PY Dietrich

Research grant : Immatics
Immunotherapy for brain tumors: From illusion to realistic prospects?

Pierre-Yves Dietrich

Director of the Center of Oncology
Geneva University Hospital
Switzerland
Brain tumors: a frequently lethal disease, with constant disability
Brain tumors

- **1\textsuperscript{st} cause of cancer related death**
  - In children

- **3\textsuperscript{rd} cause of cancer related death**
  - In young adults

- Most frequent and most devastating brain tumors are
  - **malignant glioma**
Invasion: a major cause of treatment failure
Malignant glioma: an incurable disease?

The need for novel strategies

Stupp et al, Lancet Oncol. 2009
Using the immune system?

- Incidence of cancers is increased in
  - constitutive and acquired immunodeficiencies
- T cells may kill tumor cells
- T cells invade tumors
- intra-tumoral activated T cells is a
  - favorable prognostic factor for many cancers
- spontaneous regressions are mediated by T cells
Tumor immunity: how does it work?

Cancer cells

APCs/DCs

Draining lymph node

MHC +tumor peptide

TCR

CD80 CD86 CD28

T cell
Does this model apply for tumors in the brain?

Blood-brain barrier

glioma

Cancer cells

DCs

Draining lymph node

astrocytes

tight junctions

basement membrane

No DCs

No lymphatic vessels

T cell
Immune privilege in the brain is not absolute

- Immune responses may occur or be induced against brain tumors
- Human glioma are infiltrated by clonally expanded CD8 T cells

Effector T cell infiltrate positively correlates with survival of GBM patients

How and where T cells are activated by glioma?

1. TCR-MHC/Peptide interaction + costimulation
2. Clonal expansion
3. Acquisition of effector function
T cell activation in cervical lymph nodes

Brain APCs
Microglial cells?

Cervical LN

Calzascia T, J Immunol 2003
How do T cells migrate into the brain?

Cervical LN

entry $\alpha_4\beta_1$

Calzascia T, J Immunol 2003
Calzascia T, Immunity, 2005
Masson F, J Immunol 2007
Calzascia T, Glia 2008
Does this model apply for tumors in the brain: probably YES

1. Therapeutic glioma vaccine

2. Cell therapy with engineered T cells

3. Reshaping the tumor microenvironment
Can we promote T cell activation?

1. Therapeutic glioma vaccine
Therapeutic glioma vaccines

1. synthetic peptides
   - single peptide (EFGRvIII)
   - multiple peptides (EphA2, IL-13Rα2, YKL-40, gp100, ...)

2. DC + peptides
   - multiple peptides (EphA2, IL-13Rα2, YKL-40, gp100, ...)

3. DC + tumor
   - tumor lysate
   - tumor mRNA
Tumor cell based vaccines

- Mostly DCs pulsed with
  - glioma lysates, glioma RNA or eluted peptides

- small studies, one large phase I/II study (n = 95)
  - De Vleeschouwer Cancer Immunol Immunother 2012; 61 2105 (n = 95)

- Main advantage:
  - a large repertoire of tumor antigens including private peptides
Tumor cell based vaccines

- **Potential difficulties**
  - Labor intensive
  - Manufacturing, reproducibility
  - Complex immunomonitoring
    - How to optimize vaccine schedule, dose, and composition

- **Risk of autoimmunity**
  - Contamination with normal cells
  - Shared epitopes (self peptides on tumor cells)
Peptide vaccination:
EGFRvIII as a 1st candidate

801 bp deletion
⇒ New antigenic junction
⇒ Specific for tumor cells
⇒ Ongoing phase III

Phase III ongoing
Selective loss of EGFRvIII after vaccine: Immunoediting

→ Need for several antigens

Sampson JH, J Clin Oncol 2010
### Glioma-associated antigens

*(Okada H et al., Crit Rev Immunol 2009)*

<table>
<thead>
<tr>
<th>Antigen</th>
<th>Expression</th>
<th>Expression</th>
<th>Roles in</th>
<th>HLA</th>
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<tr>
<td>IL-1</td>
<td>safe</td>
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<td>SAR</td>
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</tbody>
</table>

- safe
- Immunogenic *(IF Pollack, JCO in press)*
- Some encouraging clinical observations
  - BUT
- Which expression *in vivo*?
- Expression analyzed on cell lines
- Which expression by normal cells?

*clinical trials: peptides or DCs + peptides*
The wish list for glioma antigens

- Antigen expressed by glioma in vivo
- Presented as MHC-peptides complexes and recognized by CD8 T cells
- Wide expression in tumors and by many patients
- Little or no expression on normal tissue
Identification of glioma-associated antigens

32 HLA-A2⁺ patients

immatics biotechnologies GmbH
Tubingen, Germany

GLFVTARIL
ELVRQATIG
AAGIGILTV
GIFGTLAVL
RTAGIWYVL
TAARIGLVI
ALFVTARIL

Peptide Source protein
BCA478-486 BCAN (Brevican)
CHI10-18 CHI3L2 (Chitinase 3-like 2)
CSP21-29 CSPG4 (Chondroitin sulfate proteoglycan 4)
FABP7 118-126 FABP7 (Fatty acid binding protein 7, brain)
IGF2BP3 552-560 IGF2BP3 (Insulin-like growth factor 2 mRNA binding protein 3)
NLGN4X 131-139 NLGN4X (Neuroligin 4, X-linked)
NRCAM 692-700 NRCAM (Neuronal cell adhesion molecule)
PTP 195-203 PTPRZ1 (Protein tyrosine phosphatase, receptor-type, Z polypeptide 1)
PTP 1347-1355
TNC 3-11 TNC (Tenascin C)

3686 HLA-A2-restricted peptides

selection process
- proteins over-expressed in glioma
- absent/low expression in healthy tissues
- function associated with tumorigenesis
- immunogenic
Expression of the MHC peptide complexes

- Healthy tissues
- GBM

Relative peptide presentation

[Graph showing relative peptide presentation across various tissues including brain, colon, kidney, liver, lung, pancreas, stomach, GBM, and specific GBM samples such as GB 1008, GB 1020, GB 6010, GB 6016, GB 6019, GB 6024.]
Tolerance in GBM patients?

- The tumor might have tolerized glioma-specific CD8 T cells

### Data

<table>
<thead>
<tr>
<th>Protein</th>
<th>Percentage</th>
</tr>
</thead>
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<tr>
<td>Ge540/BCAN_{478-468}</td>
<td>8%</td>
</tr>
<tr>
<td>Ge531/CHI3L2_{10-18}</td>
<td>16%</td>
</tr>
<tr>
<td>Ge595/NLGN4X_{131-139}</td>
<td>46%</td>
</tr>
</tbody>
</table>

### Bar Chart

- BCA_{478-486}
- CHI_{10-18}
- CSP_{21-29}
- FABP7_{118-126}
- IGF2BP3_{552-560}
- NLGN4X_{131-139}
- NRCAM_{692-700}
- PTP_{195-203}
- PTP_{1347-1355}
- TNC_{3-11}

### Diagram

- T cell clones: killing properties
- HLA-A2+ Ag

Brain 2012, Dutoit et al.
Phase I/II clinical trial with the IMA950 vaccine

<table>
<thead>
<tr>
<th>Peptide code</th>
<th>Amino acid sequence</th>
<th>Gene symbol</th>
<th>Description of source protein</th>
</tr>
</thead>
<tbody>
<tr>
<td>BCA-002</td>
<td>ALWAWPSEL</td>
<td>BCAN</td>
<td>Brevican – Brain-specific chondroitin sulfate proteoglycan; mainly expressed in childhood and gliomas</td>
</tr>
<tr>
<td>CSP-001</td>
<td>TMLARLASA</td>
<td>CSPG4</td>
<td>Chondroitin sulfate proteoglycan 4 – Membrane protein; involved in tumor cell migration and invasion</td>
</tr>
<tr>
<td>FABP-007-001</td>
<td>LTFGDIVAV</td>
<td>FABP7</td>
<td>Fatty acid binding protein 7, brain – Mainly expressed in developing brain and often in gliomas</td>
</tr>
<tr>
<td>IGF2BP3-001</td>
<td>KIQTILTVVQ</td>
<td>IGF2BP3</td>
<td>Insulin-like growth factor 2 mRNA binding protein 3 – Mainly expressed during embryonic development</td>
</tr>
<tr>
<td>NLGN4X-001</td>
<td>NLDTLMTYYV</td>
<td>NLGN4X</td>
<td>Neurexin 4, X-linked – Cell adhesion protein involved in maturation and function of neuronal synapses</td>
</tr>
<tr>
<td>NRCAM-001</td>
<td>GLWHHOTEV</td>
<td>NRCAM</td>
<td>Neuronal cell adhesion molecule – Involved in neuronal cell growth; over-expressed in gliomas</td>
</tr>
<tr>
<td>PTP-003</td>
<td>AIIDGVESV</td>
<td>PTPRZ1</td>
<td>Protein tyrosine phosphatase, receptor-type, Z polypeptide 1 – receptor for the growth factor pleiotrophin</td>
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<td>PTP-005</td>
<td>KVFAIPTV</td>
<td>PTPRZ1</td>
<td>Protein tyrosine phosphatase, receptor-type, Z polypeptide 1 – receptor for the growth factor pleiotrophin</td>
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<tr>
<td>TNC-001</td>
<td>AMTOLLAGV</td>
<td>TNC</td>
<td>Tenasin C – Extracellular matrix protein involved in tumor angiogenesis; over-expressed in gliomas</td>
</tr>
</tbody>
</table>

<table>
<thead>
<tr>
<th>Peptide code</th>
<th>Amino acid sequence</th>
<th>Gene symbol</th>
<th>Description of source protein</th>
</tr>
</thead>
<tbody>
<tr>
<td>MET-005</td>
<td>TFSYVDPVITSISPKYG</td>
<td>MET</td>
<td>Met proto-oncogene (hepatocyte growth factor receptor) – over-expressed by many tumor types</td>
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<tr>
<td>BIR-002</td>
<td>TLGFLKLDREAKN</td>
<td>BIRC5</td>
<td>Baculoviral IAP repeat-containing 5 (survivin) – Apoptosis inhibitor; over-expressed by many tumor types</td>
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<tr>
<td>HBV-001</td>
<td>FLPSDFPPSV</td>
<td>n.a.</td>
<td>Hepatitis B virus core antigen – Included as a marker peptide for immunomonitoring</td>
</tr>
</tbody>
</table>

Dietrich PY, Educational ASCO 2014
Personalized glioma vaccines

GAPVAC
Glioma actively personalized vaccine consortium

Step 1: overexpressed antigens

Step 2: mutated antigens

N = 20 pts (7 centers)
Combination with adjuvant TMZ chemotherapy

PI: W. Wick, Heidelberg
Co-PI: P.-Y. Dietrich, Geneva
T cell therapy

1. Therapeutic glioma vaccine

2. Cell therapy with engineered T cells
TCR and CAR T cell engineering

introduction of a high-avidity TCR

introduction of an antibody

TCR engineering

CAR engineering
Some opportunities with TCRs from antigen-specific T cell clones (BCAN, CHI3L2, CSPG4, FABP7, NLGN4X, PTPRZ1)
CAR T cells: exploiting properties of both antibodies and T cells

- killing
- homing
- memory

- strong affinity binding
- no MHC restriction
  - for all patients
  - $\rightarrow$ MHC is allowed

Tumour-specific T cell either naturally occurring or from a transgenic mouse

Tumour antigen

scFv

CD3 complex

CD8

CD3$\zeta$ or FcεR1y

Nat Rev Cancer 2013, 13. 525 MH Kershaw
### CAR T cell trials in 2013

#### Table 1 | Recent adoptive cell therapy trials using CAR engineered T cells.

<table>
<thead>
<tr>
<th>Target antigen</th>
<th>Disease</th>
<th>CAR signaling domain</th>
<th>Clinical trial.gov identifier</th>
<th>Clinical center</th>
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<tbody>
<tr>
<td>CD19</td>
<td>B-CLL</td>
<td>CD28-CD3ζ</td>
<td>NCT00466531</td>
<td>MSKCC</td>
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<td>CD19</td>
<td>B-ALL</td>
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**Ongoing trials for GBM:**
- **NCT01454596** - EGFRvIII - NCI Bethesda
- **NCT01109095** - HER2 - Baylor College

**New opportunities**
- Recently identified antigens with cell surface expression

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MSKCC, Memorial Sloan-Kettering Cancer Center; NCI, National Cancer Institute; BCM, Baylor College of Medicine; RWMC, Roger Williams Medical Center; UP, University of Pennsylvania; MDACC, M.D. Anderson Cancer Center; FHCRC, Fred Hutchinson Cancer Research Center.

Chmielewski M, Front Immunol 2013
Reshaping the glioma microenvironment

3. Reshaping the tumor microenvironment

2. Cell therapy with engineered T cells

1. Therapeutic glioma vaccine
1. Cell-cell contact inhibition (FasL, PDL1, ...)
2. Soluble mediators (TGF-β, IL-10, PGE2, ...)
3. Cell mediators (Tregs, MDSCs, ...)
4. Rampart (IDO, ...)

Glioma is like a fortress
## TGF-β as a 1st target

Summary of TGF-β pathway targeting agents clinically evaluated in patients with high grade glioma.

<table>
<thead>
<tr>
<th>TGF-β targeting drug</th>
<th>Treatment</th>
<th>Phase</th>
<th>Patients</th>
<th>Remarks</th>
<th>Refs.</th>
</tr>
</thead>
<tbody>
<tr>
<td>Trabedersen (AP 12009) - TGF-β2-specific antisense oligo-deoxynucleotide</td>
<td>Intratumor administration</td>
<td>1/2</td>
<td>n = 24</td>
<td>Recurrent, refractory GBM, AA</td>
<td>Completed. Good tolerability and safety; indications for efficacy (7/24 responded, 2/24 complete responses)</td>
</tr>
<tr>
<td>Trabedersen - TGF-β2-specific antisense oligo-deoxynucleotide</td>
<td>10 or 80 μM trabedersen vs. standard chemotherapy</td>
<td>2</td>
<td>n = 141</td>
<td>Recurrent GBM, AA</td>
<td>Completed. In AA significant beneficial effects at 10 μM trabedersen</td>
</tr>
<tr>
<td>Trabedersen - TGF-β2-specific antisense oligo-deoxynucleotide</td>
<td>10 μM trabedersen vs. standard chemotherapy</td>
<td>3</td>
<td>n = 27</td>
<td>Recurrent, refractory GBM, AA</td>
<td>Terminated due to lack of patients</td>
</tr>
<tr>
<td>GC 1008 TGF-β neutralizing antibody</td>
<td>Intravenous administration 89Zr-GC1008</td>
<td>2</td>
<td>n = 32</td>
<td>Recurrent, refractory GBM</td>
<td>Ongoing; safety and imaging study</td>
</tr>
<tr>
<td>LY 2157299 - small molecule</td>
<td>Oral administration, alone or in combination with lomustine</td>
<td>2</td>
<td>n = 180</td>
<td>Recurrent GBM</td>
<td>Ongoing</td>
</tr>
<tr>
<td>TGF-βR kinase inhibitor</td>
<td>Combined with radio-chemotherapy with temozolomide</td>
<td>1/2</td>
<td>n = 62</td>
<td>Newly diagnosed GBM, AA</td>
<td>Ongoing</td>
</tr>
</tbody>
</table>
A Treg cell death

Chemotherapeutic agents (TMZ, CTX, etc.)

B Treg depletion or neutralization

Antibodies targeting Treg receptors (CD25, CTLA-4, GITR)

C Reduced Treg accumulation

Pharmacological agents targeting Treg trafficking (CCR4 antagonist)
What is the hierarchy in the mechanisms of immune evasion?

could we tilt the balance in favor of effective immune response by inhibiting one of them?

First results rather disappointing when used alone BUT

Strategy is more likely to provide some benefit in combination with vaccines and T cell therapy
Towards poly-immunotherapy

1. Therapeutic glioma vaccine

2. Cell therapy with engineered T cells

3. Reshaping the tumor microenvironment

Find synergistic approaches multimodality treatments

optimal homing phenotype
efficient migration to brain

Dietrich et al, Curr Op Oncol 2010
Adding immune checkpoint inhibitors

Cancer cells

DCs

PDL1 / PD1

Draining lymph node

CTLA4
From illusion to realistic prospects?

- 1994: NO
- 2004: unrealistic dream
- 2014: credible prospect for the future

Challenges:
- how to optimally use these new therapies along with existing treatment modalities
- and how to monitor both immunological and clinical outcome

➤ rigorous clinical trials that favor constant interactions between bench and bedside.
Verona Vass
Steffen Walter
Norbert Hilf
Oliver Schoor
Toni Weinschenk
Harpreet Singh

Jennifer Lohr
Judith Bucher
Katharina Dorsch
Christel Herold-Mende

Neurochirurgie
Karl Schaller and the Neurosurgery team

Service d’oncologie
Melissa Morawitz

Patients and families

The Gateway for Cancer Research