This Week in The Journal

Dopamine Transporter Is Rarely Endocytosed in Striatum

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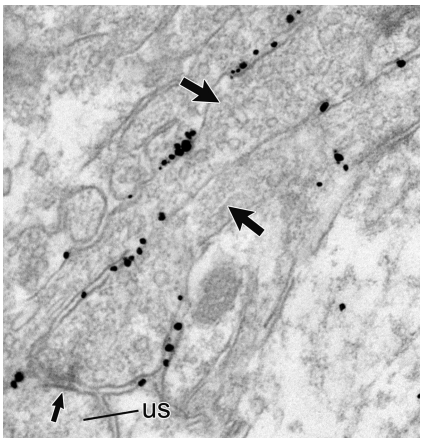
(see pages 12845–12858)

The dopamine transporter (DAT) limits the spatial and temporal spread of dopaminergic signaling by taking up extracellular dopamine. This function is thought to be modulated by endogenous signaling pathways. For example, phosphorylation of DAT by protein kinase C (PKC) reduces dopamine uptake, whereas phosphorylation by extracellular signal-regulated kinase increases uptake. Although phosphorylation might alter uptake by changing the V_{max} of the transporter, PKC-mediated phosphorylation has been proposed to reduce dopamine uptake primarily by promoting DAT endocytosis. It has further been proposed that sorting of endocytosed DAT to recycling endosomes underlies short-term reductions in uptake, whereas targeting the transporter for degradation in lysosomes underlies longer term modulation (Vaughan and Foster, 2013, Trends Pharmacol Sci 34:489).

To investigate these hypotheses and examine which endocytic pathways predominate under physiological conditions, Block et al. used knock-in mice expressing hemagglutinin-tagged DAT and examined DAT localization in acute brain slices and by electron microscopy. In the midbrain, most DAT (~75%) was associated with intracellular membranes, particularly within somata, where it was found primarily on nuclear, endoplasmic reticulum, and Golgi membranes. DAT was occasionally colocalized with markers of early, recycling, or intermediate/sorting endosomes in vesicular structures in the midbrain, but it never colocalized with markers of late endosomes or lysosomes.

Plasma membrane expression of DAT was higher in dopaminergic axons in the striatum than in the midbrain. Furthermore, DAT density was greater in axonal varicosities (putative presynaptic sites) than along intervening shafts, and most axonal DAT (~85%) was associated with the plasma membrane. Surprisingly, markers of early and recycling endosomes were rarely observed in dopaminergic axons, and DAT never colocalized with these markers. Moreover, markers of intermediate/sorting endosomes and lysosomes were never detected in dopaminergic axons. Finally, although amphetamine can induce DAT endocytosis in cultured neurons, amphetamine administration *in vivo* or in slices did not significantly change the subcellular distribution of DAT.

These results indicate that constitutive endocytosis rarely occurs in the axons of dopaminergic neurons, and when it does occur, internalized proteins are likely recycled to the plasma membrane rather being degraded in lysosomes. Thus, modulation of dopamine uptake must be regulated primarily by changes in DAT V_{max} or lateral diffusion of DAT within the membrane.



DAT is expressed primarily in the plasma membranes of axons (large arrows) in the striatum, but is excluded from synaptic active zones (small arrow). An unlabeled spine (us) is indicated. See Block et al. for details.

Transcranial Direct Current Stimulation Enhances LTP

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(see pages 12824–12832)

Transcranial direct current stimulation (tDCS) is increasingly being used to study and/or enhance human brain function. Anodal tDCS applied to different cortical regions has been reported to enhance sensory perception, motor learning, motor performance, working and episodic memory, and other cognitive functions. Importantly, these improvements often persist for several hours after stimulation. Although effects vary across studies and are often limited to select task elements and/or groups of participants, it is hoped that tDCS might ultimately be used to enhance recovery from injury, treat neuropsychiatric conditions, and improve cognitive performance.

How tDCS exerts its effects on brain function is poorly understood. Effects during the stimulation period are thought to be limited to changes in neuronal membrane potential, specifically depolarization in the case of anodal stimulation. Depending on its duration and intensity, however, anodal tDCS can also cause an increase in cortical excitability that persists for several hours after stimulation ends. This persistent effect requires activation of voltage-sensitive Na^+ channels, Ca^{2+} channels, and NMDA receptors. It has therefore been suggested that tDCS enhances long-term potentiation (LTP; reviewed in Stagg and Nitsche, 2011, Neuroscientist 17:37).

Rohan et al. confirm this hypothesis. Rats were given anodal tDCS or sham treatment for 30 min, and brain slices were taken 0.5 or 24 h later. Anodal tDCS did not affect spontaneous activity or the size of field EPSPs (fEPSPs) evoked by electrical stimulation of Schaffer collaterals in the hippocampus. But theta-burst stimulation of Schaffer collaterals caused a greater increase in the slope and amplitude of subsequently evoked fEPSPs in tDCS-treated rats than in controls. This enhancement in LTP was still present 24 h after stimulation, and it was blocked by an NMDA receptor antagonist. tDCS also enhanced paired-pulse facilitation, but this effect neither persisted for 24 h nor required activation of NMDA receptors.

These data suggest that tDCS enhances LTP and affects presynaptic release probability at Schaffer collateral terminals in hippocampus. Perhaps the most important contribution of this study, however, is the demonstration that the effects of *in vivo* tDCS can be studied in slices taken 0.5–24 h after stimulation. This should simplify future investigations into the cellular mechanisms underlying the effects of tDCS.

This Week in The Journal is written by Teresa Esch, Ph.D.