PD-1 Cancer Immunotherapy

Gordon Freeman, PhD

Dana-Farber Cancer Institute
Harvard Medical School
Disclosures

Intellectual Property related to the PD-1 / PD-1 Ligand pathway licensed non-exclusively to:

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Genentech/Roche
Merck
Merck-Serono
Boehringer-Ingelheim
Amplimmune/AstraZeneca
Novartis

Consultant: Novartis, BMS, Roche, Lilly
Immunology has offered hope for curing cancer for 100 years

What is different now?

New Strategy
Blockade of pathways used by tumors to inhibit anti-tumor immunity

Checkpoint blockade
T cells are white blood cells that can kill cancer cells: more is better

T cell clonal expansion

1000 T cells → 18 divisions (6 days) → millions of T cells
• There are positive and negative second signals
The PD-1 Pathway Inhibits T Cell Activation

Dephosphorylation

Reduced TCR signaling
Reduced cytokine production
Reduced target cell lysis
Altered lymphocyte motility
Metabolic programming

PD-1
PD-1 ligand
PD-L1 (B7-H1)
PD-L2 (B7-DC)

ITSM
SHP-2
Proximal signaling kinases

ITIM

CD3
TCR
CD8

MHC

CTLA4

B7-1

APC
Costimulation regulates T cell response to antigen dose

- **CD28**: Positive costimulatory signal
- **PD-1**: Negative coinhibitory signal
cloned from a CD3-activated T cell hybridoma undergoing activation-induced cell death (Honjo lab)

- Does not directly activate caspases and cause cell death or apoptosis; not like CD95 (Fas)

- Indirect effect on cell death by reduced cytokines, survival factors (less Bcl-xL, more BIM)
Why have negative signals like PD-1?

1. Tune down the immune response after elimination of disease
2. Prevent too strong an immune response damaging tissues
3. Maintain immune tolerance
Identify the target: block PD-1/PD-L1

Engagement of the PD-1 Immunoinhibitory Receptor by a Novel B7 Family Member Leads to Negative Regulation of Lymphocyte Activation

By Gordon J. Freeman,* Andrew J. Long,‡ Yoshiko Iwai,§ Karen Bourque,‡ Tatyana Chernova,* Hiroyuki Nishimura,§ Lori J. Fitz,‡ Nelly Malenkovich,* Taku Okazaki,§ Michael C. Byrne,‡ Heidi F. Horton,‡ Lynette Fouser,‡ Laura Carter,‡ Vincent Ling,‡ Michael R. Bowman,‡ Beatriz M. Carreno,‡ Mary Collins,‡ Clive R. Wood,‡ and Tasuku Honjo§

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PD-1 or PD-L1 Blockade Stimulates anti-tumor T cell response

Increased cytokines

IFN-γ

Increased killing

antibody drug

CD8+ CTL

TCR

MHC

PD-1

PD-L1

Tumor cell
PD-L2 is a second ligand for PD-L1 and inhibits T cell activation

Discovery may shed light on cancer’s shield against the immune system

For years, a question has tantalized cancer researchers: why is the immune system, normally so adept at unmasking and eliminating foreign invaders and abnormal cells, not always spry enough to destroy tumor cells? A new study by Dana-Farber scientists suggests an answer.

In a paper published in the March issue of Nature Immunology, investigators led by Gordon Freeman, Ph.D., of Adult Oncology report that a structure

PD-L1 on Breast cancer cell lines

<table>
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<tr>
<th>Cell number</th>
<th>MDA-231</th>
<th>SKBR-3</th>
<th>MCF-7</th>
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<tr>
<td>PD-L1</td>
<td>Log fluorescence Intensity</td>
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PD-L1 in Cancer

• Expressed on cell surface of ~30% solid tumors and selected hematologic malignancies
• Inhibits anti-tumor immune responses

Kidney tumor

Hodgkin lymphoma

Brown = PD-L1 Rodig, Signoretti, McDermott, Shipp; BWH & DFCI
Where does checkpoint blockade function?

CTLA-4 in the lymph node

PD-1 in the tumor
CTLA-4 function
CTLA-4 mAb therapeutic effects

CTLA-4 blockade → CD28, B7 → Unopposed stimulation → Ongoing immune response

CTLA-4 mAb → Treg depletion
Why doesn’t directly stimulating the immune response cure cancer?
Once the tumor gets ahead and expresses PD-L1, Immuno-inhibition is dominant and maintained by a feedback loop.
PD-1+ T cells at a PD-L1 tumor interface in melanoma

PD-L1+ melanoma

PD-1+ T cells

George Murphy, Scott Rodig, Gordon Freeman, BWH & DFCI
Agents in Clinical Trials

• Anti-PD-1
  – Nivolumab (BMS)
  – Pembrolizumab (Merck)
  – Pidilizumab (Curetech)
  – MEDI-0680 (MedImmune-AZ)
  – PDR001 (Novartis)
  – REGN2810 (Regeneron)

• Anti-PD-L1
  – Atezolimumab (MPDL3280, GNE)
  – Durvalumab (MEDI-4736 MedImmune-AZ)
  – Avelumab (MSB0010718C EMD Serono)
  – MDX-1105 (BMS)

Multiple other agents in development
PD-1 pathway immunotherapy

• 20-50% response rate in clinical trials
  Topalian et al., NEJM 366:2443 (2012)
Phase I clinical trial of anti-PD-1 antibody Nivolumab: Kidney Cancer cohort (34 patients)

- Generally tolerable: fatigue, rash, pruritus, diarrhea
  - Each line follows growth or shrinkage of tumor in one patient
  - 29% objective responses

Drake ASCO 2013
PD-1 Cancer Immunotherapy is different from chemotherapy

- Well tolerated: This is not chemotherapy or a cell poison! some nausea, no hair loss, no blood count decline.

- Good safety profile

- Most serious adverse events are autoimmune-mediated, like pneumonitis, colitis. Less than 10% of patients

- Physicians will have to learn to manage a different spectrum of adverse events than those seen in chemotherapy

- This can be community hospital medicine: half-hour intravenous drug infusion.
Broad anti-tumor efficacy of anti-PD-L1/PD-1 inhibitors: Overall Response Rates

243 clinical studies with 55,099 patients

Modified from D. Chen, BioScience Forum, 2015
PD-1 is better than chemo in melanoma

Hazard ratio for death, 0.42 (99.79% CI, 0.25–0.73)
P<0.001

Patients Who Died

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<tr>
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<th>Nivolumab</th>
<th>Dacarbazine</th>
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<tr>
<td>No. / Total No.</td>
<td>50/210</td>
<td>96/208</td>
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Median Survival

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<th>Nivolumab</th>
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<tr>
<td>Mo (95% CI)</td>
<td>Not reached</td>
<td>10.8 (9.3–12.1)</td>
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PD-1 antibodies pembrolizumab and nivolumab are now FDA approved for advanced melanoma, lung cancer, and renal cancer.
Better Quality of Life

• Reck said responding patients with advanced Squamous Non-Small Cell Lung Cancer “remaining on treatment with nivolumab returned to population health-status norm, suggesting that prolonged survival occurs with a resumption of normal life”
Quality of life: Squamous NSCLC: EQ-5D Utility Index Mean Scores Over Time While on Treatment

![Graph showing mean EQ-5D Utility Index scores over time for Nivolumab and Docetaxel treatments.](image)

- **Mean Scores Over Time While on Treatment**
  - **Lung Cancer Norm (UK-based):** 0.67
  - **Population Norm**

**Higher scores indicate better health status.** Only time points that had PRO data available for ≥5 patients in either treatment arm are plotted on the graph.

- **Nivolumab** (n = 97)
  - Week 0: 97
  - Week 12: 50
  - Week 24: 32
  - Week 30: 32
  - Week 36: 21
  - Week 42: 18
  - Week 48: 13
  - Week 54: 13
  - Week 60: 8

- **Docetaxel** (n = 89)
  - Week 0: 88
  - Week 12: 32
  - Week 24: 9
  - Week 30: 5
  - Week 36: 4
  - Week 42: 4
  - Week 48: 2
  - Week 54: 1

**References:**
Checkpoint works equally well in the aged

Meta-analysis of 6 Phase III PD-1 and CTLA-4 trials
2,078 younger patients < 65-70 years
1,224 older patients >65-70 years

Younger: Hazard Ratio, 0.73;   P<0.001
Older:    Hazard Ratio, 0.72;   P=0.004

T Funakoshi et al., SITC 2015
90 year old with metastatic melanoma and 4 brain metastases:
Treated with PD-1 mab pembrolizumab
Predictive biomarkers are essential for getting the right treatment to the right patient.
PD-L1 expression in tumor increases the likelihood of response to PD-1/PD-L1 blockade

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<td>44%</td>
<td>49%</td>
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<td>20%</td>
<td>15%</td>
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**Treatment:**
- anti-PD-1 Antibody
- anti-PD-L1 Antibody

**Assay:**
- Membranous pattern on tumor cells
- Immune infiltrate NR

Nivolumab Pembrolizumab MPDL3280A MEDI4736
A new era in PD-L1 immunohistochemistry

Now at least 5 good PD-L1 IHC mAbs available

extracellular

5H1  Chen
22C3  Merck - Dako/Quest
28-8  BMS - Dako/Quest

intracellular

SP142  Roche - Spring
E1L3N  CST
9A11  Freeman - CST
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PD-1 may be a good first line lung cancer therapy

PD-L1 as a predictive marker in the Phase 3 study comparing nivolumab with everolimus in RCC

- Confirm previous studies that higher PD-L1 expression is associated with poorer survival in RCC

- PD-L1 expression was **not** associated with a PD-1 treatment benefit in RCC

RJ Motzer et al., NEJM 2015; 373:1803
21% Discordancy between PD-L1 on Primary and Metastasis in RCC

- PD-L1 positivity was heterogeneous and almost exclusively detected in high nuclear grade areas ($P < 0.001$).

- Assessment as a predictive biomarker for PD-1 blockade may require analysis of metastatic lesions.

- Pathologists should select high grade tumor areas for PD-L1 IHC analysis to avoid false negatives.

20 positive
33 negative in primary & met
53 cases

Renal Cell Carcinoma can express PD-L1 and/or PD-L2

Tumor 1

Tumor 2

Sabina Signoretti, Scott Rodig, BWH
What does the immune system see in a tumor to attack?
The immune system recognizes protein coding changes in the tumor cell, called tumor neoantigens.

Tumors have multiple neoantigens that T cells can attack.
Two evolutionary processes in cancer:

1. DNA mutation
   - Rare driver mutations
   - Many passenger mutations

2. Immune evasion: PD-L1, IDO, TGF-b, IL-10, loss of MHC, others
Why the enthusiasm for immunotherapy?

Data from Steve Hodi & ECCO

Moderate percentage but long-term

Chapman NEJM 2011

High percentage but short-term
Clinical benefit with PD-1 blockade

- 18% in TN Bca
- 25% ORR
- 18% ORR
- 24-26% ORR
- 29-40% ORR

Mutational load correlates with response to PD-1 mAb in lung cancer

Rizvi et al, Science 2015

Mutational load

Progression free survival

n = 34

Months post therapy

p = 0.0004
Normal cell

Natural mutations

Tumor cell heterogeneity

Genetically Engineered Models (GEM)

Normal cell

Introduced Oncogene

Tumor with few neoantigens
• Genetically Engineered Models (GEM) of cancer are not the best place to test immunotherapies

• GEMs don’t have the mutational burden to generate lots of neoantigens
transplantable vs GEM tumors

Glioblastoma transplanted into brain

EGFR driven GEM mouse model of lung cancer

David Reardon

Kwok Wong
We need to know

- What GEM are responsive to checkpoint blockade?
- Is this response like a natural anti-tumor immune response: neoantigens or more innate, NK?
- Can we improve GEM with inducible neoantigens or increased mutation rates?
• Classical transplantable models are a good place to test immunotherapies: great range of immunogenicity
Understanding immunology and genetics has identified groups that respond well to PD-1/PD-L1 therapy

- Highly mutated tumors (MSI, defects in DNA repair): 62%
- Genetically amplified PD-L1 and PD-L2 (Hodgkin): 87%
- With Viral antigens (HPV, Head and neck, Merkel)
- What other cancer types might respond well??
PD-L1 and PD-L2 expression can also be upregulated by gene amplification and translocation.

Diffuse large B-cell lymphoma (DLBCL) SU-DHL-4

Primary mediastinal (thymic) large B-cell lymphoma (PMBL) Karpas 1106P

Hodgkin lymphoma HDLM-2

Gene copy number ↑

PD-L2 IHC

Min Shi, Margaretha Roemer, Bjoern Chapuy, Xiaoyun Liao, Heather Sun, Geraldine Pinkus, Margaret Shipp, Gordon Freeman, Scott Rodig
Some virally-induced tumors are highly PD-L1 positive

EBV mediated nasopharyngeal carcinoma

Rodig, Freeman, BWH, DFHCC
Why did the T cells need PD-1 blockade to attack the tumor?

The anti-tumor immune response is a years long struggle.

The T cells had tried, failed, and become “exhausted”
PD-1 pathway mediates T cell exhaustion in Chronic Viral Infections

Dan Barber
John Wherry
Rafi Ahmed
2006
PD-1 is upregulated in both acute and chronic immune responses but stays high in chronic.

PD-1+ cells are “exhausted” and produce less cytokine.
Tumor-Infiltrating T cells (TIL) behave like exhausted T cells
Human Ovarian Tumor Infiltrating T cells (TIL) express high levels of PD-1

% PD-1^+
PD-1 blockade of ovarian TILs augments cytokine secretion by tumor antigen-specific (NY-ESO-1-peptide) CD8 TILs *in vitro*: TILs are exhausted T cells that respond to PD-1 blockade.
T cell exhaustion is more than PD-1
Exhausted Tumor infiltrating lymphocytes express multiple immunoinhibitory receptors:

These are druggable targets for tumor immunotherapy.
Co-expression of PD-1 and TIM-3 immune inhibitory receptors in tumor infiltrating CD8 T-cells in ccRCC

Signoretti S and Pignon JC
CTLA-4 and PD-1 came from the study of T cell tolerance

We need to identify organ-specific mechanisms of tolerance in the prostate, colon, liver
The Future is Combination Therapy

T cell priming & activation

**DEFICIT**
- Insufficient priming/activation naïve T cells

**THERAPEUTIC APPROACH**
- Block multiple checkpoints (CTLA-4, PD-1, LAG-3, TIM-3)
- Activate stimulatory pathways (CD137, OX-40, CD27, ICOS, GITR)
- Administer stimulatory cytokines (IL-2, IL-12)

Activated APC

B7 CD28

B7

CTLA-4

Lymph node

T cell
PD-1 + CTLA-4 is better than CTLA-4 alone

T cell trafficking & tumor infiltration

**DEFICIT**

- Insufficient trafficking into tumor

**THERAPEUTIC APPROACH**

- Anti-VEGF
- Local or intravenous delivery of adoptively transferred T cells
- Cytokines
Checkpoint blockade

Recognition & killing of cancer cells

DEFICIT

- Failure to overcome T cell suppression

- Unleashing intrinsic T cell immunity is insufficient

- Tumor burden/growth exceeds capacity for immune clearance

THERAPEUTIC APPROACH

- Block multiple checkpoints (PD-1, PD-L1, LAG-3, TIM-3, CTLA-4)
- Activate stimulatory pathways (CD137, OX40, CD27, ICOS, GITR)
- Deplete/target immunosuppressive cells (Treg, MDSCs, M2)
- Target other suppressive mechanisms in microenvironment (IDO, TGF-β)

- Induce/Provide other anti-tumor immune cells (CARs, TCR-engineered T cells, NK cells)

- Reduction of tumor burden (Surgery, Radiation, Chemotherapy, Targeted therapy)
Cancer antigen release, uptake & processing

**DEFICIT**
- Non-immunogenic cell death or insufficient neoantigens
- Insufficient antigen processing/DC maturation

**THERAPEUTIC APPROACH**
- Oncolytic viruses
- Chemotherapy
- Radiation therapy
- Cryotherapy
- Targeted therapy
- Epigenetic modifiers
- Blockade of phosphatidylycerine
- Vaccines
- TLR agonists/STING
- GM-CSF
- IFN-α
- CD40 agonists
The future of cancer therapy decisions

• Tumor Immunoevasion Score:
  How much PD-L1, PD-L2, IDO, Galectin-1, Galectin-9, B7-H3, B7-H4, VISTA, HHLA2, Arginase, NKG2D-Ligands?

  Choose best immunotherapy

• Cancer Genome sequencing:
  Identify which oncogenes are drug targets?
  Which mutations are immunogenic?

  Choose best targeted therapy/vaccine
To be done

• How do we identify who will respond to PD-1 blockade?

• What are mechanisms of primary failure to respond?
  – Other immunoinhibitors?
  – Failure of immune cells to infiltrate tumor?
  – No good neoantigens?

• What are mechanisms of secondary failure to respond?
  – Expression of other immunoinhibitory receptors?
  – Loss of MHC?
It’s a great time to be an oncologist or researcher

- PD-1/PD-L1 works on a wide range of tumors with
  - moderate percentage of responders
  - good safety profile

- PD-1/PD-L1 gives us a foundation to build on

- With this success, human creativity has been unleashed and we’re learning to do better
Dana-Farber Cancer Institute

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• Tasuku Honjo