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Expansion and differentiation of T cells under defined xeno-free culture conditions

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Title: Expansion and differentiation of T cells under defined, xeno-free culture condition

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Abstract

Antigen specific suppressive immunotherapy such as Tregs therapy and highly selective targeted immunotherapy such as chimeric antigen receptor and T-cell receptor therapy have shown great benefits and therapeutic value to the patient population. These immunotherapy applications require ex vivo expansion of T cells to high cell densities without loss of T cell phenotypes and functionality for successful adoptive transfer. Moreover, a defined, xeno- and serum-free basal medium is ideal to reduce cost, limit variability and lower the risk of foreign contaminants.

By applying the quality by design approach, we report the successful development of a xeno- free basal medium containing completely defined components that can be used to activate, expand and differentiate naïve T-cells. This medium supports higher rate of growth expansion as compared to 10% bovine serum containing media. Phenotypic analysis shows the percentages of expanded CD4+ and CD8+ are comparably maintained after expansion. T cells can be re-activated and further expanded in this basal media over long term culture (≥ 21 days). This defined, xeno-free basal medium also effectively supports T cell differentiation potential into major T cell subsets such as Th1, Th2, and Tregs when introduced to polarization cocktail of rIL-12 and anti-IL-4, rIL-4 and anti-IFN γ , and TGF- β 1 and retinoic acid respectively. Potential uses for large scale cultures in spinner flasks and culture bags have also been demonstrated.

In this study, we demonstrate how the defined, xeno-free expansion and differentiation medium can be a superior alternative than serum-containing media to culture T cells on a large scale platform while maintaining T cell phenotypes and polarization potential.