S6-01
JAK2 amplifications are enriched in triple negative breast cancers (TNBCs) after neoadjuvant chemotherapy and predict poor prognosis

Dr. Balko: Nothing to disclose.
Dr. Giltinane: Nothing to disclose.
Dr. Schwarz: Nothing to disclose.
Dr. Sanders: Nothing to disclose.
Dr. Wang: Salary, Foundation Medicine; Ownership Interest, Foundation Medicine.
Dr. Harris: Nothing to disclose.
Dr. Lin: Nothing to disclose.
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Dr. Gomez: Nothing to disclose.
Dr. Arteaga: Nothing to disclose.

JAK2 amplifications are enriched in triple negative breast cancers after neoadjuvant chemotherapy and predict poor prognosis

Balko JM\textsuperscript{1,4}, Giltinane JM\textsuperscript{3,4}, Schwarz L\textsuperscript{1}, Sanders ME\textsuperscript{3,4}, Wang K\textsuperscript{5}, Harris LN\textsuperscript{6}, Lin NU\textsuperscript{7}, Miller VA\textsuperscript{5}, Stephens PJ\textsuperscript{5}, Yelensky R\textsuperscript{5}, Pinto JA\textsuperscript{8}, Gomez H\textsuperscript{9}, Arteaga CL\textsuperscript{1,7,4}

\textsuperscript{1}Departments of Medicine\textsuperscript{1}, Cancer Biology\textsuperscript{1}, and Pathology\textsuperscript{4} and Breast Cancer Research Program\textsuperscript{4}, Vanderbilt-Ingram Cancer Center; Vanderbilt University, Nashville, TN
\textsuperscript{2}Foundation Medicine, Cambridge, MA
\textsuperscript{3}Breast Cancer Program, UH Seidman Cancer Center, Cleveland OH
\textsuperscript{4}Department of Medicine, Dana Farber Cancer Center, Boston MA
\textsuperscript{5}Oncosalud, Lima, Perú; \textsuperscript{6}Instituto Nacional de Enfermedades Neoplásicas (INEN), Lima, Perú
Background

- Neoadjuvant chemotherapy (NAC) is used increasingly in triple-negative breast cancer (TNBC)
- The presence of residual disease (RD) at surgery correlates with an increased risk of metastatic recurrence
- We undertook integrated molecular analysis of the residual disease from a cohort of TNBCs after NAC to identify clinically actionable lesions that may:
  1. Be causally associated with chemotherapeutic resistance
  2. Be targetable in the adjuvant setting to eliminate residual micrometastases

Cohort

<table>
<thead>
<tr>
<th></th>
<th>Median</th>
<th>Min</th>
<th>Max</th>
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<td>3%</td>
<td></td>
</tr>
<tr>
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</tr>
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<tr>
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</tr>
<tr>
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<td>Node status</td>
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<tr>
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<td>4</td>
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</table>

Balko et al, Cancer Discovery, in press.
Deep sequencing of the residual disease in NAC-treated TNBC

- 182 oncogenes and tumor suppressors in a CLIA certified lab (Foundation Medicine, Cambridge MA)
- Data were evaluable for 81 tumors, with a sufficient coverage to determine CNAs in 72/81
- Mean depth of coverage was 635 (range: 135-1207)

Balko et al, Cancer Discovery, in press.

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JAK2

- Janus-kinase 2 (JAK2) is a receptor-coupled tyrosine kinase which transmits cytokine-mediated signals to the STAT pathway to drive proliferation and differentiation
- JAK2/STAT signaling has been shown to play a role in promoting breast cancer ‘stemness’ and driving the proliferation of CD44+/CD24-basal-like breast cancer cells.


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JAK2 gains and amplifications are confirmed by a novel JAK2-FISH assay

PT 72003

Amplified cases
Average JAK2-CEN9 ratio

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JAK2 copy number increases with treatment and metastatic progression

• JAK2 gains and amplifications were more frequent in NAC-treated TNBC than in primary untreated BLBC (TCGA)

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JAK2 amplifications are rare in untreated primary TNBC

- Overt JAK2 amplifications appear to be rare in primary breast tumors, but do exist
- Primary untreated TNBC from a patient from City of Hope>>
  - Courtesy of Jean Simpson, MD; N=100 mixed subtypes

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JAK2 amplifications are associated with high JAK2 mRNA expression and p-STAT3 in the TCGA

Data extracted from cBio data portal (TCGA)

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JAK2 amplifications are represented in cell line and patient-derived xenograft (PDX) models

### Breast Cancer Cell lines

<table>
<thead>
<tr>
<th>Cell Line</th>
<th>JAK2 Amplified</th>
</tr>
</thead>
<tbody>
<tr>
<td>KCC-3</td>
<td>*</td>
</tr>
<tr>
<td>MD4-21</td>
<td>*</td>
</tr>
<tr>
<td>MD4-364</td>
<td>*</td>
</tr>
<tr>
<td>MD4-56</td>
<td></td>
</tr>
<tr>
<td>MD4-78</td>
<td></td>
</tr>
<tr>
<td>KCC-70</td>
<td></td>
</tr>
</tbody>
</table>

Data courtesy of Melissa Landis and Jenny Chang

### TNBC PDX models (N=22)

Data courtesy of Melissa Landis and Jenny Chang

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JAK2 amplifications are a negative prognostic factor in TNBC

<table>
<thead>
<tr>
<th>N</th>
<th>Percent recurrence-free</th>
<th>Percent survival</th>
</tr>
</thead>
<tbody>
<tr>
<td>67</td>
<td>0.005</td>
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</tbody>
</table>

![Survival curves](image)

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Genes increased in JAK2-amplified patient tumors

- TGF-β-induced genes
- EMT gene signatures
- IL-6 expression

The JAK2 pathway in breast cancer

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Conclusions

- JAK2 amplifications occur in ~10% of neoadjuvant chemotherapy-treated TNBCs and are enriched during therapy and metastatic progression.
- Amplifications in JAK2 are represented in cell line and PDX models, representing useful tools for translational experiments.
- JAK2 amplifications correlate with higher expression of JAK2 and activation of downstream STAT3 which is sensitive to ruxolitinib treatment.
- JAK2 amplifications are associated with very poor prognosis and gene expression signatures of EMT.
- Ruxolitinib is a clinically approved targeted therapy which may be of benefit in JAK2-amplified patients, and this hypothesis is currently under investigation in translational and clinical studies.

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- Jenny Chang, MD

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