COMPOUND SCREEN IDENTIFIES INHIBITOR OF APOPTOSIS PROTEINS (IAP) ANTAGONIST AS AN INDUCER OF T CELL PROLIFERATION AFTER CROSS-PRESENTATION BY DENDRITIC CELLS

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Background Cross-presentation of tumor antigens by dendritic cells (DCs) is crucial to prime and (re-)stimulate CD8+ T cells. This process is important in initiating and maintaining an anti-tumor response. Previously, we found that tumor presence of conventional type 1 DCs (cDC1), a subtype that excels at cross-presentation, correlates with response to neoadjuvant immune checkpoint blockade (ICB) in melanoma. This led us to hypothesize that patients failing to respond to ICB could benefit from enhanced cross-presentation of tumor antigens.

Methods A cross-presentation assay to screen over 5,500 compounds for improvement of CD8+ T cell proliferation after cross-presentation of tumor antigens by DCs was established. For this purpose, bone marrow derived GM-CSF DCs were cultured in the presence of irradiated B16F10-OVA257-264-/- tumor cells, CpG ODN class B and naïve CTV-labelled CD8+ T cells that have a TCR specificity for OVA257-264 (SIINFEKL) in the context of H2-Kb (OT-I TCR). In this setting, tumor antigens (OVA) that have been processed into SIINFEKL peptide and are presented in the context of H2-Kb by the DCs can induce CD8+ T cell proliferation and activation.

Results A total 145 compounds were identified that significantly improved T cell proliferation after cross-presentation of tumor antigens by DCs. Subsequently, we selected compounds that also increased IFN-γ production. A total of 11 compounds were confirmed to significantly enhance CD8+ T cell proliferation and IFN-γ production. A particular strong effect was observed for AZD5582, an antagonist of inhibitor of apoptosis proteins (IAPs) cIAP1, cIAP2 and XIAP. AZD5582 treatment led to DC activation of the non-canonical nuclear factor kappa B (NF-κB) pathway, enhanced antigen import from endolysosomes into the cytosol and increased expression of genes involved in cross-presentation. Furthermore, it upregulated expression of CD80, CD86, MHC class II, CD70 and secretion of TNF by DCs. This enhanced DC activation and maturation program was observed also in tumor-bearing mice upon AZD5582 treatment, culminating into an increased frequency of systemic tumor antigen-specific CD8+ T cells.

Conclusions We identified several compounds that enhance T cell proliferation after cross-presentation. Addition of the identified AZD5582 to (combinations of) ICB might improve outcome in unfavorable patients.

Ethics Approval The protocol and amendments of the OpACIN-neo and PRADO trial were reviewed and approved by the appropriate review boards and ethics committees of each institute. All participating patients provided written informed consent before enrollment.

All animal procedures were approved by the Animal Welfare Committee of the Netherlands Cancer Institute, in accordance with institutional and national guidelines.