Human/Mouse/Rat Tyrosine Hydroxylase Antibody

Antigen Affinity-purified Polyclonal Sheep IgG
Catalog Number: AF7566

DESCRIPTION

Species Reactivity	Human/Mouse/Rat
Specificity	Detects human, mouse and rat Tyrosine Hydroxylase in Western blots. In direct ELISAs, less than 5% cross-reactivity with recombinant human TPH1 is observed.
Source	Polyclonal Sheep IgG
Purification	Antigen Affinity-purified
Immunogen	*E. coli*-derived recombinant human Tyrosine Hydroxylase Ala278-Tyr401, predicted Accession # P07101
Formulation	Lyophilized from a 0.2 µm filtered solution in PBS with Trehalose. See Certificate of Analysis for details. *Small pack size (-SP) is supplied as a 0.2 µm filtered solution in PBS.

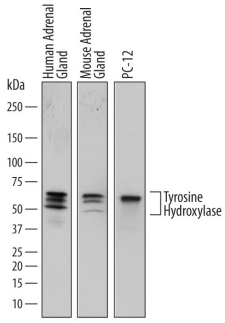
APPLICATIONS

Please Note: *Optimal dilutions should be determined by each laboratory for each application. General Protocols are available in the Technical Information section on our website.*

	Recommended Concentration	Sample
Western Blot	1 µg/mL	See Below
Immunocytochemistry	5-15 µg/mL	See Below

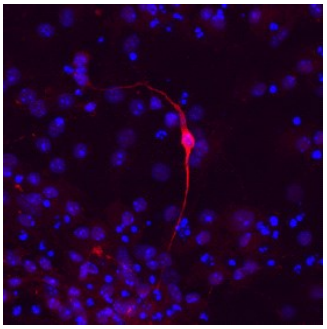
DATA

Western Blot



Detection of Human, Mouse, and Rat Tyrosine Hydroxylase by Western Blot. Western blot shows lysates of human adrenal gland tissue, mouse adrenal gland tissue, and PC-12 rat adrenal pheochromocytoma cell line. PVDF membrane was probed with 1 µg/mL of Sheep Anti-Human Tyrosine Hydroxylase Antigen Affinity-purified Polyclonal Antibody (Catalog # AF7566) followed by HRP-conjugated Anti-Sheep IgG Secondary Antibody (Catalog # HAF016). Specific bands were detected for Tyrosine Hydroxylase at approximately 56-60 kDa (as indicated). This experiment was conducted under reducing conditions and using Immunoblot Buffer Group 1.

Immunocytochemistry



Tyrosine Hydroxylase in D3 Mouse Embryonic Stem Cells. Tyrosine Hydroxylase was detected in immersion fixed D3 mouse embryonic stem cells using Sheep Anti-Human Tyrosine Hydroxylase Antigen Affinity-purified Polyclonal Antibody (Catalog # AF7566) at 10 µg/mL for 3 hours at room temperature. Cells were differentiated into dopaminergic neurons using R&D Systems Human/Mouse Dopaminergic Neuron Differentiation Kit (Catalog # SC001B). Cells were stained using the NorthernLights™ 557-conjugated Anti-Sheep IgG Secondary Antibody (red; Catalog # NL010) and counterstained with DAPI (blue). Specific staining was localized to dopaminergic neurons. View our protocol for Fluorescent ICC Staining of Cells on Coverslips.

PREPARATION AND STORAGE

Reconstitution	Sterile PBS to a final concentration of 0.2 mg/mL.
Shipping	The product is shipped at ambient temperature. Upon receipt, store it immediately at the temperature recommended below. *Small pack size (-SP) is shipped with polar packs. Upon receipt, store it immediately at -20 to -70 °C
Stability & Storage	**Use a manual defrost freezer and avoid repeated freeze-thaw cycles.** ● 12 months from date of receipt, -20 to -70 °C as supplied. ● 1 month, 2 to 8 °C under sterile conditions after reconstitution. ● 6 months, -20 to -70 °C under sterile conditions after reconstitution.

BACKGROUND

TH (Tyrosine 3-hydroxylase; also tyrosine 3-monooxygenase) is a 60-62 kDa member of the biopterin-dependent aromatic amino acid hydroxylase family of molecules. It is expressed by neurons of the dopamine and autonomic nervous system, plus the neuroendocrine cells of the adrenal medulla. TH is considered the rate limiting enzyme for catecholamine synthesis, and serves to catalyze the hydroxylation of L-tyrosine. It maintains stores of catecholamines following secretion, and its activity is regulated by targeted site phosphorylation. Human TH is 528 amino acids (aa) in length. It contains an N-terminal ACT domain (aa 69-190) that binds small molecules and regulates enzyme activity, and a C-terminal enzymatic region (aa 196-493). There are three significant utilized phosphorylation sites. Two at Ser31 and Ser40 increase enzyme activity, while phosphorylation at Ser19 promotes subsequent Ser40 phosphorylation. TH functions as a 240 kDa noncovalent homotetramer. There are four potential splice variants. One shows a deletion of aa 31-34, a second shows a deletion of aa 31-61, while a third contains a Met substitution for aa 1-34. A fourth isoform variant shows a deletion of aa 35-61. Over aa 278-401, human TH shares 94% aa sequence identity with mouse TH, a molecule that most closely resembles the fourth human isoform variant described above.