

MHC-based dissection of antigen-specific T cell immunity

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Peptide-MHC tetramers and other types of pMHC multimers have become an essential tool for the analysis of T cell immunity. However, the inefficiency of the classical strategy for pMHC generation has precluded the development of large libraries of MHC multimers for high-throughput monitoring of immune responses. In the past years we have aimed to address this issue by designing MHC class I molecules occupied with UV-sensitive 'conditional' peptide ligands. This technology that can be utilized to generate collections of thousands of pMHC reagents was first developed for HLA-A2.1 and has subsequently been extended to human MHC alleles HLA-A1, -A3, -A11 and -B7.

While the UV-induced ligand exchange technology is valuable for the parallel production of massive numbers of pMHC complexes, limitations on patient sample size can preclude a comprehensive analysis of T cell immunity by classical MHC tetramer flow cytometry. To tackle this issue, we have developed a novel 'combinatorial coding' strategy that allows the parallel detection of a large number of different T cell populations within a single sample. This technology is based on the concept that a given T cell specificity is not encoded by a single identifier (in this case fluorescent proteins or quantum dots), but is encoded by a defined combination of two or more identifiers. Using such combinatorial coding, the number of specificities that can be encoded increases dramatically. The combined use of MHC ligand exchange and combinatorial coding forms a robust platform for the high-throughput dissection of disease- and therapy-induced CTL immunity