Evaluating prediction strategies for identification of T cell responsive mutation-derived neoepitopes in cancer

Increasing evidences point to an important role of mutation-derived antigens in immune recognition of cancer. Current strategies for prediction of immunogenic neoepitopes results in large personalized peptide libraries, but only a minority (<1%) elicit T cell responses at detectable levels. Neoepitopes are of potential valuable as predictors of response to therapy and targets for personalized immunotherapeutic approached. Consequently, there is an unmet need to understand the rules identifying immunogenic neoepitopes. Both tumor mutation mapping via exome sequencing and mass-spectrometry-based elution for MHC class I presented peptides has been applied in different studies, combined with RNA sequencing to determine the expression level of relevant transcripts. Additionally, neoepitopes may be defined based on either autologous tumor cell lines or snapfrozen tumor material. We present here a study in which all the above mentioned strategies are assessed in three melanoma patients. Predicted large peptide libraries matching the HLA expression of the patients was identified and selected based on any of the strategies given above. This resulted in a total of ~3000 peptides for the three patients. We investigated the T cell recognition of these personalized peptide libraries using a new technology based on DNA-barcode labeled MHC multimers to detect multiple, potentially > 1000, different neoepitope specific T cell populations in a single sample. Through this unbiased comparison, we evaluate selection strategies for prediction of immunogenic cancer-associated neoepitopes, and identify rules for precise prediction. Precise prediction is essential for future application of neoepitopes both as predictors of responses to therapy and immunotherapeutic targets.