EX VIVO CHEMOTHERAPY RESISTANCE OF METABOLICALLY REPROGRAMMED AUTOLOGOUS TH1/TC1 CELLS (RAPA-201): TOWARDS THE CLINICAL TRANSLATION OF IMMUNE-SPARING HOST CONDITIONING

1Jee Hyun Park*, 2Deborah Glass, 1Martin Gutierrez, 1Tania Felizardo, 1Daniel H Fowler. 1Rapa Therapeutics, Rockville, MD, USA; 2Hackensack Meridian Health, Hackensack, NJ, USA

Background

Manufactured RAPA-201 cell therapy products are enriched for central-memory, polarized to Th1/Tc1, and rendered immune checkpoint-deficient and homeostatic cytokine-responsive. RAPA-201 safely mediates multiple myeloma remissions when administered after lymphodepleting conditioning and induces remissions in solid tumor patients in the post-PD-(L)-1 setting using immune-sparing conditioning [carboplatin (CBCCA) plus paclitaxel (PTX)] (separate abstract submitted by Gutierrez et al, SITC 2023; clinicaltrials.gov, NCT05144698). This latter clinical translation was motivated in part by our finding, described herein, that RAPA-201 demonstronstrate resistance to CBCCA plus PTX.

Methods

RAPA-201 were manufactured using one-week culture in temsirolimus- and IFN-α-containing media and tested for sensitivity to chemotherapy inhibition (temsirolimus, TEM; etoposide, VP-16; paclitaxel, PTX; carboplatin, CBCCA; and topotecan, TOPO; see table 1/figure 1: concentrations and exposure intervals). Peripheral blood mononuclear cells (PBMC) served as negative control whereas positive controls consisted of pancreatic cancer cells (MIA-PaCa-2 line) and lung cancer cell lines (NCI-H1975 and NCI-H820). Multidrug resistance (MDR) activity factor (MAF) values were determined by flow cytometry efflux assays (Abcam, ab294534).

Results

Relative to MIA-PaCa-2, RAPA-201 had increased resistance to TEM (table 1, EXPT #1: 9.8% vs. 69.0% yield, respectively), VP-16 (16.3% vs. 82.8% yield), and PTX (0.0% vs. 55.2% yield) and similar resistance to CBCCA (205.6% vs. 103.4% yield). Similar results were obtained in a second experiment (table 1, EXPT #2). Relative to PBMC, RAPA-201 had increased resistance to combination CBCCA + PTX (table 1, EXPT #1: 29.7% vs. 61.7% yield; EXPT #2: 63.4% vs. 138.2% yield). Relative to cellular controls (NCI-H1975, NCI-H820, PBMC), RAPA-201 had increased topotecan resistance (figure 1). MDR activity was not detected in control PBMC (MAF score 0); in contrast, in two separate experiments, RAPA-201 had functional MDR activity for MDR1 (median MAF: 5.2, 15.9), MRPI/2 (median MAF: 6.1, 24.7), and BCRP (median MAF: 8.7, 8.2).

Conclusions

RAPA-201 are globally resistant to numerous chemotherapy agents, including carboplatin, paclitaxel, temsirolimus, etoposide, and topotecan. The mechanism of RAPA-201 resistance may involve MDR activity; however, as we have previously demonstrated, modulation of intrinsic apoptotic signals is also likely operational. It is important to harness RAPA-201 chemotherapy resistance for clinical translation, as we have now successfully achieved through use of CBCCA/PTX immune-sparing conditioning to safely facilitate RAPA-201 regression of post-PD-(L)1 solid tumors. Finally, given RAPA-201 resistance to topotecan, which is in the topoisomerase I drug class commonly utilized as payload for antibody-drug-conjugates (ADC), it will be important to clinically translate combination therapy involving RAPA-201 and ADC agents.

Abstract 1028 Table 1

<table>
<thead>
<tr>
<th>Specific Chemotherapy Agent Tested</th>
<th>(% Yield Relative to No Inhibition)</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>TEM</td>
</tr>
<tr>
<td>EXP 41</td>
<td>MIA-PaCa-2</td>
</tr>
<tr>
<td>EXP 42</td>
<td>MIA-PaCa-2</td>
</tr>
</tbody>
</table>

Abstract 1028 Figure 1

The NCI-H1975 and NCI-H820 lung cancer cell lines were maintained in exponential growth phase and then subjected to further culture in complete media with or without addition of topotecan (30 nM). At 72-hours of culture with or without topotecan, absolute numbers of viable lung cancer cells were enumerated using flow cytometry and nucleocounter instruments, with results expressed as the percent of viable cells relative to the media control condition. For the T cell cultures, non-manufactured CD4+ and CD8+ T cells (‘Control’) or ex vivo manufactured RAPA-201 T cells from a patient with non-small cell lung cancer were brought into growth phase using anti-CD3/anti-CD28 co-stimulation combined with cytokine support (IL-2, IL-7, IL-15). After 48-hours, control or RAPA-201 T cells were then subjected to further culture in cytokine-containing media with or without addition of topotecan (30 nM). At 72-hours of culture with or without topotecan, absolute numbers of viable T cells were enumerated using flow cytometry and nucleocounter instruments, with results expressed as the percent of viable cells relative to the media control condition.

http://dx.doi.org/10.1136/jitc-2023-SITC2023.1028