

T cell Optimization of Trial Design

A Quick Recap

Overview

- 14 investigators reviewed studies
- Long term data comes from early NCI TIL studies with long term remissions that now extend out about 6 years
- More recent protocols, largely in CD19 malignancies and NY-ESO-1, have also shown early efficacy responses, but also demonstrate that there may be disease-specific factors that influence efficacy, for example the greater efficacy seen with ALL
- A number of cases of cytokine release syndrome reported, many of which were very responsive to steroids and seemed to occur more often with bulky disease

Host Preparation

- Goal of host preparation
 - Eliminate cytokine sinks
 - Eradicate T regs
 - Enhance APC activation
- Both mouse data and clinical data with TIL demonstrate that efficacy increases with lymphodepletion
 - Increases in IL7 and IL15 level important for persistence
- NCI has used a cyclophosphamide/fludarabine and found this combination has been relatively safe with most patients recovering lymphocytes within 7-10 days
- Animal data indicate that elimination of T regs and continued depletion of these cells may be critical for efficacy - This is more difficult to prove in clinical studies

Host Preparation (2)

- Some interesting data from mice shows that administration of fluoroquinolones decreases antitumor efficacy, raising the possibility of other factors that may be affected by lymphodepletion and contribute to efficacy
- Not all studies use the same preparation, in CD19 protocols some patients have received just cyclophosphamide or disease specific chemotherapy
- Some pediatric patients with ALL have been successfully treated without any lymphodepletion

Co-stimulatory Domains and Persistence

- Most studies are being done with second generation CARs but there are some 3rd generation
- Limited data, if any to compare the efficacy or safety of different signaling domains
- However, the question is more complicated than just signaling domains as the “same CARs”, i.e. same target and signaling domains can perform quite differently – small changes in the design can have big impact
- In addition to signaling domains, there are variable epitope binding, Sc-Fv linkage, variable CAR expression and finally patient specific factors

Co-stimulatory Domains and Persistence

- Finally, do we understand what is the optimum cell mix, for example are central memory cells preferred or other cell types
- Is the freezing cells altering the mix of cells in unexpected or variable ways?
- Is the answer to move to 3rd generation or to develop more targeted approaches to try to control the proliferation of cells?
- One approach may be to have split activation - antigen target for CAR and another antigen receptor for co-stimulation, where by antigen recognition and co-stimulation are not linked

Use of Cytokine Support

- Both animal and clinical data in TIL demonstrate improved T cell persistence and efficacy with the use of IL-2, at least for solid tumors
- IL-2 has not been as critical in hematologic malignancies likely due to continued signaling due to higher antigen presentation
- Virus-specific CARs have also not used IL2 consistently as the signaling through the viral receptor may provide adequate support

TCR design

- One of the main challenges is understanding the biology that leads to durable responses in some patients but not all patients.
- In addition, strategies to limit mispairing between endogenous and modified receptors are being explored, such as leucine zippers.

Gene Delivery

- To date, most protocols use lentiviral vectors or gamma retroviral vectors
 - While lentiviral vectors would be preferred in CD34+ cells data is lacking on comparisons between lenti and gammaretroviruses in mature T cells
- Some groups working with transposons which may offer a more cost-effective method to generate new constructs
- Similarly, electroporation of mRNA may provide another strategy to test new antigens
- An alternative is to use CD34+ hematopoietic stem cells to get long term expression of T cell receptors
 - Do we understand the kinetics of expression of TCRs or CARs after infusion of CD34+ cells and should NHP studies be done?



Target Issues – the Achilles Heel?

- Limited number of tumor specific antigens that are not expressed at some level on normal tissues
 - Focus on unique antigens to cancer
 - Mutated antigens
- Is a more personalized approach looking at unique mutations in the a patient's tumor an avenue to pursue
 - Data from TIL removed from melanoma patients provides some data in support of this approach
- The MAGE 3 on-target, off tissue cardiac toxicity demonstrates several issues
 - The potential for potency creates greater risk for toxicity
 - This toxicity is not easily predicted with traditional preclinical models
 - In-vitro screens not able to predict
 - Mouse models were unable to identify

Target Issues

- For new antigens, are Phase I studies ultimately the only way we will be able to determine off tissue toxicity with slow dose escalation or are there other preclinical models to explore, e.g. iPS or other libraries/bioinformatics, or larger animal models for appropriate targets and diseases
- Can this off-tissue toxicity be addressed in part by more tumor specific targeting without trying to get peripheral expansion?
- Use of combination that would recognize the tumor antigen in the context of the tumor – i.e. the tumor antigen and another tumor environment antigen?

Dosing Strategies

- Published CD19 studies had doses ranging from 3×10^6 and 3×10^7
- The published results in hematologic tumors do not show a clear correlation between dose and efficacy
- Published reports in hematologic malignancies do not show that toxicity is necessarily related in increased dose
- Split dosing is done at some institutions but no data to say that there is enhanced safety with this approach
 - This may be a strategy for certain novel targets but ability to avoid toxicity will depend upon how immediate any toxicity might arise
- Some protocols allow second infusion of T cells if patient has response and acceptable side effects, but data on whether this approach will improve response remains to be seen