Anti-Tryptophan Hydroxylase 1 (TPH1) Antibody [SC53-07]

ET1610-37

Product Type:	Recombinant Rabbit monoclonal IgG, primary antibodies
Species reactivity:	Human, Mouse, Rat
Applications:	WB, IF-Cell, IF-Tissue, IHC-P, IP, FC
Molecular Wt:	51 kDa
Clone number:	SC53-07

Description: Phenylalanine hydroxylase (PAH), tyrosine hydroxylase (TH) and tryptophan hydroxylase (TPH) comprise a small family of monooxygenases that use tetrahydropterine as a cofactor during the catabolism of aromatic L-amino acids. PAH, TH and TPH all contain catalytic domains with an amino-terminal regulatory domain and a short carboxy-terminal tetramerization domain. Each of these enzymes also contains a single ferrous iron atom, which is bound to two histidines and a glutamate and is likely to be involved in the formation of the hydroxylating intermediate. TPH is the first and rate-limiting step in the biosynthesis of serotonin in the central nervous system and melatonin in the pineal gland. Alteration of TPH function may be a key factor in the pathology of several neuropsychiatric disorders associated with serotonin, including depression, aggression, alcoholism and schizophrenia. For instance, L-DOPA, which is used as a common therapy for Parkinson's disease (PD) patients, inhibits TPH function, which subsequently, is thought to contribute to the onset of depression in PD patients.

Immunogen: Synthetic peptide within Human TPH1 aa 372-419 / 444.

Positive control: HL-60 cell lysate, Hela cell lysate, rat brain tissue, mouse cerebellum tissue, Hela.

Subcellular location: Cytosol, neuron projection.

Database links: SwissProt: P17752 Human | P17532 Mouse | P09810 Rat

Recommended Dilutions:

WB	1:500
IF-Cell	1:50-1:200
IF-Tissue	1:50-1:200
IHC-P	1:50-1:200
FC	1:50-1:100
IP	Use at an assay dependent concentration.

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℃ or -80℃. Avoid repeated freeze / thaw cycles.

Purity: Protein A affinity purified.

Images

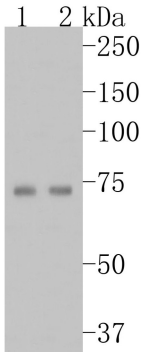

Fig1: Western blot analysis of Tryptophan Hydroxylase 1 (TPH1) on different lysates. Proteins were transferred to a PVDF membrane and blocked with 5% BSA in PBS for 1 hour at room temperature. The primary antibody (ET1610-37, 1/500) was used in 5% BSA at room temperature for 2 hours. Goat Anti-Rabbit IgG - HRP Secondary Antibody (HA1001) at 1:5,000 dilution was used for 1 hour at room temperature.
Positive control:
Lane 1: HL-60 cell lysate
Lane 2: Hela cell lysate

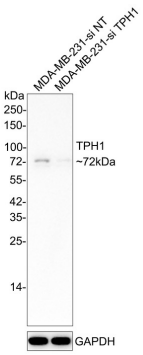

Fig2: Western blot analysis of TPH1 on different lysates with Rabbit anti-TPH1 antibody (ET1610-37) at 1/1,000 dilution.

Lane 1: MDA-MB-231-si NT cell lysate
Lane 2: MDA-MB-231-si TPH1 cell lysate

Lysates/proteins at 10 µg/Lane.

Predicted band size: 51 kDa
Observed band size: 72 kDa

Exposure time: 3 minutes;

4-20% SDS-PAGE gel.

ET1610-37 was shown to specifically react with TPH1 in MDA-MB-231-si NT cells. Weakened band was observed when MDA-MB-231-si TPH1 sample was tested. MDA-MB-231-si NT and Hela-si TPH1 samples were subjected to SDS-PAGE. Proteins were transferred to a PVDF membrane and blocked with 5% NFDM in TBST for 1 hour at room temperature. The primary antibody (ET1610-37, 1/1,000) and Loading control antibody (Rabbit anti-GAPDH, ET1601-4, 1/10,000) 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.

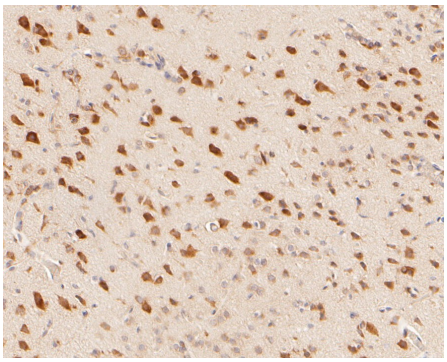

Fig3: Immunohistochemical analysis of paraffin-embedded rat brain tissue using anti-Tryptophan Hydroxylase 1 (TPH1) antibody. The section was pre-treated using heat mediated antigen retrieval with Tris-EDTA buffer (pH 9.0) for 20 minutes.The tissues were blocked in 5% BSA for 30 minutes at room temperature, washed with ddH_2O and PBS, and then probed with the primary antibody (ET1610-37, 1/50) for 30 minutes 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.

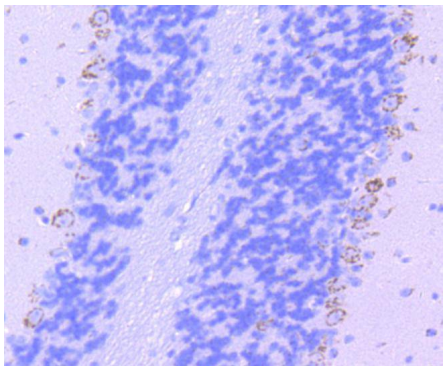

Fig4: Immunohistochemical analysis of paraffin-embedded mouse cerebellum tissue using anti-Tryptophan Hydroxylase 1 (TPH1) antibody. The section was pre-treated using heat mediated antigen retrieval with Tris-EDTA buffer (pH 8.0-8.4) for 20 minutes.The tissues were blocked in 5% BSA for 30 minutes at room temperature, washed with ddH_2O and PBS, and then probed with the primary antibody (ET1610-37, 1/50) for 30 minutes 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.

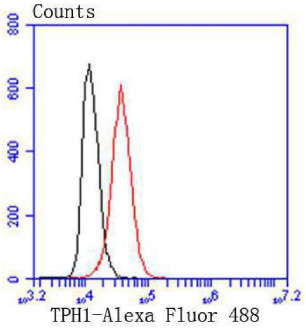

Fig5: Flow cytometric analysis of Tryptophan Hydroxylase 1 (TPH1) was done on Hela cells. The cells were fixed, permeabilized and stained with the primary antibody (ET1610-37, 1/50) (red). After incubation of the primary antibody at room temperature for an hour, the cells were stained with a Alexa Fluor 488-conjugated Goat anti-Rabbit IgG Secondary antibody at 1/1000 dilution for 30 minutes.Unlabelled sample was used as a control (cells without incubation with primary antibody; black).

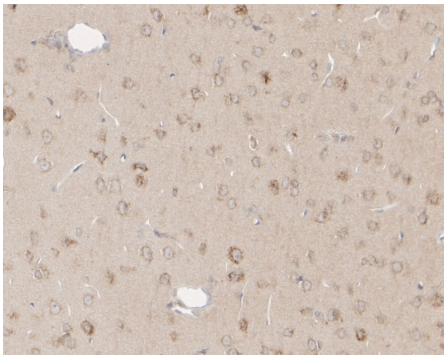

Fig6: Immunohistochemical analysis of paraffin-embedded rat brain tissue with Rabbit anti-Tryptophan Hydroxylase 1 (TPH1) antibody (ET1610-37) at 1/200 dilution.

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_2O and PBS, and then probed with the primary antibody (ET1610-37) 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.

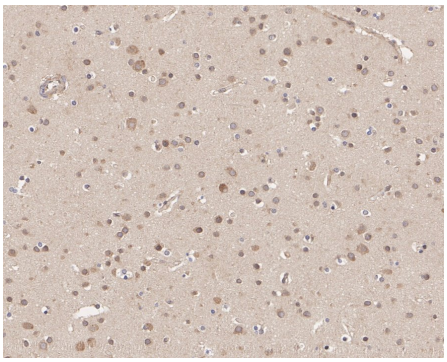

Fig7: Immunohistochemical analysis of paraffin-embedded human brain tissue with Rabbit anti-Tryptophan Hydroxylase 1 (TPH1) antibody (ET1610-37) at 1/200 dilution.

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_2O and PBS, and then probed with the primary antibody (ET1610-37) 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.

Background References

1. Cheng HH et al. Quiescent and proliferative fibroblasts exhibit differential p300 HAT activation through control of 5-methoxytryptophan production. PLoS One 9:e88507 (2014).
2. Dempsie Y et al. Dexfenfluramine and the oestrogen-metabolizing enzyme CYP1B1 in the development of pulmonary arterial hypertension. Cardiovasc Res 99:24-34 (2013).