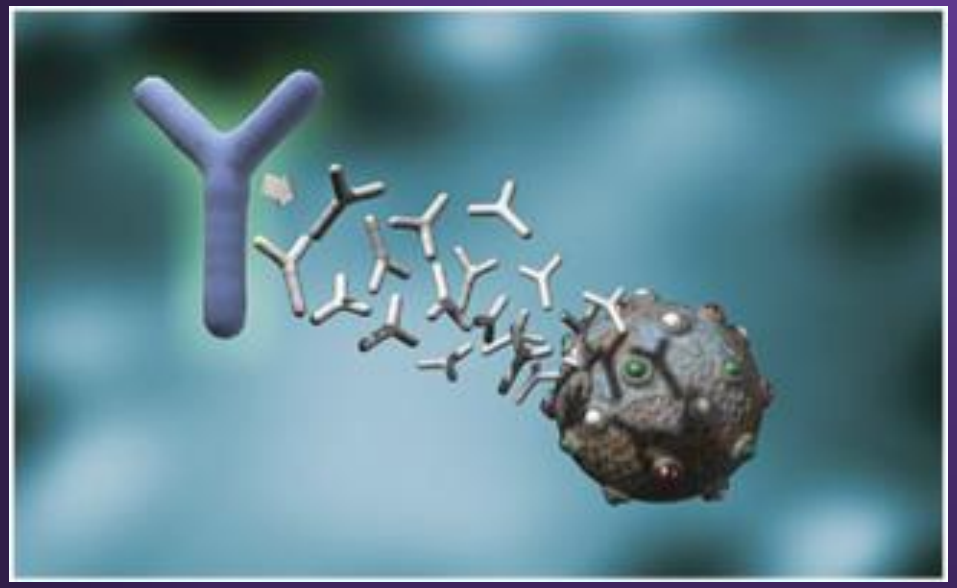
Developing a First in Man Carbohydrate Mimetic Peptide Vaccine for Cancer: A Translational Story



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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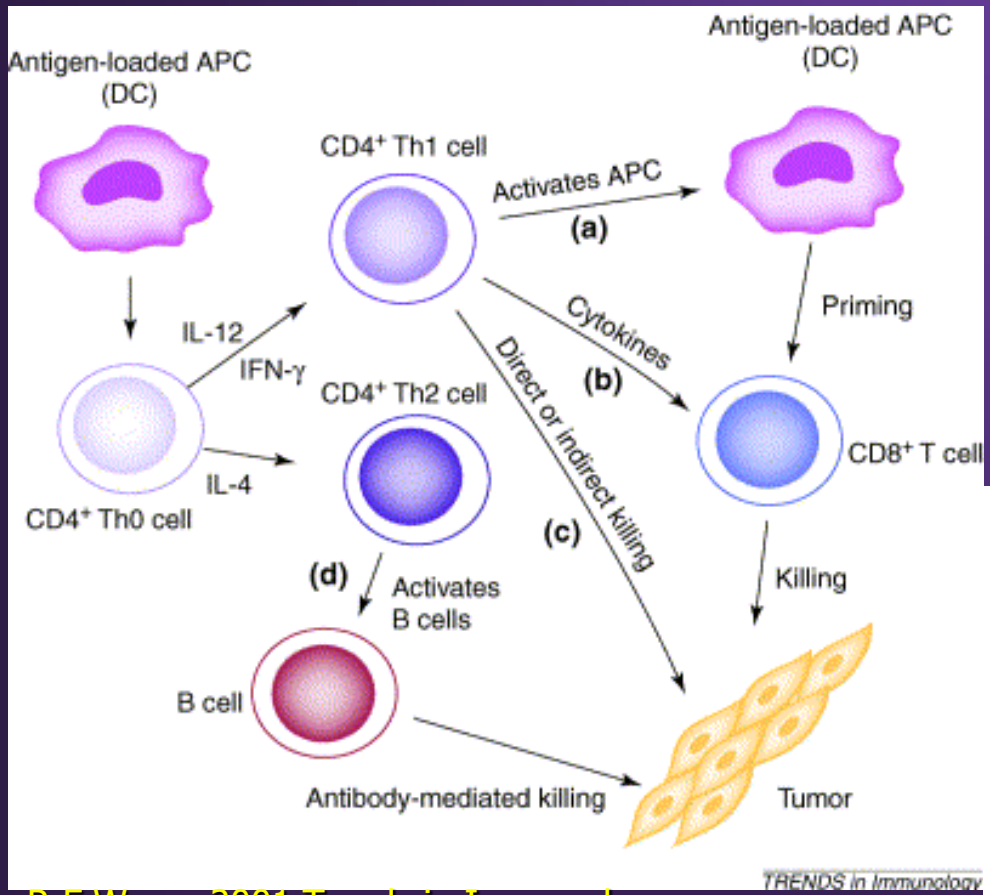
Different from Carbohydrate Vaccines



R-F Wang 2001 Trends in Immunology

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.

Each CMP is a Pan antigen

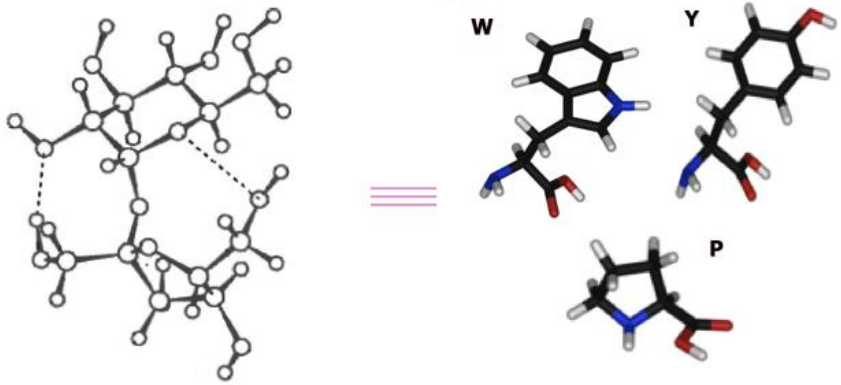
Generate poly reactive or Panspecific antibodies

Generate anti-Tumor T cells

Provides for multiple platforms

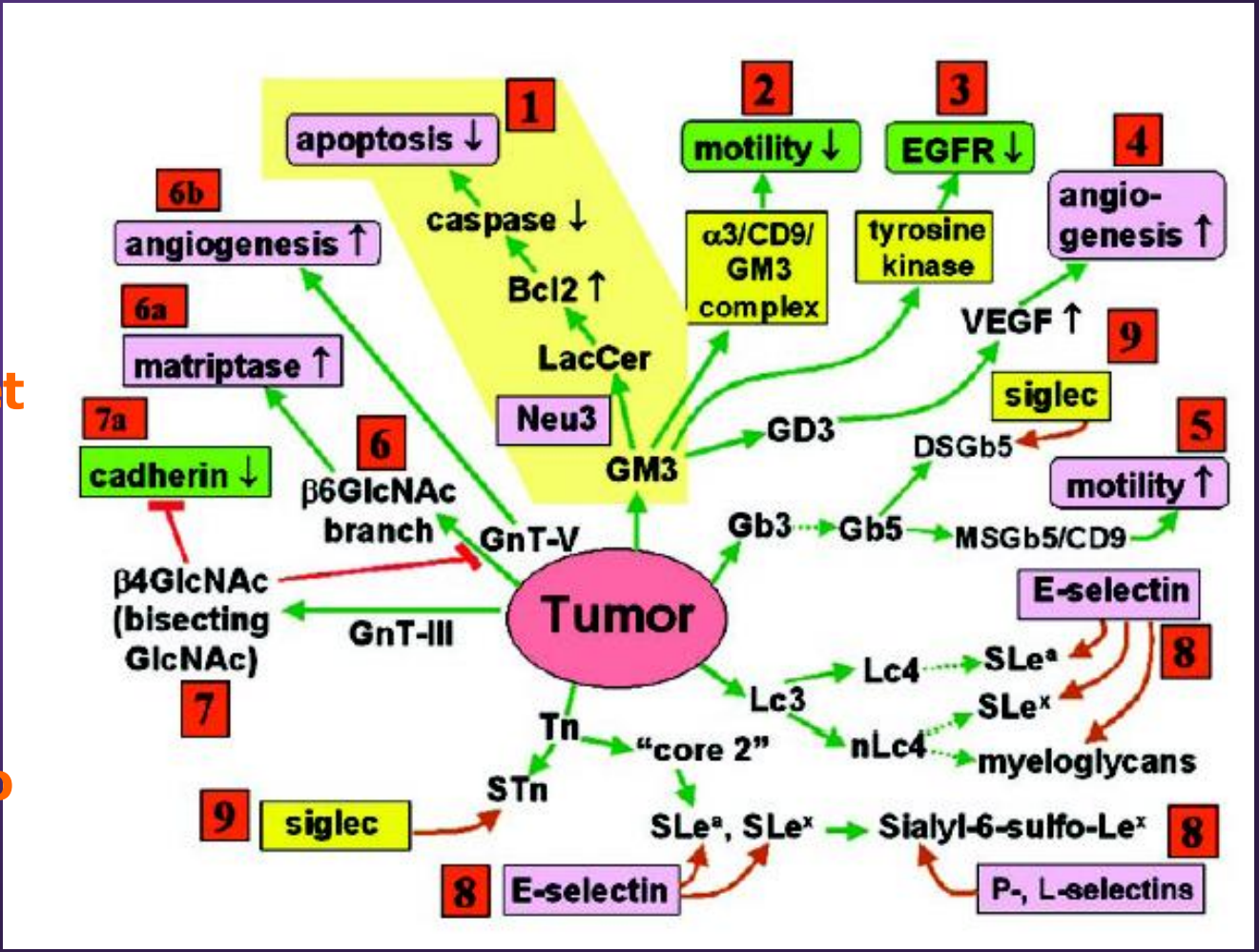
Generates long term memory

Carbohydrate mimetic peptides as TACA surrogates



Rational for Harnessing the Inherent Polyspecificity of Antibodies To Target Multiple TACA

- 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

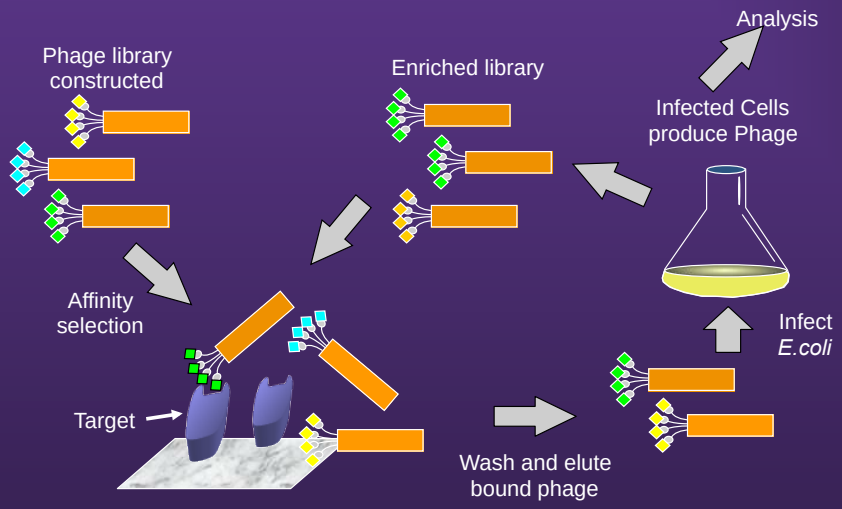
Binding apoptosis

1

Lectins (WGA-GS-I)

Mab BR55-2/ME36.1

Phage display peptide library



4

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

Binding peptide selection

Molecular Modeling

2

Carbohydrate mimetic peptide

Immunization

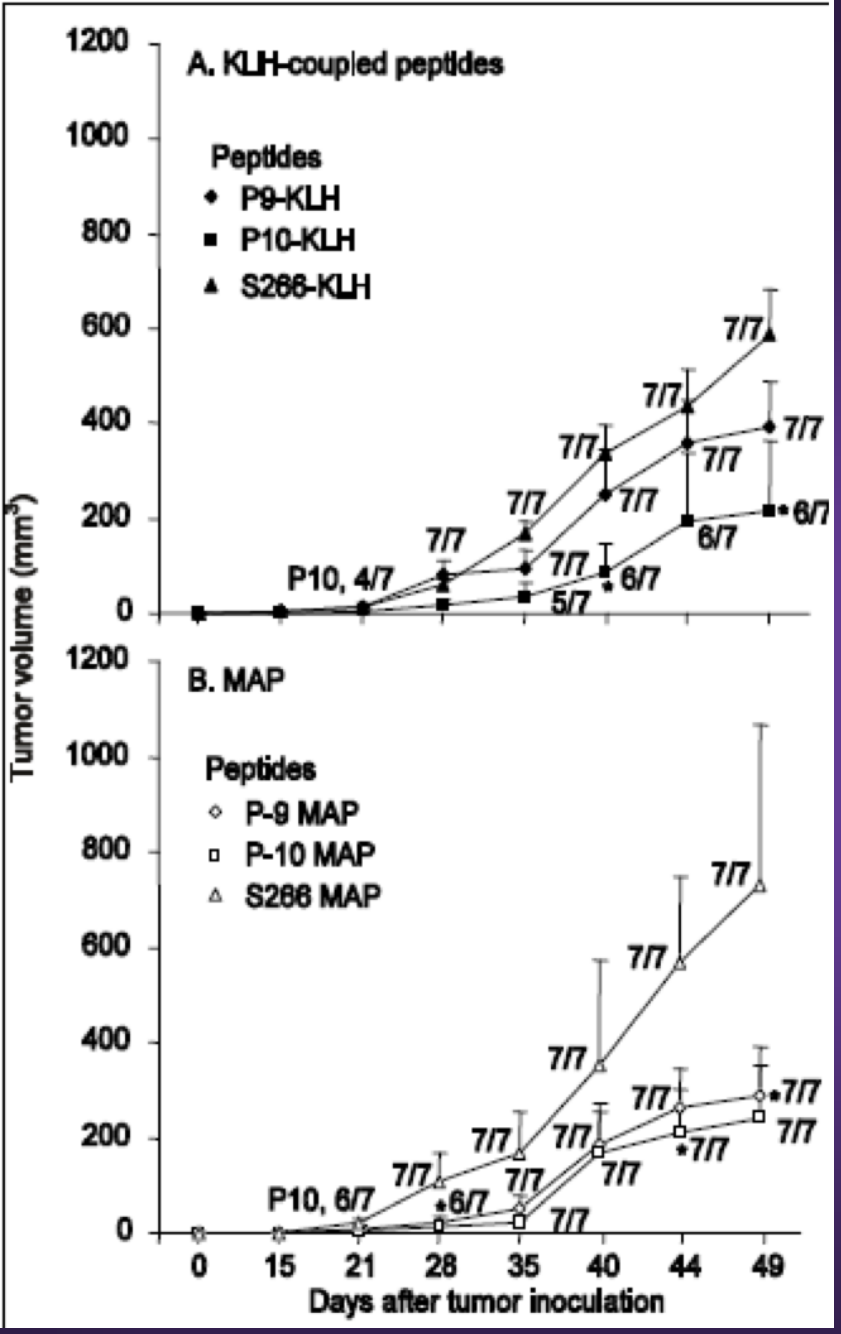
3

Anti-TACA response

P10: GVVWRYTAPVHLGDG

P10s: WRYTAPVHLGDG

Inhibition of tumor growth in C57BL/6 mice



Peptides used for immunization

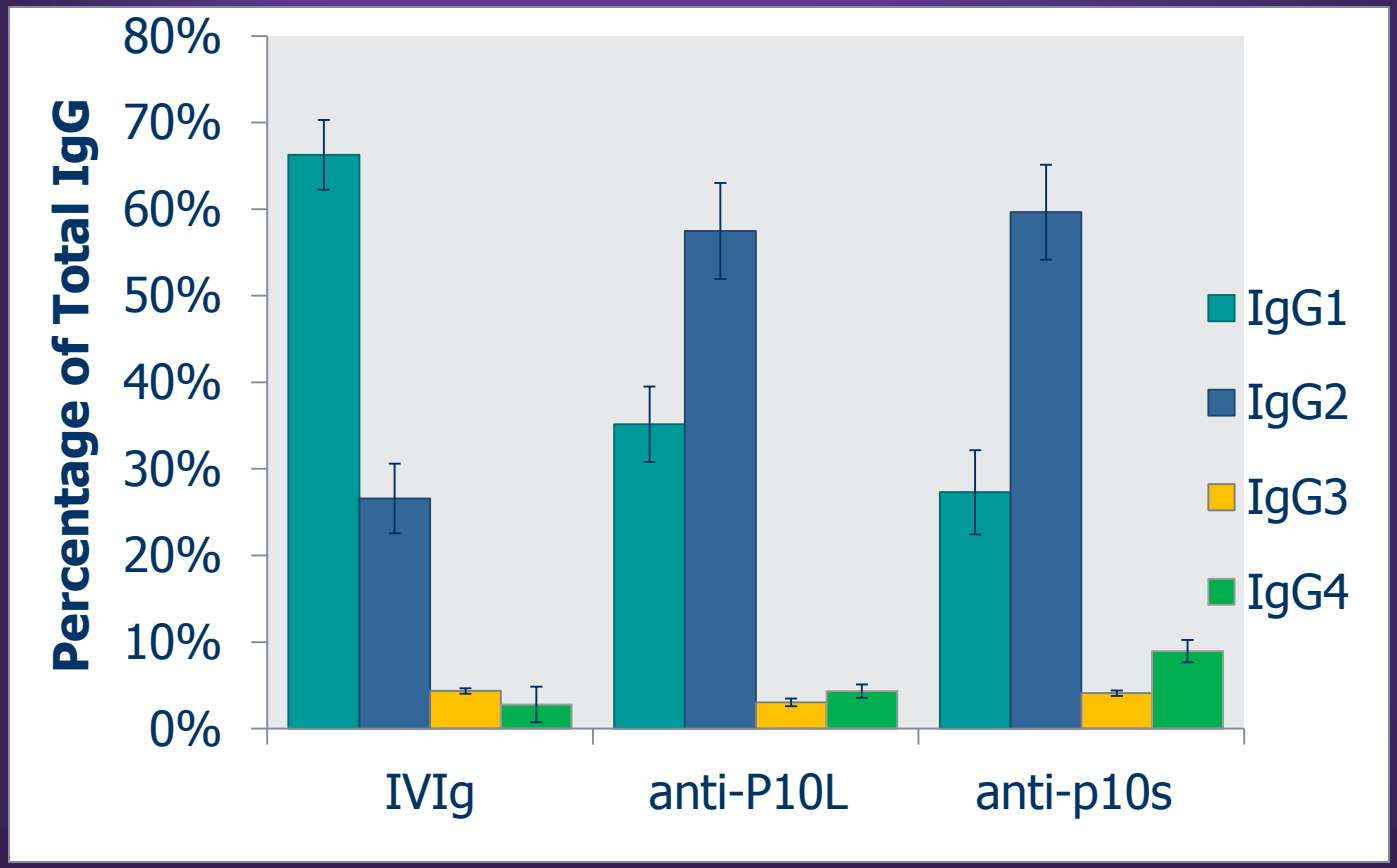
P9: LDVVLAWRDGLSGAS

P10: GVVWRYTAPVHLGDG

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

Wondimu et al CII (2008) 57:1079–1089

Natural Antibodies are Reactive with CMPs

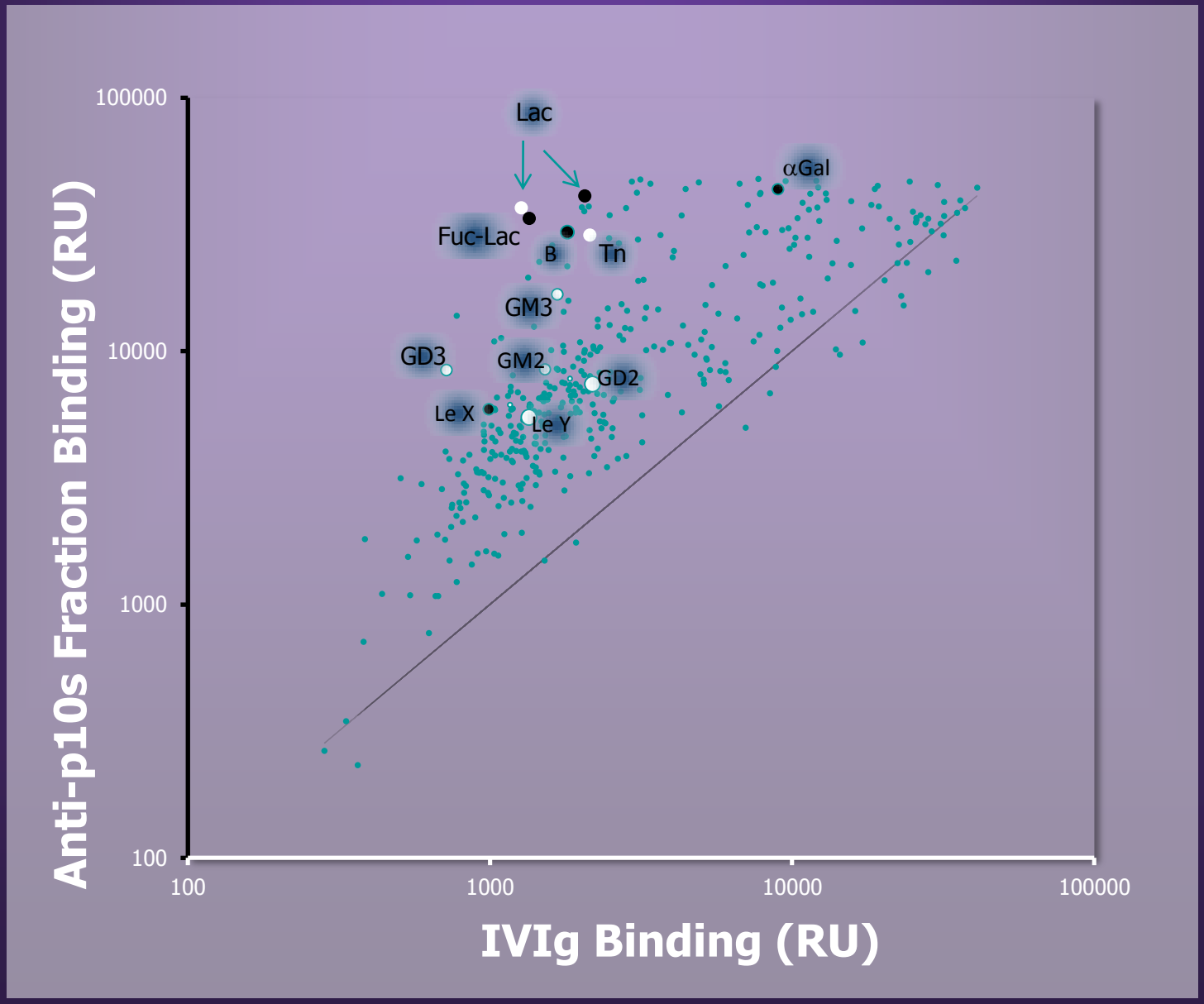


P10: GVV**WRY**TAPVHLGDG

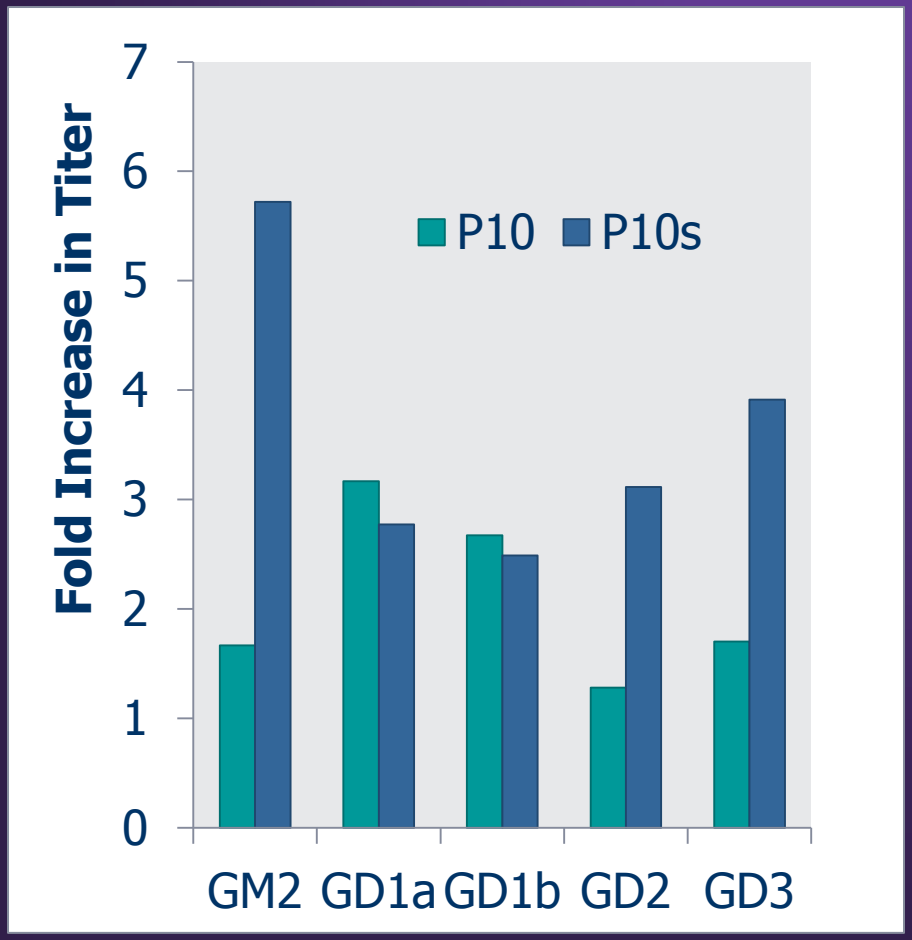
P10s: **WRY**TAPVHLGDG

Pashov, A., et al. (2009) Vaccine. 27: 3405-3415

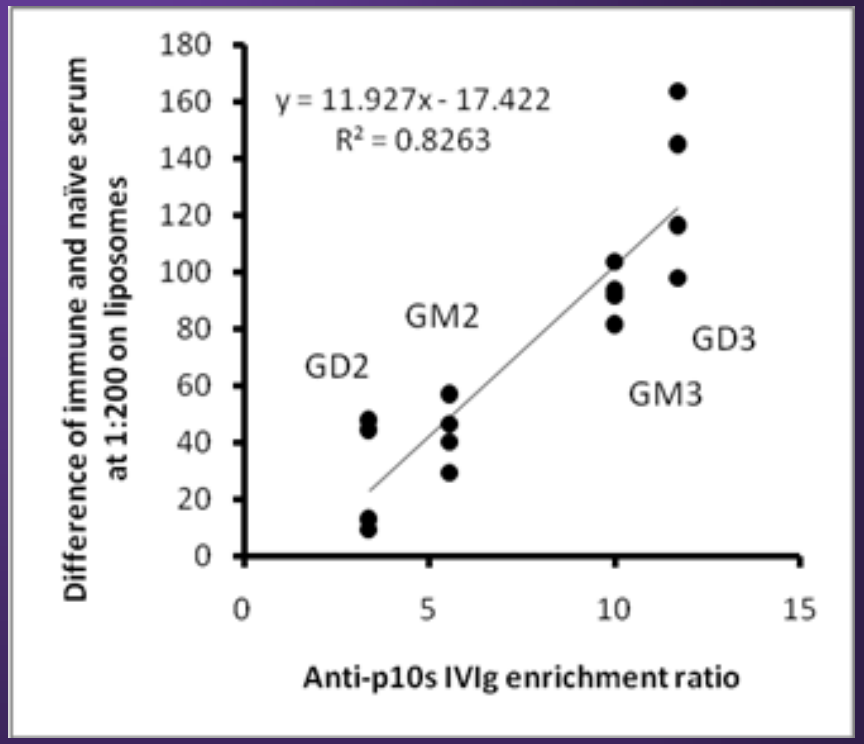
Pan TACA Mimicry



CMPs selectively enhance ganglioside antibodies

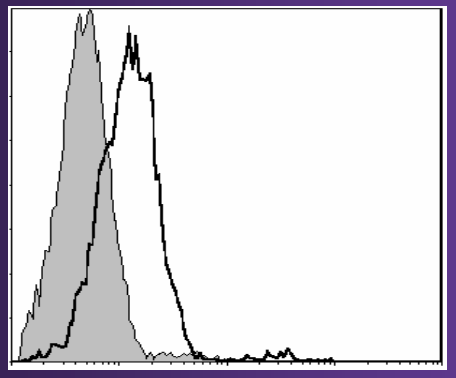


P10: GVV**WRY**TAPVHLGDG
P10s: **WRY**TAPVHLGDG



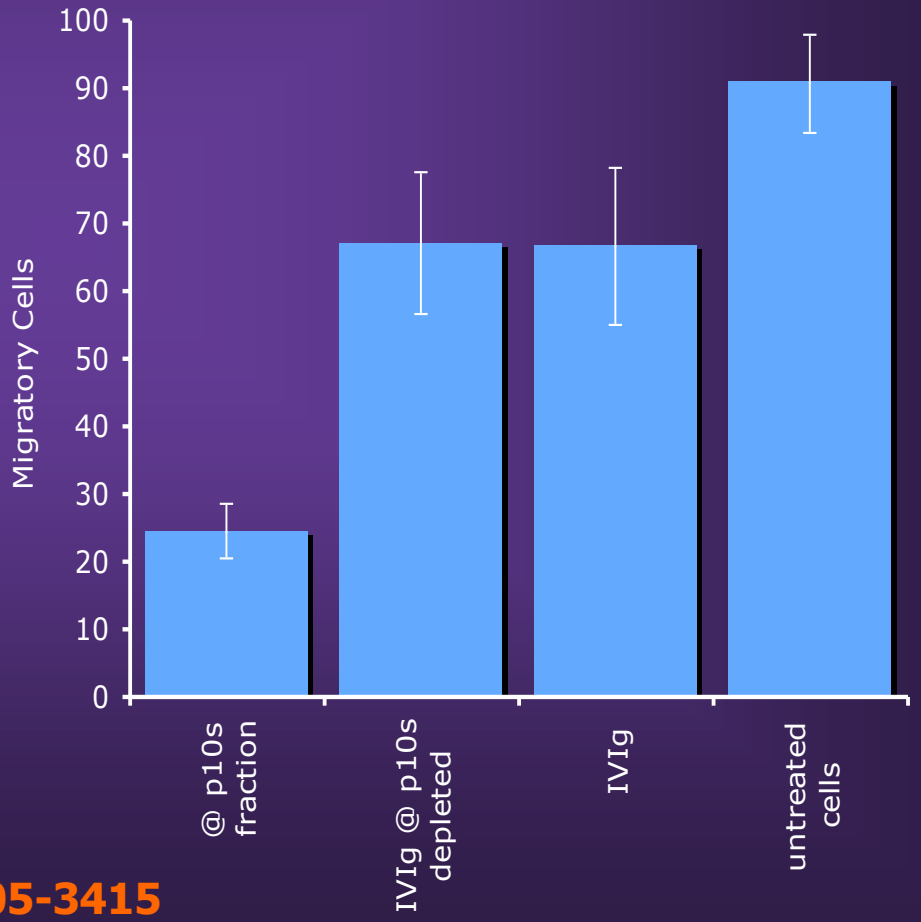
Comparison between induced mouse serum and human serum antibodies to P10s
Monzavi-Karbassi, et al. (2007) Vaccine 25: 3022-3031.

Human Preimmune IgG that Bind P10s MAP Suppress Migration of MDA-MET Cells *In Vitro*



Binding of anti-p10s to MDA-MET

P10s: WRYTAPVHLGDG



Pashov, A., et al. (2009) Vaccine. 27: 3405-3415

Discovery – Structural Designed

Pre-Clinical

Efficacy Assessment

Set up GLP condition

SOPs, Animal Housing

FDA regulation

IND Preparation

>1600 pages

→ FDA

GLP Study Report	Study Protocol	Chem/Man/Con
Safety – 295 pages	Vaccine Candidate	Vaccine Candidate

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

PRMC
IRB approval

FDA approval

Phase I

No adverse effect observed

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

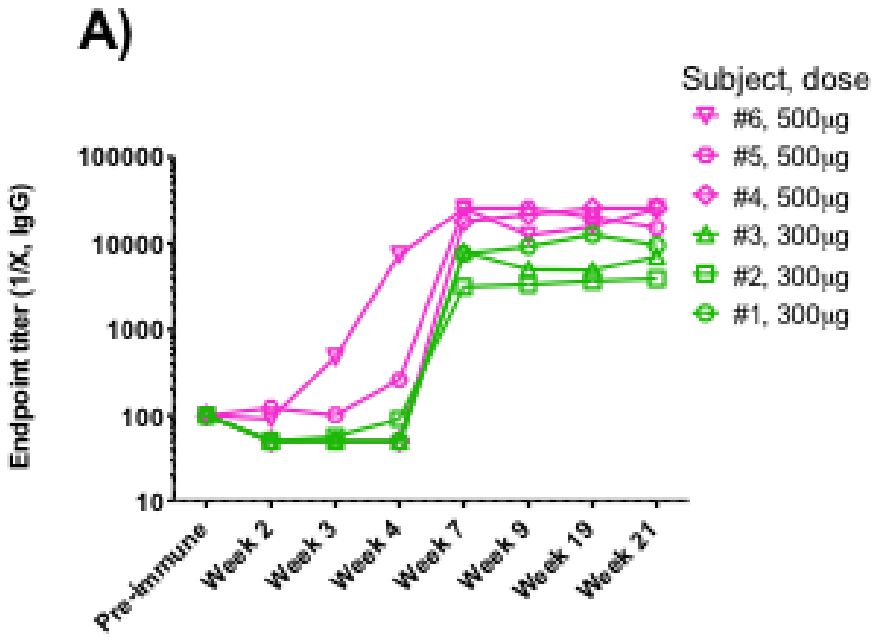
Protocol Summary

- 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.

Protocol Summary – Cont.

- **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



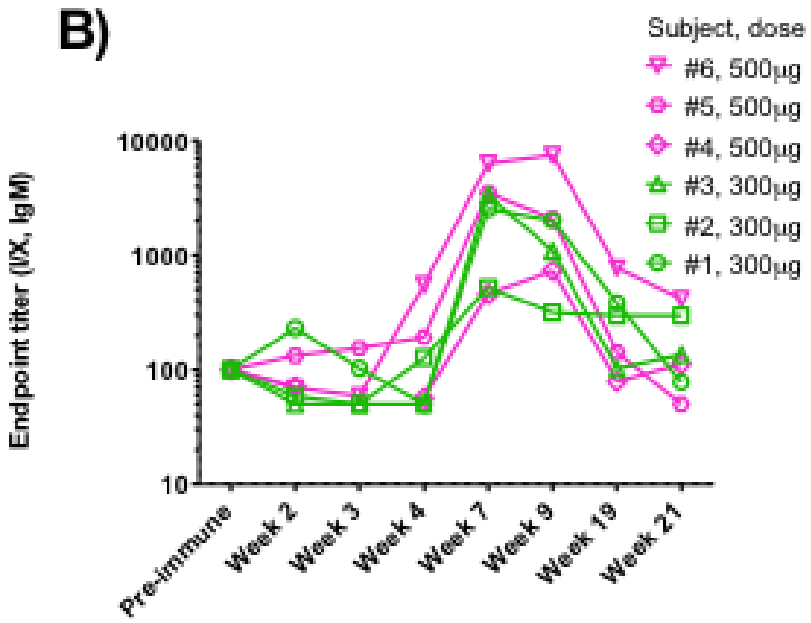
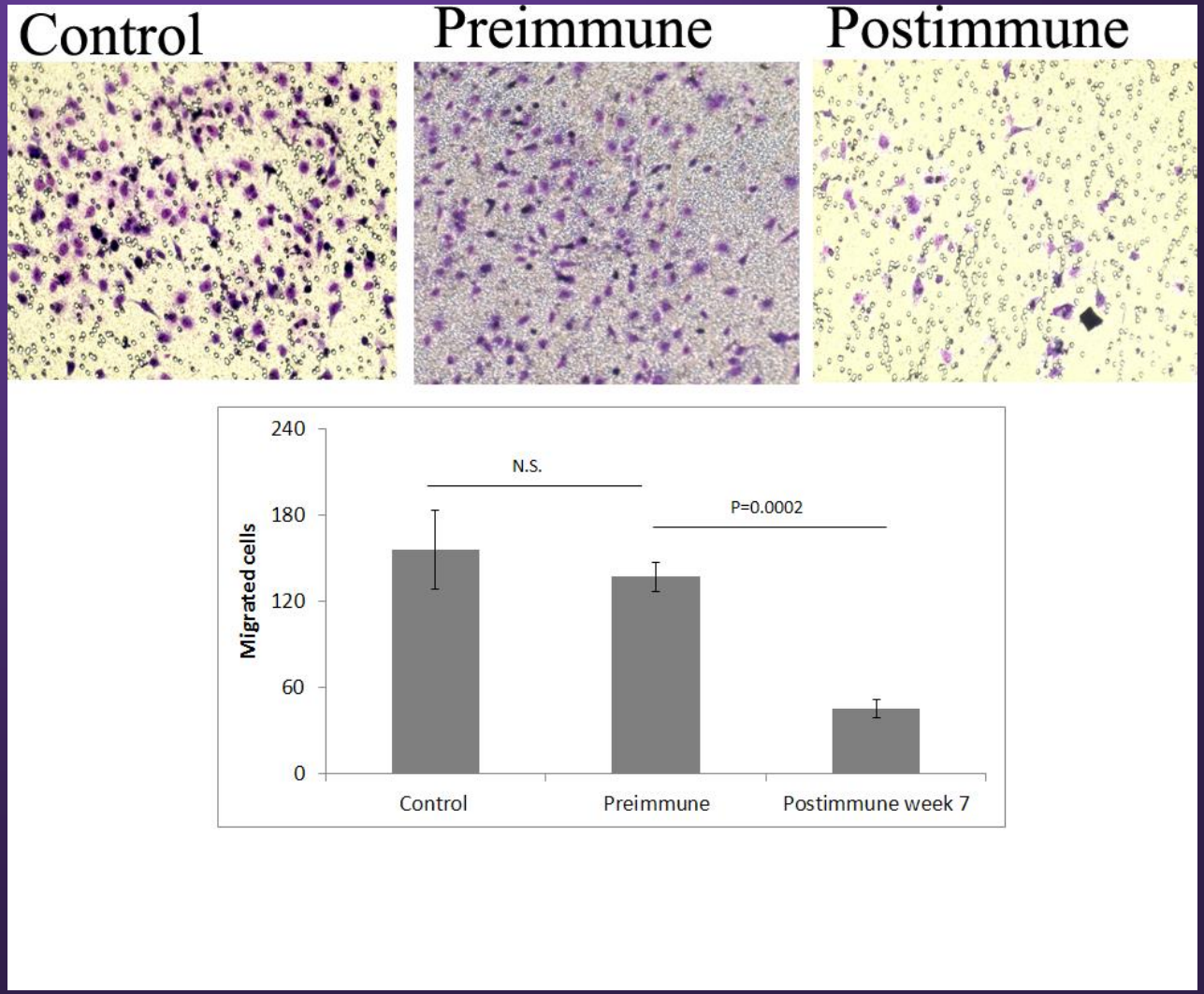


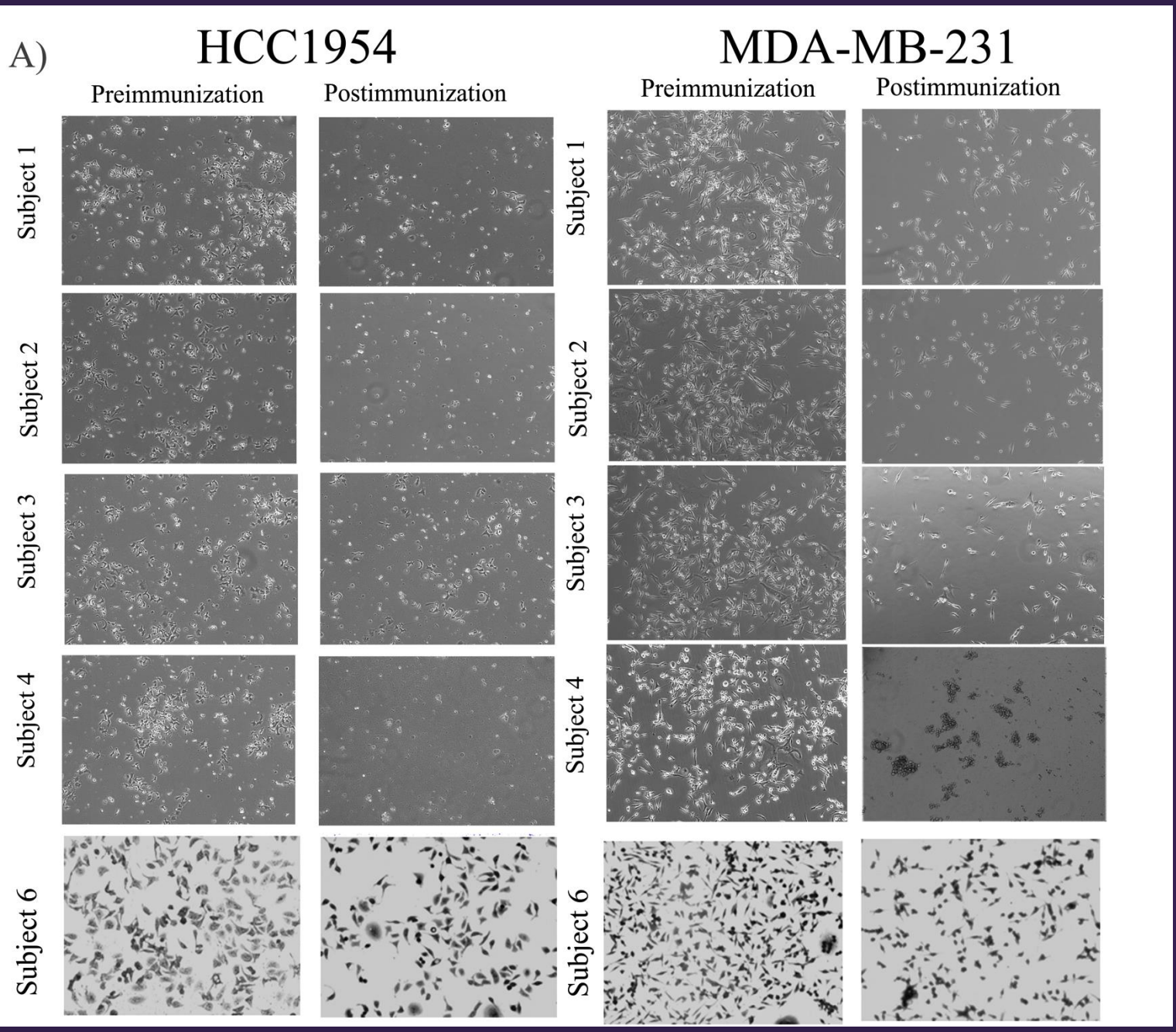
Table 4. Anti-GD2 antibody titer in vaccinated subjects.

Subject	Endpoint titer in post immune serum	Fold increase in endpoint titer after immunization
1	160	4
2	160	8
3	80	4
4	160	8
5	160	4
6	160	4

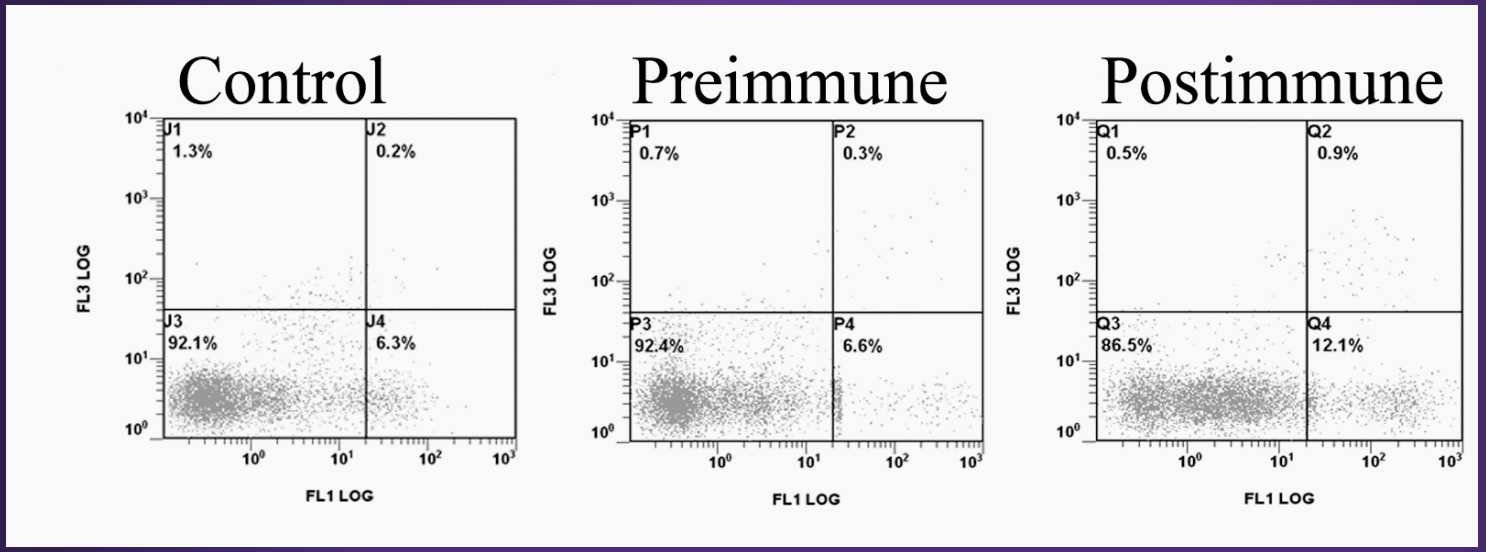
Anti-P10s serum inhibits migration of HCC1954 cells

- 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.




A)
HCC1954
MDA-MB-231
Preimmunization
Postimmunization
Preimmunization
Postimmunization
Subject 1
Subject 2
Subject 3
Subject 4
Subject 6
Subject 1
Subject 2
Subject 3
Subject 4
Subject 6

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.

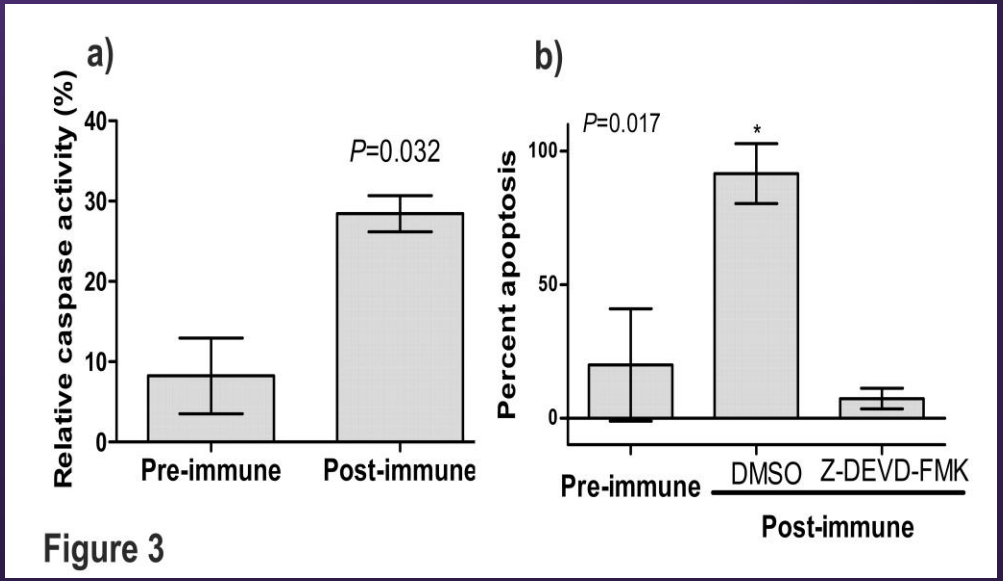
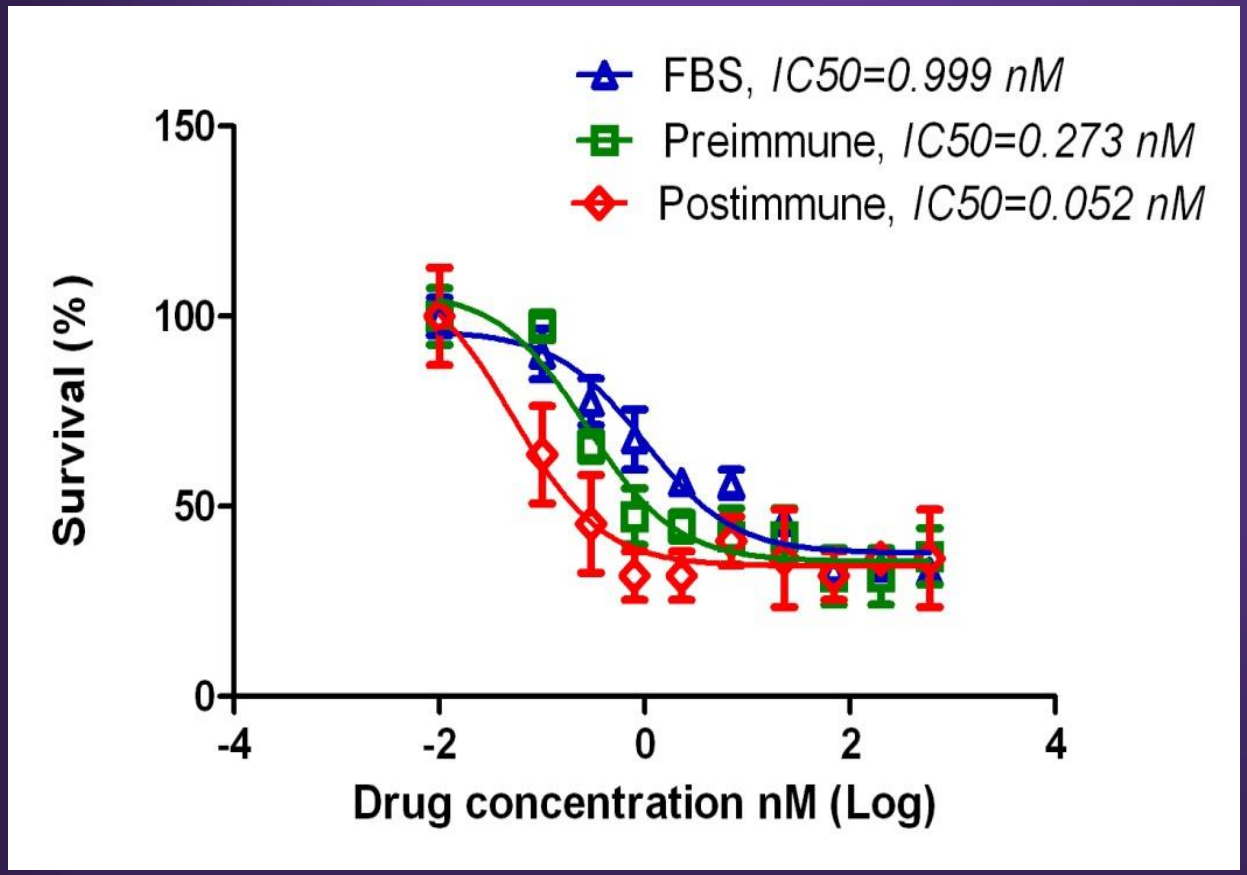


Figure 3

Combination Therapy?

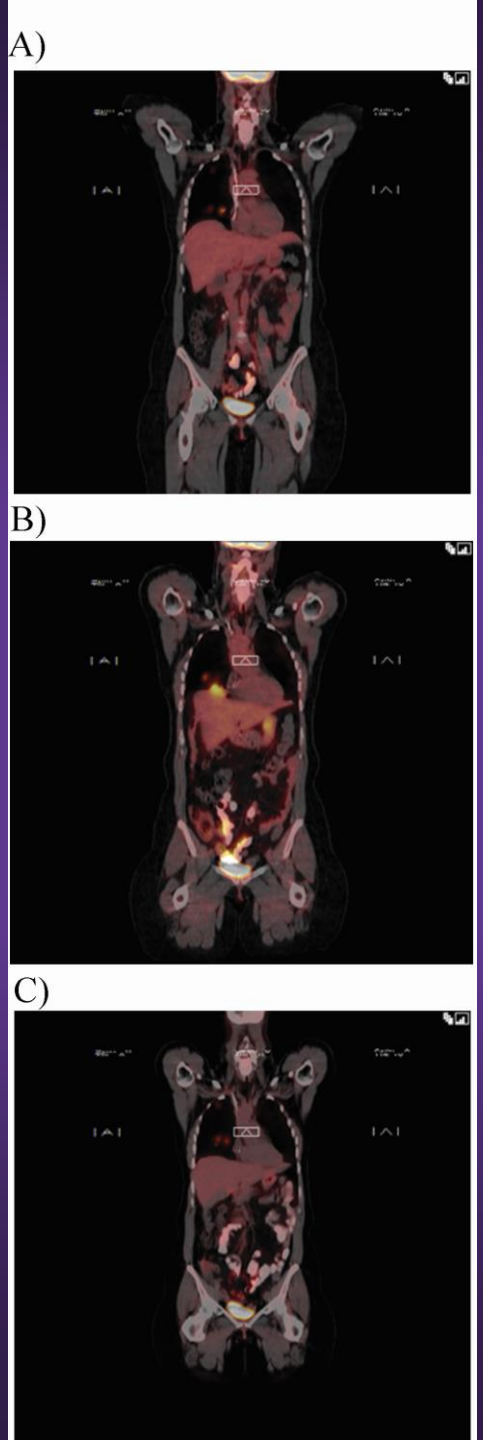


Preincubation with subject's serum sensitized tumor cells to docetaxel toxicity. Cell survival was determined and IC50 were estimated. Postimmune IC50 is significantly different than FBS and Preimmune IC50s, with P values of 6.21E-08 and 0.002, respectively.

(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.

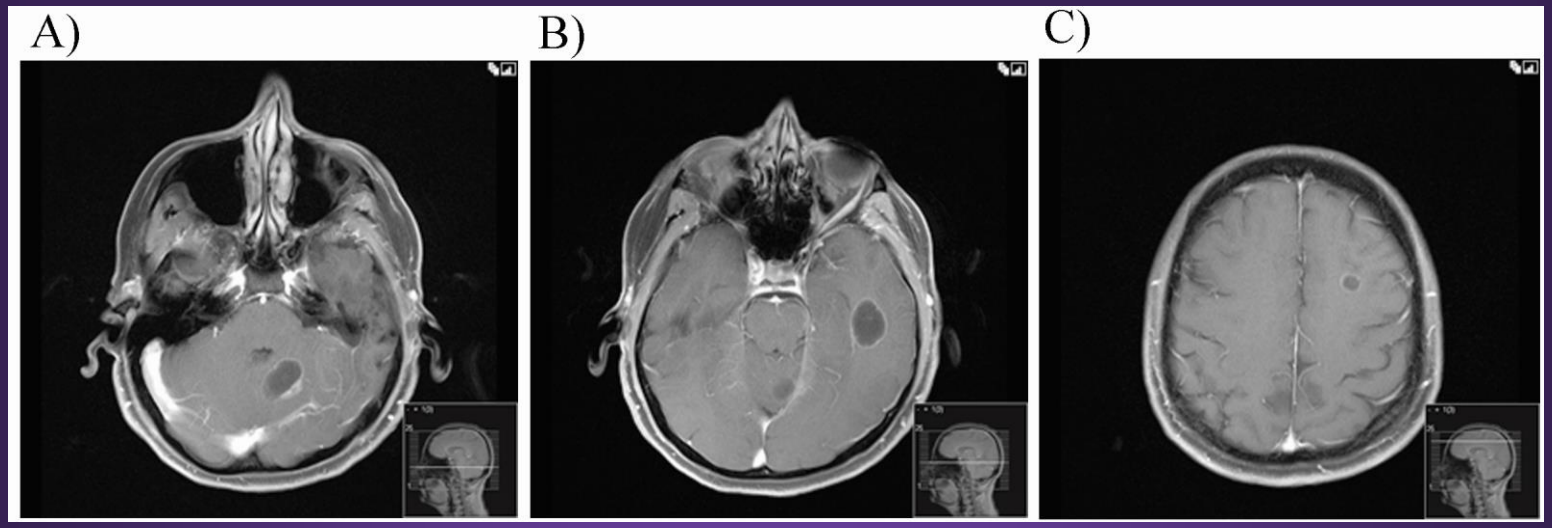
(C) PET scan six months later showed return to baseline with no new lesions elsewhere.



- 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**

MRI of the brain



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.

Phase II - Sequential combination therapy effects – with Immune suppression and pCR endpoints

- Week 1
- Administer vaccine
- AC Chemotherapy – cyclophosphamide 600 mg/m2 and doxorubicin 60 mg/m2
- Review of side effects
- Clinic visit for history and
- physical exam
- Blood work for research
- Weeks 2 & 3
- Administer vaccine
- Review of side effects

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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C.Fred Brewer- Einstein
Tom VanCott - HJ Found
Moon Nahm- U. Alabama
Jeff Alexander –Epimmune
Julie Westerink –Ohio
Dorothee Herlyn –Wistar

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