830 INHIBITION OF MACROPHAGE MIGRATION INHIBITORY FACTOR (MIF) TO OVERCOME IMMUNE CHECKPOINT RESISTANCE IN MELANOMA

[1]Gabriela Athziri Sanchez-Zuno*, [2]Jasmine Caulfield, [1]Lin Leng, [2]Lin Zhang, [2]Lucia Jilaveanu, [1,3]Harriet Kluger, [1,2]Richard Bucala, [4]Thuy Tran. *[1]Yale University, School of Medicine, New Haven, CT, USA; [2]Yale Smilow Cancer Center, New Haven, CT, USA; [3]Yale Cancer Center, New Haven, CT, USA; [4]Yale University, New Haven, CT, USA*

Background Immune checkpoint inhibitors (ICIs) have shown efficacy in melanoma treatment, but resistance and toxicities remain ongoing challenges. Macrophage migration inhibitory factor (MIF) is an immunoregulatory cytokine implicated in melanoma progression and angiogenesis. This study investigates targeting MIF in combination with anti-programmed cell death 1 (PD-1) to overcome immune resistance and elucidates the underlying molecular mechanisms of anti-tumor activity.

Methods We evaluated the in vivo effects of anti-MIF using subcutaneous YUMMER1.7 melanoma and MC38 colorectal carcinoma models. Animals were treated with isotype antibody, anti-PD-1, anti-MIF, and anti-PD-1/anti-MIF and evaluated for tumor growth, survival, plasma cytokines/chemokines, and tumor immune cell infiltration by flow cytometry and immunohistochemistry. Intratumoral blood vessels were also evaluated by CD31 staining. YUMMER1.7 cells were also implanted in wild-type or MIF knock-out mice and evaluated for tumor growth and survival.

Results Anti-PD-1/anti-MIF treatment delayed YUMMER1.7 growth ($p < 0.01$) and prolonged survival ($p < 0.0001$) compared to monotherapy or control groups. Similar results were observed in the MC38 colorectal model.

Treated mice demonstrated robust anti-tumor memory responses off therapy when rechallenged.

Anti-PD-1/anti-MIF treatment enhanced TH1 signaling, as evidenced by increased markers of T cell stimulation (MIG and IL-1a), particularly TH1 cytokines (GM-CSF, IL-12p40, IL-12p70, and IFN-g), and markers of macrophage activation (MIP-1a, MIP-1B, MIP-2, and M-CSF). Flow cytometry analysis revealed an increase in the cDC1 population ($CD45^{hi}$/$Ly6C^{lo}$/$CD3^{lo}$/$CD19^{lo}$/TCR^{lo}/$CD11c^{hi}$/$MHCII^{hi}$/$CD172^{lo}$/$XCR1^{hi}$) within YUMMER1.7 tumors upon combined anti-PD-1/anti-MIF therapy. Anti-PD-1/anti-MIF treatment was associated with a trend towards higher CD8+ T cells and fewer CD31+ vessels compared to monotherapy or control groups ($p=0.08$ and $p<0.01$, respectively). As YUMMER1.7 and MC38 produced high levels of excreted and intracellular MIF, we evaluated the impact of MIF loss on immune cells and showed YUMMER1.7 had reduced growth when implanted into MIF-knockout mice ($p=0.0095$).

Conclusions Dual inhibition of PD-1 and MIF overcomes immune checkpoint resistance by modulating the tumor microenvironment. Combined anti-PD-1/anti-MIF results in additive responses to enhance cDC1 populations, known for their role in antigen presentation, CD8+ T cell activation, and IL-12 secretion, thus promoting anti-tumor immunity. Targeting MIF in addition to PD-1 blockade represents a promising strategy to overcome immunotherapy resistance and improve therapeutic outcomes.

Ethics Approval All animal studies were done under Dr. Richard Bucala's IACUC animal protocol 2020–10992 (approved 7/28/2020)

http://dx.doi.org/10.1136/jitc-2023-SITC2023.0830