René Bartz

4SC-202 plus Anti-PD1: Breaking PD1-refractoriness to increase efficacy of checkpoint inhibition in patients with advanced melanoma
With reference to this presentation, I hereby declare

that I do not conduct activities that would involve a conflict of interest with CME-accreditable training, but that in the past 2 (two) years I have been a paid employee of 4SC AG.
Introduction

• Current strategies in the I/O space
• Histone deacetylase inhibitor (HDACi) 4SC-202 as combination partner
  o Rationale and preclinical evidence
• Translation of findings into the clinic
  o Study design and biomarker program
Checkpoint Inhibitors have revolutionized Cancer Therapy

- Checkpoint inhibitors (CIs) are the new paradigm for the treatment of cancer patients in many indications
- CIs overcome tumor-specific immune escape mechanisms

**Immune escape**
Evasion strategies by tumor cells; expression of cell surface PD-L1 molecules

**Checkpoint blockade**
Pharmacological intervention of PD-1/PD-L1 binding

**Tumor Elimination**
Inhibition of T-lymphocytes is abrogated; elimination of tumor cells

PD-1: Programmed cell death protein 1
PD-L1: Programmed death-ligand 1
CD8: Cytotoxic T-Cell (CD8+)
Checkpoint inhibitors: a success story

• First approved in 2011 (ipilimumab; CTLA-4) in melanoma
  o Since then: mostly PD-1/ PD-L1 antibodies
• Quite dramatic responses in some patients
  o Increasing number of indications; some even as First-Line-Treatment
Additional anti-cancer therapies in the immuno-oncology space required

- High unmet medical need demands alternative/ additional treatment options
  - Low response rates in some cancers

### Anti-PD-1/PD-L1 – Overall response rates

<table>
<thead>
<tr>
<th>Indication</th>
<th>Response Rate [%]</th>
</tr>
</thead>
<tbody>
<tr>
<td>Non-small cell lung cancer (NSCLC), squamous and non-squamous</td>
<td>15-20%</td>
</tr>
<tr>
<td>Small cell lung cancer (SCLC)</td>
<td>15%</td>
</tr>
<tr>
<td>Renal cell carcinoma</td>
<td>15-20%</td>
</tr>
<tr>
<td>Bladder cancer</td>
<td>25%</td>
</tr>
<tr>
<td>Gastric cancer</td>
<td>20%</td>
</tr>
<tr>
<td>Ovarian cancer</td>
<td>15%</td>
</tr>
<tr>
<td>Triple negative breast cancer (TNBC)</td>
<td>20%</td>
</tr>
<tr>
<td>Melanoma</td>
<td>40%</td>
</tr>
</tbody>
</table>

Adapted from: Curie Institute; Bryan, Garnier & Co.
Combining therapies to address the needs adequately

- Possible approaches:
  - Novel checkpoint inhibitors
  - Combination therapies (chemotherapy, radiotherapy, other entities…)
    - Modulation of multiple pathways
    - Variable targeting (e.g. tumor microenvironment/ tumor/ immune cells)
    - Sequence of existing therapies

Explosion in combinatorial therapies
The Cancer-Immunity Cycle

- Complex relationship between tumor, tumor microenvironment and immune system
- Leaves multiple ways for pharmacological intervention

- Chemotherapy
- Radiotherapy
- Targeted Therapy

- Anti-CTLA4
- Anti-VEGF
- Vaccines
- CARs
- Anti-PD-1
- Anti-PD-L1
Why do some patients respond to CIs – and some not?

- Degree of inflammation may play a critical role in facilitating bodies own immune system to fight cancer
- Identification of biomarkers
  - Tumor microenvironment
    - Number, type and location of immune cells
    - Other immunological parameters (mutational load, cytokines)
    - Predisposition of tumor microenvironment rendering it more or less susceptible to CI treatment?

Conversion from noninflamed to inflamed tumor by epigenetic intervention?
Epigenetic Modulation in Oncology

• What is Epigenetics?
  o Heritable changes in gene expression (active versus inactive genes)
  o No change in the underlying DNA sequence
  o Example: histone acetylation/deacetylation

• Epigenetic regulation is often disturbed in cancer cells
  o Vorinostat/ SAHA (CTCL).
  o Romidepsin (CTCL).
  o Panobinostat multiple myeloma
Pleiotropic effects upon epigenetic intervention

• Current evidence suggests that epigenetic mechanisms play an important part in tumor immune evasion

• Changes in expression with immunological relevance
  o Neoantigen expression (e.g. ‘TINATS’)
  o MHC presentation machinery
  o Rejuvenating exhausted effector cells
  o Release of proinflammatory chemokines
  o Direct anti-tumor effects

Enter 4SC-202

- Small molecule HDAC class 1 inhibitor (HDAC 1, 2 and 3)
- Orally available
- Phase I data in 24 patients ‘TOPAS’
  - study in patients with hematological malignancies
  - completed 2015
  - safe, well tolerated with anti-cancer activity
    - 1CR, 1 PR

Important: Checkpoint inhibitors still required
4SC-202 in preclinical mouse models

- Comparison of models with/without functional immune system

CT26 mouse model

Anti-tumor activity observed with 4SC-202 only syngenic mouse tumor model (murine colon carcinoma).

How does the tumor microenvironment compare +/- 4SC-202 treatment?
Increased influx of immune effector cells

- 4SC-202 stimulates infiltration of CD8⁺/CD4⁺ immune cells into tumor
- This is not due to an increased proliferation of cells
Increased infiltration CD8$^+$ T-Cells

Vehicle control [CD3/ CD8]

4SC-202 [CD3/ CD8]
Enhanced expression of MHC

- 4SC-202 increases the expression of MHC molecules
Effect of HDAC inhibitors on T-Cell viability

T-Cell viability [IC50; µM]

<table>
<thead>
<tr>
<th>HDAC inhibitor</th>
<th>CD4⁺ T cells*</th>
<th>CD8⁺ T cells*</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>naive</td>
<td>memory</td>
</tr>
<tr>
<td>Vorinostat</td>
<td>1.5</td>
<td>1.4</td>
</tr>
<tr>
<td>Mocetinostat</td>
<td>1.3</td>
<td>1.4</td>
</tr>
<tr>
<td>Entinostat</td>
<td>2.1</td>
<td>2.7</td>
</tr>
<tr>
<td>4SC-202</td>
<td>&gt;10</td>
<td>&gt;10</td>
</tr>
</tbody>
</table>

* T cells were stimulated with CD3/28 beads; different subpopulations were isolated using appropriate Miltenyi kits according to manufacturer's instructions; cells were treated with indicated compounds for 48 h; viability was determined using Vialight Kit (Lonza); ** up to 10 µM

4SC-202 appears to be less toxic to immune effector cells when compared to other HDAC inhibitors.
Immune modulatory effects of 4SC-202

- Epigenetic modulation changes the tumor microenvironment
  - Increased infiltration of immune cells into tumor
  - Enhanced expression of MHC molecules
  - Induction of tumor associated antigen expression
  - Increases expression of chemokines like IFN-γ in TME

- Some 'stand-alone' anti-tumor effects by 4SC-202
  - Less toxic to effector cells when compared to other HDACi

- Synergistic effects in combination with checkpoint inhibitors?
4SC-202 synergizes with PD-1 blockade in C38 tumor models

- Some single agent activity with PD-1 blockade and 4SC-202
- Combination of both results ins stronger anti-tumor activity
4SC-202 in combination with CIs

Combination of 4SC-202 and checkpoint inhibitor reduces tumor burden and increases survival in animal models.
Translation into the clinic: SENSITIZE

- Phase Ib single arm study in patients with unresectable stage III or stage IV cutaneous melanoma
- Patients must be primary refractory or non-responding to anti-PD-1 monotherapy
- 3 dose cohorts [100, 200, 2 x 200 mg 4SC-202 + Pembrolizumab 2 mg/kg q3w]
SENSITIZE Study Objectives

• Primary
  o The primary objective of the study is to determine safety and tolerability of combination treatment with 4SC-202 and Pembrolizumab
  o AEs, lab tests, vital signs, ECG, ECOG PS, physical examination, concomitant medication

• Secondary
  o Examine preliminary efficacy of combination treatment with 4SC-202 and Pembrolizumab
  o Determine
    • Non-tolerated dose (NTD)
    • Maximum tolerated dose (MTD)
    • Recommended phase 2 dose (RPTD)
  o Characterize pharmacokinetics (PK) of 4SC-202
SENSITIZE dosing scheme and exploratory objectives

- Exploratory Endpoint Biomarker assessment
  - Gene expression tumor and blood
  - IHC Analysis (tumor; skin biopsies)
  - Exosome collection and analysis
- PK/ PD sampling
  - PK/PD relationship analysis planned for some of the patients

**Dosing scheme for dose finding part**

<table>
<thead>
<tr>
<th></th>
<th>4SC-202</th>
<th>Treatment pause</th>
</tr>
</thead>
<tbody>
<tr>
<td>Days 1-14</td>
<td>Days 15-21</td>
<td>Cycle 1</td>
</tr>
<tr>
<td>Days 1-14</td>
<td>Days 15-21</td>
<td>Cycle 2 and 3</td>
</tr>
<tr>
<td>Days 1-14</td>
<td>Days 15-21</td>
<td>Cycle 4</td>
</tr>
<tr>
<td>Days 1-14</td>
<td>Days 15-21</td>
<td>Cycle x</td>
</tr>
</tbody>
</table>

- biopsy sample for biomarker analysis
- administration of pembrolizumab

*Important: translation of preclinical findings into the clinic*
SENSITIZE is currently recruiting

- 6 cancer centers in Germany
- PI: Dirk Schadendorf, Essen
Reserves
Clinical trials with comparator molecules

• Entinostat NCT02437136 (ENCORE 601)
  o Ph1b/2 dose-escalation study combining entinostat and pembrolizumab in NSCLC
  o Expansion Cohorts in Melanoma and Colorectal Cancer