


5. Jerby-Arnon L, Shah P, Cuoco MS, Rodman C, Su MJ, Melms JC, Leeson R, Kano-ries against melanoma-associated and cancer testis antigens NY-ESO-1, SSX2 and MAGEA10. Antibodies were observed in 13 of 4 patients identified several putative antigens. Immuno-precipitation with IVTT candidate proteins confirmed antibod-ies against melanoma-associated and cancer testis antigens NY-ESO-1, SSX2 and MAGEA10. Antibodies were observed in 1 of 1 (100%) patient with a complete response, 2 of 4 (50%) with a partial response, 1 of 1 (100%) with stable disease, and 0 of 6 (0%) with progressive disease. Antibody levels varied over the course of therapy, with previously undetectable specificities arising during treatment response in patients #1–3. Patient #1 with a complete tumor regression developed antibodies to SSX2 and MAGEA10 that were absent before treatment. Further, detection of these antibodies coincided with diagnosis of IRAEs (anti-SSX2 with pancreatitis and anti-MAGEA10 with dermatitis). In contrast, patient #3, initially with a partial tumor regression, demonstrated a loss of detect-able anti-NY-ESO-1 antibodies upon disease progression, and subsequent metastasectomy demonstrated loss of NY-ESO-1 protein expression in the progressing tumor. Testing sera from all 12 patients with IVTT products for NY-ESO-1, SSX2 and MAGEA10 did not reveal additional humoral responses.

Conclusions Our comprehensive screening platform detected circulating antibodies specific to multiple melanoma-associated and cancer testis antigens in patients deriving clinical benefit from ICI. Expanded investigations of the evolution of antibody production over the course of ICI therapy, associated with tumor response to treatment and development of IRAEs, are warranted.

Acknowledgements This study was supported by the Johns Hopkins Bloomberg-Kimmel Institute for Cancer Immunother-apy, and NIH P30-AR070254.

Ethics Approval This study was approved by the Johns Hop-kins Institutional Review Board, approval #NA_00090257.

http://dx.doi.org/10.1136/jitc-2020-SITC2020.0231

Abstract 231 Table 1 Antibodies detected in the serum or plasma of patients with metastatic melanoma treated with ICI therapy. Treatment response indicates best overall response according to RECIST v1.1. Post-treatment blood collections were drawn during or after ICI therapy.

<table>
<thead>
<tr>
<th>Antibody</th>
<th>NY-ESO-1</th>
<th>SSX2</th>
<th>MAGEA10</th>
<th>NY-ESO-1 + SSX2</th>
<th>NY-ESO-1 + MAGEA10</th>
<th>NY-ESO-1 + SSX2 + MAGEA10</th>
<th>NY-ESO-1 + SSX2 + MAGEA10 + NY-ESO-1</th>
<th>NY-ESO-1 + SSX2 + MAGEA10 + NY-ESO-1 + SSX2</th>
</tr>
</thead>
<tbody>
<tr>
<td>F</td>
<td>++</td>
<td>++</td>
<td>+</td>
<td>++</td>
<td>++</td>
<td>++</td>
<td>++</td>
<td>++</td>
</tr>
<tr>
<td>M</td>
<td>++</td>
<td>++</td>
<td>+</td>
<td>++</td>
<td>++</td>
<td>++</td>
<td>++</td>
<td>++</td>
</tr>
</tbody>
</table>

Background Tumor-specific antibodies have been reported in patients with cancers responding to immune checkpoint inhibitors (ICI), and there is an increasing appreciation for the potential role of B cells in mediating ICI responses. However, the humoral immune response to melanoma remains incompletely defined. We hypothesized that screening sera for antibodies by immunoprecipitation with lysates of cultured melanoma cells would increase the likelihood of detecting circulating antibodies in melanoma patients receiving ICI, and potentially identify novel antibody targets associated with treatment response and/or immune-related adverse events (IRAEs).

Methods Pre-and on/post-treatment sera or plasma from 12 clinically-annotated patients with advanced metastatic melano-ma receiving ICI were assayed for tumor-specific antibodies with an established immunoprecipitation platform. 353-methio-nine-labeled lysates from cultured 624Mel cells were used for immunoprecipitation. 624Mel expresses several shared non-mutated melanoma antigens (e.g., MAGEA3, tyrosinase, MART-1/Melan-A, gp75, and gp100). Antigen identity was determined using on-bead digests followed by mass spectrometry, and was confirmed by immunoprecipitation with in vitro transcription/translation (IVTT) products.

Results Antibodies reactive against 624Mel proteins were detected in 4 of 12 (33%) patients (table 1). Mass spectromet-ric sequence performed on proteins captured with sera from 3 of 4 patients identified several putative antigens. Immuno-precipitation with IVTT candidate proteins confirmed antibod-ies against melanoma-associated and cancer testis antigens NY-ESO-1, SSX2 and MAGEA10. Antibodies were observed in 1

232

THE EPITHELIAL-TO-MESENCHYMAL TRANSITION (EMT) CONTRIBUTES TO IMMUNOSUPPRESSION IN BREAST CARCINOMAS AND REGULATES THEIR RESPONSE TO IMMUNE CHECKPOINT BLOCKADE

Anushka Dongre1, Robert Weinberg2, Mohammad Rashidian3, Eliron Eaton4, Ferenc Reinhart1, Prat Thius5, Maria Zagoruyko6, Sunita Nepal6, Tibba Banaz7, Anna Martner8, Stefani Spranger9. Whitehead Institute for Biomedical Research, Cambridge, MA, USA; Stanford University School of Medicine, Stanford, CA, USA; Koch Institute MIT, Cambridge, MA, USA; University of Gothenburg, Gothenburg, Sweden

Background Immune checkpoint blockade (ICB) has generated some dramatic responses in certain types of human tumors, most notably, melanomas. However, the response of breast tumors has been largely limited. We have previously demonstrated that the residence of breast cancer cells in the epithelial or mesenchymal phenotypic states can itself be used as an important determinant of the success or failure of ICB. Specifically, we have shown that while epithelial tumors are sensitive

http://dx.doi.org/10.1136/jitc-2020-SITC2020.0231