# PRODUCT INFORMATION



### Dopamine Transporter (Phospho-Thr<sup>53</sup>) Polyclonal Antibody

Item No. 29261

#### **Overview and Properties**

This vial contains 100 µl of affinity-purified polyclonal antibody from pooled serum. Contents:

Synonyms:

Immunogen: Phosphopeptide corresponding to amino acid residues surrounding phospho-Thr<sup>53</sup> of

Molecular Weight: ~55 kDa Species Reactivity: (+) Mouse, rat Storage: -20°C (as supplied)

Stability: ≥1 vear

Storage Buffer: 10 mM HEPES, pH 7.5, with 150 mM sodium chloride, 100 µg/ml BSA, and 50%

glycerol

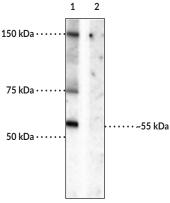
Host: Rabbit

**Applications:** Western blot (WB); the recommended starting dilution is 1:1,000. Other applications

were not tested, therefore optimal working concentration/dilution should be

determined empirically.

#### **Image**



Lane 1: DAT protein phosphorylated at Thr<sup>53</sup>

Lane 2: Preadsorption of the phosphopeptide used as the antigen

WB of rat striatal lysate showing specific immunolabeling of the ~55 kDa glycosylated form of the DAT protein phosphorylated at Thr $^{53}$  in Lane 1. Phosphospecificity is shown in Lane 2 where immunolabeling is blocked by preadsorption of the phosphopeptide used as the antigen, but not by the corresponding non-phosphopeptide (not shown).

WARNING
THIS PRODUCT IS FOR RESEARCH ONLY - NOT FOR HUMAN OR VETERINARY DIAGNOSTIC OR THERAPEUTIC USE.

This material should be considered hazardous until further information becomes available. Do not ingest, inhale, get in eyes, on skin, or on clothing. Wash thoroughly after handling. Before use, the user must review the complete Safety Data Sheet, which has been sent via email to your institution.

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#### Description

The dopamine transporter (DAT) is a member of the SLC6 family of transporters and is encoded by the *SLC6A3* gene in humans.<sup>1,2</sup> DAT is expressed in dopaminergic neurons and localizes to perisynaptic sites, where it functions to translocate the neurotransmitter dopamine (DA; Item No. 21992) into presynaptic neurons from the extracellular space.<sup>1,3</sup> It is comprised of 12 transmembrane helices flanked by large cytoplasmic N- and C-terminal tails that are subject to protein-protein interactions with DAT binding proteins as well as posttranslational modifications.<sup>1,3</sup> DAT can be phosphorylated by ERK at threonine 53 (Thr<sup>53</sup>) *in vitro*.<sup>4</sup> Phosphorylation of DAT at Thr<sup>53</sup> (phospho-Thr<sup>53</sup>) is increased in rat striatal synaptosomes in response to phorbol 12-myristate 13-acetate (PMA; Item No. 10008014), okadaic acid (Item No. 10011490), amphetamine, or methamphetamine.<sup>4,5</sup> *In vivo*, phospho-Thr<sup>53</sup> in rat striatum is increased following administration of methamphetamine.<sup>5</sup> LLC-PK1 cells expressing rat DAT (rDAT) with non-phosphorylatable or phosphomimetic Thr<sup>53</sup> mutations exhibit defects in DA uptake and amphetamine-induced efflux of the substrate [<sup>3</sup>H]MPP+ compared to cells expressing wild-type rDAT, indicating that phosphorylation of Thr<sup>53</sup> has roles in regulating the uptake and efflux functions of DAT.<sup>4</sup> Cayman's Dopamine Transporter (Phospho-Thr<sup>53</sup>) Polyclonal Antibody can be used for Western blot (WB) applications. The antibody recognizes DAT (phospho-Thr<sup>53</sup>) at approximately 55 kDa from mouse and rat samples.

#### References

- 1. Vaughan, R.A. and Foster, J.D. Mechanisms of dopamine transporter regulation in normal and disease states. *Trends Pharamacol. Sci.* **34(9)**, 489-496 (2013).
- 2. Michelhaugh, S.K., Fiskerstrand, C., Lovejoy, E., et al. The dopamine transporter gene (SLC6A3) variable number of tandem repeats domain enhances transcription in dopamine neurons. J. Neurochem. 79(5), 1033-1038 (2001).
- 3. Torres, G.E. The dopamine transporter proteome. J. Neurochem. 97(Suppl. 1), 3-10 (2006).
- Foster, J.D., Yang, J.-W., Moritz, A.E., et al. Dopamine transporter phosphorylation site threonine 53 regulates substrate reuptake and amphetamine-stimulated efflux. J. Biol. Chem. 287(35), 29702-29712 (2012).
- 5. Challasivakanaka, S., Zhen, J., Smith, M.E., *et al.* Dopamine transporter phosphorylation site threonine 53 is stimulated by amphetamines and regulates dopamine transport, efflux, and cocaine analog binding. *J. Biol. Chem.* **292(46)**, 19066-19075 (2017).

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