ESMO 2012 – MONDAY OCTOBER 1st
Special symposium. Melanoma therapy: from frustration to enthusiasm
Chemotherapy and immunity : Friends or Foes?

Nathalie Chaput, PharmD, PhD
Centre of Clinical Investigation Biotherapy 507
Institut Gustave Roussy, France
Immunostimulatory effects of conventional anti-cancer therapies

- Dacarbazine Promotes Stromal Remodeling and Lymphocyte Infiltration in Cutaneous Melanoma Lesions
  - Alessandra Nardin¹, Wing-Cheong Wong², Charlene Tow¹, Thierry Jo Molina³,⁸, Frédérique Tissier⁴,⁵,⁶,⁸, Anne Audebourg⁴, Marylene Garcette⁵,⁶, Anne Caignard⁵,⁶, Marie-Francoise Avril⁵,⁶,⁷, Jean-Pierre Abastado¹,⁹ and Armelle Prévost-Blondel⁵,⁶,⁹
- Dacarbazine Treatment before Peptide Vaccination Enlarges T-Cell Repertoire Diversity of Melan-A –Specific, Tumor-Reactive CTL in Melanoma Patients
  - Belinda Palermo, Duilia Del Bello, Alessandra Sottini, et al.
  - Cancer Res 2010;70:7084-7092. Published OnlineFirst September 7, 2010.

Chemotherapy Induces Intratumoral Expression of Chemokines in Cutaneous Melanoma, Favoring T-cell Infiltration and Tumor Control

- Michelle Hong, Anne-Laure Puaux, Caleb Huang, et al.
- Cancer Res 2011;71:6997-7009. Published OnlineFirst September 26, 2011.
### Endogenous danger signals that can lead to activation of innate immunity

**Dying cell**

- **Endogenous danger signals**
  - **Damage associated molecular pattern**
  - HMGB1, HSP
  - DNA
  - RNA
  - ATP, uric acid
  - SAP130

**Innate immunity**

- **PRR**
  - TLR-2, -4
  - TLR-9
  - TLR-3
  - NLRP3
  - CLEC4A

From Chen GY, Nat Rev Immunol, 2010
Can conventional anticancer treatments lead to immunogenic cell death?

Oxaliplatin, anthracyclins, Radiotherapy

Living tumor cell

IMMUNOGENIC SIGNALS?
Molecular events leading to immunogenic cell death

Oxaliplatin, anthracyclins, Radiotherapy

Living tumor cell → Dying cell

Calreticulin exposure → HMGB1

TLR4

ATP

NLRP3

pro-IL-1β

IL-1β

IFNγ

LT CD8+

NK

P2RX7
Calreticulin exposure dictates the immunogenicity of cancer cell death

Toll-like receptor 4–dependent contribution of the immune system to anticancer chemotherapy and radiotherapy

Activation of the NLRP3 inflammasome in dendritic cells induces IL-1β–dependent adaptive immunity against tumors

Apetoh, Nat Med 2007
Temozolomide and sorafenib combination in advanced melanoma patients

Schema of IGR Phase II Trial: Dr Caroline ROBERT, 2006-2009

European Union Drug Regulating Authorities clinical trial EudraCT 2007-000527-18

A

IM

Day 1

TMZ

Sorafenib

Day 7

TMZ

Day 14

Day 21

Day 28

IM

B

IM

T0

Cycle 1

M1

Cycle 2

M2

Cycle 3

M3

IM: immunomonitoring
TMZ: temozolomide
T0: before therapy
M: month
### Table 1. Characteristics of Patients (n=45)

<table>
<thead>
<tr>
<th>Characteristic</th>
<th>Value</th>
</tr>
</thead>
<tbody>
<tr>
<td>Gender (male/female)</td>
<td>24/21</td>
</tr>
<tr>
<td>Age (mean ± SD)</td>
<td>48.6 ± 13.8 [22; 75]</td>
</tr>
<tr>
<td>Type</td>
<td></td>
</tr>
<tr>
<td>SSM (1)</td>
<td>12</td>
</tr>
<tr>
<td>Nodular</td>
<td>8</td>
</tr>
<tr>
<td>Lentigo maligna</td>
<td>1</td>
</tr>
<tr>
<td>Acral lentiginous</td>
<td>2</td>
</tr>
<tr>
<td>Mucous</td>
<td>6</td>
</tr>
<tr>
<td>Ophthalmologique</td>
<td>4</td>
</tr>
<tr>
<td>Other</td>
<td>12</td>
</tr>
<tr>
<td>Metastasis (n=43)</td>
<td></td>
</tr>
<tr>
<td>Number of metastases lesions per patient (mean ± SD)</td>
<td>5.1 ± 2.0 [1-10]</td>
</tr>
<tr>
<td>Metastatic sites</td>
<td></td>
</tr>
<tr>
<td>Nodes</td>
<td>40</td>
</tr>
<tr>
<td>Liver</td>
<td>11</td>
</tr>
<tr>
<td>Lung</td>
<td>22</td>
</tr>
<tr>
<td>Peritoneum</td>
<td>8</td>
</tr>
<tr>
<td>Skin</td>
<td>19</td>
</tr>
<tr>
<td>Bone</td>
<td>7</td>
</tr>
<tr>
<td>Muscle</td>
<td>4</td>
</tr>
<tr>
<td>LDH level U/l (mean)</td>
<td>288.3 U/l</td>
</tr>
<tr>
<td>LDH &lt; 250 U/l</td>
<td>25</td>
</tr>
<tr>
<td>LDH &gt; 250 U/l</td>
<td>15</td>
</tr>
<tr>
<td>ND</td>
<td>5</td>
</tr>
<tr>
<td>Treatment schedule</td>
<td></td>
</tr>
<tr>
<td>sorafenib 400 mg/l, temozolomide 100 mg/m²</td>
<td>3</td>
</tr>
<tr>
<td>sorafenib 400 mg/l, temozolomide 150 mg/m²</td>
<td>5</td>
</tr>
<tr>
<td>sorafenib 800 mg/l, temozolomide 150 mg/m²</td>
<td>37</td>
</tr>
<tr>
<td>Previous chemotherapy</td>
<td></td>
</tr>
<tr>
<td>No</td>
<td>7</td>
</tr>
<tr>
<td>One line</td>
<td>15</td>
</tr>
<tr>
<td>Two lines</td>
<td>15</td>
</tr>
<tr>
<td>Three lines</td>
<td>7</td>
</tr>
<tr>
<td>Four lines</td>
<td>1</td>
</tr>
<tr>
<td>3-month evaluation</td>
<td></td>
</tr>
<tr>
<td>Objective response or stabilization</td>
<td>18</td>
</tr>
<tr>
<td>Progressive disease or death</td>
<td>27</td>
</tr>
</tbody>
</table>

(1) SSM: Spreading Superficial Melanoma; ND: Not determined
Accumulation of a CD4⁺ NKG2D⁺T cell subset in MM patients…

These cells are correlated with OS after two cycle of treatment

Only patients with OS > median survival had an augmentation in the proportion of CD4⁺NKG2D⁺ T after treatment.
CD4⁺ NKG2D⁺ T cells produce Th1 cytokines after stimulation through TCR or CD122 in synergy with NKG2D triggering.

CD4⁺ NKG2D⁺ T cells constituted a Th1 polarized T-cell subset with a potential to react in a TCR independent fashion when stimulated by IL-15 along with sMIC.
Sorafenib-induced IL-15Ra expression in the tumor
Sorafenib–induced shedding of MICA/B leading to accumulation of sMIC in these MM

sMIC being associated with clinical activity
Putative scenario during MM treatment by Sorafenib & Temozolomide

**Tumor cell**

- **Sorafenib**
  - c-KIT
  - VEGFR
  - PDGFR
  - FLT-3
  - Raf-MEK-ERK

**Augmentation**

- ADAM9/ADAM17
- IL-15Ra/IL-15

**Temozolomide**

- Induces lymphopenia
- **Lymph**
  - NKG2D
  - CD122
  - **sMICA/B**

**Augmentation**

- ADAM9/ADAM17
- IL-15Ra

**Sorafenib**

- c-KIT
- VEGFR
- PDGFR
- FLT-3
- Raf-MEK-ERK

**Temozolomide**

- Induces lymphopenia
- **Lymph**
  - NKG2D
  - CD122
  - **sMICA/B**

**Augmentation**

- ADAM9/ADAM17
- IL-15Ra

**IL-15**

- **Lymph**
  - NKG2D
  - CD122

**IL-7**

- **Lymph**
  - NKG2D
  - CD122

**NKG2D**

- **Lymph**
  - CD122
Anti-cancer treatments: A new point of view

A better comprehension of these mechanisms should help to determine which treatment should be combine with immunoregulators and to select groups of patients that could benefit from this chemo/immuno-approaches.
Sophie Caillat Zücman  
*St Vincent de Paul*

Antoine Toubert  
*Saint Louis*
Immunohistochemistry stainings of melanoma: No significant modulation of T cell infiltrates nor NKG2DL expression with the combo therapy.

<table>
<thead>
<tr>
<th>Clinical response</th>
<th>CD3</th>
<th>CD4</th>
<th>CD8</th>
<th>MICA/B</th>
</tr>
</thead>
<tbody>
<tr>
<td>Nr. 1</td>
<td>++</td>
<td>+/-</td>
<td>++</td>
<td>++</td>
</tr>
<tr>
<td>Nr. 2</td>
<td>-</td>
<td>-</td>
<td>-</td>
<td>++</td>
</tr>
<tr>
<td>Nr. 3</td>
<td>++</td>
<td>+</td>
<td>+</td>
<td>+</td>
</tr>
<tr>
<td>Nr. 4</td>
<td>+</td>
<td>-</td>
<td>+</td>
<td>+</td>
</tr>
<tr>
<td>Nr. 5</td>
<td>+</td>
<td>-</td>
<td>-</td>
<td>+</td>
</tr>
<tr>
<td>Nr. 6</td>
<td>+</td>
<td>-</td>
<td>-</td>
<td>++</td>
</tr>
<tr>
<td>Nr. 7</td>
<td>+</td>
<td>-</td>
<td>++</td>
<td>+</td>
</tr>
<tr>
<td>Nr. 8</td>
<td>+</td>
<td>-</td>
<td>++</td>
<td>++</td>
</tr>
</tbody>
</table>

Before therapy

After therapy
Can drugs in melanoma treatment lead to immunity?

Dacarbazine Promotes Stromal Remodeling and Lymphocyte Infiltration in Cutaneous Melanoma Lesions

Alessandra Nardin\textsuperscript{1}, Wing-Cheong Wong\textsuperscript{2}, Charlene Tow\textsuperscript{1}, Thierry Jo Molina\textsuperscript{3,8}, Frédérique Tissier\textsuperscript{4,5,6,8}, Anne Audebourg\textsuperscript{4}, Marylene Garcette\textsuperscript{5,6}, Anne Caignard\textsuperscript{5,6}, Marie-Francoise Avril\textsuperscript{5,6,7}, Jean-Pierre Abastado\textsuperscript{1,9} and Armelle Prévost-Blondel\textsuperscript{5,6,9}

Dacarbazine Treatment before Peptide Vaccination Enlarges T-Cell Repertoire Diversity of Melan-A –Specific, Tumor-Reactive CTL in Melanoma Patients

Belinda Palermo, Duilia Del Bello, Alessandra Sottini, et al.


Chemotherapy Induces Intratumoral Expression of Chemokines in Cutaneous Melanoma, Favoring T-cell Infiltration and Tumor Control

Michelle Hong, Anne-Laure Puaux, Caleb Huang, et al.