

Neuroblastoma

Major finding: A GPC2-targeting antibody–drug conjugate promotes tumor regression in a high-risk neuroblastoma PDX.

Approach: An RNA-seq pipeline identifies candidate cell-surface targets specifically expressed in neuroblastoma.

Impact: Immunotherapeutic GPC2 targeting may be beneficial in high-risk neuroblastoma and other embryonal tumors.

GPC2 MAY BE AN IMMUNOTHERAPEUTIC TARGET IN HIGH-RISK NEUROBLASTOMA

Activating mutations in druggable targets are rare in neuroblastoma, supporting investigation of other therapeutic strategies. Neuroblastoma arises from neural crest progenitor cells in the developing sympathetic nervous system, and thus expresses targets not found on mature cells, suggesting the possibility for the development of immunotherapies. Using an RNA-sequencing (RNA-seq)-based pipeline to analyze 126 primary high-risk neuroblastomas, Bosse and colleagues identified candidate genes for optimal immunotherapeutic targeting. The putative targets were cell-surface proteins (or proteins with extracellular epitopes) highly expressed in neuroblastoma compared with normal tissues, and expressed in the majority of tumors. The extracellular glycosylphosphatidylinositol anchored signaling co-receptor glypican 2 (GPC2) was selected as a top hit. MYCN bound to the GPC2 promoter to increase its expression in neuroblastoma, thereby upregulating GPC2 in MYCN-amplified tumors. Further, in approximately 40% of high-risk neuroblastomas somatic gain of chromosome 7q, which harbors the GPC2 locus, promoted enhanced GPC2 expression. GPC2 was highly expressed on the plasma membrane of the majority of neuroblastoma tumors, and its expression was limited in normal tissues, suggesting the possibility for therapeutic targeting with

minimal toxicity. Moreover, GPC2 was required for neuroblastoma cell proliferation, and its depletion resulted in apoptosis. The dependency of neuroblastoma cells on GPC2 expression may prevent the emergence of immune escape mechanisms in response to therapeutic targeting. A GPC2-targeting antibody–drug conjugate was developed and was cytotoxic to neuroblastoma cells expressing GPC2 *in vitro*. Further, the antibody–drug conjugate promoted durable tumor regression in a patient-derived xenograft (PDX) model of MYCN-amplified neuroblastoma, extending survival with minimal toxicity. Additionally, GPC2 is expressed in other pediatric embryonal malignancies including medulloblastoma and retinoblastoma, suggesting that GPC2 may be an immunotherapy target in other tumor types. Collectively, these findings suggest that GPC2 may be an optimal target for immunotherapy in high-risk neuroblastoma and other embryonal tumors, supporting further development of GPC2-directed immunotherapeutics. ■

Bosse KR, Raman P, Zhu Z, Lane M, Martinez D, Heitzeneder S, et al. Identification of GPC2 as an oncoprotein and candidate immunotherapeutic target in high-risk neuroblastoma. *Cancer Cell* 2017;32:295–309.e12.

Leukemia

Major finding: Tigecycline inhibits mitochondrial oxidative phosphorylation to target leukemic stem cells (LSC).

Concept: Tigecycline targets CD34⁺CD38[−] CML cells with little effect on nonleukemic CD34⁺CD38[−] cells.

Impact: Imatinib plus tigecycline may target LSCs and differentiated CML cells to suppress CML relapse.

TIGECYCLINE MAY SELECTIVELY TARGET LEUKEMIC STEM CELLS IN CML

ABL-specific tyrosine kinase inhibitors (TKI), including imatinib mesylate, have improved outcomes in patients with chronic myeloid leukemia (CML). However, these drugs primarily target differentiated cells, and persistent residual leukemic stem cells (LSC) can promote resistance or relapse. In order to identify potentially targetable metabolic vulnerabilities in CML LSCs, Kuntz and colleagues performed metabolic profiling to detect steady-state levels of 70 metabolites in CD34⁺ stem-cell enriched CML cells and CD34⁺ differentiated CML cells from four patients with CML, compared with normal hematopoietic CD34⁺ cells from healthy donors. The CD34⁺ CML population exhibited an increase in oxidative metabolism with an increase in glycerol-3-phosphate, carnitine, and acylcarnitine derivatives, and a decrease in free fatty acids, suggesting an increase in lipolysis and fatty-acid oxidation. Moreover, CD34⁺ cells had increased mitochondrial oxygen consumption, an increase in glucose oxidation, and an increase in anaplerosis (reactions that replenish tricarboxylic acid cycle intermediates). Based on these findings, CML LSCs were treated with tigecycline, an FDA-approved antibiotic that inhibits



the synthesis of mitochondrial proteins required for oxidative phosphorylation. Single-agent tigecycline inhibited oxidative metabolism and anaplerosis in CD34⁺ CML cells and suppressed their proliferation. Combined treatment with imatinib and tigecycline eliminated colony formation in CD34⁺ CML cells and stringent stem cell assays, but exhibited relatively little toxicity to nonleukemic cells. In a human CML xenotransplantation model, treatment with imatinib plus tigecycline eliminated CD34⁺CD38[−] CML LSCs and reduced signs of relapse after drug withdrawal compared with single-agent imatinib. The finding that CML LSCs are sensitive to disruption of oxidative phosphorylation, whereas normal CD34⁺CD38[−] cells are not, suggests the possibility for combination therapy with tigecycline and imatinib to eliminate LSCs and differentiated CML cells to potentially prevent relapse in patients with CML. ■

Kuntz EM, Baquero P, Michie AM, Dunn K, Tardito S, Holyoake TL, et al. Targeting mitochondrial oxidative phosphorylation eradicates therapy-resistant chronic myeloid leukemia stem cells. *Nat Med* 2017 Sep 18 [Epub ahead of print].