IRISKit™MH010101, www.luminiris.cn). The section was incubated in six rounds of staining: in the order of ET1602-12(1/5,000 dilution), ET1705-78 (1/2,000 dilution), ET1601-23 (1/5,000 dilution), ET1604-29 (1/1,000 dilution), HA500177 (1/5,000 dilution) and ET1606-11 (1/2,000 dilution) for 20 mins at room temperature. Each round was followed by a separate fluorescent tyramide signal amplification system. Heat mediated antigen retrieval with Tris-EDTA buffer (pH 9.0) for 30 mins at 95℃. DAPI (blue) was used as a nuclear counter stain. Image acquisition was performed with Olympus VS200 Slide Scanner.

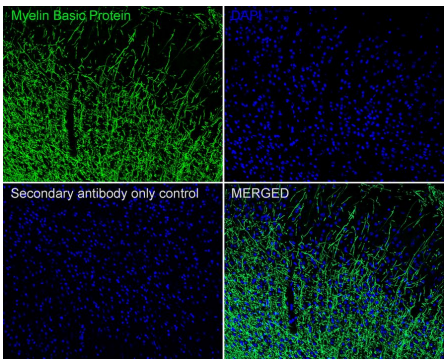

Fig3: Immunofluorescence analysis of frozen mouse cerebrum tissue with Rabbit anti-Myelin Basic Protein antibody (ET1702-15) at 1/500 dilution.

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 (ET1702-15, green) at 1/500 dilution overnight at 4 ℃, washed with PBS. Goat Anti-Rabbit IgG H&L (iFluor™ 488, HA1121) was used as the secondary antibody at 1/1,000 dilution. Nuclei were counterstained with DAPI (blue).

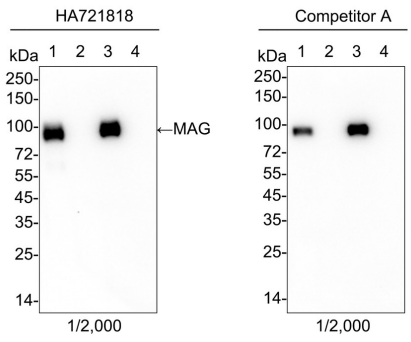

Fig4: Western blot analysis of MAG on different lysates with Rabbit anti-MAG antibody (HA721818) at 1/2,000 dilution and competitor's antibody at 1/2,000 dilution.

Lane 1: Mouse cerebellum tissue lysate
Lane 2: Mouse liver tissue lysate (negative)
Lane 3: Rat cerebellum tissue lysate
Lane 4: Rat liver tissue lysate (negative)

Lysates/proteins at 20 µg/Lane.

Predicted band size: 69 kDa
Observed band size: 100 kDa

Exposure time: 2 minutes; ECL: K1801;
4-20% 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 (HA721818) at 1/2,000 dilution and competitor's antibody at 1/2,000 dilution were used in 5% NFDM/TBST at 4℃ overnight. Goat Anti-Rabbit IgG - HRP Secondary Antibody (HA1001) at 1/50,000 dilution was used for 1 hour at room temperature.

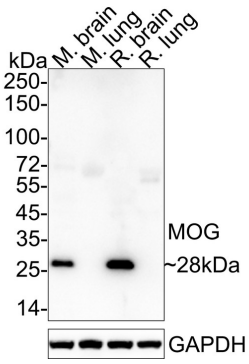

Fig5: Western blot analysis of Myelin oligodendrocyte glycoprotein on different lysates with Rabbit anti-Myelin oligodendrocyte glycoprotein antibody (ET1705-16) at 1/1,000 dilution.

Lane 1: Mouse brain tissue lysate (40 µg/Lane)
Lane 2: Mouse lung tissue lysate (negative) (40 µg/Lane)
Lane 3: Rat brain tissue lysate (40 µg/Lane)
Lane 4: Rat lung tissue lysate (negative) (40 µg/Lane)

Predicted band size: 28 kDa
Observed band size: 28 kDa

Exposure time: 1 minute; ECL: K1802;

4-20% 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 (ET1705-16) at 1/1,000 dilution was used in 5% NFDM/TBST at 4℃ overnight. Goat Anti-Rabbit IgG - HRP Secondary Antibody (HA1001) at 1/50,000 dilution was used for 1 hour at room temperature.

Background References

1. Harauz G, Boggs JM. Myelin management by the 18.5-kDa and 21.5-kDa classic myelin basic protein isoforms. J Neurochem. 2013 May;125(3):334-61.
2. Molina-Gonzalez I, Holloway RK, Jiwaji Z, Dando O, Kent SA, Emelianova K, Lloyd AF, Forbes LH, Mahmood A, Skripuletz T, Gudi V, Febery JA, Johnson JA, Fowler JH, Kuhlmann T, Williams A, Chandran S, Stangel M, Howden AJM, Hardingham GE, Miron VE. Astrocyte-oligodendrocyte interaction regulates central nervous system regeneration. Nat Commun. 2023 Jun 8;14(1):3372.