Basic Immunotherapy for current and future cancer treatment strategies

John Haanen, MD PhD

ELCC, Geneva April 2016
My disclosures

• Research grant funding:
  – BMS, GSK, MSD, NEON Therapeutics

• Advisory role:
  – BMS, Novartis, MSD, Pfizer, Roche, NEON Therapeutics
Revolution in cancer treatment
Immunotherapy in scientific news

2013

Science

Breakthrough of the Year
Cancer Immunotherapy
T cells on the attack

2014

nature

Immune-checkpoint blockade in cancer

2015

Science

Special issue
Cancer Immunology and Immunotherapy

Netherlands Cancer Institute
Antoni van Leeuwenhoek
Jimmy Carter says his brain cancer is gone — here’s how his age may have helped him

Carter said in a statement that a recent brain MRI indicated that the four melanoma lesions on his brain were gone and no new ones had formed. The former president said that he will continue to receive doses of Keytruda, a recently approved immunotherapy drug.
Immunotherapy in the public news

What if your immune system could be taught to kill cancer?

Inside the brutally selective, hugely expensive, lifesaving trials of immunotherapy.

By Alice Park

April 2016
Immunotherapy of cancer

William Bradley Coley (1862-1936)

Surgeon, specialized in bone cancer

Worked at the Memorial Hospital, NYC (MSKCC)
Coley Fluid: bacterial toxins

<table>
<thead>
<tr>
<th>Coley Fluid Therapy – advanced breast cancer</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>Years survival:</strong></td>
</tr>
<tr>
<td>----------------------</td>
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<tr>
<td><strong>Effective formulations</strong></td>
</tr>
<tr>
<td><strong>Ineffective formulations</strong></td>
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</tbody>
</table>

*Patients treated for less than 3 months*

<table>
<thead>
<tr>
<th>Effective formulations</th>
<th>0</th>
<th>0</th>
<th>16</th>
<th>6</th>
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</thead>
<tbody>
<tr>
<td>Ineffective formulations</td>
<td>0</td>
<td>0</td>
<td>10</td>
<td>1</td>
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</table>

*Patients treated for 3 months or longer*

<table>
<thead>
<tr>
<th>Effective formulations</th>
<th>4</th>
<th>5</th>
<th>3</th>
<th>3</th>
</tr>
</thead>
<tbody>
<tr>
<td>Ineffective formulations</td>
<td>0</td>
<td>1</td>
<td>10</td>
<td>1</td>
</tr>
</tbody>
</table>
FDA approval of immunotherapies

Coley Toxin: 1896
IL-2: 1992
sipuleucel-T: 1995
IFN-a: 2010
ipilimumab: 2011
nivolumab: 2014
pembrolizumab: 2015
T-Vec: 2015

*: no FDA approval for Coley Toxin

NETHERLANDS CANCER INSTITUTE
ANTONI VAN LEEUWENHOEK
Tumor Infiltrating Lymphocytes (TIL) (HNSCC)

Diffuse infiltration with CD8+ TILs in HNSCC

Absence of TILs in HNSCC

Keck et al., Clin Canc Res 2014
Role for T cells in cancer

Intratumoral T Cells, Recurrence, and Survival in Epithelial Ovarian Cancer

Lin Zhang, M.D., Jose R. Conejo-Garcia, M.D., Ph.D., Dionysios Katsaros, M.D., Ph.D., Phyllis A. Gimotty, Ph.D., Marco Massobrio, M.D., Giorgia Regnani, M.D., Antonis Makrigiannakis, M.D., Ph.D., Heidi Gray, M.D., Katia Schlienger, M.D., Ph.D., Michael N. Liebman, Ph.D., Stephen C. Rubin, M.D., and George Coukos, M.D., Ph.D.

Objective Measurement and Clinical Significance of TILs in Non–Small Cell Lung Cancer

Kurt A. Schalper, Jason Brown, Daniel Carvajal-Hausdorf, Joseph McLaughlin, Vamsidhar Velcheti, Konstantinos N. Syrigos, Roy S. Herbst, David L. Rimm

Type, Density, and Location of Immune Cells Within Human Colorectal Tumors Predict Clinical Outcome


Immunotype and Immunohistologic Characteristics of Tumor-Infiltrating Immune Cells Are Associated with Clinical Outcome in Metastatic Melanoma

TIL and cancer outcome

Fridman et al., NRC 2012
Treatment with tumor-infiltrating lymphocytes
Patient: continuing complete remission > 4 year

Haanen et al., unpublished)
The Cancer-Immunity Cycle

1. Release of cancer cell antigens (cancer cell death)
2. Cancer antigen presentation (dendritic cells/ APCs)
3. Priming and activation (APCs & T cells)
4. Trafficking of T cells to tumors (CTLs)
5. Infiltration of T cells into tumors (CTLs, endothelial cells)
6. Recognition of cancer cells by T cells (CTLs, cancer cells)
7. Killing of cancer cells (immune and cancer cells)
The Cancer-Immunity Cycle

Chen & Mellman, Immunity 2012
T cell activation requires at least 2 signals

Tumor releases antigen

Antigen-presenting cell (APC)

Interaction of the CD28 receptor on the T-cell with ligands on the APC leads to activation of T-cell attack

Release of apoptosis-inducing proteins

Active T-cells

APCs activate T-cells that proliferate and migrate to the tumor to attack cancer cells

www.immunooncologyhcp.bmsinformation.com
CTLA4 ligation dampens an induced T cell response
Loss of CTLA-4 Leads to Massive Lymphoproliferation and Fatal Multiorgan Tissue Destruction, Revealing a Critical Negative Regulatory Role of CTLA-4

Elizabeth A. Tivol, * Frank Borriello,*
A. Nicola Schweitzer*, William P. Lynch,*
Jeffrey A. Bluestone,† and Arlene H. Sharpe*
CTLA4 blockade renders T cells in an active state
Anti-CTLA4: ipilimumab and tremelimumumab

• Activity of ipilimumab observed in:
  – Advanced stage melanoma (approved)\textsuperscript{1,2}
  – Advanced stage ccRCC\textsuperscript{3}
  – Advanced (N)SCLC in combination with chemotherapy\textsuperscript{4,5}

• Activity of tremelimumumab observed in:
  – Advanced stage melanoma\textsuperscript{6} (phase 3 trial failed to show OS benefit)\textsuperscript{7}
  – Mesothelioma\textsuperscript{8}

\textsuperscript{1}Hodi et al. NEJM 2010; \textsuperscript{2}Robert et al., NEJM 2011; \textsuperscript{3}Yang et al., J Immunother 2007; \textsuperscript{4}Lynch et al. J Clin Oncol 2012; \textsuperscript{5}Reck et al., Ann Oncol 2013; \textsuperscript{6}Comacho et al. J Clin Oncol 2009; \textsuperscript{7}Ribas et al., J Clin Oncol 2013; \textsuperscript{8}Guazzelli et al., Exp Opin Biol Ther 2015
**Ipilimumab**

### Pre-treated pts +/- gp100
- **HLA-A2**
- **3mg/kg**
- Re-induction possible

### naive pts + DTIC
- **10 mg/kg**
- Maintenance possible

<table>
<thead>
<tr>
<th></th>
<th>1 Year</th>
<th>2 Year</th>
</tr>
</thead>
<tbody>
<tr>
<td>Lpi + gp100 N=403</td>
<td>44%</td>
<td>22%</td>
</tr>
<tr>
<td>Lpi + pbo N=137</td>
<td>46%</td>
<td>24%</td>
</tr>
<tr>
<td>gp100 + pbo N=136</td>
<td>25%</td>
<td>14%</td>
</tr>
</tbody>
</table>

**Hodi et al 2010 NEJM**

<table>
<thead>
<tr>
<th></th>
<th>1 Year</th>
<th>2 Year</th>
<th>3 Year</th>
</tr>
</thead>
<tbody>
<tr>
<td>Ipilimumab+ DTIC N=250</td>
<td>47.3</td>
<td>28.5</td>
<td>20.8</td>
</tr>
<tr>
<td>Placebo+ DTIC N=252</td>
<td>36.3</td>
<td>17.9</td>
<td>12.2</td>
</tr>
</tbody>
</table>

**Robert et al NEJM 2011**
Pooled OS Analysis of ipilimumab treated 4846 patients (incl EAP)

Mediane OS (95% CI): 9.5 (9.0–10.0)

3-year OS rate (95% CI): 21% (20–22%)

Schadendorf et al., J Clin Oncol 2015
Immune related adverse events upon anti-CTLA-4 mAb treatment

colitis

hypophysitis

thyroiditis
hepatitis
meningitis
etc.

vitiligo

dermatitis
Immune checkpoints
PD1/PDL1

One way that tumors can evade normal immune attack is through exploitation of the PD-1 checkpoint pathway via the PD-1 receptor, a key regulator of T-cell activity, by converting active T cells to inactive T cells.\textsuperscript{5-8}

Both PD-L1 and PD-L2 ligands on the tumor cells bind to the PD-1 receptor on T cells to exploit the immune checkpoint pathway. This inhibits activated T cells and suppresses T-cell attack.\textsuperscript{5-9}
PD1/PDL1 blockade reinvigorates inactivated T cells at the tumor site

Ribas. NEJM 2012
Mechanisms of tumor PDL1 expressions

Topalian et al. Cancer Cell 2015
Activity of anti-PD1/PDL1 over many tumor types

Melanoma¹ (N=411) KEYNOTE-001
NSCLC² (N=262) KEYNOTE-001
H&N³ (N=61) KEYNOTE-012
Urothelial⁴ (N=33) KEYNOTE-012
Gastric⁵ (N=39) KEYNOTE-012
TNBC⁶ (N=32) KEYNOTE-012
cHL⁷ (N=29) KEYNOTE-013
Mesothelioma⁸ (N=25) KEYNOTE-028

¹ Daud A et al. 2014 SMR; ² Garon EB et al. ESMO 2014; ³ Chow LQ et al. ESMO 2014; ⁴ O'Donnell P et al. 2015 Genitourinary Cancers Symposium; ⁵ Muro K et al. 2015 Gastrointestinal Cancers Symposium; ⁶ Nanda R et al. SABCS 2014; ⁷ Moskowitz C et al. 2014 ASH Annual Meeting; ⁸ Alley EA et al. 2015 AACR.

Courtesy of J Eid
Nivolumab: Overall survival in NSCLC
(>1st line therapy with cisplatin doublet)

Overall Survival in Trial 2 (Intent-to-Treat Analysis)

<table>
<thead>
<tr>
<th></th>
<th>Nivolumab (n=135)</th>
<th>Docetaxel (n=137)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Events (%)</td>
<td>86 (64%)</td>
<td>113 (82%)</td>
</tr>
<tr>
<td>Median survival in months (95% CI)</td>
<td>9.2 (7.3, 13.3)</td>
<td>6.0 (5.1, 7.3)</td>
</tr>
<tr>
<td>p-value</td>
<td>0.00025</td>
<td></td>
</tr>
<tr>
<td>Hazard ratio (95% CI)</td>
<td>0.59 (0.44, 0.79)</td>
<td></td>
</tr>
</tbody>
</table>

http://packageinserts.bms.com/pi/pi_opdivo.pdf
Updated OS results from CheckMate 066 trial in BRAF wt advanced melanoma

Atkinson et al. abstract 3774 SMR 2015
Potential biomarkers of response to immunotherapy

- CD8 T cell infiltrates
- Expression of PDL1 on:
  - Tumor cells
  - Immune cells
- Tumor mutational burden
Clinical response to anti-PD1 accompanied by increase in CD8 T cell influx

Hamid et al., NEJM 2013
CD8 T cells in invasive tumor margins correlates with response to anti-PD1

Tumeh et al. Nature 2014
Expression of PD-L1 co-localizes with TILs

Taube et al. Science Transl Med 2012
Loss of PTEN promotes resistance to T cell mediated immunotherapy

Peng et al., Cancer Disc 2015
LETTER

Melanoma-intrinsic β-catenin signalling prevents anti-tumour immunity

Stefani Spranger¹, Riyue Bao² & Thomas F. Gajewski¹,³
Potential biomarkers of response to immunotherapy

- CD8 T cell infiltrates
- **Expression of PDL1 on:**
  - Tumor cells
  - Immune cells
- Tumor mutational burden
OS in Checkmate-066 according to PDL1 expression

- **NIVO ≥5% (N = 59)**
  - Median OS, mo (95% CI): NR (NR, NR)
  - HR (95% CI): 0.56 (0.32, 0.98); P = 0.0399

- **NIVO <5% (N = 127)**
  - Median OS, mo (95% CI): NR (16.6, NR)
  - HR (95% CI): 1.16 (0.79, 1.68); P = 0.451

- **DTIC ≥5% (N = 61)**
  - Median OS, mo (95% CI): 9.7 (6.7, 13.5)

- **DTIC <5% (N = 116)**
  - Median OS, mo (95% CI): 11.6 (9.3, 13.0)

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**Note:** Hazard ratios expressed as PD-L1 ≥5% over PD-L1 <5%

CI = confidence interval; HR = hazard ratio; mo = month; NR = not reached

Atkinson et al. abstract 3774 SMR 2015
(MSD) PD-L1 NSCLC Sample IHC staining

PD-L1 = 0% positive
Negative

PD-L1 = 2% positive
Weak Positive
(1%-49%)

PD-L1 = 100% positive
Strong Positive
(50%-100%)
OS of pembrolizumab vs docetaxel in pretreated PDL1+ NSCLC (KN-010)

Herbst et al., Lancet 2015

≥ 50% PDL1 expression
# PD-L1 and response in Mel and NSCLCs

## Melanoma

<table>
<thead>
<tr>
<th>ORR – “PDL1+”</th>
<th>Pembrolizumab</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>1% cut-off: 49%</td>
</tr>
<tr>
<td></td>
<td>10% cut-off: 52%</td>
</tr>
<tr>
<td>ORR – PDL1-</td>
<td>1% cut-off: 13%</td>
</tr>
<tr>
<td></td>
<td>10% cut-off: 23%</td>
</tr>
</tbody>
</table>

## NSCLCs

<table>
<thead>
<tr>
<th>ORR – “PDL1+”</th>
<th>Pembrolizumab</th>
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<tbody>
<tr>
<td></td>
<td>1% cut-off: 25%</td>
</tr>
<tr>
<td></td>
<td>50% cut-off: 37%</td>
</tr>
<tr>
<td>ORR – PDL1-</td>
<td>1% cut-off: 7%</td>
</tr>
<tr>
<td></td>
<td>50% cut-off: 11%</td>
</tr>
</tbody>
</table>

Daud, AACR 2014
Garon, WCLC 2013, #2416
Ghandi, AACR 2014
Limitations in use of PDL1 expression as biomarker

- PD-L1 expression is dynamic
- PD-L1 is heterogeneous within tissue
- Unclear what level of expression is important
- On which cell type: tumor or immune cells?
- Importance of co-localization with TILs
- Scalability and reliability of assays
- Archived material often used
- Variability collection time of tissue
- Variability in mAb used for IHC
Potential biomarkers of response to immunotherapy

• CD8 T cell infiltrates
• Expression of PDL1 on:
  – Tumor cells
  – Immune cells
• Tumor mutational burden
Does the extent of DNA damage correlate with the clinical effects of cancer immunotherapy?

Alexandrov et al, Nature 2013
Mutational load and outcome to ipilimumab
Tobacco exposure and PD-1 response in NSCLC

<table>
<thead>
<tr>
<th></th>
<th>Never smokers</th>
<th>Smokers or Ex-smokers</th>
<th>Reference</th>
</tr>
</thead>
<tbody>
<tr>
<td>Pembrolizumab</td>
<td>5/60 (8%)</td>
<td>33/129 (26%)</td>
<td>Garon et al, ASCO 2014</td>
</tr>
<tr>
<td>MPDL3280A</td>
<td>1/10 (10%)</td>
<td>11/43 (26%)</td>
<td>Soria et al, WCLC 2013</td>
</tr>
<tr>
<td>Nivolumab</td>
<td>0/13 (0%)</td>
<td>20/75 (25%)</td>
<td>Hellmann et al, ESMO 2014</td>
</tr>
</tbody>
</table>

Courtesy of Dr Rizvi
Mutational load correlates with clinical outcome to anti-PD-1 in NSCLC

Rizvi et al, Science 2015
Mutational load correlates with clinical outcome to anti-PD-1 in NSCLC

Rizvi et al, Science 2015
Mutational load correlates with clinical outcome to anti-PD-1 in NSCLC

Mutational load

Progression free survival

- High
- Low

p = 0.0004

n = 34

Rizvi et al, Science 2015
Response to anti-PD1 of mismatch repair deficient tumors

Le et al., NEJM 2015
Analyzing the neo-antigen-specific T cell repertoire in human cancer

Generate map of tumor-specific mutations (ExomeSeq)

Determine which mutated genes are expressed (RNASeq)

Predict epitopes for each mutation/ each HLA-allele \textit{in silico}

Screen for T cell recognition of mutated epitopes
Pt 002: Partial response upon anti-CTLA4 treatment

**pre-treatment**

August 2010

- 80.39 mm
- 40.11 mm

**post-treatment**

December 2010

- 64.30 mm
- 27.75 mm

![Bar chart showing S100 (mg/L) days after start of therapy](chart.png)
Pt 002: Partial response upon anti-CTLA4 treatment

Resected tumor material

Isolate tumor cells

Identify tumor-specific mutations

Predict potential epitopes

Isolate tumor-infiltrating T cells

Screen with MHC multimer technology

[Diagram showing processes involving resected tumor material, isolation of tumor cells and tumor-infiltrating T cells, identification of tumor-specific mutations, and prediction of potential epitopes.]
Strong T cell response against an ATR_{S>L} neo-epitope within the tumor

- Resected tumor material
- Isolate tumor cells
  - Isolate tumor-infiltrating T cells
  - Screen with MHC multimer technology
  - Identify tumor-specific mutations
  - Predict potential epitopes

Diagram:
- pMHC QD 625 (ATR_{S>L})
- pMHC PE-Cy7 (ATR_{S>L})

3.3%
Increased magnitude of neo-antigen-specific T cell response under anti-CTLA4

% CD8+ MHC-multimer+ cells of total CD8+ cells

Days after start of therapy

van Rooij, van Buuren JCO 2013
Induction of neo-antigen specific T cell reactivity in a patient with NSCLC upon PD-1 blockade
Induction of neo-antigen specific T cell reactivity in a patient with NSCLC upon PD-1 blockade

Rizvi et al, Science 2015
Likelihood of response to immunotherapy is correlated with mutational burden.

Schumacher & Schreiber. Science 2015
PD-L1 negative status is associated with lower mutation burden, differential expression of immune-related genes, and worse survival in stage-III melanoma

Madore et al., CCR 2016
How to integrate this information for future immunotherapy strategies?
The Cancer – Immunogram

Describing the state of Cancer - Immune interaction
high tumor foreignness
*mutational load*

- **sensitivity to immune effector mechanisms**
  - pMHC expression
  - IFNγ sensitivity

- **absence of inhibitory tumor metabolism**
  - Low LDH, glucose utilization

- **absence of local inhibitory factors:**
  - soluble mediators
  - Low IL6 -> CRP/ESR

- **high immune infiltration capacity**
  - CD8 intratumoral

- **general immune status**
  - Normal lymphocyte count
  - Right microbiome

- **absence of local inhibitory factors:**
  - Checkpoints
    - PD-L1
high tumor foreignness
mutational load

sensitivity to immune effector mechanisms
pMHC expression
IFNγ sensitivity

absence of inhibitory tumor metabolism
Low LDH, glucose utilization

absence of local inhibitory factors:
soluble mediators
Low IL6→CRP/ESR
Right microbiome

absence of local inhibitory factors:
Checkpoints
PD-L1

high immune infiltration capacity
CD8 intratumoral

general immune status
Normal lymphocyte count
Combination therapy guided by biomarkers

- Co-stimulatory mAbs targeting:
  - CD137
  - OX40
  - CD40
  - GITR

- Conventional agents inducing immunogenic cell death:
  - Chemotherapy
  - Radiotherapy
  - Anti-angiogenics
  - Targeted therapies

- Other checkpoint inhibitory molecules:
  - CTLA4
  - LAG3
  - TIM3
  - BTLA
  - TIGIT

- Cancer vaccines considering individual neoantigens

- Functional modification of immunosuppressive enzymes such as:
  - IDO1
  - iNOS

- $T_{reg}$ cell targeting or inhibition

- Adoptive cell therapy

- Myeloid cell modulation

PD1 or PD-L1 blockade

Melero, .., Haanen, Nat Rev Canc 2015