Developing a First in Man Carbohydrate Mimetic Peptide Vaccine for Cancer: A Translational Story

Building a vision

• Foundation for the story – rationale for therapy
  • Discovery Phase – panspecific response
• Phase I clinical trial in Stage 4 Metastatic BC
  • Does it work – what are the expectations?

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Different from Carbohydrate Vaccines

Each CMP is a Pan antigen
Generate poly reactive or Panspecific antibodies
Generate anti-Tumor T cells
Provides for multiple platforms
Generates long term memory

Carbohydrate mimetic peptides as TACA surrogates

To circumvent lack of cooperation we have developed carbohydrate mimetic peptides (CMPs) with overlapping B and T cell epitopes to link TACA reactive humoral responses with anti-tumor cellular responses.

R-F Wang 2001 Trends in Immunology
TACA expressed on multiple glycolipids and proteins - impact on biology and cell death.

Polyspecificity has the potential to target multiple TACA.

Targeting TACA has the potential to synergize with other chemotherapeutics to enhance cell death.
From Lectin/Antibody to Vaccine
CMPs provide a multifacet approach to target for panspecific immunotherapy against categories of TACA.

**Induction of Apoptosis – PanSpecific**

**TACA target**

1. Phage display peptide library

   - Lectins (WGA-GS-I)
   - Mab BR55-2/ME36.1

2. Binding peptide selection

   - Molecular Modeling

3. Carbohydrate mimetic peptide

4. Immunization

   - Br55-2 – LeY
   - ME36.1- GD2
   - Lectins

**Phage display peptide library**

- Phage library constructed
- Affinity selection
- Infected Cells produce Phage
- Wash and elute bound phage

**Analysis**

**Immunization**

- Anti-TACA response
  - P10: GVVWRYTAPVHLGDG
  - P10s: WRYTAPVHLGDG
Inhibition of tumor growth in C57BL/6 mice

Peptides used for immunization

- **P9**: LDVVLAWRDGLSGAS
- **P10**: GVVWRYTAPVHLGDG

- Peptides identified as mimic for GD2 from random peptide library screen using the anti-GD2/GD3 antibody ME361

Natural Antibodies are Reactive with CMPs

Percentage of Total IgG

- IVIg
- anti-P10L
- anti-p10s

P10: GVVWRYTAPVHLGDG
P10s: WRYTAPVHLGDG

Pan TACA Mimicry

Anti-p10s Fraction Binding (RU)

IVIg Binding (RU)
CMPs selectively enhance ganglioside antibodies

Comparison between induced mouse serum and human serum antibodies to P10s
Human Preimmune IgG that Bind P10s MAP Suppress Migration of MDA-MET Cells *In Vitro*

Binding of anti-p10s to MDA-MET

**P10s**: WRYTAPVHLCGDG

Discovery – Structural Designed

Pre-Clinical
Efficacy Assessment
- Set up GLP condition
- SOPs, Animal Housing
- FDA regulation

Safety study – Protocol Number 2935

4 groups: 3 doses (500ug, 300ug, 100ug), 1 control – (32 mice per group)

5 Immunization: Weeks 1, 2, 3, 7 & 19
3 Euthanasia: Weeks 3, 9, 21
General Health Monitoring: Weight meas. Inj. site reaction, Morbidity
Immunological assessment: Serum binding to CMP, CH-expressing cells
Clinical pathology: Urine & Blood analysis
Necropsy: Weighted Organs: Kidney, Liver, Spleen, Heart
Gross pathology: Tissue processing H&E staining

No adverse effect observed

IND Preparation
>1600 pages
- GLP Study Report
  Safety – 295 pages
- Study Protocol
  Vaccine Candidate
- Chem/Man/Con
  Vaccine Candidate

FDA

Phase I

PRMC
IRB approval

FDA approval

- P10s-PADRE: WRYTAPVHLGDG-aK-Cha-VAAWTLKAAa + (QS-21) Montanide ISA 51
Protocol Summary

- A Phase I dose escalation trial followed a rule-based 3+3, with 3 subjects at each dose if no toxicities were observed.

- Patients with advanced Breast Cancer underwent CMP vaccination (subcutaneous injections) on weeks 1, 2, 3, 7 and 9.

- The primary endpoint was the safety of CMP vaccination.

- The secondary endpoints were immune response, as measured by antibody titer to P10s and reactivity to TACA expressing human breast cancer cell lines.
Investigational product:
- P10s-PADRE administered with MONTANIDE ISA 51 VG- 1.0 mL subcutaneous (SC) injections

Inclusion Criteria:
- Females
- Stage IV breast
- Who have not required a treatment change for 2 months
- DTH Response to Recall Antigen

Exclusion Criteria:
- Pregnant, breast-feeding
- Autoimmune disease
- Immunosuppression
Immunization leads to anti-P10s and enhanced GD2 Responses

Table 4. Anti-GD2 antibody titer in vaccinated subjects.

<table>
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<th>Subject</th>
<th>Endpoint titer in post immune serum</th>
<th>Fold increase in endpoint titer after immunization</th>
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<tr>
<td>6</td>
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Anti-P10s serum inhibits migration of HCC1954 cells

- Cells incubated overnight with FBS or indicated sera on transwell membranes.
- Membranes fixed, stained. Migrated cells were visualized by light microscope and counted.
- Average over three replications with SD are shown.
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P10s serum induced apoptosis in Breast Cancer cells lines

Overall survival among the vaccinated subjects had a mean ±SE (median) of 908±116 (928) days compared to 583±126 (312) days among the unvaccinated, consented subjects.
Preincubation with subject’s serum sensitized tumor cells to docetaxel toxicity. Cell survival was determined and IC50 were estimated. Postimmune IC50 is significantly different than FBS and Preimmune IC50s, with P values of 6.21E-08 and 0.002, respectively.
(A) Baseline PET scan shows two small lesions in the lower lobe of the right lung.

(B) The lesions FDG uptake on PET scan that was done 7 weeks after vaccination.

(C) PET scan six months later showed return to baseline with no new lesions elsewhere.

- The subject was receiving Vinorelbine and Trastuzumab (VT) for two months prior to study eligibility.

- The subject was switched to Docetaxel, Pertuzumab and Trastuzumab and her last PET scan showed a response in the lungs and lymph nodes.
Cystic lesions were seen in the cerebellum (A), temporal (B) and frontal (C) lobes.

Resection of the two large lesions showed no viable tumor on pathology specimens. PET scan done around the same time showed return of the lung lesions to baseline suggesting maximal response in the lung and possibly in the brain, which might explain the absence of viable tumor on pathology.
Tumor was 1.3 cm as compared to 4 cm before we started the treatment (this represents almost 90% drop in volume). This is after one cycle of AC!!!!
Where are we going Clinically?

- DCIS
- Fibrosarcoma
- Lung
- Pancreatic
- Melanoma/glio/astro (Neuronal)
  - Primary objectives: Continue to monitor safety, tolerability, immunogenicity and cytotoxicity.
  - Secondary – CTC, T cell responses
Where are we going translationally?

- B Cell compartment and Biomakers
- CTCs
- Role for NK cells
- Role of T cells – new peptide design
The Workers

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