specifically expressed in terminally exhausted cells. These data suggest the balance of Batf and Nr4a2 may modulate the enhancer landscape to promote terminal exhaustion, while hypoxia simultaneously promotes hypermethylation and gene repression.

Conclusions Our study defines for the first time the features of epigenetic dysfunction in tumor-mediated T cell exhaustion and deepens our understanding of the epigenetic regulation of gene expression. These observations are the bases for future work that will elucidate that factors that drive progression towards terminal T cell exhaustion at the epigenetic level and identify novel therapeutic targets to restore effector function of tumor T cells and mediate tumor clearance.

<http://dx.doi.org/10.1136/jitc-2020-SITC2020.0518>

519

DIACYLGLYCEROL KINASE ζ LIMITS IL-2-DEPENDENT CONTROL OF PD-1 EXPRESSION IN TUMOR-INFILTRATING T LYMPHOCYTES

¹Javier Arranz-Nicolas*, ¹Cristina Rodríguez-Rodríguez, ¹Rosa Liébana, ²Judith Leitner, ¹Antonia Ávila-Flores, ²Peter Steinberger, ¹Isabel Mérida. ¹Spanish National Centre for Biotechnology (CNB-CSIC), Madrid, Spain; ²Medical University of Vienna, Vienna, Austria

Background Tumors evade T cell responses targeting them through the upregulation of tolerance-inducing mechanisms. One of the best characterized is that of PD-1/PD-1L engagement, that in healthy CD8+ T cells limits cytotoxic responses against self-antigens and that tumors employ to neutralize T cell attack. Antibody-based therapies aimed to block the PD-1/PD-1L axis have rendered notable results, but most patients eventually develop resistance. This failure is attributed to CD8+ T cells achieving an exhausted phenotype where recovery is hardly feasible. The dysfunctional phenotype of tumor-infiltrating T cells is largely triggered by the unbalance of diacylglycerol (DAG)- and Ca²⁺-regulated signals that results in alteration of the transcriptional T cell program. DAG kinase (DGK) ζ -dependent DAG consumption contributes to hypofunctional T cell states while DGK ζ deficiency facilitates tumor rejection in mice without apparent adverse autoimmune effects. In spite of its therapeutic potential, little is known about DGK ζ function in human T cells and there are not isoform-specific inhibitors targeting this DGK isoform.

Methods Here we used a human triple parameter reporter (TPR) cell line to examine the consequences of DGK ζ depletion in the transcriptional restriction imposed by PD-1 ligation. We also investigated the effect of DGK ζ deficiency in the expression dynamics of PD-1, as well as the impact of the absence of this DGK isoform in the in vivo growth of a MC38 adenocarcinoma cell line.

Results We demonstrate that DGK ζ depletion enhances DAG-regulated transcriptional programs, favoring IL-2 production and limiting PD-1 expression. Diminished PD-1 expression and enhanced expansion of cytotoxic CD8+ T cell populations is also observed even in the context of immunosuppressive milieus and correlates with the failure of MC38 adenocarcinoma cells to form tumors in DGK ζ -deficient mice.

Conclusions Our results suggest the relevance of DGK ζ as a therapeutic target on its own as well as a biomarker of CD8+ T cell dysfunctional states.

<http://dx.doi.org/10.1136/jitc-2020-SITC2020.0519>

520

THE IMMUNE LANDSCAPE OF PEDIATRIC TUMORS

¹Shimaa Sherif*, ²Jessica Roelands, ²William Mifsud, ²Blessing Dason, ²Darawan Rinchai, ²Adrian Charles, ²Ayman Saleh, ²Chiara Cugno, ²Khalid Fakhro, ¹Borbala Mifsud, ²Davide Bedognetti, ²Wouter Hendrickx. ¹Hamad Bin Khalifa University, Doha, Qatar; ²Sidra Medicine, Doha, Qatar

Background It is now well established that the immune system has a substantial role in controlling cancer growth and progression. Immunotherapy is quickly coming to the forefront of cancer treatment, however the implementation of immunotherapy in pediatric solid cancers, which classically display a low mutational load, is hindered by insufficient understanding of the determinants of cancer immune responsiveness in children. In order to better understand tumor-host interplay, we sought to characterize solid pediatric cancers based on immunological parameters¹ using analytes extracted from gene expression data.

Methods We performed single sample GeneSet Enrichment Analysis for 105 immune signatures previously described on 5 pediatric tumors (410 patients) from TARGET dataset¹ to identify coherent signature modules. Then we clustered samples according to representative signatures¹ and compared survival across clusters. We completed the analysis by analyzing the enrichment of immune subpopulations and the expression of the immune checkpoints. The degree of dysregulation of

Abstract 520 Figure 1 Immune subtypes of pediatric solid tumors

A. Spearman Correlation matrix of 105 cancer immune signatures showing 5 main modules.

B. Spearman's correlation of the 105 cancer immune signatures, identifies separation of the 5 immune signatures in different clusters

C. Distribution of cancer types within immune subtypes. The percentage of samples belonging to each tumor is shown in colors.

D. Distribution of immune subtypes within TARGET pediatric tumors.

The percentage of samples belonging to each immune subtype is shown in colors.

E. Distributions of signature scores within the six immune subtypes (rows), with dashed line indicating the median.

F. Kaplan-Meier OS curve for the 6 immune subtypes (S1-S6) showing different outcomes.