This Week in The Journal

Tracking the Cellular Roots of Associative Learning in Flies

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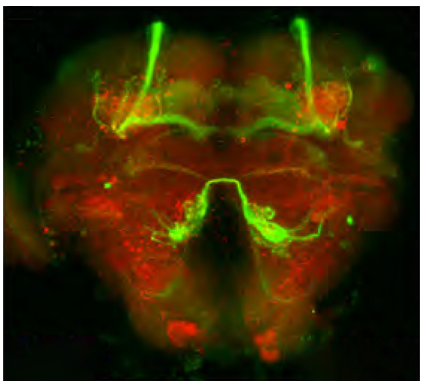
(see pages 5274–5286)

Neurofibromin (NF1) is a cytoplasmic protein with a variety of cellular regulatory functions; mutations in *NF1* result in neurofibromatosis, a developmental disorder that causes cognitive impairment among other symptoms. In *Drosophila melanogaster*, loss of the fly ortholog dNf1 leads to deficits on tasks requiring conditioned responses to aversive odors. Georganta et al. investigated the cells and signaling pathways mediating these effects and discovered unexpected contributors.

Mushroom body (MB) neurons are crucial to olfactory learning in flies, and they contain dNf1, so researchers suspected them as the critical site for dNf1 loss. But restoration of dNf1 to MB neurons alone did not rescue the deficits seen in dNf1-null animals; other neurons known to be involved in olfactory learning, including cholinergic neurons, GABAergic anterior-paired-lateral neurons, and antennal lobe projection neurons were also excluded as the critical cells. Nevertheless, when dNf1 was expressed exclusively in neurons that express the marker OK72, learning was restored in adult flies. Conversely, knocking down dNf1 in OK72-expressing neurons in wild-type flies led to a potent learning deficit.

OK72 is expressed in a limited subset of neurons in various brain regions, including the MB. Importantly, restoration of dNf1 in non-MB OK72 neurons was sufficient to rescue associative learning. Restoration of dNf1 in all GABAergic neurons also rescued learning deficits in dNf1-null flies, as did knocking down glutamic acid decarboxylase, the enzyme required for GABA production. The authors conclude that a novel circuit including GABAergic OK72-expressing neurons synapsing on MB neurons contributes to associative learning. They hypothesized that learning was compromised by elevated GABA production or release by these neurons when dNf1 was depleted.

Previous work has suggested that cAMP, which is regulated by dNf1, is responsible for developmental delays in dNf1 mutants. But elevating cAMP by expressing constitutively active protein kinase A or by inhibiting phosphodiesterase in either OK72-expressing or MB neurons did not restore learning in mutant flies. In contrast, expressing a dominant negative form of the receptor tyrosine kinase dAlk or knocking down the transcriptional-control protein Ras1 in OK72 neurons restored learning in mutant flies. These results suggest that hyperactivation of dAlk-to-Ras1 signaling in the absence of dNf1 leads to increased GABA release, which impairs learning.



Staining of an optical slice of fly brain shows presynaptic OK72 GABAergic neurons (green) and postsynaptic MB neurons (red). See Georganta et al. for details.

Cortical Astrocytes Modulate Diabetic Neuropathic Pain in Rats

Jingshan Lu, Lan Yang, Ying Xu, Lijing Ai, Jian Chen, et al.

(see pages 5287–5302)

Approximately one-third of patients with type 2 diabetes develop neuropathic pain, a debilitating condition with few treatment options. Astrocytes play a role in pain modulation in the spinal cord, but supraspinal glia have received less attention. Now Lu, Yang, Xu, Ai, et al. have used a chemogenetic strategy to demonstrate a surprising role for motor cortex astrocytes in pain behaviors in diabetic rats.

Rats injected with a drug that induces diabetes developed persistent neuropathic pain, as indicated by increased sensitivity to a poke of the paw. Immunostaining showed that diabetic rats had increased staining for glial fibrillary acid protein, a marker of astrocytes and some other glial cells. Chronic inhibition of motor cortex astrocytes with a designer receptor exclusively activated by designer drugs (DREADD) alleviated mechanical hypersensitivity in diabetic rats. Conversely, DREADD-mediated activation of astrocytes produced mechanical hypersensitivity in naïve rats.

Neuronal activity (indicated by c-Fos labeling) was reduced after DREADD-mediated inhibition of astrocytes, whereas neuronal activity was increased when astroctyes were activated. Notably, ~80% of cells activated in response to astrocyte activity expressed calcium/calmodulin-dependent protein kinase II, a marker of excitatory neurons. Moreover, DREADD-mediated inhibition of excitatory motor cortex neurons attenuated mechanical hypersensitivity in diabetic rats, whereas DREADD-mediated activation of these neurons produced mechanical hypersensitivity in naive rats. Pain-like behaviors developed in rats expressing the excitatory DREADD in both glia and excitatory neurons, but not in rats expressing an excitatory glial receptor and an inhibitory neuronal receptor.

Neuroinflammation in the motor cortex may underlie the modulation of pain-related behaviors by astroctyes. Indeed, expression of the proinflammatory cytokines TNF-α and IL-β were higher in diabetic rats than in controls. Levels decreased after DREADD-mediated inhibition of astrocytes and increased after astrocyte activation. Together, the data suggest that astrocyte activation leading to increased excitatory neural activity and neuroinflammation contributes to diabetic neuropathic pain.

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