recombine their TCR α and β chains in discrete steps: TCRβ followed by TCRα. If a developing T cell successfully recombines its β chain, it receives weak functional signals that prompt progression to α-chain recombination and functional maturity[9]. However, if it receives an unexpectedly strong signal during early development, the cell responds as if it has recombined a TCR using the alternate γ and δ chains, and is diverted to an alternative γδ-T-cell-like developmental pathway[10].

Schmitt *et al.*[2] take advantage of the fact that triggering of the γδ pathway—and the associated strong signaling via the TCR—can be tracked with cell surface markers. Thus, T cells that carry a fully formed αβ TCR and signal at an earlier-than-expected stage of T-cell development after pMHC engagement can be readily isolated. The authors harness this process to select for strongly reactive, tumor-antigen-specific TCRs (**Fig. 1**). They introduce a recombined TCRα chain derived from a previously validated tumor-specific TCR into developing T cells and allow the cells to mature on feeder cells in the presence of the tumor antigen. Cells that show a strong TCR signal during TCRβ rearrangement are sorted, and their TCRβ chains are sequenced and tested to identify those that confer higher affinity than the parental one.

The authors demonstrate their method with a mouse antitumor TCR both *in vitro* and *in vivo* and with a human antitumor TCR *in vitro* using T-cell progenitors. They find that the T cells do indeed recombine TCRβ chains specific for the intended tumor antigens. As hoped, the isolated T cells contain a diverse collection of TCR Vβ regions and a collection of peptide-binding CDR3 loops that are diversified in both sequence and length. The newly generated TCRβ chains recognize the intended antigen, and some bind their antigens with higher affinity than the parental chain does and signal more potently.

When vetting a new TCR for clinical development, a chief concern is unwanted recognition of healthy cells expressing non-target antigens that are recognized by the tumor-specific TCR. This is a particular challenge because each TCR has its own cross-reactivity profile, and even extensive vetting against cell lines derived from multiple organs cannot rule out a negative interaction in patients. In principle, optimizing a TCR for better function through a native T-cell pathway while simultaneously conserving most of the TCR interface should mitigate potential cross-reactivity. Indeed, when tested for function, the newly identified TCRs show no signs of unwanted self-reactivity in a mouse model.

With the principle demonstrated, there are some considerations in moving the approach forward. First, the system optimizes only TCRβ chains, which account for half of the antigen-binding interface[4], and reliance on the V(D)J recombination machinery imposes biases on the peptide-contacting junctions, limiting diversity. These issues may explain why the observed affinity gains were modest. Additionally, it remains to be seen whether the improvements observed *in vitro* translate into improved antitumor activity *in vivo*. Finally, while modifying only the peptide-binding CDR3 loops should help prevent peptide-independent reactivity, and limiting TCR selection to β chains with high germline-encoded similarity to the parental chain should help limit off-target reactivity, any improved TCR will have to undergo extensive vetting for cross-reactivity on the path to clinical development.

These caveats aside, the method of Schmitt *et al.*[2] could prove important for future TCR engineering efforts. In relying on T-cell development, it should be applicable to any TCR. It also eliminates much of the effort and TCR-specific optimizations of other methods. Having been generated by the natural diversification and signaling machinery, the resulting TCRs already pass several of the requirements for function and specificity needed for therapeutic applications.

COMPETING FINANCIAL INTERESTS
The authors declare no competing financial interests.

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