Targeting HSP90 sensitizes pancreas carcinoma to PD-1 blockade

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ABSTRACT
Interferon gamma (IFNG/IFNγ)-induced adaptive immune resistance remains a challenge for tumor therapy. We observed that the chaperone heat shock protein 90 (HSP90) stabilizes the transcription factor signal transducer and activator of transcription 1 (STAT1), resulting in IFNγ-induced expression of immunosuppressive indoleamine 2,3-dioxigenase 1 (IDO1) and programmed death-ligand 1 (PD-L1/CD274). Pharmacological inhibition of HSP90 enhances the efficacy of programmed cell death 1 (PDCD1/PD-1) targeting immunotherapy in suitable mouse models without any toxicity.

Next, we used the KRAS and tumor protein p53 (TP53, best known as p53)-mutated human PDAC cell line CFPAC1 to screen for compounds that inhibit IFNγ-induced IDO1 as well as expression of CD274 molecule (best known as programmed death-ligand 1 [PD-L1]). Of note, we found that 24 compounds (used at 10 µM) that blocked IFNγ-induced IDO1 and PD-L1 expression in CFPAC1 cells. The vast majority (71%) of these agents were HSP90 inhibitors. In addition, nanomolar amounts of six HSP90 inhibitors (luminespib, ganetespib, SNX-2112, PF-04929113, HSP990 and XL888) suppressed IFNγ-induced IDO1 and PD-L1 expression in 16 human tumor cell lines (corresponding to 11 different types of cancer) and primary PDAC cells from patients and KPC mice. Mechanistically, we demonstrated that the binding of HSP90 to its partner SGT1 homolog, MIS12 kinetochore complex assembly cochaperone (SUGT1) resulted in increased protein stability of STAT1, a key transcription factor for the expression of immune checkpoint molecules. Expression of dominant-negative HSP90 (D88N) led to inhibition of STAT1-mediated IFNγ signaling, suggesting that the aforementioned HSP90 inhibitors act on target. Altogether, these results point to a broad role for HSP90 in mediating the expression of inducible immune checkpoint molecules.

IDO1 is a rate-limiting metabolic enzyme that converts the essential amino acid tryptophan to a downstream immunosuppressor, kynurenine, thereby inhibiting the proliferation and function of cytotoxic CD8+ T cells. However, the mechanism of action of IDO1-mediated kynurenine production and secretion remains poorly understood. We found that tetraspanin 5 (TSPAN5), a transmembrane protein of the tetraspanin family, plays a critical role in mediating IFNγ-induced kynurenine secretion, but not IFNγ-induced IDO1 expression. We also
verified that the enzymatic activity of IDO1 requires iron (but not other metal ions) to trigger kynurenine production. Consequently, iron-enhanced kynurenine release (but not kynurenine synthesis) was inhibited in TSPAN5-deficient CFPAC1 cells. These findings reveal a role for iron in promoting IDO1-dependent kynurenine production and subsequent TSPAN5-mediated kynurenine release.

Finally, we evaluated the efficacy and safety of combinations of aPD-1 with the HSP90 inhibitor ganetesplib, the IDO1 inhibitor BMS-986205, or the iron chelator desferoxamine in the treatment of transplanted or a transgene-induced PDAC. Compared with the aPD-1 alone group, the combination of aPD-1 with ganetespib, BMS-986205, or desferoxamine was more efficient in reducing tumor growth and enhancing the infiltration of PDAC by CD8+ T cells and dendritic cells (but not CD4+ T cells, macrophages, and natural killer cells). Depletion of CD8+ T cells abolished desferoxamine and aPD-1-mediated tumor suppression, demonstrating that this combination therapy relies on the contribution of tumor-specific cytotoxic T lymphocytes. This new combination therapy had an acceptable safety profile and did not affect liver and kidney function in mice.

In summary, our study uncovers a new strategy through which PDAC cells hide from the immune system. Moreover, IDO1 emerges as a potential biomarker for predicting treatment responses to anti-PD-1. We provide proof-of-concept for future clinical applications of HSP90 inhibitors, IDO1 inhibitors, or iron chelators to enhance the anticancer activity of PD-1 blockade. In addition to STAT1, the expression of immune checkpoint molecules is controlled by other transcriptional factors, such as TP53, hypoxia inducible factor 1 subunit alpha, and MYC. It is important to further define the crosstalk of gene transcription and protein degradation pathways in coordinating the expression of immune checkpoint molecules by PDAC cells. Potentially, distinguishing different clients of the HSP90 chaperone machinery in tumor immunity remains a challenge. Regardless, the hypothesis that inducers of immunogenic stress and death, including HDP90 inhibitors, may sensitize PDAC cells to immunotherapy should be explored in future clinical assays.

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