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Control of neuronal survival, migration and outgrowth by GDNF and its receptors

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ABSTRACT

Glial cell line-derived neurotrophic factor (GDNF) is the prototypical member of a family of growth factors that are indispensable in nervous system development and maintenance. GDNF signals by binding to a multi-component receptor complex comprised of the ligand-binding subunit GFRα1 and the signaling subunit RET or NCAM. While initial interest in GDNF was merely focused on its potential therapeutic effects in Parkinson's disease, it has rapidly become evident that the mechanisms by which GDNF and its receptors regulate diverse physiological events are incomplete. Thus, we have demonstrated new insights into the control of neuronal survival, migration, and outgrowth by GDNF and its receptors.

To begin clarifing the complexity of GDNF signaling, we examined RET intracellular tyrosine residues which become phosphorylated upon receptor activation, and serve as docking sites for downstream signaling effectors. The functions of most of these phosphorylated tyrosine residues are still unknown. In paper I, we identified the protein-tyrosine phosphatase SHP2 as a novel direct interactor of RET and as the first effector known to bind to phosphorylated Tyr^{687}. Furthermore, we found that activation of protein kinase A (PKA) by forskolin reduced the recruitment of SHP2 to RET, negatively affecting ligand-mediated neurite outgrowth. Together, these findings establish SHP2 as a novel positive regulator of RET function and reveal Tyr^{687} as a critical platform for integration of RET and PKA signals.

To continue to understand GDNF signaling diversity, we examined RET signaling in a clinical context. Most patients with medullary thyroid carcinoma (MTC) and type 2A multiple endocrine neoplasia (MEN2A) exhibit cysteine residue mutations in the juxtamembrane region of RET which result in unexplained oncogenic activation. Thus, in paper II we identified and described the self-association determinants of the RET transmembrane (RET-TM) domain underlying such oncogenic activation by mutations found in these patients. We found that strong propensity for RET-TM self-association underlies - and may be required for – dimer formation and oncogenic activation by juxtamembrane cysteine mutations localized in close proximity to the plasma membrane in MTC and MEN2A syndromes.

To further dissect the many facets of GDNF signaling, we alternatively investigated the functions of GDNF and GFRα1 independent of RET. GDNF promotes the differentiation and migration of GABAergic neuronal precursors from the medial ganglionic eminence (MGE). These functions are dependent on GFRα1, but are independent of the two known receptor partners RET and NCAM. In paper III, we revealed that soluble GFRα1 is able to promote GABAergic differentiation and migration, but requires endogenous GDNF production. Furthermore, we showed that MET signaling inhibition promoted the same physiological response as GDNF. Finally, we justified the existence of a novel transmembrane receptor for the GDNF/GFRα1 complex and uncovered an unexpected interplay between GDNF/GFRα1 and HGF/Met signaling in the early diversification of GABAergic MGE interneuron subtypes.