319 IMMUNOMETABOLIC MODULATION OF ENGINEERED IPSC-NK CELLS FOR IMMUNOTHERAPY OF SOLID TUMORS

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Background Natural killer (NK) cells have efficient intrinsic recognition capabilities towards abnormal cells. This fact makes them particularly attractive as safe effector cells for cancer immunotherapy. Despite being innately tumor-killing cells, sourcing them from human donors is a tedious and highly donor-specific process.[1] As an alternative, induced pluripotent stem cells (iPSCs) represent a universal source of NK cells. iPSC-NK cells can be prepared with homogenous quality and can be easily genetically modified to modulate their specificity and activity.[2] Despite their demonstrated potential, differentiation processes to generate NK cells from iPSCs require improvements in terms of NK cell yield. Our group has reported that chemically defined and serum- and feeder-free differentiation generates NK cells that are predominantly CD56+/CD16+/CD3- and which express NK activation markers NKG2D, NKp30, NKp44, NKp46, and DNAM-1. We also recently engineered these cells to express multi-specific CAR constructs and showed that they mediate strong anti-tumor activity and lead to improved survival in mouse xenografts of glioblastoma.[3]

Methods However, we also found that modulating the differentiation and genetic engineering processes can yield further improvements in terms of NK cell quality, yield, and activity. Here, we report our recent work on evaluating iPSC-NK protocol advancements to improve reproducibility and yield, as well as the generation of more efficient immunometabolically-reprogrammed CAR-iPSC-NK cells. These involve the addition of Interleukin-15 in the second stage of differentiation, where differentiating embryoid bodies transitions to the generation of NK cells. We are also modulating the activity of multi-specific CAR-iPSC-NK cells with small molecule inhibitors of metabolic activity, by targeting mitochondrial dysfunction.

Results The involvement of interlukin-15 and inhibition/silencing of metabolic acitivity with small molecule inhibitors generated metabolically resilient and functionally robust CAR-NK cells. In addition, these cells showed robust cell-mediated effector functions including cytotoxicity, degranulation, and IFN-γ production against patient-derived cancer cells.

Conclusions These advancements lead to improved NK cell quality and activity and represent a significant step toward off-the-shelf immunotherapies for solid tumors.

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