

dCODE Dextramer® Reagents Identify Antigen-specific Populations and Their TCR Clonotypes at Single Cell Level

Adapted from Jacobsen *et al.* SITC 2019 poster P186

BACKGROUND

dCODE Dextramer® (10x compatible) reagents are DNA barcoded MHC Dextramer reagents designed for use with 10x Genomics Feature Barcode protocol for Single Cell Immune Profiling (**Fig. 1**). Each unique barcode is specific for the MHC-peptide displayed on the dCODE Dextramer®.

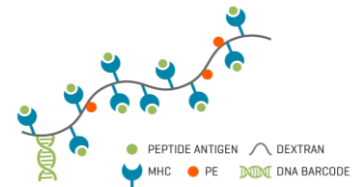


Fig. 1. dCODE Dextramer®

STUDY DESCRIPTION

Goal: Detect antigen-specific T cells and their cognate T-cell receptors sequences in a human PBMC sample using a highly multiplexed panel of dCODE Dextramer® reagents.

1. Healthy donor PBMC sample was stained with a pool of 50 different MHC I dCODE Dextramer® reagents displaying different viral and cancer epitopes
2. dCODE Dextramer® positive cells were sorted by flow cytometry and loaded onto 10X Chromium controller
3. Generation of three DNA libraries: 1) dCODE Dextramer® binders; 2) V(D)J sequences; 3) RNA expression. Each library sequenced by next generation sequencing (Illumina)

RESULTS

Four antigen-specific T-cell populations were identified: one for influenza (Flu), two for Epstein-Barr virus (EBV-1 and EBV-2), one for MART-1 (**Fig. 2**). For each population, the paired clonal TCR sequences were directly obtained and quantified.

Multiple TCR clones were identified for each antigen-specific T-cell population (**Table 1**). Highly expanded TCR clones were found in viral T-cell populations (EBV-1 and EBV-2). Fewer expanded TCR clones were found for MART-1. To confirm whether the identified MART-1-specific T-cells represent a naïve population, it is possible to interrogate the available gene expression profile.

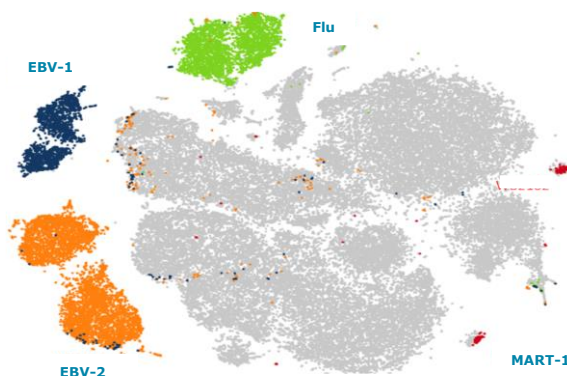


Fig. 2. Four antigen-specific T-cell populations identified after analysis of sequencing data

Specificity (MHC-peptide)	Flu	EBV-1	EBV-2	MART-1
Positive cells (number)	2594	1846	4472	187
Specific clones (number)	732	166	389	180
Most represented clone (number of cells)	110	1241	2373	8
Most represented clone (frequency)	4%	67%	53%	4%
Clone with >1 cell (number)	251	19	44	1
Clone with 1 cell (number)	481	147	345	179

Table 1. TCR clones for each antigen-specific population

CONCLUSIONS

- dCODE Dextramer® technology enables the generation of highly multiplexed antigen-specificity data in a single experiment
- Combining dCODE Dextramer® and 10x Chromium represents a powerful tool for deep phenotyping of immune relevant cells
- Personalized dCODE Dextramer® libraries allow profiling of patients' T cells and give a new understanding of T-cell immunity