# Affinity Purified Rabbit Anti-Dopamine Transporter, C-Terminus Certificate of Analysis

#### ORDERING INFORMATION

Catalog Number: PPS068

Lot Numbers: 1686740

Size: 100 µL (sufficient for 10 mini-blots)

Storage: ≤ -20° C

- Specificity: Human and monkey ~88 kDa Dopamine Transporter (DAT)
- Immunogen: Peptide from the C-terminus region of human DAT, conjugated to keyhole limpet hemocyanin (KLH)

## Ig Type: rabbit IgG

Applications: Western blot Immunohistochemistry



Western blot of human caudate lysate showing specific immunolabeling of the ~88 kDa DAT protein.

#### Description

The dopamine transporter (DAT) is a 70 - 80 kDa member of the Na<sup>+</sup>-neurotransmitter symporter family of transmembrane (TM) proteins. DAT plays a crucial role in the synaptic clearance of dopamine (DA). It mediates the uptake of dopamine by the presynaptic terminal, thus limiting the strength of the dopamingeric response. One molecule of DA is accompanied by two Na<sup>+</sup> and one Cl<sup>-</sup> ion. Molecules such as amphetamine both competitively inhibit DA uptake, and induce DA release through the DAT, increasing the rewarding property of DA. Human DAT is a 620 amino acid (aa), 12 TM phosphoglycoprotein with an N- and C-terminal cytoplasmic domain. It exists as a disulfide-linked oligomer on the cell surface. Phosphorylation of the N-terminus (S7/12) promotes DA release. The C-terminus binds CaMKII, as well as Hic5, Pick1, and synuclein which regulate receptor trafficking and expression. There is an extended 71 aa extracellular (EC) loop between TM segments 3 and 4. Glycosylation at this site is necessary for oligomer expression on the cell membrane. The human 42 aa DAT C-terminus is 93% aa identical to the mouse DAT C-terminus. The human EC loop is also 93% aa identical to the rhesus monkey EC loop.

## Preparation

Prepared from rabbit serum by affinity purification using a Sulfo-Link<sup>®</sup> column matrix to which the peptide immunogen was coupled.

# Formulation

100 µL in 10 mM HEPES (pH 7.5), 150 mM NaCl, 100 µg/mL BSA and 50% glycerol.

#### Storage

For long-term storage,  $\leq$  -20° C is recommended. Product is stable at  $\leq$  -20° C for at least 1 year.

## Specificity

Specific for the ~88 kDa DAT protein in Western blots of SDS-solubilized human and mouse striatal samples and in IHC applications with formaldehyde-fixed human and monkey (Macaque) brain sections.

# Applications

Western blot - 1:1000 Immunohistochemistry - 1:1000 (frozen sections; unpublished observations)

Optimal dilutions should be determined by each laboratory for each application.

### References

- 1. Torres, G.E. (2006) J. Neurochem. 97(Suppl. 1):3.
- 2. Sotnikova, T.D. et al. (2006) CNS Neurol. Disord. Drug Targets. 5:45.
- 3. Fog, J.U. et al. (2006) Neuron 51:417.
- 4. Giros, B. et al. (1992) Mol. Pharmacol. 42:383.
- 5. Cervinski, M.A. et al. (2005) J. Biol. Chem. 280:40442.

**Quality & Regulatory Affairs** 

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