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A RABL6A-PP2A-AKT Pathway Drives Pancreatic Neuroendocrine Tumor Growth

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BACKGROUND: Hyperactivated AKT/mTOR signaling is a hallmark of pancreatic neuroendocrine tumors (PNETs). Drugs targeting this pathway are used clinically but tumor resistance invariably develops. A better understanding of factors regulating AKT/mTOR signaling and PNET pathogenesis is needed to improve current therapies. Our data suggests that RABL6A, a new oncogenic driver of PNET proliferation, is a key regulator of this clinically relevant pathway.

METHODS: Transcript levels were assayed by microarray and RNA-Seq in human cells and tissues, proteins by immunoblotting and IHC, and cell proliferation/survival by cell counts, trypan blue exclusion and commercial kits. RABL6A was silenced in cells by lentiviral shRNAs, AKT overexpressed by retroviral delivery, and PP2A activity inhibited with okadaic acid or activated with SMAP. BON-1 xenografts in mice were used for in vivo drug studies with SMAP and MK-2206 (AKT inhibitor).

RESULTS: We compared gene expression profiles from patient tumors and RABL6A depleted cells and found that RABL6A expression correlated with AKT pathway activation. Silencing RABL6A caused PNET cell cycle arrest that coincided with selective loss of AKT-S473 (not T308) phosphorylation, AKT/mTOR inactivation, and desensitization to an AKT inhibitor. Mechanistically, loss of AKT-S473 phosphorylation in RABL6A depleted cells resulted from increased

activation of protein phosphatase 2A (PP2A), a powerful tumor suppressor and known inhibitor of AKT. Inactivation of PP2A with okadaic acid restored AKT-S473 phosphorylation in RABL6A depleted cells whereas PP2A reactivation using a specific small molecule activator of PP2A (SMAP) abolished that phosphorylation. Importantly, SMAP treatment killed PNET cells in a RABL6A-dependent manner and suppressed PNET growth in vivo.

CONCLUSION: This work identifies RABL6A as a novel activator of the AKT-mTOR pathway and inhibitor of the PP2A tumor suppressor. Expression of RABL6A dictates PNET cell response to clinically relevant drugs targeting those pathways. Our findings offer novel targets, PP2A and RABL6A, for anticancer therapy in PNET patients.