

RAC # 1007-1050

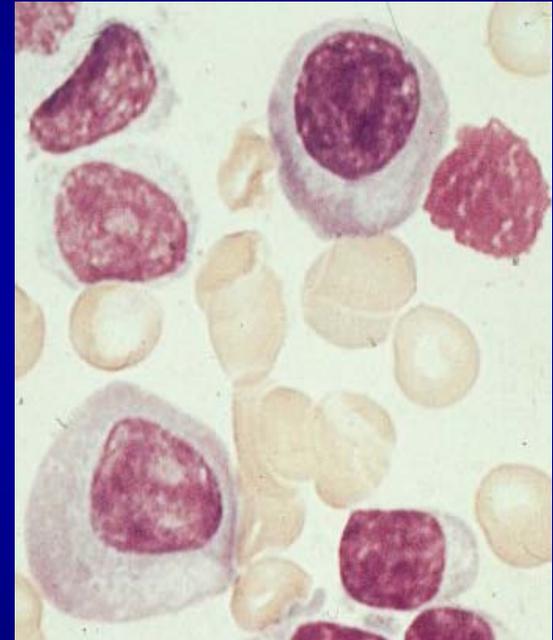
Phase I Study of an Active
Immunotherapy for Asymptomatic
Phase Lymphoplasmacytic
Lymphoma with DNA Vaccines
Encoding Antigen-Chemokine Fusion

Sheeba Thomas, M.D. Principal Investigator

Larry Kwak, M.D., Ph.D. Co-Principal Investigator

Lymphoplasmacytic Lymphoma

- Low grade B-cell lymphoproliferative disorder
 - Bone marrow infiltration with lymphoplasmacytic cells
 - Monoclonal gammopathy
- Median age at diagnosis: 63 years
- Median overall survival from start of Rx: 4-10 years
- Indications for treatment
 - Hemoglobin level < 10 g/dL
 - Platelet count < $100 \times 10^9/L$
 - Bulky adenopathy/organomegaly
 - Symptomatic hyperviscosity
 - Severe neuropathy
 - Amyloidosis
 - Cryoglobulinemia
 - Cold agglutinin disease
 - Evidence of disease transformation



Lymphoplasmacytic Lymphoma

- Current recommendations:
 - No survival advantage to starting treatment early
 - Standard of care is to follow asymptomatic patients on a program of observation.
- When treatment is required:
 - Overall response rates have been high
 - Complete responses are infrequent
 - Eventual relapse from disease is inevitable
 - Disease remains incurable

Objective of Vaccine

- Lengthen interval before chemotherapy is required to maintain disease control.
 - Improve quality and quantity of patients' lives
- Well tolerated
- Without inducing cross resistance to available agents

Protocol Objectives

- **Primary**
 - Safety and feasibility of vaccine
 - Maximum tolerated dose (MTD)

- **Secondary**
 - Assess immunogenicity of vaccine to generate humoral and cellular immune responses

Study Population

- Age \geq 18 years
- Asymptomatic phase LPL with surface IgG, IgA or IgM phenotype.
- Previously untreated
- ECOG performance status of 0 or 1.
- Able to provide at least 5 million CD20 and/or CD38+ cells by Bone Marrow aspirate or else excisional Lymph Node biopsy

Treatment Schema

- Formulation and Administration:
 - 0.5ml intramuscular injection rotated between thighs
- Dosing Cohorts:
 - Cohort 1: 500 μg
 - Cohort 2: 2500 μg
- Schedule of Administration:



Statistical Design

- Standard 3+3 Design
- Maximum Sample Size = 12 patients
- DLT: any grade 3 or 4 toxicity except for fever
 - evaluated for DLT at the end of first cycle (4 weeks)
 - grade 4 fever requires 50% dose reduction
- MTD: Highest dose level in which 6 patients have been treated with <2 DLT

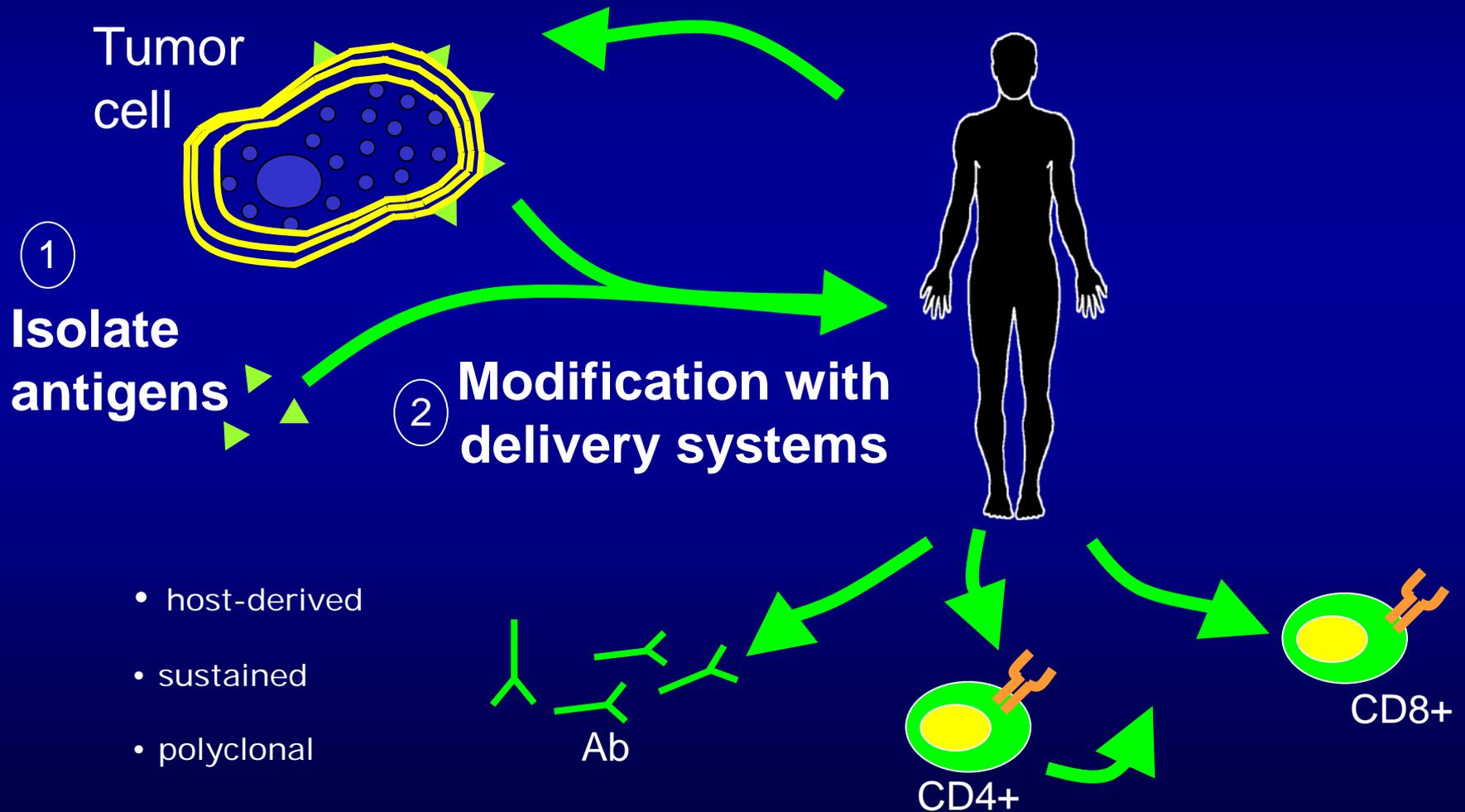
Statistical Design

- Cellular immune response
 - $\geq 3x$ rise in precursor frequency of tumor-reactive T cells at 12 weeks
- Humoral immune response
 - $\geq 4x$ rise in idiotypic specific antibody

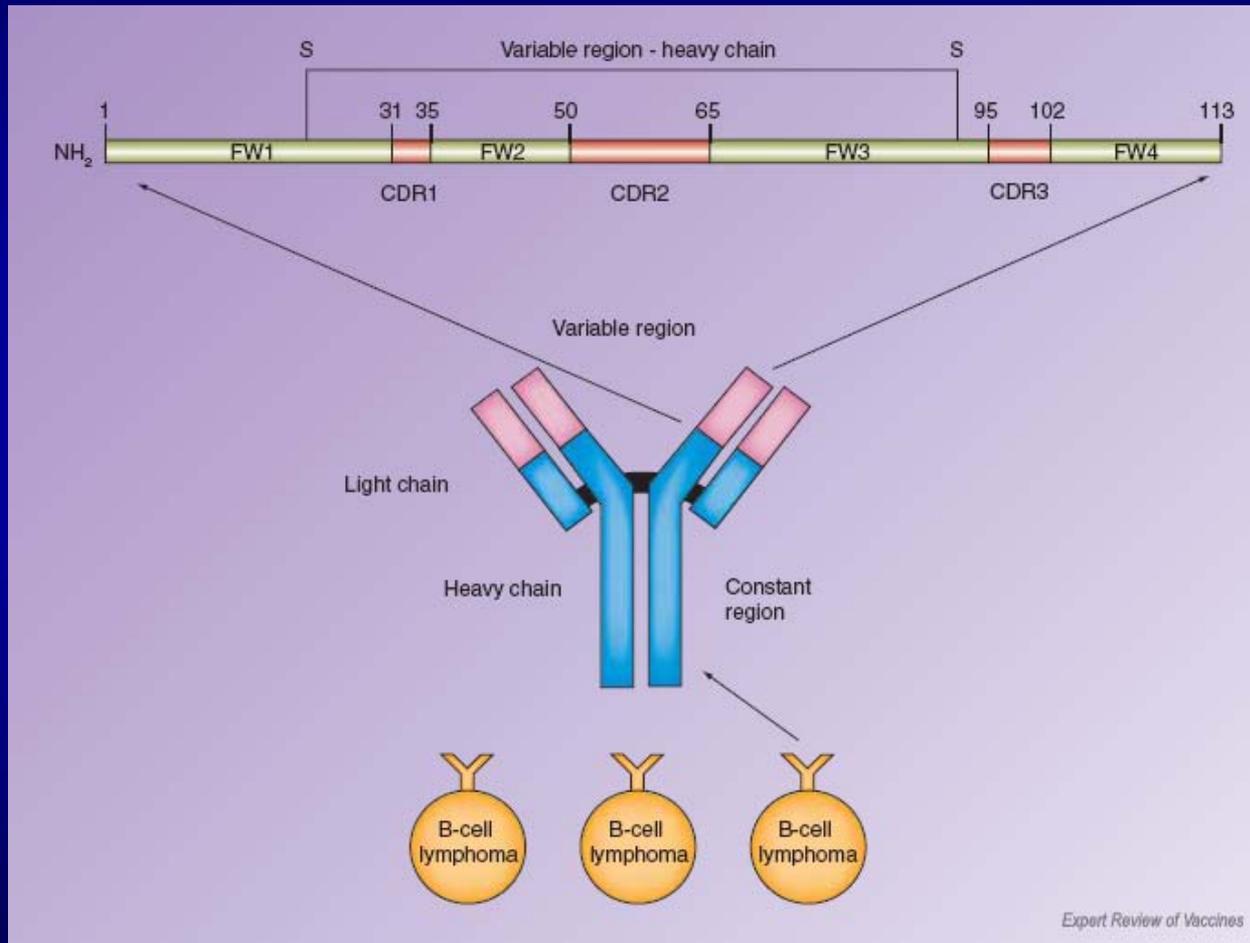
Management of Potential Toxicity

- Anticipated reactions
 - local erythema and induration injection site
 - transient flu-like symptoms
- Management
 - Acetaminophen , meperidine and antihistamines may be used
 - Steroids and NSAIDs are to be avoided
 - May suppress the immune response induced by the vaccine
 - Steroids may also reduce burden of lymphoma

Cancer Vaccine Strategies



Idiotypic as a Tumor-Specific Antigen for B-Cell Lymphoma



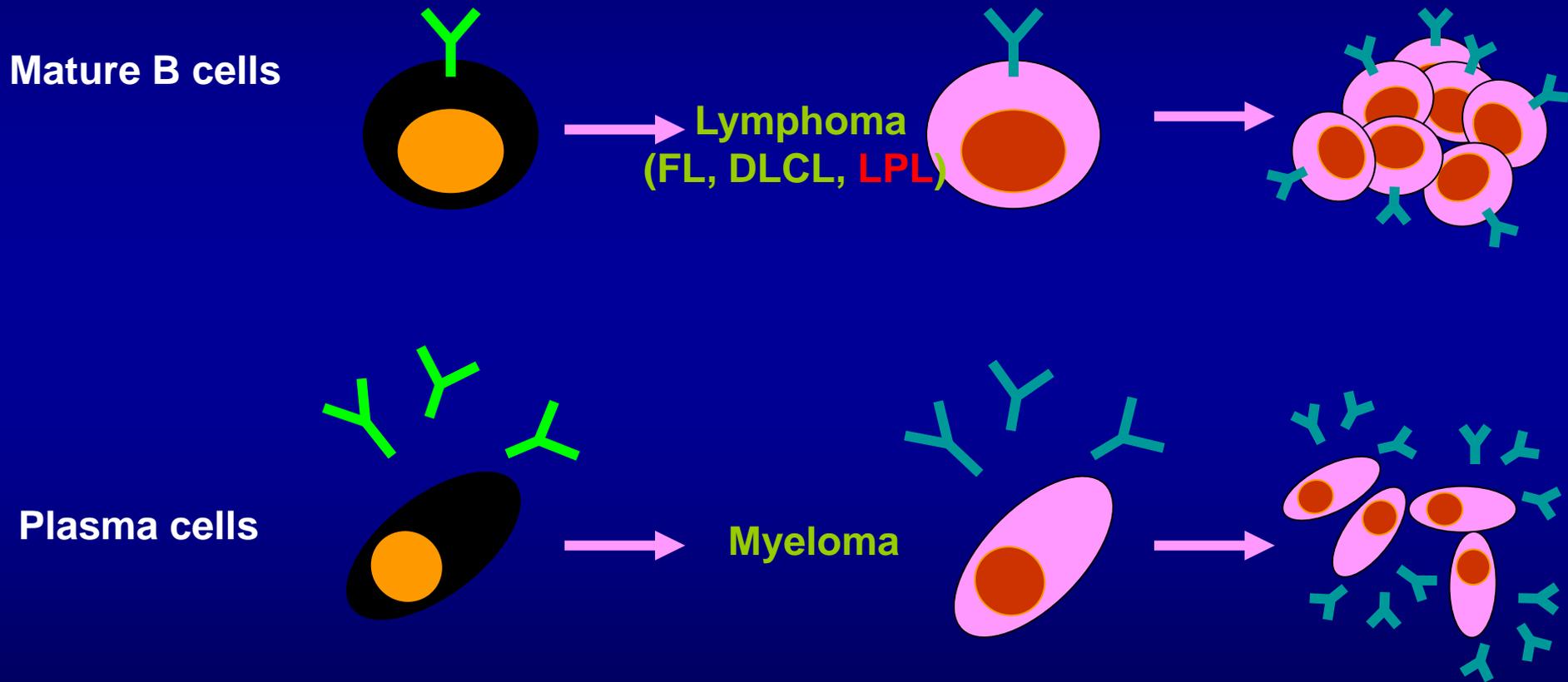
Reproduced with permission from Neelapu et al. *Expert Rev Vaccines*. 2006;5:381-394.

Kwak et al. *N Engl J Med*. 1992;327:1209-1215.

Stevenson et al. *Fed Proc*. 1977;36:2268-2271.

NIH-OBA 1007-1050

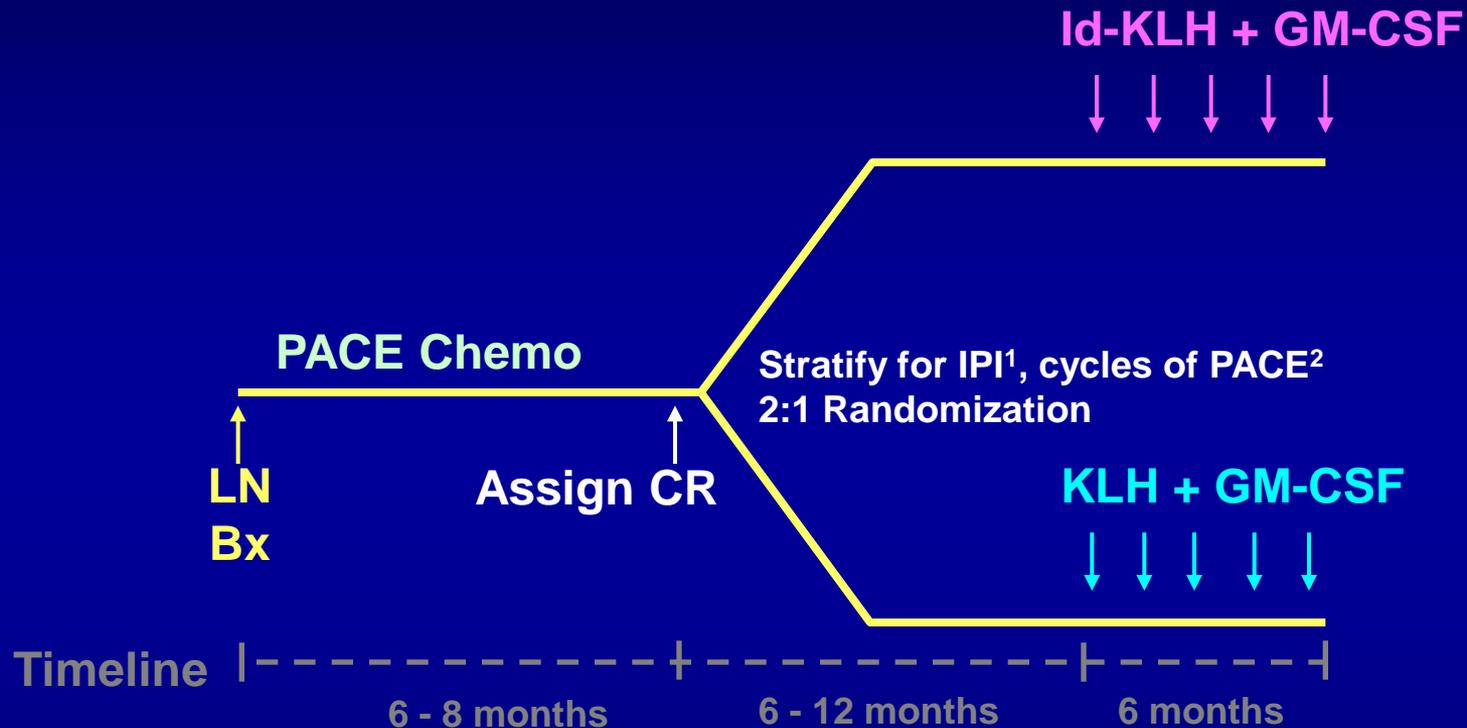
Idiotype (Id): A clonal marker and model tumor antigen



Id Vaccination: Early Phase Clinical Trials

<u>Publication</u>	<u>Vaccine</u>	<u>No. Patients</u>	<u>Histology</u>	<u>Phase I</u>	<u>Phase II</u>	<u>Phase III</u>
Kwak, NEJM 1992 Hsu, Blood 1997	Id-KLH + Adjuvant	41	FL	Phase I		
Bendandi, Nat Med 1999	Id-KLH + GMCSF	20	FL	Phase I	Phase II	
Timmerman, Blood 2002	Id - DC	35	FL	Phase I		
Timmerman, Clin Can Res 2002	Plasmid DNA	12	FL	Phase I		
Barrios, Hematologica 2002	Id-KLH + adjuvant	9	FL	Phase I		
Neelapu, Nat Med 2005	Id – KLH + GMCSF	26	MCL	Phase I	Phase II	
Inoges, JNCI 2006	Id – KLH + GMCSF	25	FL	Phase I		
Bertinetti, Can Res 2006	Fab + MF59 + GMCSF	18	various	Phase I		
Redfern, JCO 2006	Id – KLH + GMCSF	31	indolent	Phase I		

NCI/Biovest Phase III Vaccine Study Design



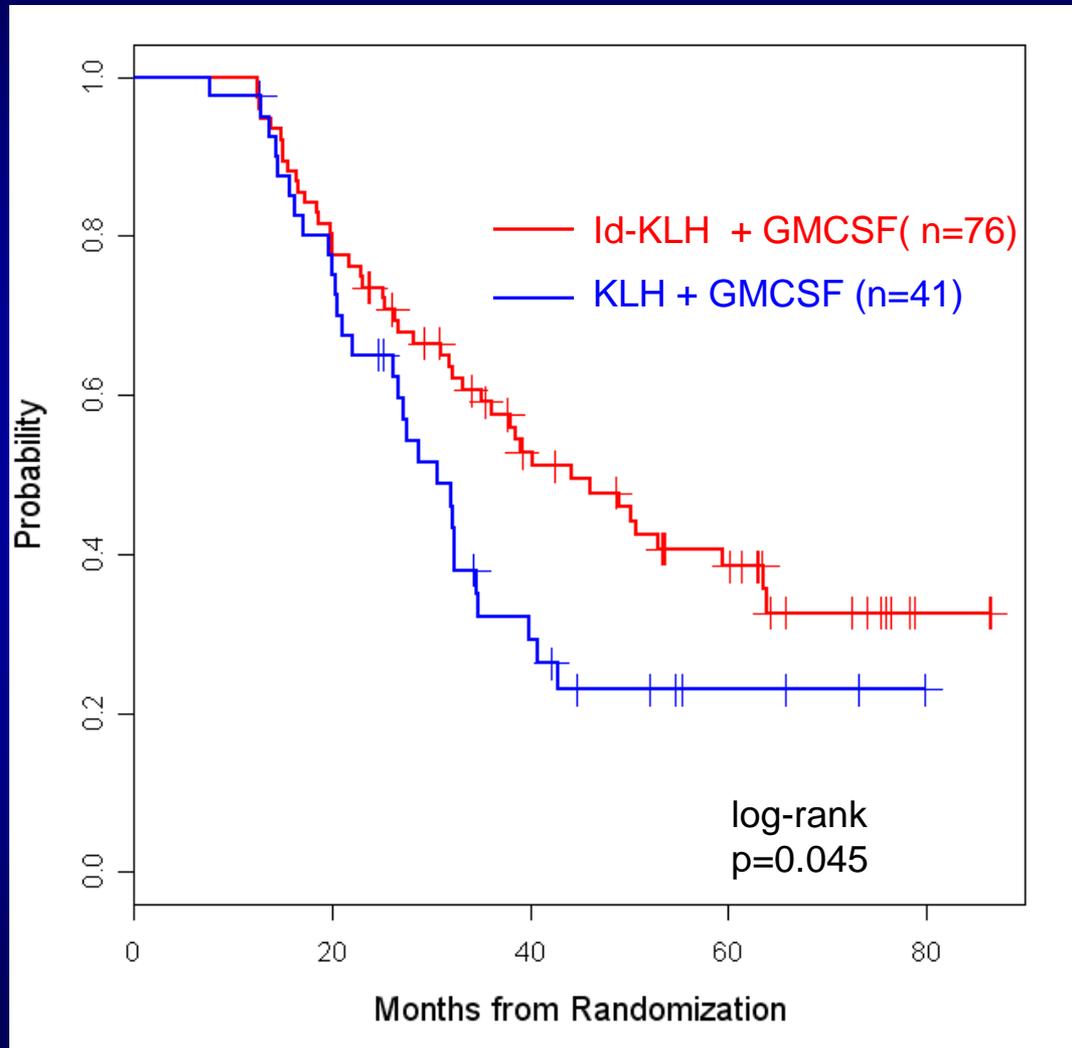
- Primary endpoint: disease-free survival
- 14 sites enrolled patients from 2000-2007

¹low, low-intermediate or high-intermediate, high groups
² < 8 or ≥ 8 cycles

Statistical Design: Two Prospective Efficacy Analyses

- Intent-to-Treat Analysis (ITT) compared DFS in treatment arms for all randomized pts
- Modified Intent-to-Treat Analysis (mITT) compared DFS in treatment arms for randomized pts who remained in CR/CRu and received either Id-KLH or control vaccine

Disease Free Survival from Randomization (mITT)



Median Follow-up

56.6 mo (range 12.6 – 89.3)

Median DFS

Id-KLH = 44.2 mo

Control vaccine = 30.6 mo

Events

Id-KLH = 44

Control vaccine = 29

Cox PH Model

HR = 0.62; [95% CI: 0.39,0.99]
(p=0.047)

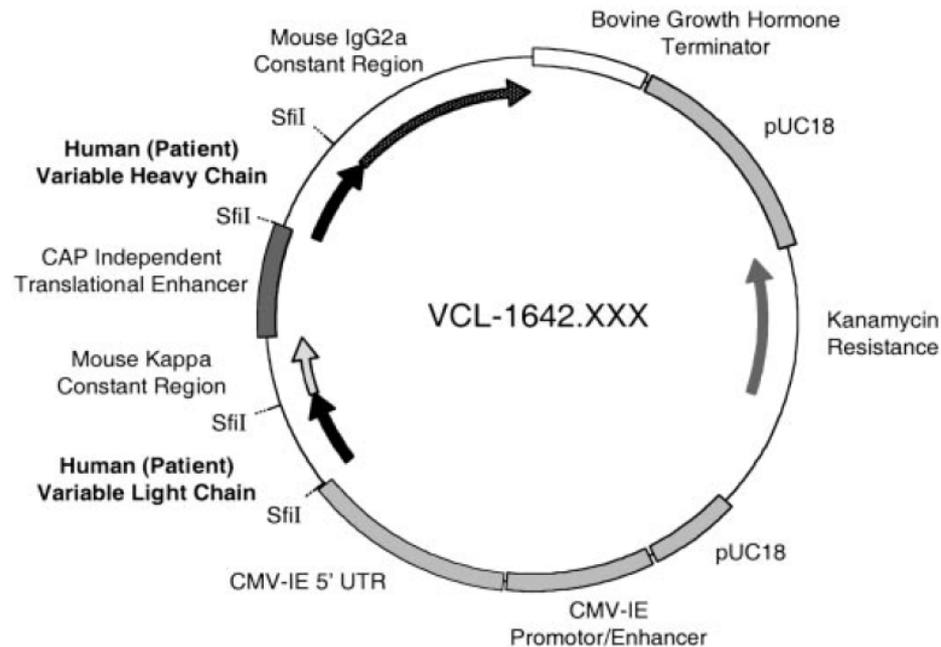
Safety summary: Local Injection Site Reactions

Maximum Local Reaction	Severity	KLH + GMCSF (%)	Id-KLH + GMCSF (%)
Erythema	No reaction	12%	3%
	< 1 cm	0%	0
	1-10 cm	39%	51%
	> 10 cm	49%	46%
Induration	No reaction	24%	14%
	< 1 cm	2%	4%
	1-10 cm	46%	62%
	> 10 cm	27%	20%
Ulceration	No reaction	98%	93%
	< 1 cm	2%	3%
	1-10 cm	0%	3%
	> 10 cm	0%	1%

Immunogenicity of a Plasmid DNA Vaccine Encoding Chimeric Idiotype in Patients with B-Cell Lymphoma¹

John M. Timmerman,² Gita Singh, Gary Hermanson, Peter Hobart, Debra K. Czerwinski, Behnaz Taidi, Ranjani Rajapaksa, Clemens B. Caspar, Adrienne van Beckhoven, and Ronald Levy³

Division of Oncology, Department of Medicine, Stanford University School of Medicine, Stanford, California 94305 [J. M. T., D. K. C., B. T., R. R., C. B. C., A. v. B., R. L.], and Vical Corporation, San Diego, California 92121 [G. S., G. H., P. H.]



(VL-mouse kappa constant region) – (VH-mouse IgG2a constant region)

Study Design

Tumor biopsy, Chemotherapy,
Tumor Id DNA cloning and GMP production



Series #1 vaccinations

Dose escalation Id DNA (200, 600, or 1800 μg)

Needle & syringe, i.m.

Monthly x 3



17 months

Series # 2 vaccinations

1800 μg Id DNA

Biojector; 80% i.m., 20% i.d.

Monthly x 3



14 months

Series # 3 vaccinations

1800 μg Id DNA + 500 μg GM-CSF DNA

Biojector; 80% i.m., 20% i.d.

Monthly x 3

4 patients per cohort

(12 patients total)

Safety of Idiotype DNA vaccines

DNA vaccines at all three doses were well-tolerated

No acute or long-term toxicity was found in over 4 years of follow-up

Modest, transient elevation of serum rheumatoid factor in 5 patients, however, no patient developed clinical rheumatologic manifestations.

Immunogenicity of idiotype DNA vaccines

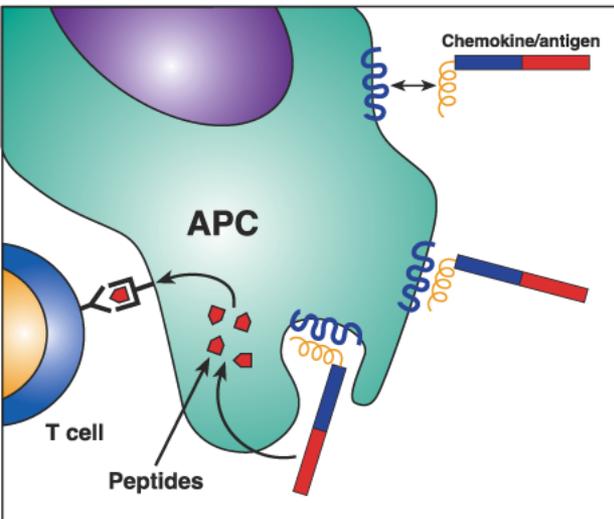
Table 2 *Summary of immune responses in vaccination series 1–3*

	Antibody responders		T-cell responders		Total responders anti-MsIg ^a
	MsIg	Id	MsIg	Id	
Series 1 Dose escalation	4/12	0/12	4/12	1/12 ^b	7/12
Series 2 1800 μ g Biojector 80% i.m., 20% i.d.	6/12	2ns/12	6/12	4ns/12 1/12 ^b	9/12
Series 3 As Series 2, plus GM-CSF DNA (500 μ g)	7/12	3ns/12	4/12	2ns/12	8/12

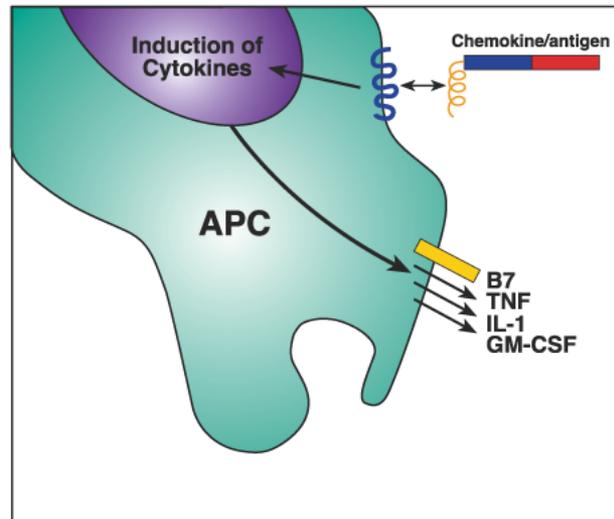
^a MsIg, mouse IgG2a/ κ ; Id, autologous tumor idiotype; ns, not specific for autologous Id.

^b Specific for autologous Id.

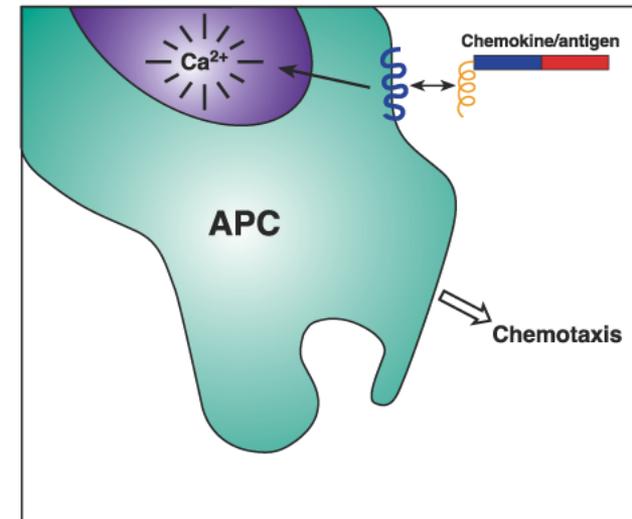
Possible Modes of Action of Chemokine-Ag Fusions In Vivo



APC Receptor Targeting

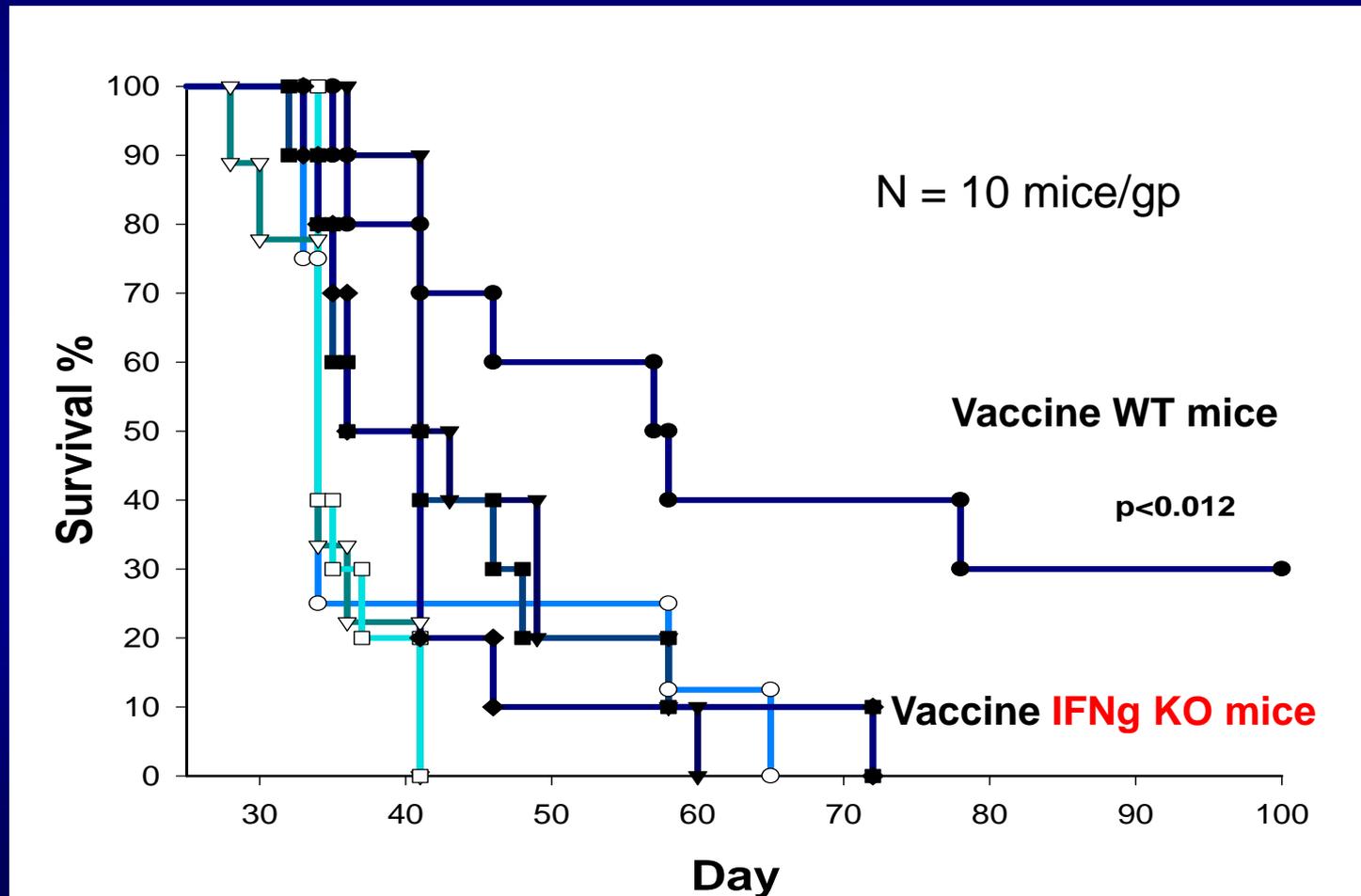


APC Maturation/Activation

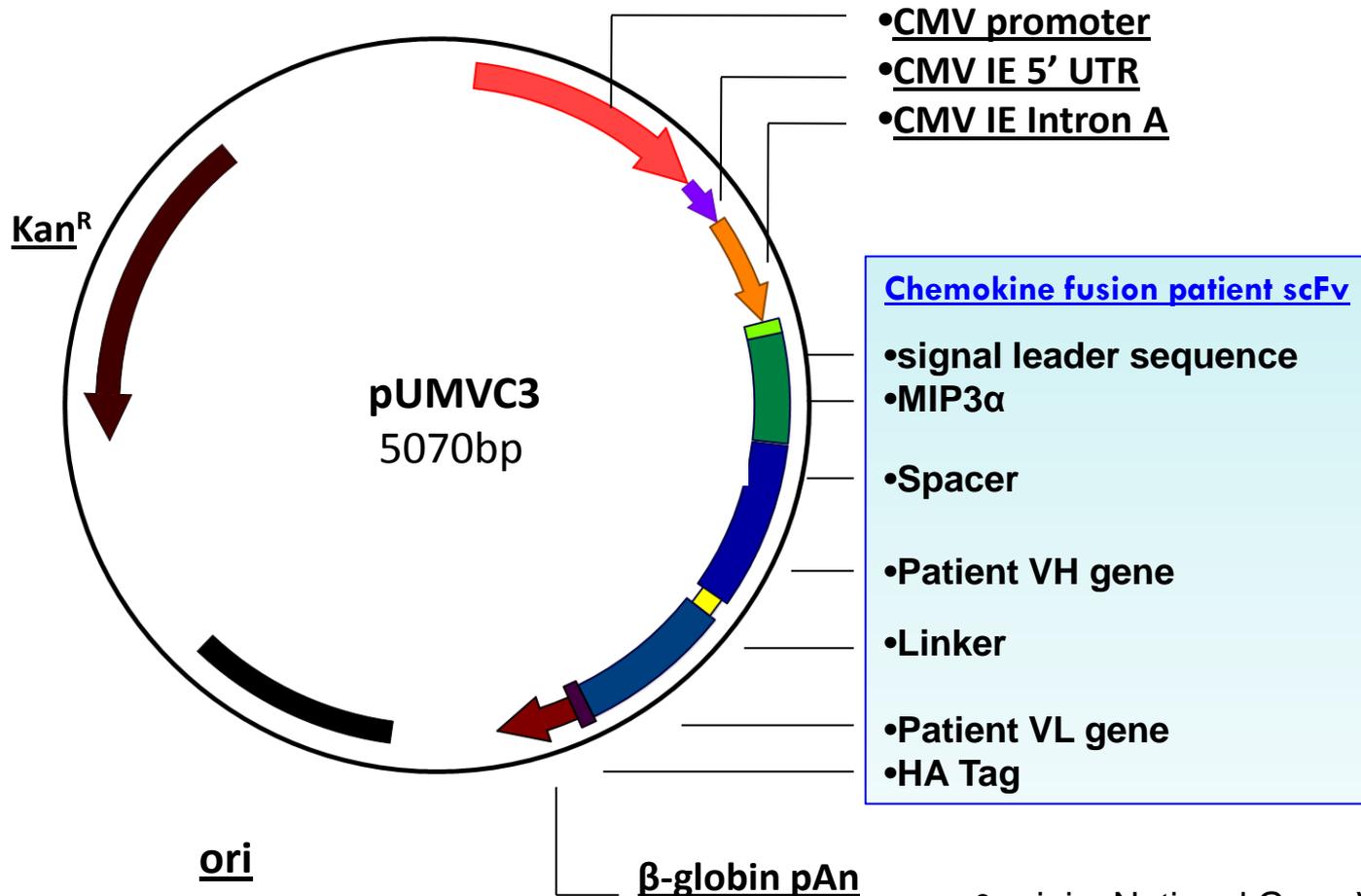


Chemotaxis of APC and Other Cells

Therapeutic anti-lymphoma immunity elicited by Idiotypic vaccines requires IFN-gamma.



Backbone DNA vaccine construct



- origin: National Gene Vector Laboratory (licensed to Aldevron)

- Daud et al JCO 26: 5896, 2008

Proposed release criteria: Filled, patient-specific plasmid DNA

Test	Method	Specification	Detection Limit
Appearance	Visual observation	Clear, colorless solution	
DNA Concentrations	Spectrophotometry A_{260}	0.40 ± 0.04 mg/mL 1.20 ± 0.12 mg/mL 3.60 ± 0.36 mg/mL	1 µg/mL
Pyrogen	USP Rabbit Pyrogen	Nonpyrogenic at a dose (per kg body weight) equivalent to the maximum human dose	
Sterility	USP Sterility	No growth through 14 days	No growth
Gene Expression	ELISA Assay	Plasmid expresses chemokine	
Electrophoretic migration of the expression product	Native polyacrylamide gel/ western blot	Ig approximately 150,000 MW	
	Denaturing polyacrylamide gel/ western blot	Heavy chain approximately 50,000 MW	
		Light chain approximately 25,000 MW	

Conclusions

- LPL is a low-grade lymphoma with an indolent clinical course, but with no curative treatment currently available
- Phase II/III trials showed that idiotype as a lymphoma antigen is immunogenic, and that protein vaccination was safe and improved disease-free survival following chemotherapy in patients in complete remission
- A previous Phase I lymphoma idiotype DNA vaccine trial demonstrated feasibility and safety