Comprehensive Profiling of Metaplastic Breast Carcinoma Reveals Frequent Over-Expression of PD-L1

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Abstract (updated)
Background: Metaplastic breast carcinoma (MBC) is a rare subtype of breast carcinoma less responsive to conventional chemotherapy relative to usual breast carcinomas such as ductal and lobular types. MBC usually consists of triple negative breast carcinoma (TNBC), but MBCs portray a worse prognosis in comparison with TNBC. Published studies investigating MBCs for specific biomarkers of therapy response are rare and limited by the methodological approaches.

Methods: 297 samples [MBC (n=75), triple-negative breast cancer of no-special-type (TNBC-NOS, n=106), HER2-positive breast cancers (n=32) and luminal breast carcinomas (n=84)] were profiled using direct sequencing analysis (Illumina MiSeq Next Generation Sequencing (NGS)) and immunohistochemistry for PD-L1 (SP142, Spring Bioscience) and PD-1 (EH121, Pharmingen) was performed using automated procedures.

Results (updated)

Mean age of patients: 57 years (range, 35-93 years).

Histology: MBCs exhibited various morphologic features including squamous, myoepithelial, spindle, and rhabdoid morphology with heterologous elements: bone (n=3) and cartilage (n=22 cases).

ER, PR and HER2 status were available for 71 patients; 63 cases (89%) of MBCs were HER2-negative, 4 cases (6%) of MBCs were HER2-positive, and 4 cases (6%) of MBCs were HER2-negative and HER2-positive.

PD-L1 and TILs status; The proposed stratification is a framework for tailored immunotherapy against tumor microenvironment (TME); PD-L1 positive tumors infiltrating lymphocytes (TILs) varied greatly in MBCs (0 to 400/mm², mean: 67.3). No significant association was found between the number of TILs and PD-L1 status (p=0.209).

Overall PD-L1 positivity among 75 metaplastic breast carcinomas was 46%, highest of all types.

Conclusions
Metaplastic carcinomas are characterized by increased PD-L1 expression in carcinoma cells, and PD-1 expression in TILs, which can be exploited in clinical trials utilizing immune check point inhibitors in this hard-to-treat subtype of breast cancer.

Comprehensive mutational profiling of MBC highlighted predominance of TP53 and PIK3CA mutations and a wild type EGRF gene expression.

References
4. Zhang Y, Toy KA, Kleer CG. Metaplastic breast carcinomas are enriched in markers of tumor-initiating cells and are more responsive to anti-EGFR therapy. Cancer Res 2011;71:5486-95.
6. Figure 1A-D, 2A-D. Despite various morphologic appearances (A, C) of metaplastic carcinomas, strong PD-L1 overexpression (B, D) was observed in nearly 50% of the cases.

Table 2. The stratification of MBCs into 4 types (S) on the basis of PD-L1 and TILs status. The proposed stratification is a framework for tailored immunotherapy against tumor microenvironment (TME).

<table>
<thead>
<tr>
<th>Type</th>
<th>TME (PD-L1/TIL)</th>
<th>Frequency (%)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Type 1 (PD-L1+/TIL-)</td>
<td>29</td>
<td>40.8%</td>
</tr>
<tr>
<td>Type 2 (PD-L1-, TIL+)</td>
<td>14</td>
<td>19.7%</td>
</tr>
<tr>
<td>Type 3 (PD-L1-, TIL-)</td>
<td>4</td>
<td>5.6%</td>
</tr>
<tr>
<td>Type 4 (PD-L1+, TIL+)</td>
<td>24</td>
<td>33.8%</td>
</tr>
<tr>
<td>Total</td>
<td>71</td>
<td>100%</td>
</tr>
</tbody>
</table>

Table 1. PD-L1 expression was significantly higher in metaplastic carcinomas in comparison with other breast cancer subtypes (46% vs. 6-9%, p<0.001).

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Methods
The study included 297 samples [MBC (n=75), triple-negative breast cancer of no-special-type (TNBC-NOS, n=106), HER2-positive breast cancers (n=32) and luminal breast carcinomas (n=84)]. The latter properties can be exploited in clinical trials with triple negative (ER-/PR-/Her2-) breast cancer (TNBC), and luminal (ER+/Her2+) breast cancers (n=84)). The samples were profiled using direct sequencing analysis (Illumina MiSeq Next Generation Sequencing (NGS)) using formalin-fixed paraffin embedded tissue blocks. Immunohistochemistry for PD-1 (SP142, Spring Bioscience) and PD-1 (EH121, Pharmingen) was performed using automated procedures.

Results
PD-L1 expression was significantly higher in metaplastic carcinomas in comparison with other breast cancer subtypes (46% vs. 6-9%, p<0.001).

Figure 1 Figure 2

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Figure 2

SDMA4 genes. None of the cases harbored EGRF mutation.

Mutational profiling of metaplastic carcinoma

72 MBCs were tested by NGS of which 57 cases had interpretable results. Mutations were detected in 16 out of 45 tested genes affecting 48 out 57 metaplastic carcinomas (84%). TP53 mutation was the most frequent mutation (32/57, 56%). In 20 cases, TP53 was the sole mutation detected in the tumor tissue while in the remaining 12 cases other mutations co-occurred. PIK3CA mutation was the second most commonly detected mutation (13/72, 23%) while HRAS mutations were detected in 4 cases (7%).

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Conclusions
Metaplastic carcinomas are characterized by increased PD-L1 expression in carcinoma cells, and PD-1 expression in TILs, which can be exploited in clinical trials utilizing immune check point inhibitors in this hard-to-treat subtype of breast cancer.

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References