Characterization of a novel clonal neoantigen reactive T cell (cNeT) product through a comprehensive translational research program

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Introduction

Achilles Therapeutics has developed a personalised Adoptive Cell Therapy (ACT) to identify and target multiple clonal neoantigens present on all cancer cells (Figure 1). Achilles has two ongoing phase I/IIa clinical trials using this approach; CHIRON (NCT04032847), for the treatment of Non-Small Cell Lung Cancer, and THETIS (NCT03997474), for the treatment of metastatic melanoma.

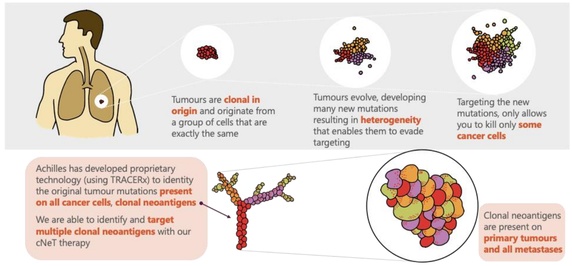


Figure 1: Targeting multiple clonal neoantigens may provide a precise mechanism to attack all cancer cells in a patient's tumour[1]

Achilles' proprietary VELOS™ manufacturing process and PELEUS™ bioinformatics platform produces Clonal Neoantigen T cells (cNeT) that target clonal neoantigens unique to each patient (Figure 2).

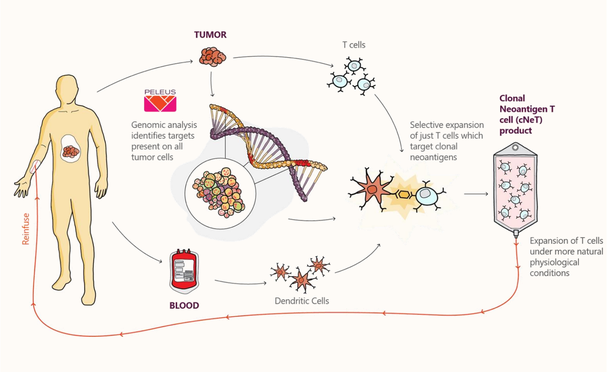
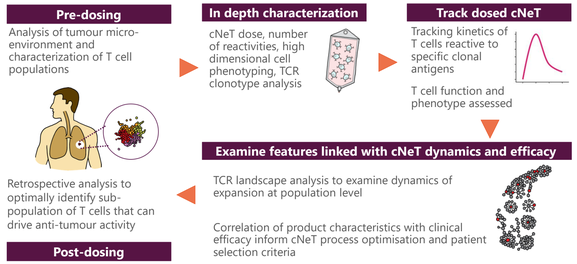


Figure 2: Schematic diagram of the VELOS™ manufacturing process[2]: Tumour and blood samples undergo genomic analyses to identify clonal neoantigens. In parallel, TIL are expanded from tumour fragments and monocyte-derived dendritic cells are generated from whole blood. In the final step, the co-culture of TIL with neoantigen peptide-pulsed dendritic cells drives the expansion of cNeT.

The Translational Program

Achilles has established a comprehensive Translational Science Program to analyse cNeT and their behaviour and dynamics in treated patients. The Program will:

- establish the product phenotype, functionality and specific reactivity of the manufactured ATL001 product in Achilles' clinical trials.
- monitor persistence, expansion and reactivity of cNeT in the peripheral circulation of treated patients.
- derive associations between product characteristics, clinical biomarkers and cNeT efficacy.



These investigations require patients to be monitored extensively throughout the trial (Figure 3). A multitude of assays at monitoring points provide deep and complementary information to measure multiple aspects of cNeT cell activity. For example:

- ELISpot assays measure product and peripheral reactivity to allow tracking of cNeT.
- TCRseq provides an orthogonal method to characterize cNeT by assessing the TCR composition of product and peripheral samples.

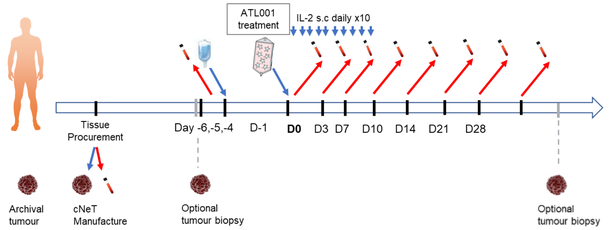


Figure 3: Translational Program patient sampling plan: Tumour, product and pre and post dosing peripheral blood are sampled from patients.

Patient T-05 Case Study

Patient T-05 enrolled in the THETIS trial with an initial diagnosis of BRAF wildtype cutaneous melanoma in 2006. The patient had previously received a three cycle combination of ipilimumab in 2017 which was stopped due to toxicity. The patient remained off treatment and had recurrent cutaneous lesions resected in the years following therapy. A soft tissue lesion was excised from the patient's abdomen in Feb 2020 and was taken forward into cNeT manufacturing (Figure 4).

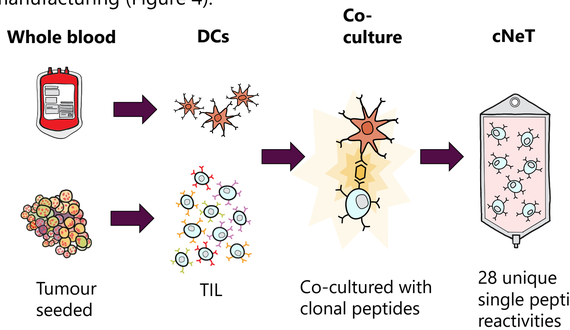


Figure 4: Manufacturing for patient T-05: 28 single peptide reactivities were identified by product restimulation with clonal peptides.

The T cell function of the manufactured product was measured by intracellular cytokine secretion of IFN-γ and TNF-α using flow cytometry (Figure 5). This shows, along with the multiple reactivities identified by ELISpot analysis, the presence of single as well as multi-functional cytokine secreting cNeT.

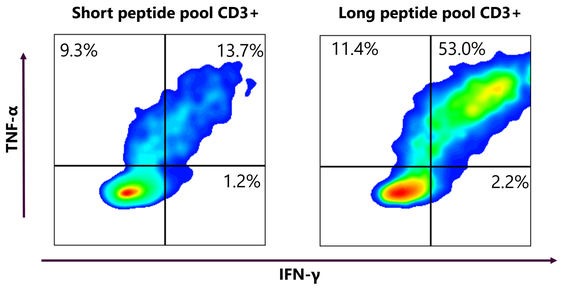


Figure 5 Cell function for patient T-05: Function is measured by cytokine secretion using flow cytometric analysis. Left: CD3+T cell cytokine secretion in response to short peptide pools. Right: CD3+T cell cytokine secretion in response to long peptide pools.

cNeT were tracked pre- and post-dosing using the long and short peptide pools that incorporate the identified clonal mutations (Figure 6A). Adjusting for the impact of immune system reconstitution (Figure 6B) allows normalisation for T cell frequency in the ELISpot assay (Figure 6C) and provides an estimate of the cNeT count/ml in peripheral circulation (Figure 6D). This shows expansion and detection of cNeT post dosing and provides an estimate of the quantity of reactive T cells in circulation.

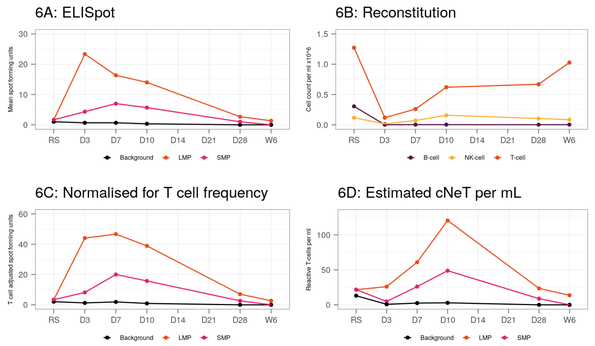


Figure 6 Tracking cNeT in peripheral circulation allows estimation of the reactive T cell component pre and post-dosing. RS is the patient rescreening visit, D are visit days post-dosing, W are visits weeks post-dosing. There is detectability of both short and long peptide reactivity.

Conclusions

- Achilles Therapeutics has a comprehensive Translational Science Program that allows quantification, and characterization of our active component (cNeT) prior to and post dosing.
- The ability to characterize and track the active component of our product uniquely positions Achilles Therapeutics as it:
  - Enables us to develop and deliver a reliable manufacturing potency assay.
  - Offers insight into the *in-vivo* dynamics of cNeT and its correlations with patient outcomes in association with product and clinical factors.

References

1. McGranahan N., et al. Clonal neoantigens elicit T cell immunoreactivity and sensitivity to immune checkpoint blockade. *Science.* 6280: 1463-1469 (2016)
2. Kotsiou E, et al. Next generation clonal neoantigen targeting T cells, generated using the PELEUS™ bioinformatics platform and the VELOS™ manufacturing method show superior reactivity and phenotypic characteristics than classical TIL products. Proceedings: AACR Annual Meeting (2020)

Acknowledgements

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Disclosures

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