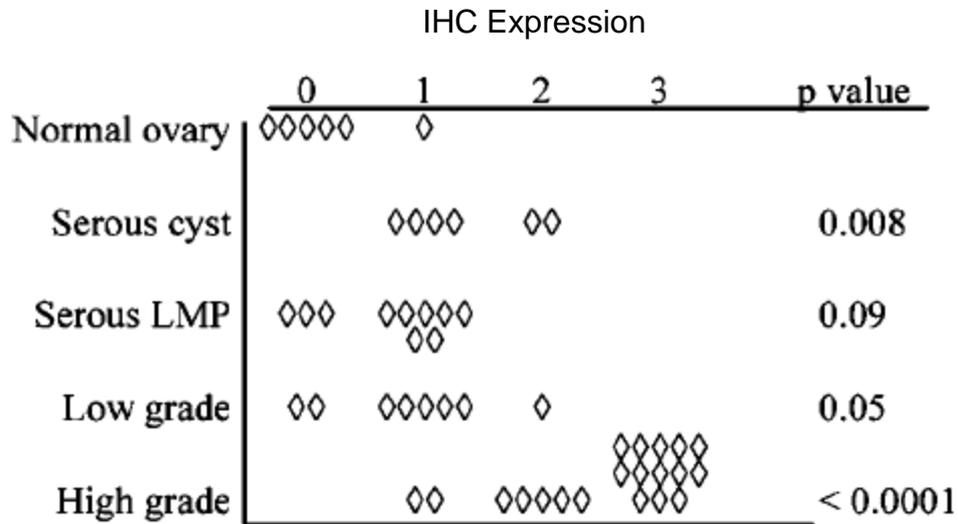


**A Phase I Trial of the Safety and Immunogenicity of a DNA Plasmid Based Vaccine Encoding the Amino Acids 1-163 of Insulin-Like Growth Factor Binding Protein-2 (IGFBP-2) in Patients with Advanced Ovarian Cancer**

## **Rationale for targeting IGFBP-2**

# IGFBP-2 expression in ovarian cancer



Lee et al, Mol Ca, 2005

Incidence of expression

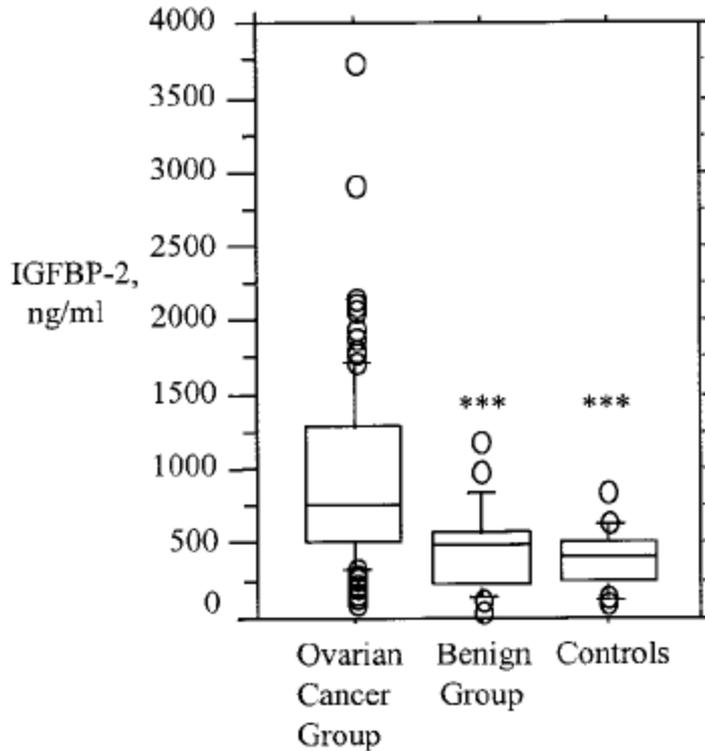
Ovarian Tumor	Number Samples	% with 2-3+
Serous	173	81%
Endometrioid	26	72%
Mixed	66	68%
Stage I	20	63%
Stage II	19	70%
Stage III	197	73%
Stage IV	61	82%

Wang et al , Mod Pathol, 2006

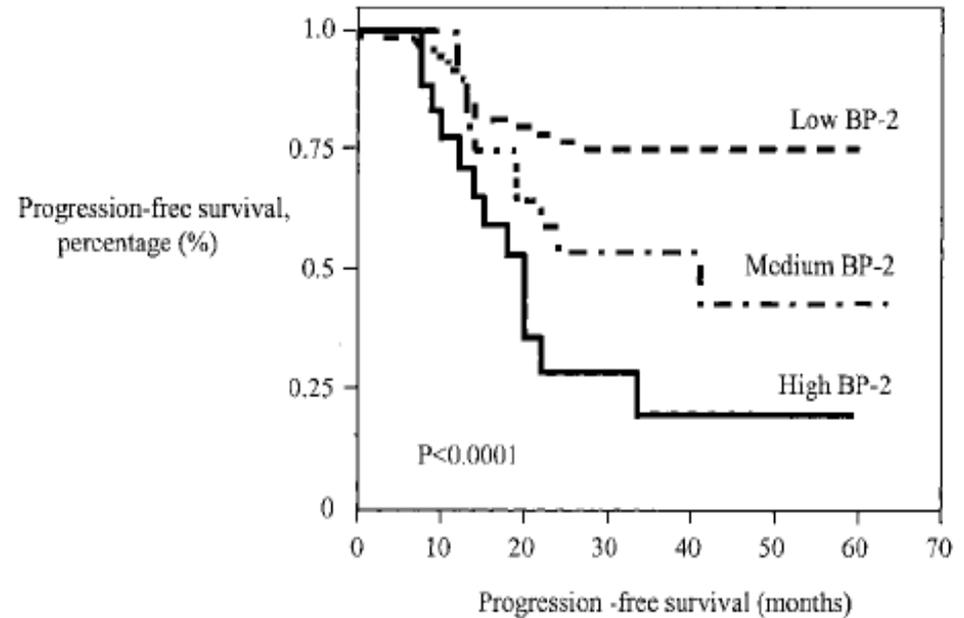
All ovarian cancer (n=113) have upregulated IGFBP-2 RNA as compared to normal ovarian tissue (34 fold)

Lancaster et al , In J Gyne Ca, 2006

# IGFBP-2 is a negative prognostic indicator in ovarian cancer

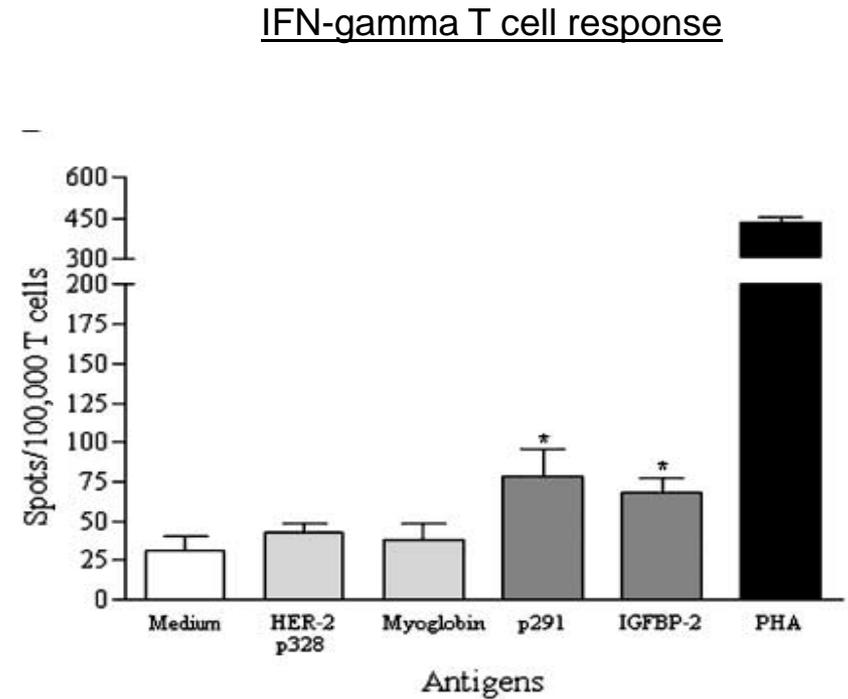
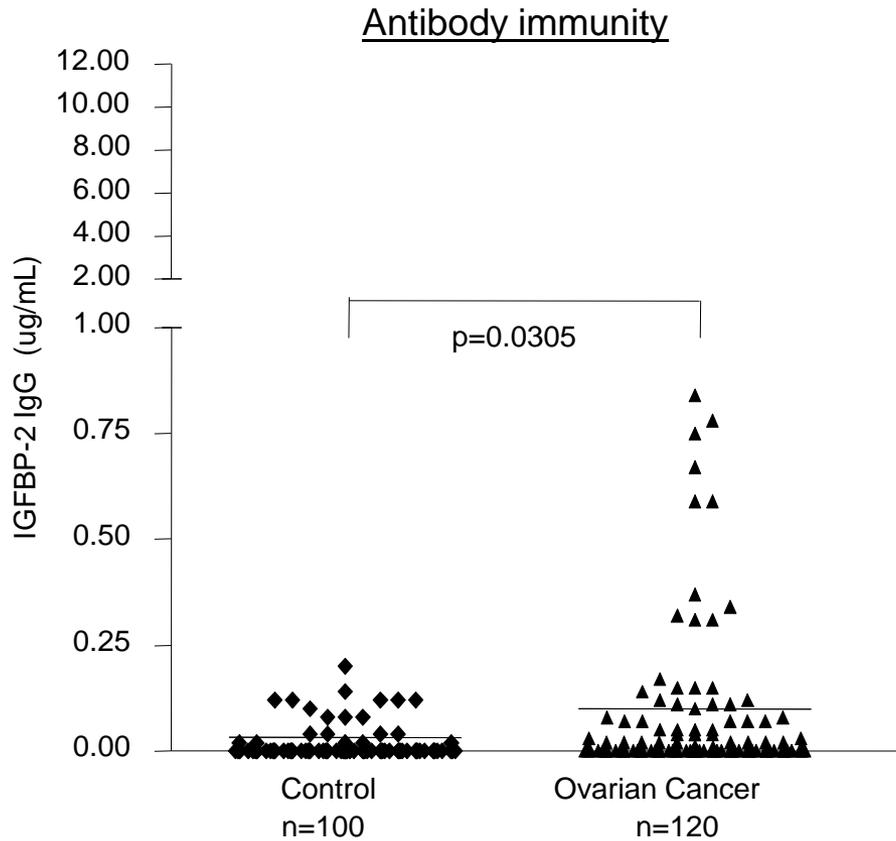


Elevated serum levels in invasive disease



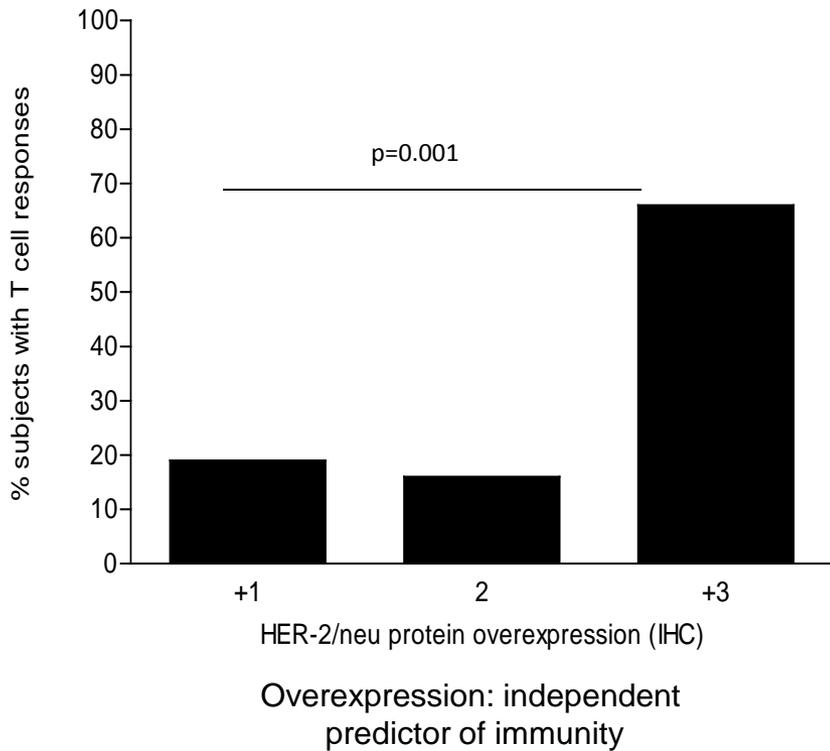
Elevated IGFBP-2 associated with poor survival

# IGFBP-2 is an ovarian cancer antigen

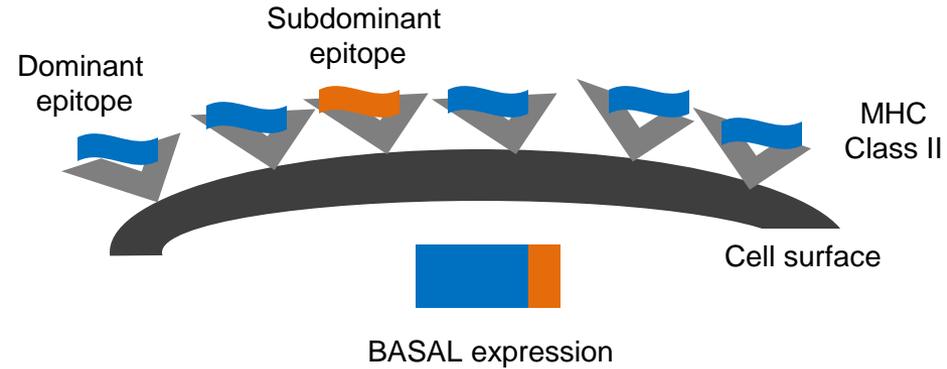


\* $p < 0.05$

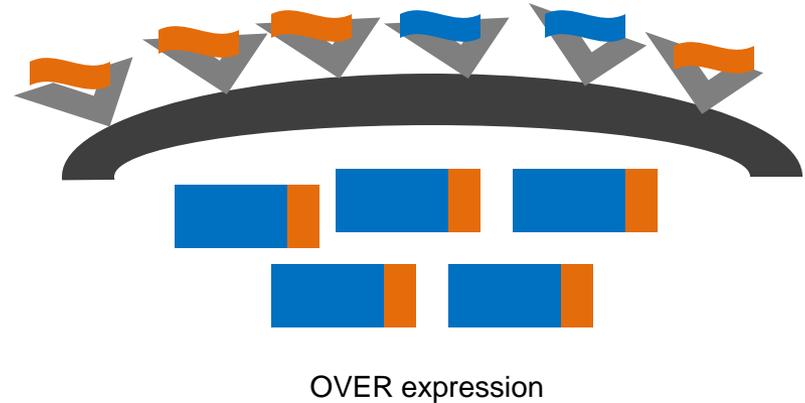
# T cell specificity for overexpressed self proteins



## TOLERANCE

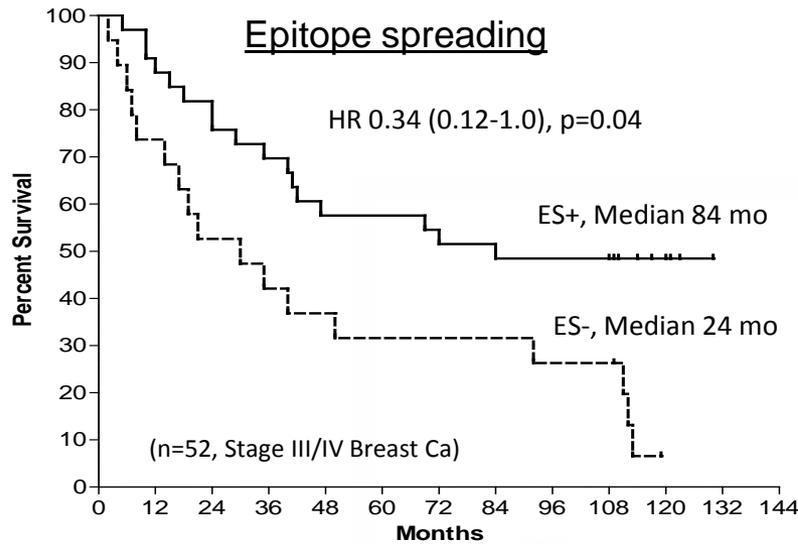


## IMMUNE RESPONSE

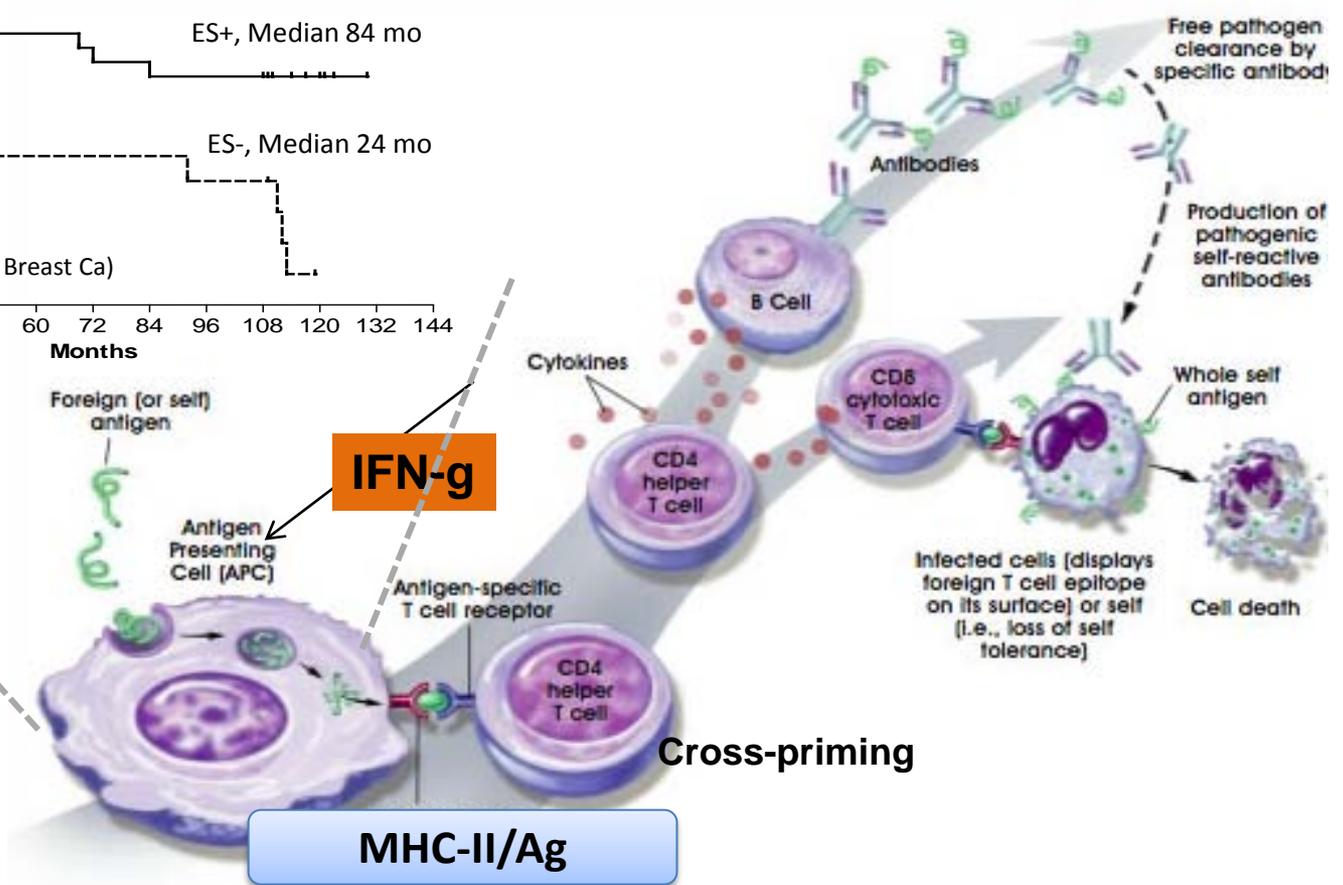


# **Rationale for vaccine design**

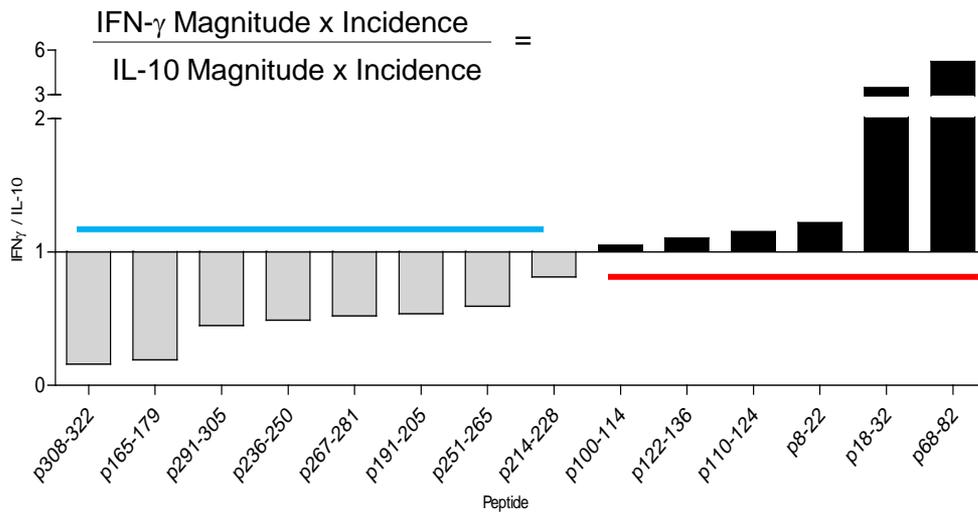
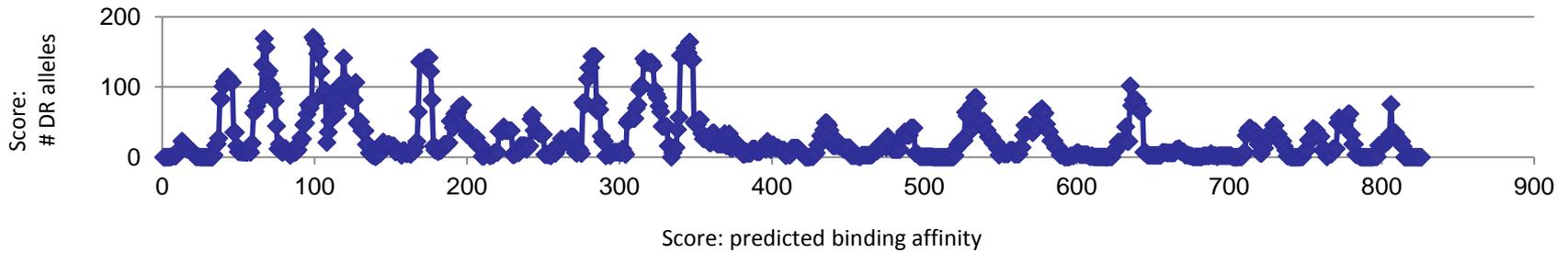
# Vaccines designed to stimulate IGFBP-2 specific CD4<sup>+</sup> T cells



Salazar et al, 2011



# IGFBP-2 T cell epitopes that preferentially induce IFN-gamma secretion

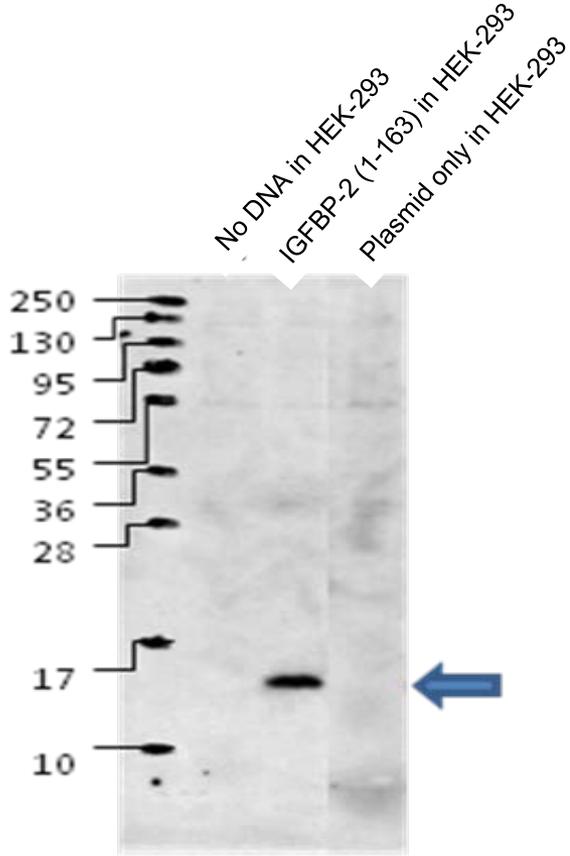
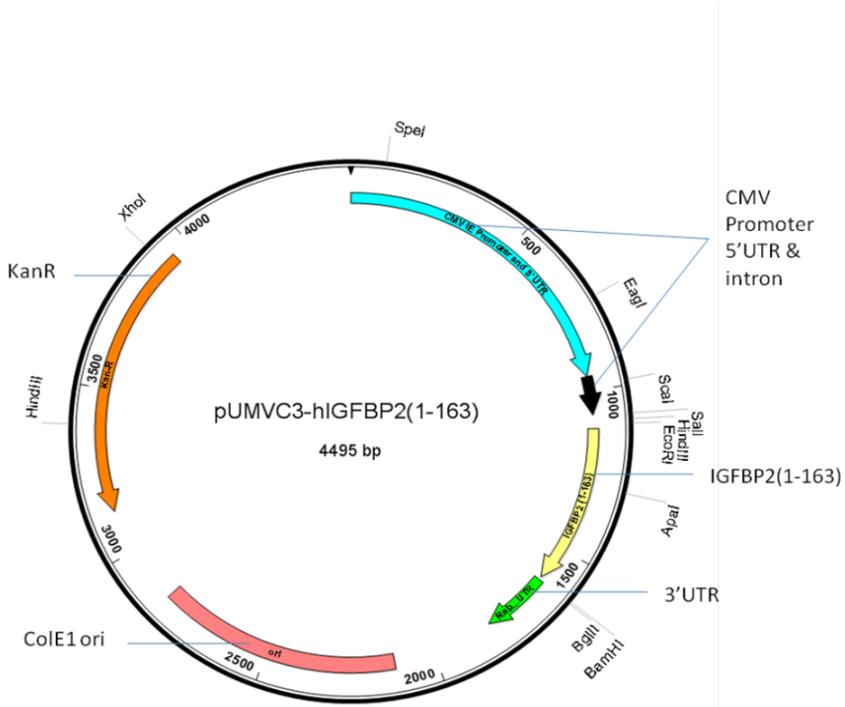


## N terminus

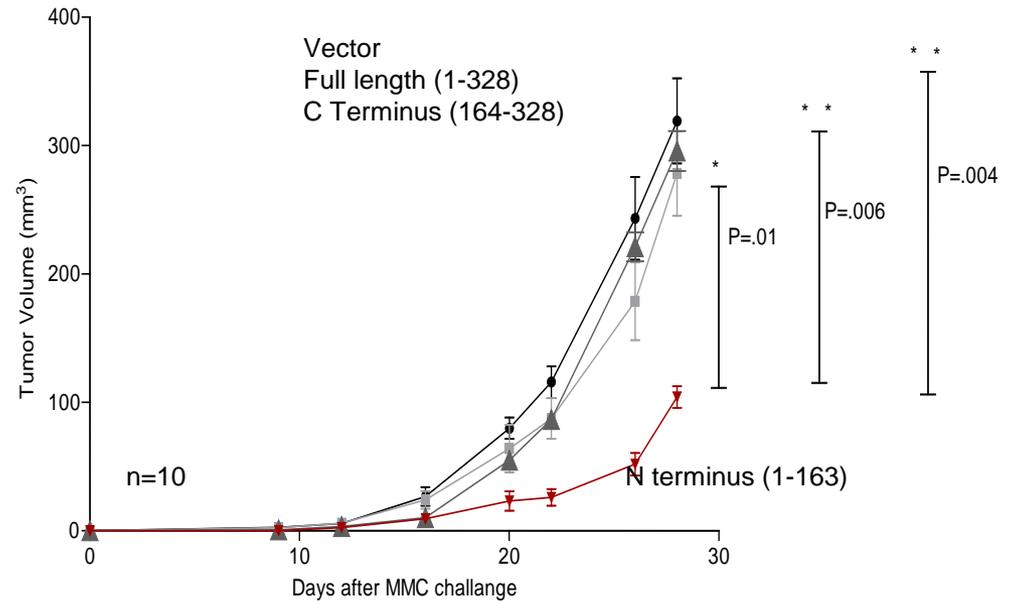
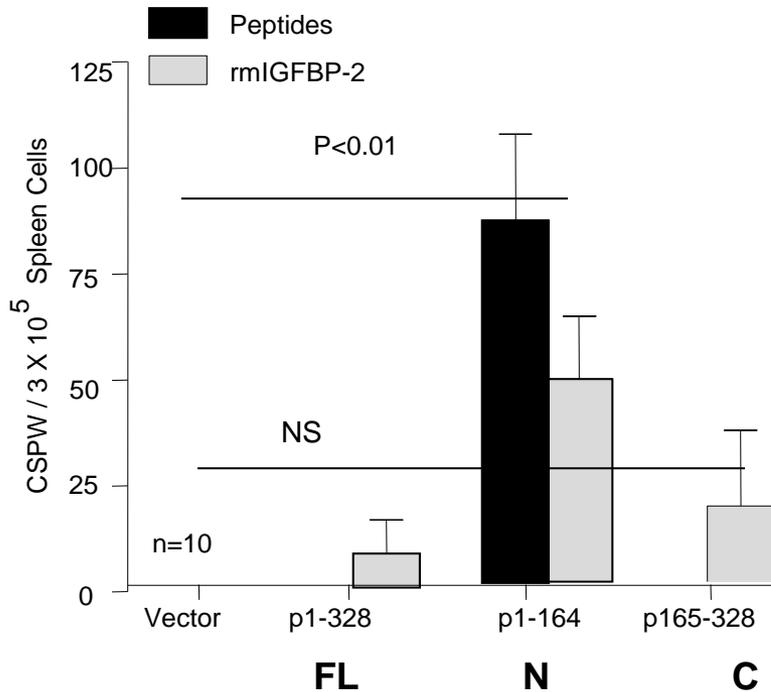
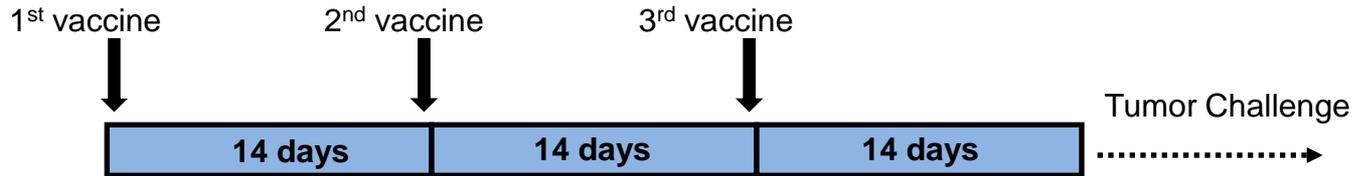
MLPRVGC**PALPLPPP**LLPLLLLLLLL**GASGGGGGA**  
 RAEVLFRCPPCTPERLAACGPPPVAPPA**AAVAVAGG**  
**ARMPCAELV**REPGCGCCSVCARLEG**EACGVYTPRC**  
**GQGLRCYPHPGSELPLQALVMGEGTCE**KRRDAEY**G**  
 ASPEQVADNGDDHSEGLVEN**NHVDSTMNMLGGGG**  
**SAGRKPLKSGMKELAVFREK**VTEQHRQM**GKGGKHH**  
**LGLEPKKLRPPP**ARTPCQQELDQVLER**ISTMRLPDE**  
**RGP**LEHLYSLHIPNCDKHGLYN**LKQCKMSLNPNTCKL**  
**IQCAP**TIRGD**PECHLFYNEQQE**ARGVHTQ**RMQ**

## C terminus

# IND 14906: pUMVC3-hIGFBP-2 (1-163)



# Immunogenicity and anti-tumor efficacy of hIGFBP-2\*\* N-terminus vaccine



Cecil et al, 2011

\*\*IGFBP-2 highly homologous mouse/human: immunity is species cross reactive

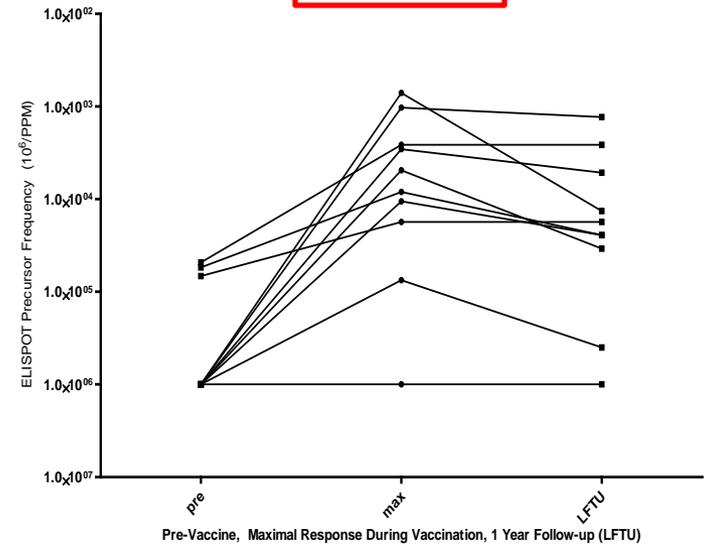
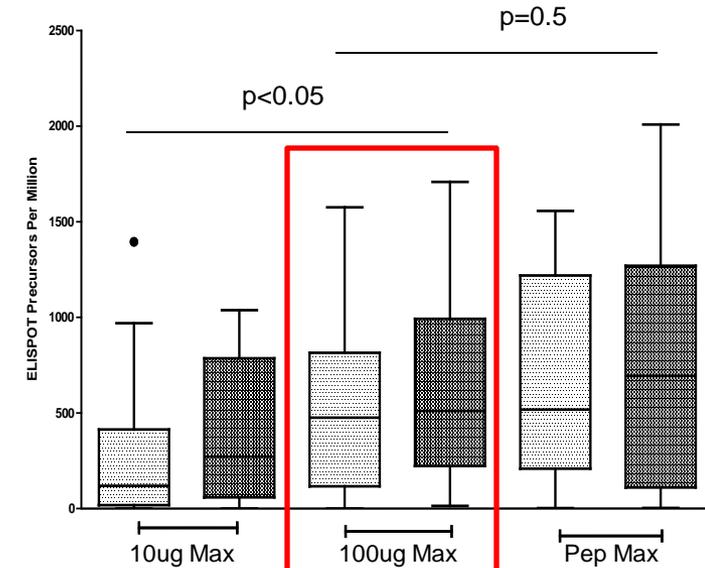
# pUMVC3-hIGFBP-2 (1-163) tumor and toxicity studies

- 45 MMTV-neu mice were immunized with pUMVC3-hIGFBP2 to assess immunogenicity and tumor protection
  - No clinical signs of toxicity
  - Average decrease in tumor growth 75% (n=15)
  - Every mouse had anti-tumor response
  - None demonstrated tumor growth acceleration
  - Immunogenic, mIGFBP-2 compared to HIV,  $p=0.026$  (n=10)
- 20 FVB mice were immunized with pUMVC3-hIGFBP2 for acute and chronic toxicity studies
  - Compared to Vector alone, Adjuvant alone, PBS
  - Complete blood count, electrolytes, and glucose WNL in all groups
  - Renal and liver function WNL in all groups
  - No evidence of autoimmunity in any group by pathologic survey of organs
  - No evidence of malignancy at vaccine site

# **Rationale for clinical trial design**

# Previous experience with plasmid DNA vaccine targeting the ICD of HER2

Trial Design
Phase I dose study, HER2 ICD plasmid DNA (10, 100, 500 ug)
66 patients
Stage III, IV HER2+ breast cancer
CR, SD (bone only)
Concurrent trastuzumab allowed
3 vaccines, id, 1 month apart
GM-CSF as adjuvant (100ug)



80% with persistence

# Grade I/II toxicity (99%) after HER2 ICD plasmid based DNA immunization

## Minimal systemic toxicity

Adverse Events Experienced by Study Subjects (n = 876)							
Event Grade	Arm 1 (n = 336)		Arm 2 (n = 275)		Arm 3 (n = 265)		Possibly, Probably, or Definitely Related (%)
	No. of events	%	No. of events	%	No. of events	%	
1	289	86%	234	85%	219	83%	91%
2	46	13%	40	14%	44	16%	8%
3	0	0%	1	4%	2	<1%	<1%
4	1	<1%	0	0%	0	0.00%	<1%

4 Grade 3-4 toxicities unrelated to vaccine: vomiting (virus), dyspnea (PE), appendectomy, syncope

## No lasting serologic conversion events

Subjects with an Abnormal Autoimmune Assay Result								
Autoimmunity Marker	Combined (n = 66)		Arm 1 (n = 22)		Arm 2 (n = 22)		Arm 3 (n = 22)	
	No. subjects	%	No. subjects	%	No. subjects	%	No. subjects	%
Anti Nuclear Antibodies	3	5%	1	5%	0	0%	2	9%
Anti Double-Str, DNA	0	0%	0	0%	0	0%	0	0%
Complement C3	5	6%	3	14%	1	5%	1	5%

All serologies low titer, clinically asymptomatic, and resolved by study end

# Clinical trial design and endpoints

Trial Design
Phase I study pUMVC3-hIGFBP-2 100 ug dose
22 patients
Stage III, IV ovarian cancer
NED
3 vaccines, id, 1 month apart
GM-CSF as adjuvant (100ug)

Phase II: Prevent relapse

## PRIMARY ENDPOINT

Determine **safety**

## SECONDARY ENDPOINTS

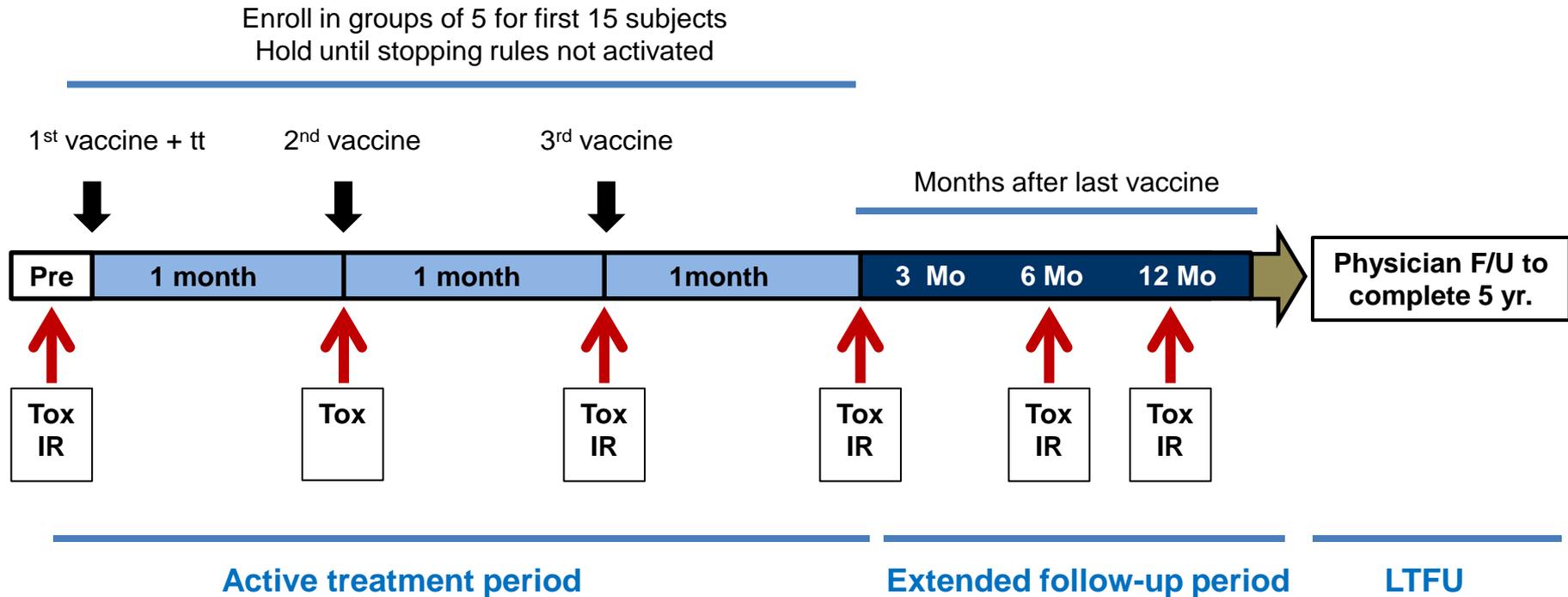
1. Determine the **immunogenicity** of the vaccine
2. Determine if intermolecular **epitope spreading** occurs
3. Determine if vaccination modulates **T regulatory cells**

## STATISTICAL DESIGN

If no toxicities occur, the probability of such an occurrence is at least 90% if the true toxicity rate is 10% or more

80% confidence that the estimated immune response rate is within at least .14 of the true immune response rate

# Current clinical trial schema\*



## Revisions to RAC submitted protocol:

- Describe study as 3 separate evaluation periods
- Increase and extend toxicity testing
- Replace up to 5 patients in active and extended periods to ensure adequate toxicity evaluation
- Collect data on remission duration and compare to historical control (SEER)

\* IND approved 12/4/2011, represents FDA suggested revisions

# Evaluation of toxicity

## Basal HER and IGFBP-2 in normal tissues

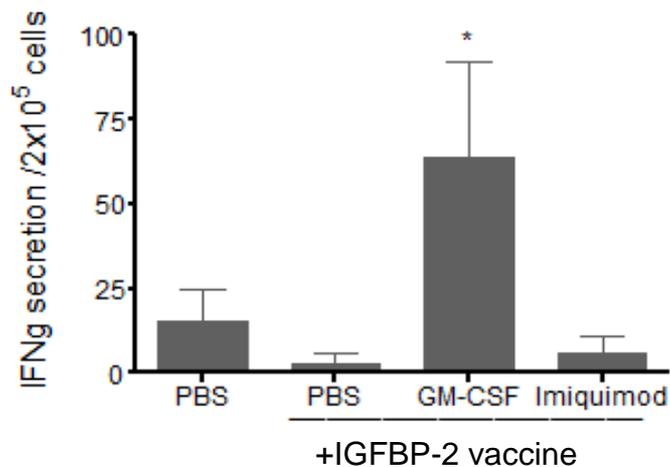
HER2	IGFBP-2
Gastrointestinal	Gastrointestinal
Respiratory	Respiratory
Reproductive	Reproductive
Urinary	Urinary
Skin	-----
Breast	-----
Cardiac	-----
Placenta	Placenta
-----	Thyroid gland
-----	Ocular

## APPROACH

- Physical examination/ROS by physician and collection of AEs
- CBC to assess hematologic toxicities
- BUN, Cr, electrolytes: renal
- Glucose: endocrine
- LFTs: gastrointestinal
- Thyroid function tests: thyroid
- Autoimmune panel: generalized autoimmunity
- CA-125: inflammation/disease progression
- Request for medical records yearly during LTFU

# Evaluation of immunogenicity

## GM-CSF as an adjuvant



## Evaluation of IGFBP-2 specific immunity:

- IFN-gamma ELISPOT for IGFBP-2 peptides and protein (CD4<sup>+</sup>)
- Antibody levels and avidity

## Evaluation of ES:

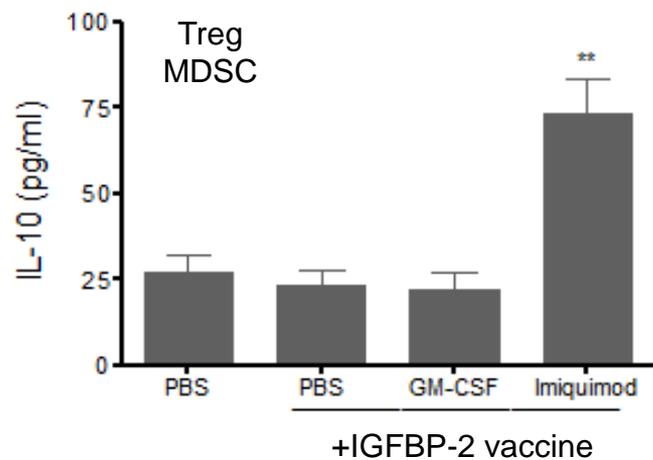
- IFN-gamma ELISPOT for p53, IGF-IR, topoisomerase IIa

## Modulate self regulatory elements

- Changes in Treg levels over time

## T cell persistence

- T cell kinetics through active treatment and extended follow-up
- T cell memory populations by flow cytometry

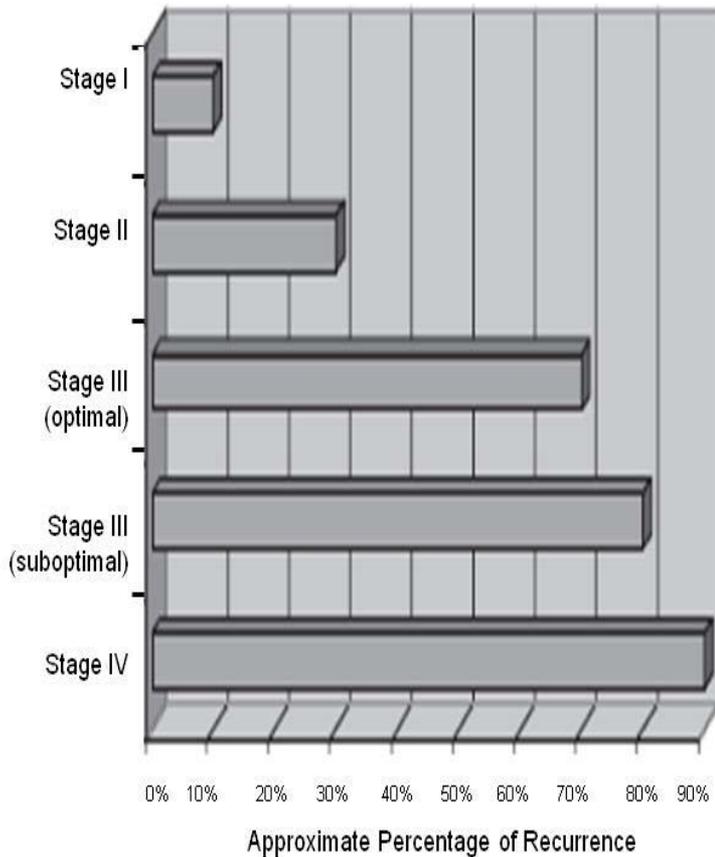


\*p<0.05

**A Phase I Trial of the Safety and Immunogenicity of a DNA Plasmid Based Vaccine Encoding the Amino Acids 1-163 of Insulin-Like Growth Factor Binding Protein-2 (IGFBP-2) in Patients with Advanced Ovarian Cancer**

# Patient population to prevent disease recurrence

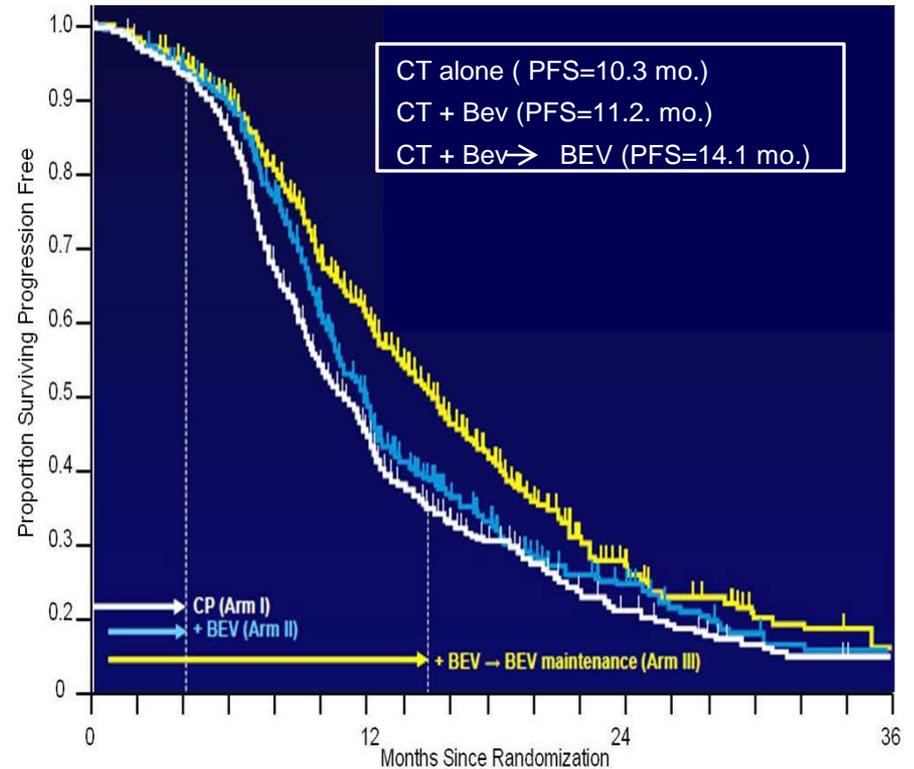
Risk of recurrence by stage at dx



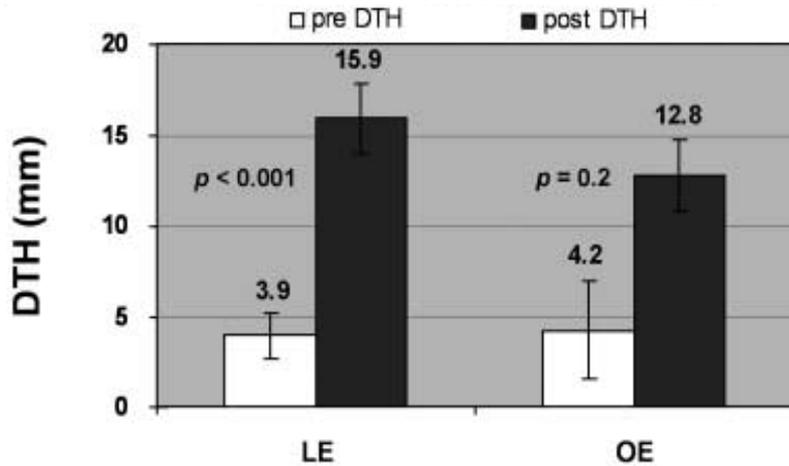
## GOG Randomized Phase III trial in Stage III/IV Ovarian Cancer

1873 patients randomized to 3 arms

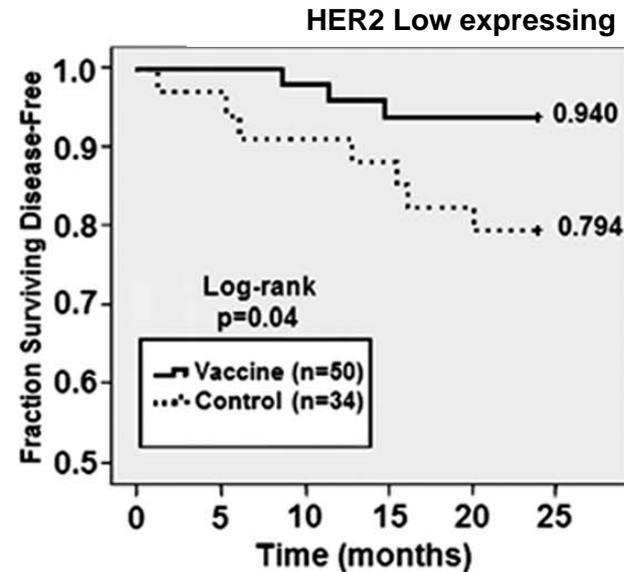
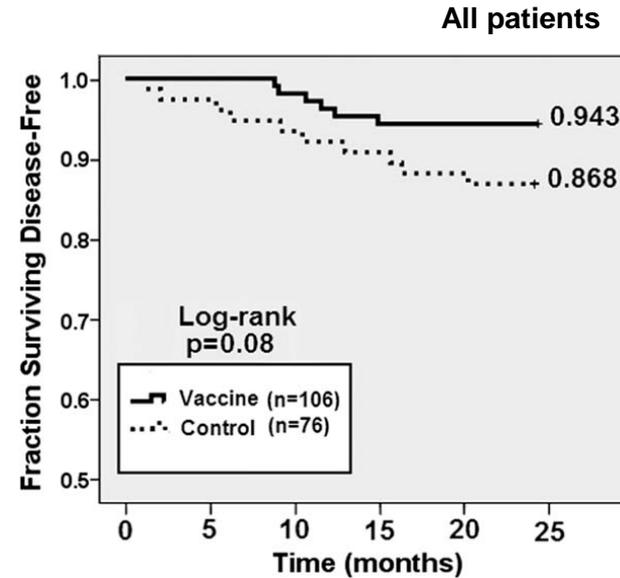
- (1) CT (IV paclitaxel + carboplatin) (n=625; 472 Stage III)
- (2) CT + concurrent BEV (n=625; 461 Stage III)
- (3) CT + concurrent BEV + maintenance BEV (n=623; 458 Stage III)



# Immunity, response at all levels of protein overexpression after HER2 vaccination



Benavides et al, Clin Ca Res, 2009



Mittendorf et al, Cancer 2011

# Overexpressed oncoproteins are effective vaccine targets

Antigens Associated with Clinical Response

Foreign Antigens	Self Antigens	
LMP2	HER2	GD2
HPV	WT1	CEA
HepB	MUC1	MART-1
	MAGE A2	gp100
	NY-ESO-1	PR1
	PSMA	Tyrosinase
	PSA	PAP
	PSCA	NA17

Cheever et al, Clin Ca Res, 2009

## Examples of vaccines targeting self antigens:

- **p53:** 54 mixed tumor types  
50% immune response  
All Grade 1-2 toxicities  
+Clinical response
- **EGFR:** 80 Stage III/IV NSCLCa  
51% immune response  
All Grade 1-2 toxicities  
+Clinical response
- **Telomerase:** 25 NSCLCa  
46% immune response  
All Grade 1-2 toxicities
- **HER2:** 192 non-metastatic breast Ca  
48% immune response  
Grade 1-2 toxicities  
Angioedema of tongue (1 Gr. 3)  
+ Clinical response