Assessment of sensitivity to a PD-1 check point inhibitor and cisplatin in bladder cancer patient-derived xenografts with various levels of PD-L1 expression in HuCD34NCG mice

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INTRODUCTION

Bladder cancer is the ninth most common cancer in the US, and the ninth most common cancer worldwide. Treatment of bladder cancer has evolved over time to encompass traditional modalities of chemotherapy and surgery, but has been particularly impacted by the recent use of immunotherapy. Modern immunotherapy has focused on checkpoint protein inhibitors that impede immune function. The cisplatin in a panel of bladder patient-derived xenografts (PDX) with distinct patterns of PD-L1 expression in CD34+ stem cell humanized ngs (HuCD34 NCG) mice.

METHODS

Generation and Maintenance of Bladder PDX tumor models

Three bladder PDX models, PNX0428, PNX0434 and PNX1028 have been established under informed consent from the patients at the Fox Chase Cancer Center, Philadelphia. The impersonalized patient information is presented in Table 1.

RESULTS: PD-L1 Staining

Figure 1. IH staining of PNX0428 tumor tissue with PD-L1 antibody SP263 (Ventana). A -1 and B -20x magnification. An immunostaining with anti-PD-L1 rabbit monoclonal antibody SP263 determined islands of PD-L1 positive cells in PNX0428 bladder PDX tissues. The staining estimated 70% of the cells were positive for PD-L1 protein. IH staining of PNX0434 and PNX1028 tumor tissues with anti-PD-L1 antibody were considered negative (data not shown).

RESULTS: Humanization Levels of HuCD34 NCG mice

Table 2. PDX Model: Characterization

<table>
<thead>
<tr>
<th>Model</th>
<th>Type</th>
<th>Disease</th>
<th>Tumor Implantation</th>
<th>Survival (Weeks)</th>
<th>Functional endpoint</th>
</tr>
</thead>
<tbody>
<tr>
<td>PNX0428</td>
<td>Bladder</td>
<td>Bladder Cancer</td>
<td>Subcutaneously</td>
<td>12</td>
<td>50 %</td>
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RESULTS: Tumor Study

Figure 3. Efficacy study in three Bladder PDX xenografts identified PD-L1 negative PDX models as the most sensitive to treatment with check point inhibitor Pembrolizumab. HuCD34 NCG mice and NCG mice were implanted with bladder PDX xenografts and treated with Pembrolizumab and Cisplatin. Treatment began on Day 0 in mice with established PDX tumors (tumor size ~100 mm3). The study endpoint was at Day 21 or when tumors reach a volume of 2000 mm3. Data are shown as Median ± SD.

Table 3. Analysis of treatment efficacy in HuCD34 NCG and NCG mice.

<table>
<thead>
<tr>
<th>Group</th>
<th>Treatment</th>
<th>Tumor Volume (mm3)</th>
<th>TTE (Days)</th>
<th>Mean BW (g)</th>
<th>Percent of NCG Mice Positive</th>
<th>p-value</th>
<th>%TGI</th>
</tr>
</thead>
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<tr>
<td>PNX0428</td>
<td>Pembrolizumab</td>
<td>720 ± 23</td>
<td>28</td>
<td>20 ± 2</td>
<td>70%</td>
<td>&lt;0.001</td>
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CONCLUSION

- Confirmed ability of three proprietary bladder PDX models to form tumors in HuCD34 NCG mouse model.
- Evaluated immuno-oncology therapy Pembrolizumab and Standard of Care chemotherapy regimen. Pembrolizumab in three bladder PDX models with various levels of PD-L1 expression in HuCD34 NCG and standard NCG mice.
- Tumor treatment with Pembrolizumab developed new adverse effects during the study. Treatment with Pembrolizumab was associated with significant weight loss in both HuCD34 NCG and standard NCG mice.
- Treatment with Pembrolizumab produced significant tumor growth inhibition in all three PDX models and the effect was not associated with the type of NCG mice.
- Treatment of PNX0434 and PNX1028 models with Pembrolizumab in HuCD34 NCG mice produced statistically significant tumor growth inhibition that is demonstrative of a stable disease phenotype in patients.
- Data indicate that abundant expression of PD-L1 protein in tumors should not be used as the only biomarker for patient stratification for the treatment with PD-1/PD-L1 check point inhibitors.
- The HuCD34 NCG mouse model is an effective tool for supporting tumor growth and evaluating immunotherapies.

Acknowledgements: The authors thank Dr. Qi Cai from the animal pathology facility at Fox Chase Cancer Center for her time and efforts in anti-PD-L1 IHC analysis and Dr. Christopher Steele from Charles River Discovery for performing flow cytometry.