Background Poly (ADP-ribose) polymerase inhibitors (PARPi) have improved outcomes of BRCA-associated breast cancer (BC); however, treatment responses are often not durable. Our preclinical studies demonstrated PARPi activates the cGAS/STING pathway inducing recruitment of anti-tumor CD8+ T-cells, required for tumor clearance. These studies led to clinical trials testing PARPi plus ICB. However, single-arm and randomized trials of PARPi + ICB have not yet demonstrated efficacy superior to PARPi monotherapy, underscoring the need to characterize the tumor microenvironment (TME) during PARPi therapy to identify optimal strategies to enhance T-cell activation. We recently showed that PARPi induce CSF-1R+ suppressive tumor associated macrophages (TAMs) that restrict anti-tumor immune responses, contributing to PARPi resistance. Removing TAMs with CSF-1R-blockade significantly enhanced overall survival (OS) when combined with PARPi compared to PARPi monotherapy in preclinical models, which was CD8-T-cell-dependent. Here, we investigate if combined PARPi + CSF-1Ri blockade and PARPi can be further enhanced by ICB.

Methods Mice bearing Brca1-deficient TNBC (K14-Cre;Brca1f/f; Tp53f/f) tumors were treated with PARPi (talazoparib) ± small molecule inhibitor of CSF-1R (ARRAY-382; CSF-1Ri) ± anti-PD-1 and followed for survival. Flow cytometry and Nanostring gene expression analysis were employed to elucidate changes in the TME following short-term treatment.

Results PARPi + CSF-1Ri-treated mice cleared 7/10 tumors by day 98, compared to 2/8 mice treated with PARPi monotherapy. PARPi + CSF-1Ri-treated mice had 100% protection upon tumor rechallenge, whereas PARPi monotherapy provided 50% protection. The addition of anti-PD-1 to PARPi did not enhance OS compared to PARPi monotherapy. Triple combination of anti-PD-1 + PARPi + CSF-1Ri demonstrated similar clearance of tumor by day 98 (7/10) compared to PARPi + CSF-1Ri. Triple combination led to superior long-term tumor clearance; whereat day 301, 5/10 mice were tumor-free compared to 2/10 treated with PARPi + CSF-1Ri.

Flow cytometry revealed an increasing trend of infiltrating Granzyme B+CD8+T-cells in triple therapy-treated tumors, compared to PARPi + CSF-1Ri-treated tumors. Triple therapy-treated tumors had increased expression of genes associated with anti-tumor immune response (CD80, CD86, PD-L1, IL-1β, IFNβ, IFNγ), antigen presentation, and T-cell chemotaxis (CXCR3 ligands) compared to PARPi + CSF-1Ri.

Conclusions These data confirm targeting immunosuppressive TAMs induces favorable anti-tumor responses that enhance PARPi therapy, which is associated with immunologic memory and can be further enhanced by ICB, with improved long-term durability of tumor clearance and further favorable modulation of the TME. Trials combining CSF-1R-blockade and PARPi are under development for BRCA-associated BC; our results suggest addition of ICB may further improve efficacy.