IDENTIFICATION OF CD320, CD92, AND CD267 AS POTENTIAL NOVEL THERAPEUTIC TARGETS FOR CAR-T THERAPY IN MULTIPLE MYELOMA

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Introduction

Multiple myeloma (MM) is a hematological malignancy characterized by the clonal expansion of plasma cells (PCs) in the bone marrow, leading to significant therapeutic challenges and high relapse rates. Despite advancements in immunotherapies, including chimeric antigen receptor T-cell (CAR-T) therapy (1,2), treatment resistance, antigen escape, and severe toxicities remain major obstacles.

Aim

Our study aimed to identify genes encoding surface proteins specifically expressed in PCs, with the goal of discovering potential targets for CAR T-cell therapies in MM. By streamlining the search for tumor-specific markers, we sought to highlight the most promising candidates for the development of more effective and personalized treatments tailored to the molecular profiles of individual patients.

Objectives

The objectives of this study were to identify surface protein-encoding genes specifically expressed on PCs in MM through single-cell RNA sequencing (scRNA-seq), to validate their expression using flow cytometry, and assess their potential prognostic significance by integrating data from scRNA-seq and bulk RNA-seq datasets of MM patients.

Methods

This study aimed to identify novel molecular targets for CAR-T therapy through single-cell RNA sequencing (scRNA-seq) analysis of 147 patients with monoclonal gammopathies (25 with monoclonal gammopathy of undetermined significance (MGUS), 14 with smoldering MM (SMM), 76 with MM, and 32 with relapsed/refractory MM (RRMM), as well as 52 healthy bone marrow samples, integrating publicly available datasets (GEO accession numbers: GSE271107, GSE271915, GSE120221, GSE189460, GSE223060, GSE210079, GSE145977, GSE124310, GSE161801, GSE163278, GSE161722).

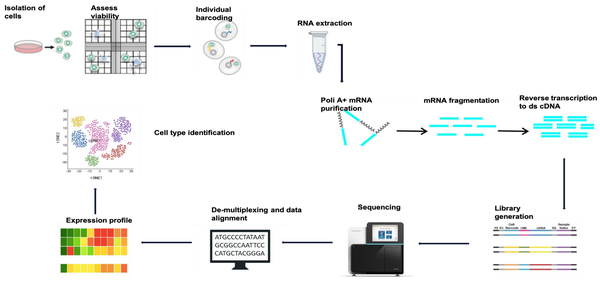


Fig. 1 scRNA-seq workflow. It begins with isolating and preparing single cells.The cells are lysed and RNA is extracted with subsequent library preparation. The sequencing of the cDNA libraries is followed by data preprocessing. The data analysis is performed with reduction techniques to visualize and cluster cells based on their expression profiles.

Results

We identified 15 genes encoding surface proteins (TNFRSF17, SDC1, CD63, FCRL5, CD38, SLAMF7, TNFRSF13B, CD59, BST2, BSG, SLC44A1, FCGR2B, CD79B, CD320, ICAM3) overexpressed in PCs, of which five—CD267 (TNFRSF13B), CD59, CD92 (SLC44A1), CD32B (FCGR2B), and CD320—were prioritized due to their limited clinical exploration. Notably, CD320 was significantly upregulated in PCs from MM and RRMM patients, while prognostic analysis of ~2,000 MM patients from the Multiple Myeloma Research Foundation (MMRF) CoMMpass study (NCT01454297) revealed that CD320 and CD92 expression correlated with poorer outcomes, whereas CD267, CD59, and CD32B were associated with better prognosis. Flow cytometry validation confirmed the expression of CD267, CD59, CD92, and CD320 in MM cell lines and patient samples, while CD32B expression remained inconclusive.

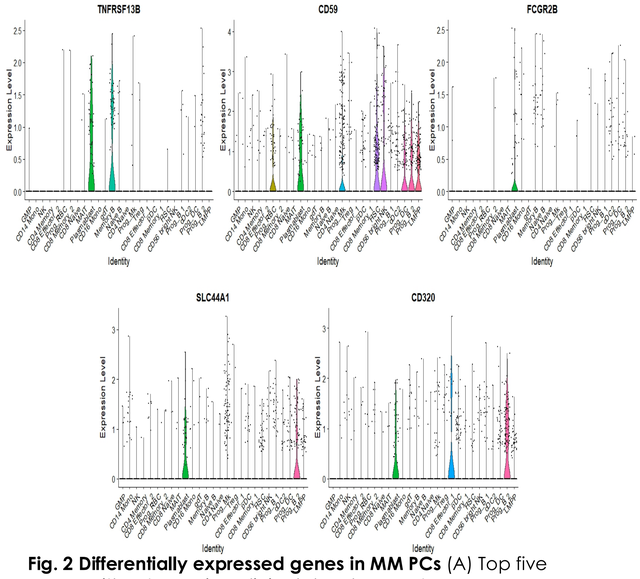


Fig. 2 Differentially expressed genes in MM PCs (A) Top five genes without ongoing clinical development

Summary

This approach led us to identify a series of potential new targets for MM patients, depicting a new scenario where each patient could be "screened" to identify the best molecule to be targeted. CD320 and SLC44A1 can be utilized as a marker for monitoring disease progression and predicting worse outcomes.

Discussion

These findings provide valuable insights into MM biology and highlight multiple potential targets—particularly CD320, CD92, and CD267—for CAR-T therapy, offering new strategies to enhance treatment efficacy and overcome resistance in MM patients. Inhibiting CD59 may increase tumor cell susceptibility to complement-mediated lysis, but this strategy poses significant risks and demands precise targeting to prevent damage to healthy tissues.

References

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