Small G Proteins and Neurologic Disease

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Tuberous sclerosis and Rheb

This is a fascinating story of research that unfolds a biochemical process in regulating growth. Tuberous sclerosis is a multisystemic tumor syndrome inherited as an autosomal dominant trait manifested by numerous benign tumors (hamartomas) that develop most often in the brain, kidneys, skin, heart and lungs. The most critical morbidity results from these tumors in the brain resulting in epilepsy, autism and mental retardation; angiomyolipomas in kidney; and lymphangioleiomyomatosis in lung. The cause is a mutation in either of the tuberous sclerosis complex genes, *TSC1* (OMIM #605284) or *TSC2* (OMIM #191092). These were first identified as tumor suppressor genes.

The gene products, hamartin from *TSC1* and tuberin from *TSC2*, associate to form what is thought to be a heterodimeric complex, TSC1–TSC2. TSC1 is required to stabilize TSC2 and prevent its ubiquitin-mediated degradation while TSC2 acts as a GTPase-activating protein (GAP). Missense mutation in the TSC2 GAP domain affects its GAP function and mutations in either TSC1 or TSC2 commonly destabilize the complex, thereby leading to TSC2 ubiquitination and degradation. The main function of the TSC1–TSC2 complex is the negative regulation of the mTORC1 (mammalian target of rapamycin complex 1), which itself promotes cell growth via several processes including activation of S6K (ribosomal S6 kinase) and inhibition of 4E-BP (a protein translation inhibitor) (Huang & Manning, 2008). TSC1–TSC2 complex overexpression inhibits mTORC1-mediated phosphorylation of S6K1 and 4E-BP1 while this phosphorylation is increased by the lack of TSC1–TSC2 complex.

How does TSC1–TSC2 control mTORC1 activation? Through the biochemistry and

genetic studies of several groups, the molecular switch in this case was identified as the small G protein Rheb (Ras Homologue Enriched in Brain). The GAP domain of TSC2 in the TSC1–TSC2 complex stimulates the GTPase activity of Rheb converting Rheb-GTP to Rheb-GDP. Rheb-GTP leads to activation of mTORC1 so that the conversion to Rheb-GDP decreases the activation. Thus, in the absence of functional TSC1–TSC2 complex, both the ratios of Rheb-GTP to Rheb-GDP and of active to inactive mTORC1 remain high. However, the molecular mechanism by which Rheb-GTP controls mTORC1 activity is not yet clear.

Since Rheb-GTP-mTORC1 appears to be the main switching locus for stimulating tumor growth, potential therapies for tuberous sclerosis, and possibly other tumors, may ensue from discovery of inhibitors of mTORC1, such as rapamycin, or inhibitors of the Rheb-GTP activating effect on mTORC1. For the interested reader, there is an excellent, detailed review of the biochemical and genetic work elucidating the biochemical pathways discussed above (Huang & Manning, 2008).

Hereditary sensory-motor neuropathy (HSMN) and Rab

HSMN is a heterogeneous group of inherited peripheral neuropathies with length-dependent sensory, motor and autonomic nerve dysfunction in various combinations caused by mutations in a number of different genes (see Ch. 38). One such syndrome, HMSN2 (Charcot-Marie-Tooth type 2B), is characterized by dominantly inherited peripheral nerve axon degeneration without demyelination and with normal conduction velocities, in contrast to HMSN1 in which there is demyelination and slowed conduction.

HMSN2 produces sensory and autonomic neuropathy, which leads to ulcerations and mutilation. It is caused by mutations in the gene for Rab7 [OMIM #602298], a member of the Rab family of about 70 proteins in the Ras super family. Rab proteins regulate vesicular

transport, maturation and fusion of vesicles with membranes. Activation and deactivation of the downstream effectors ensue from the cycling of Rab through its active GTP-bound form, hydrolysis of GTP by its GTPase function and to its inactive GDP-bound form. This Rab cycling is tightly regulated by other regulatory proteins so that its signaling is orchestrated with other signaling pathways. Rab regulation is involved in ensuring cargo unloading at correct locations and vesicle recycling at specialized target membranes, such as in presynaptic nerve terminals and in retrograde transport of neurotrophins (types of nerve growth factors). Rab7 is located in the late endosome and controls vesicular transport to late endosomes and lysosomes in the endocytic pathway (Vanlandingham & Ceresa, 2008; Zweifel et al., 2005).

Disease-associated mutations in Rab7 result in increased activation of the downstream signaling pathway, increased GTP binding to Rab, and dysregulation of GDP–GTP exchange on the protein with increased proportions of GTP- relative to GDP-bound Rab which is the probable cause of excessive and dysregulated activation. There is hydrolysis-independent inactivation, abnormal retention of Rab7 on target membranes, and dysregulated, hydrolysis-independent membrane cycling. The abnormal Rab7 function leads to spatiotemporal dysregulation of vesicle traffic and cycling (Cogli et al., 2009; McCray et al., 2010).

These genetic disturbances help elucidate the specific molecular reactions critical to axonal function and which may be dysfunctional in acquired as well as inherited neuropathies. These biochemicals may be targeted for pharmacologic therapy.