612 PERIPHERAL IMMUNE SIGNATURE OF RESPONSIVENESS TO ADOPTIVE CELL THERAPY WITH EX VIVO EXPANDED TILS USING HIGH DIMENSIONAL SINGLE CELL ANALYSIS

Cecilie Oelvang Madsen*, Marta Velasco Santiago, Troels Holz Borch, Inge Marie Svane, Morten Hansen. *The National Center of Cancer Immune Therapy (CCIT-DK), Herlev, Denmark*

Background Adoptive cell therapy (ACT) with *ex-vivo* expanded tumor infiltrating lymphocytes (TILs) has shown remarkable results in patients with metastatic melanoma (MM).[1, 2] There is a crucial need for noninvasive biomarkers for screening patients which could improve the selection of patients responsive to therapy. Most studies have until now investigated phenotype, numbers, and persistence of infused TILs, while the role of peripheral immune cell in this process is still poorly understood. Here, we sought to elucidate potential peripheral immune signatures before and early on-treatment to predict patients with clinical outcome of ACT-TIL therapy, as well as characterize its impact on circulating immune cells.

Methods Here we retrospectively analyzed peripheral blood mononuclear cells (PBMCs) and serum on-treatment changes from 45 patients with MM treated at our cancer center in one of three clinical trials: Study I (NCT00937625),[3] Study II (NCT02379195),[4] and Study III (NCT02354690).[5] Three multicolor flow cytometry panels were developed, two focused on T cells and one designed to measure all major mononuclear subsets and their characteristics. Data were evaluated by algorithm-based clustering and manual gate-setting of major immune cell subsets. Multiplex Luminex and ELISA was used to analyze serum samples to further reveal specific cytokine profiles of responsiveness to TIL therapy.

Results Higher frequency of $CD8^+$effector memory (EM) T cells was the only subset characterizing a peripheral immune signature of responsiveness pre-TIL. However, post-TIL therapy multiple immune subsets were associated with clinical outcome. Non-responders were characterized by mobilization of monocytic and NK subsets at the expense of $CD4^+$ T cells and several subsets of dendritic cells (DCs). While major pro-inflammatory cytokines, IL-6 and IL-8, decreased post-TIL in responding patients they persisted in non-responding patients. In contrast, responders post-TIL therapy was characterized by higher frequency of both cDC2s and pDCs, $CD4^+$ T cells and a subpopulation of $CD4^+$ EM T cells expressing CD28, CD29, CD95, and PD-1.

Conclusions While TIL therapy encompasses adoptive transfer of *ex-vivo* expanded preexisting TILs such therapy seems to modify both lymphoid and myeloid cell populations in peripheral blood. This indicates that early on-treatment peripheral immune signature of responsiveness to TIL therapy is characterized by higher frequency of $CD4^+$ T cells and subsets of DCs, and a decrease in inflammatory cells and their mediators.

Trial Registration NCT00937625, NCT02379195, NCT02354690

REFERENCES

1. Rosenberg, Steven A, *et al*. Durable Complete Responses in Heavily Pretreated Patients with Metastatic Melanoma Using T-Cell Transfer ImmunotherapyComplete Regressions in Melanoma. *Clinical cancer research*. 2011;**17**(13):4550-4557.
2. Dafni, U, *et al*. Efficacy of adoptive therapy with tumor-infiltrating lymphocytes and recombinant interleukin-2 in advanced cutaneous melanoma: a systematic review and meta-analysis. *Annals of Oncology*. 2019;**30**(12):1902-1913.
3. Andersen, Rikke, *et al*. Long-Lasting Complete Responses in Patients with Metastatic Melanoma after Adoptive Cell Therapy with Tumor-Infiltrating Lymphocytes and an Attenuated IL2 Regimen TIL Therapy for Melanoma Patients with Attenuated IL2 Dose. *Clinical cancer research*. 2016;**22**(15):3734-3745.
4. Andersen R, *et al*. T cells isolated from patients with checkpoint inhibitor-resistant melanoma are functional and can mediate tumor regression. *Annals of Oncology*. 2018;**29**(7):1575-1581.
5. Borch, Troels Holz, *et al*. Clinical efficacy of T-cell therapy after short-term BRAF-inhibitor priming in patients with checkpoint inhibitor-resistant metastatic melanoma. *Journal for ImmunoTherapy of Cancer*. 2021;**9**(7).

Ethics Approval The Ethical Committee of the Capital region of Denmark, the Danish Data Protection Agency and the Danish Medical Agencies approved Study I (NCT00937625: EudraCT no. 2008-008141-20), Study II (NCT02379195: EudraCT no. 2014-001420-29), and Study III (NCT02354690: EudraCT no. 2014-001419-38).

http://dx.doi.org/10.1136/jitc-2022-SITC2022.0612