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Paul Jerard Layug

Phonetic spelling of your (full) name

pawl zhuh-RARD lah-YOOG

Presenting Author Category

PhD Student

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GBD Audio File

GBD_Layug.m4a

Headshot

PL.jpg

Short Bio (100 words)

Paul Jerard Layug is a PhD candidate in the Department of Biochemistry and Medical Genetics at the University of Manitoba and the Paul Albrechtsen Research Institute at CancerCare Manitoba. Working in the laboratory of Dr. Yale Michaels, his research focuses on synthetic biology and fine-tuning gene expression to develop safer, longer-lasting CAR T-cell therapies for pediatric leukemia. Driven by a commitment to patient-centred research, Paul also leads Co-PILOT, an initiative connecting cancer researchers with patient partners. Through his work, he aims to translate innovative cell engineering into life-saving treatments for children fighting cancer.

Research Category

Basic Science

Abstract Title

Precise tuning of CAR expression mitigates excessive inflammation and dysfunction in CD19-CAR T cell therapy against B-ALL

Authors

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Background

B-cell acute lymphoblastic leukemia (B-ALL) is the most common childhood cancer. Chimeric Antigen Receptor (CAR) T therapy has revolutionized the immunotherapy landscape for relapsed and/refractory B-ALL. CARs are engineered fusion proteins comprising an extracellular ligand-binding domain that recognizes a cancer antigen, such as CD19, linked to an intracellular activation domain that primes the T cell to kill. Despite clinical success, suboptimal safety and durability limit their use as first-line treatments. Such limitations stem from supraphysiological levels of CAR expression upon lentiviral transduction of the CAR into the T-cell genome, leading to excessive activation and lethal side effects, including cytokine release syndrome (CRS) and rapid dysfunction.

Objective

We set out to identify a “Goldilocks” level of CAR expression that dampens excessive pro-inflammatory cytokine release and mitigates T-cell exhaustion whilst maintaining effective tumour killing.

Methods

To fine-tune CD19-CAR expression, we used our previously established microRNA silencing-mediated fine-tuners (miSFIT) technology. miSFITs are engineered target sites for endogenous microRNAs, enabling precise control over gene expression levels. We assessed the effect of CAR expression on T-cell activation, T-cell cytotoxicity and exhaustion in vitro and in vivo.

Results

Our in vitro co-culture assays demonstrated that reducing CAR expression strikingly dampened cytokine release across all cytokines implicated in CRS (e.g., IL-6 by at least 1.5-fold, IFN- γ by at least 2-fold). Notably, lowering CAR levels also significantly decreased the expression of the exhaustion markers PD-1 and LAG3 by at least 1.3-fold, while maintaining robust tumour killing across multiple re-challenges. Supporting these findings, our tuned CAR T cell product exhibited similar tumour control in our preclinical B-ALL xenograft mouse model, when compared to the representative commercial product.

Conclusion

Collectively, these findings demonstrate that CAR expression influences T cell fate and function, and that precisely tuning CAR expression with miSFITs may enable the production of safer and longer-lasting CAR T products for childhood B-ALL.