Developing CLL-1 and MSLN Immunogens for Dual CAR-T Cell Therapy for AML

Liu, Andrew (School: Ponte Vedra High School)

Purpose: Acute myeloid leukemia (AML) is aggressive cancer with a poor prognosis. Most patients with AML ultimately relapse and succumb to cancer progression after receiving standard treatments. Thus, developing novel therapy for AML is critically needed. Chimeric antigen receptor (CAR) – T cell therapy is a new revolutionary cancer treatment involving engineered receptors that function to redirect T lymphocytes to recognize and eradicate antigen-specific cancer cells. C-type lectin-like molecule-1 (CLL-1) and Mesothelin (MSLN) are highly expressed on AML cancer cells with no expression on normal tissues, making these two molecules ideal therapeutic targets for AML. In this study, we developed two novel immunogens, CLL-1 and MSLN-expressing NIH/3T3 or 239FT cells, for developing antibodies used to construct dual CAR - T cell therapy of AML. Procedure: The cDNAs of CLL-1 or MSLN were subcloned into a pCDH lentiviral expression system, and vectors encoding CLL-1 or MSLN were generated. Then, 293FT and NIH/3T3 cells were transduced with vectors expressing CLL-1 and MSLN. The CLL-1 or MSLN over-expressed cell clones were selected using puromycin and established as stable cell lines. The expressions of CLL-1 or MSLN on cell lines were validated by flow cytometry. Data analysis: The expressions of CLL-1 on NIH/3T3 and 293FT cells are 98.8 % and 98.6%. The expression of MSLN is 58.1% on NIH/3T3 cells and 99.1% on 293FT cells. Conclusions: Our data indicate that the CLL-1 and MSLN immunogens are well established and can be further developed into CLL-1 and MSLN specific dual CAR -T cell therapy to treat AML patients.