Carfilzomib Impairs the Innate Antiviral Immune Response and Promotes Cytotoxic T-cell Expansion in Oncolytic Virus Treated Multiple Myeloma Patients

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Introduction

Pelareorep is an infusible form of human Reovirus (RV) Serotype 3 – Dearing Strain, a naturally occurring, ubiquitous, non-enveloped double-stranded RNA virus. Our single-agent phase 1 RV trial in relapsed multiple myeloma (MM) showed it selectively infected MM cells but not the bone marrow (BM) stroma. However, apoptosis of cancer cells was not observed (PMID: 25294913). Our ongoing phase 1 trial, which combines the proteasome inhibitor carfilzomib (CFZ) with RV, has demonstrated RV infection, apoptosis, and clinical responses (NCT02101944) and recent published data have shown that in a mouse model PI resistant MM cells still respond to RV/BTZ combination treatment in terms of decreased tumor burden and improved overall survival (PMID: 30850386).

Here we further investigated the molecular mechanisms behind the beneficial effect of a PI (carfilzomib) in this setting.

Methods

- Flow Cytometry for Reovirus capsid detection—PBMCs or MM cells were fixed with 1% of formalin then stained with Purified Antibody (Protein G) Reovirus T3D for 30 min at room temp and incubated with an anti-goat IgG PE conjugated as a secondary antibody.
- Phagocytosis assay—Total PBMC isolated from different HDs were treated overnight with RV (10 MOI) and incubated with an anti-goat IgG PE conjugated as a secondary antibody. After 24h, flow cytometry analysis using FACS gaiting was used to assess phagocytic activity of the monocytes against infected target cells.
- PBMCs obtained from two patients enrolled on NCI 6033 (NCT02101944) were used for assessing immunological changes upon RV/CFZ treatment at different time points after RV infusion. Flow cytometry analysis was used to assess CD14+, CD8+ and CD4+ frequency in total PBMCs.

The combination CFZ plus RV is not able to induce a synergistic effect on Apoptosis of MM cell lines, in vitro

Conclusion

- Carfilzomib enhances reovirus entry, infection, and killing of myeloma cells through its effect on the CD14+ fraction. Reovirus infection and replication within CD14+ cells is augmented because Carfilzomib inhibits the early innate pro-inflammatory immune response.
- Reovirus significantly increases CD14+ frequency and activation/polarization in the monocytic fraction (CD14+), increasing the phagocytic activity against MM cells.
- Our data suggests that the combination Carfilzomib plus the oncolytic virus Pelareorep (Reovirus) increases the total frequency of cytotoxic T-cells.