Phase Ia/Ib Trial of Anti-PSMA Designer T Cells in Advanced Prostate Cancer after Nonmyeloablative Conditioning

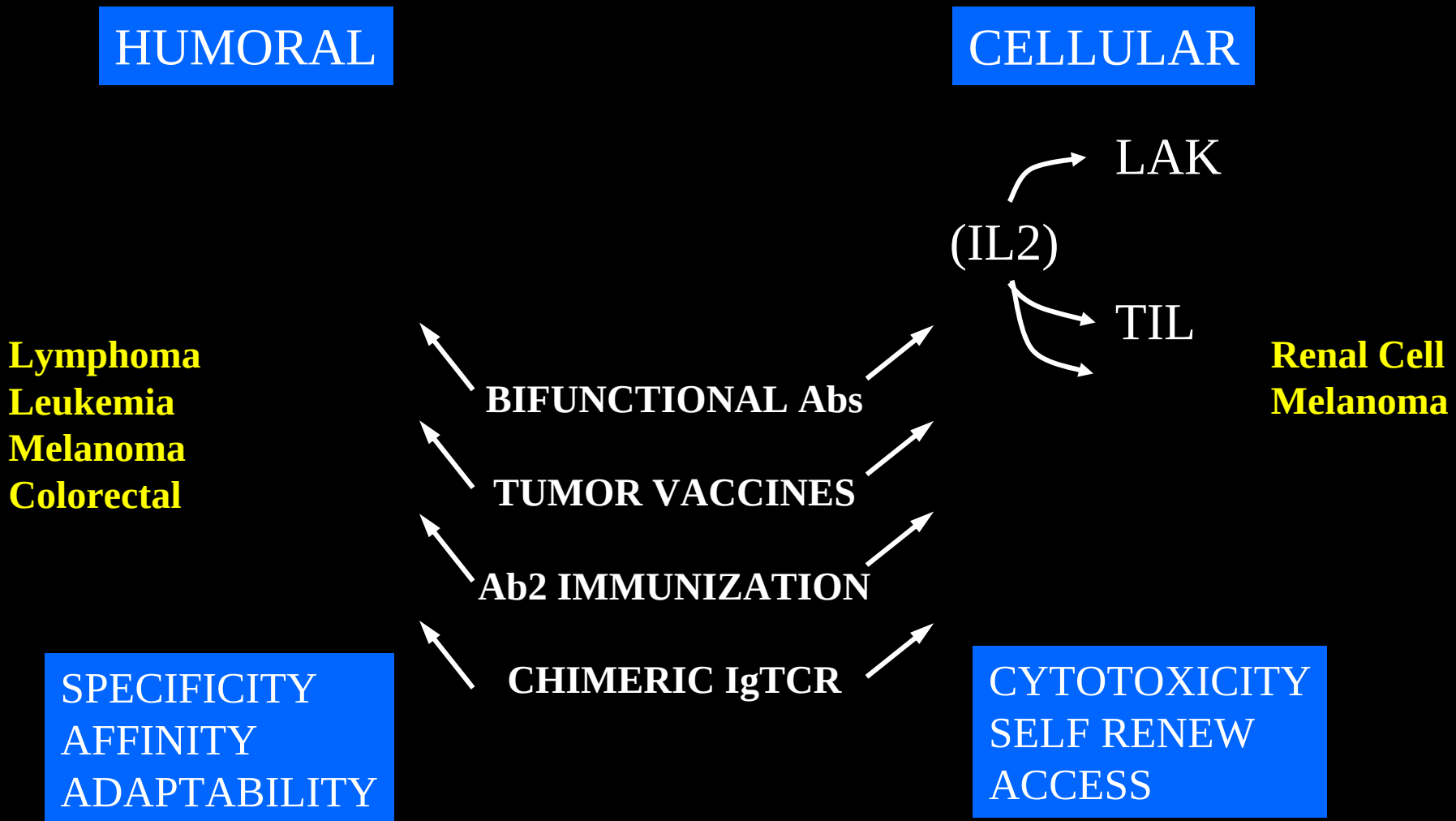
Richard P Junghans, PhD, MD

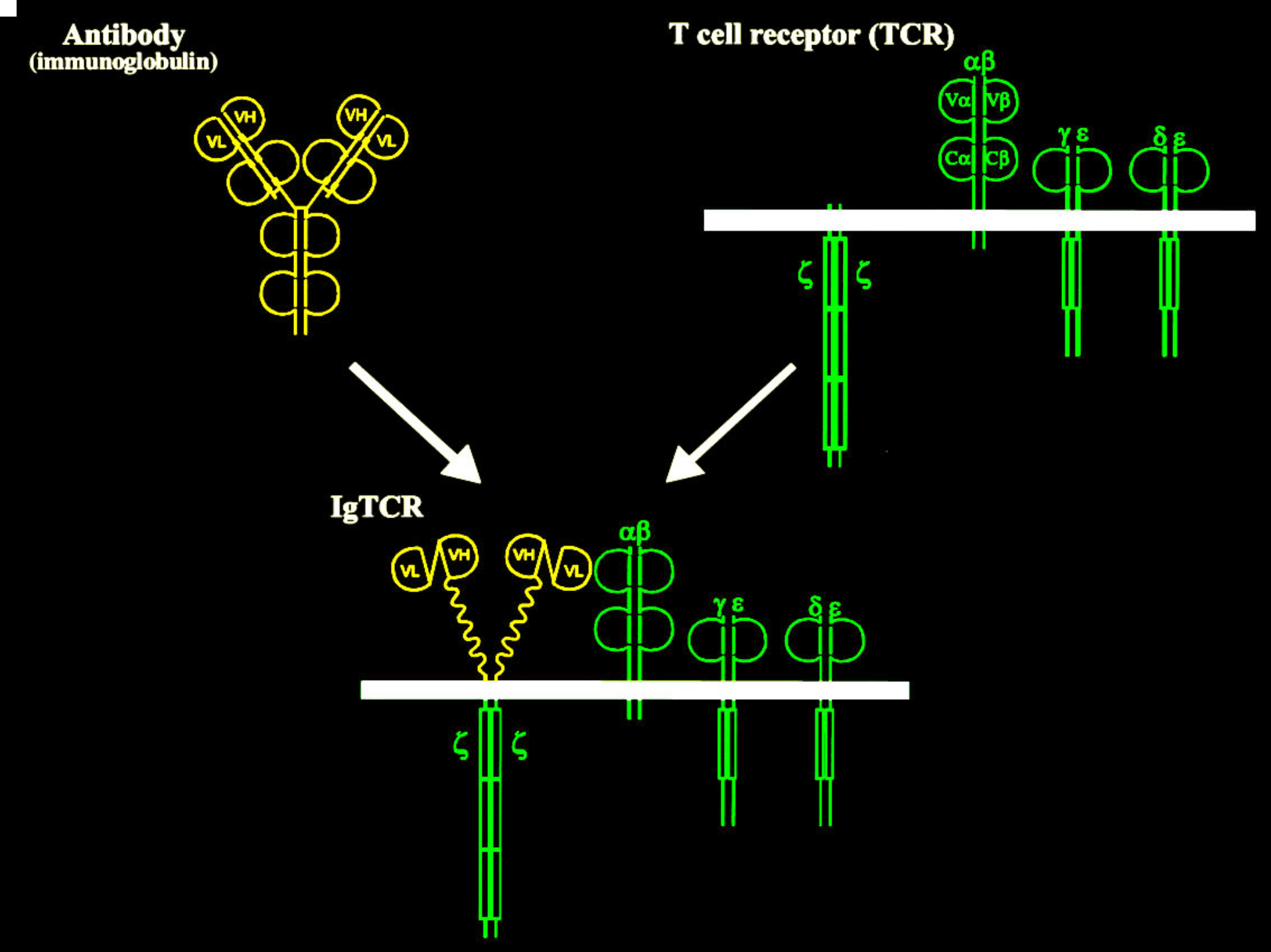
Director, Biotherapeutics Development Lab

Associate Professor of Surgery and Medicine
Boston University School of Medicine

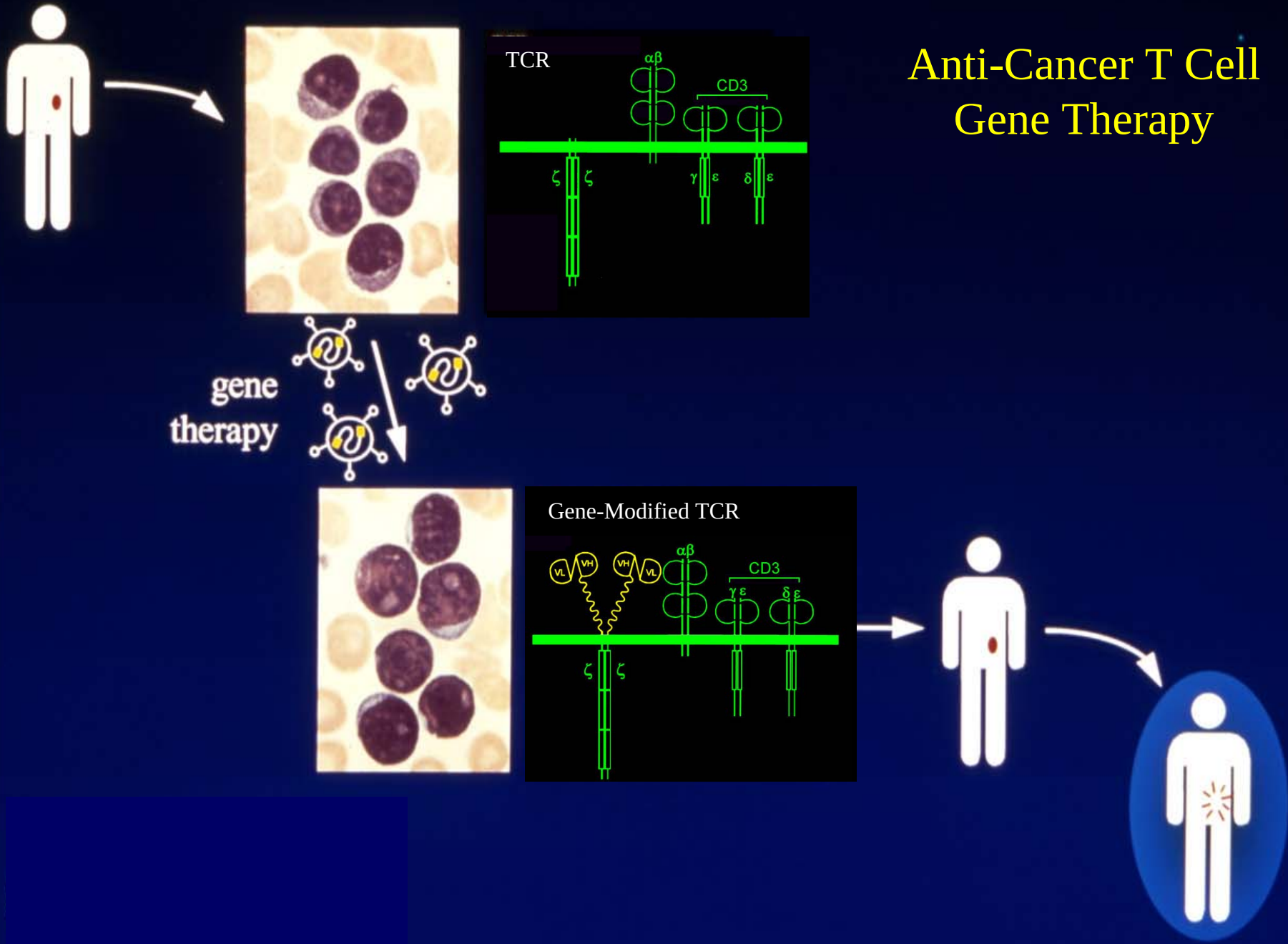
Chief, Division of Surgical Research
Roger Williams Medical Center
Providence, RI, USA

History of Biotherapies




Antibody
(immunoglobulin)
VH
VL
VH
VL
T cell receptor (TCR)
αβ
Vα
Vβ
Cα
Cβ
γ ε
δ ε
ζ ζ
IgTCR
VL
VH
VH
VL
αβ
γ ε
δ ε
ζ ζ

Anti-Cancer T Cell Gene Therapy



Prostate Specific Membrane Antigen (PSMA)

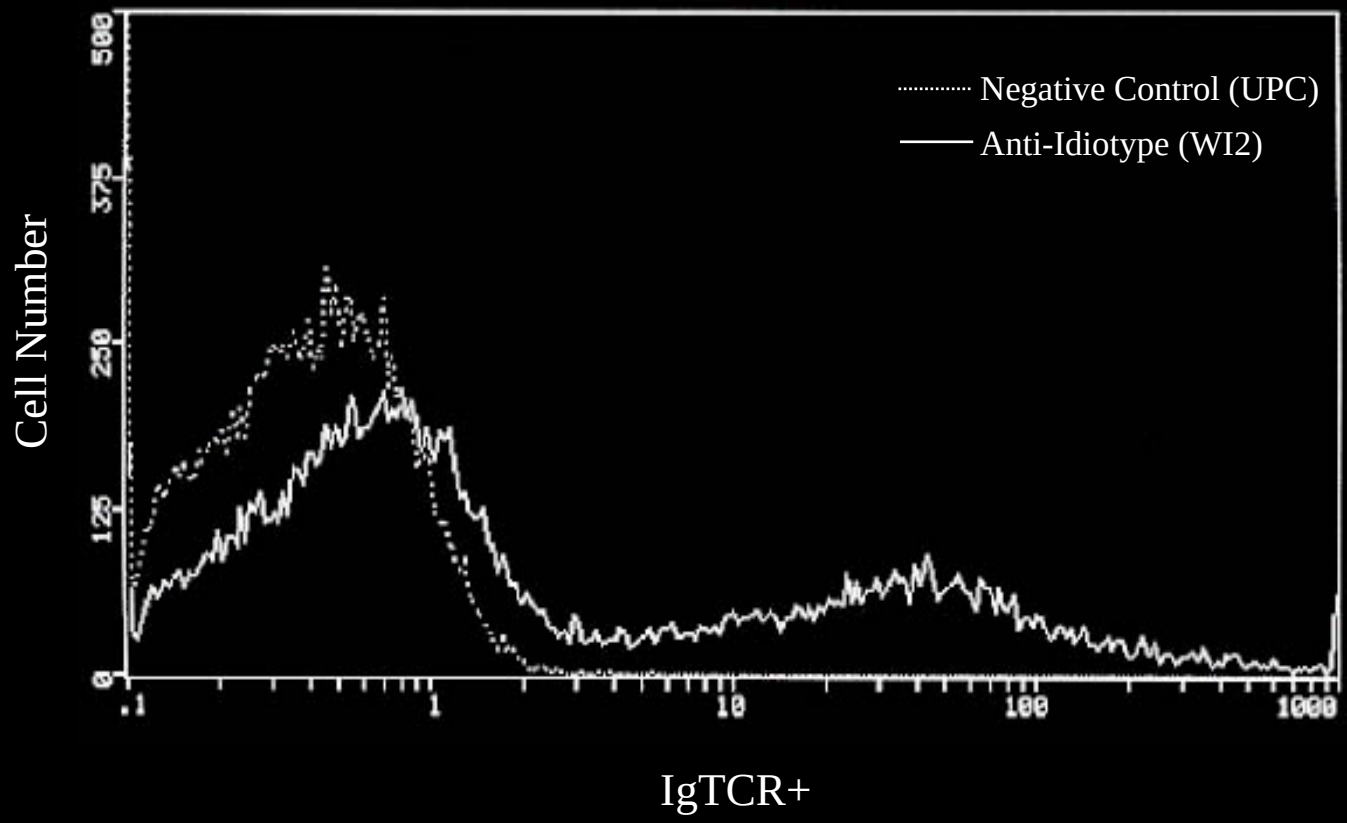
- Surface membrane glycoprotein 100,000 Daltons
- Normal prostate epithelium and prostatic vasculature
- Elevated expression in metastatic lesions and hormone refractory disease
- High clinical relevance:
  - 25,000 deaths per year from PSMA+ prostate tumors
- Antibody (3D8) from G. Murphy and A. Boynton

Clinical Retroviral Vector

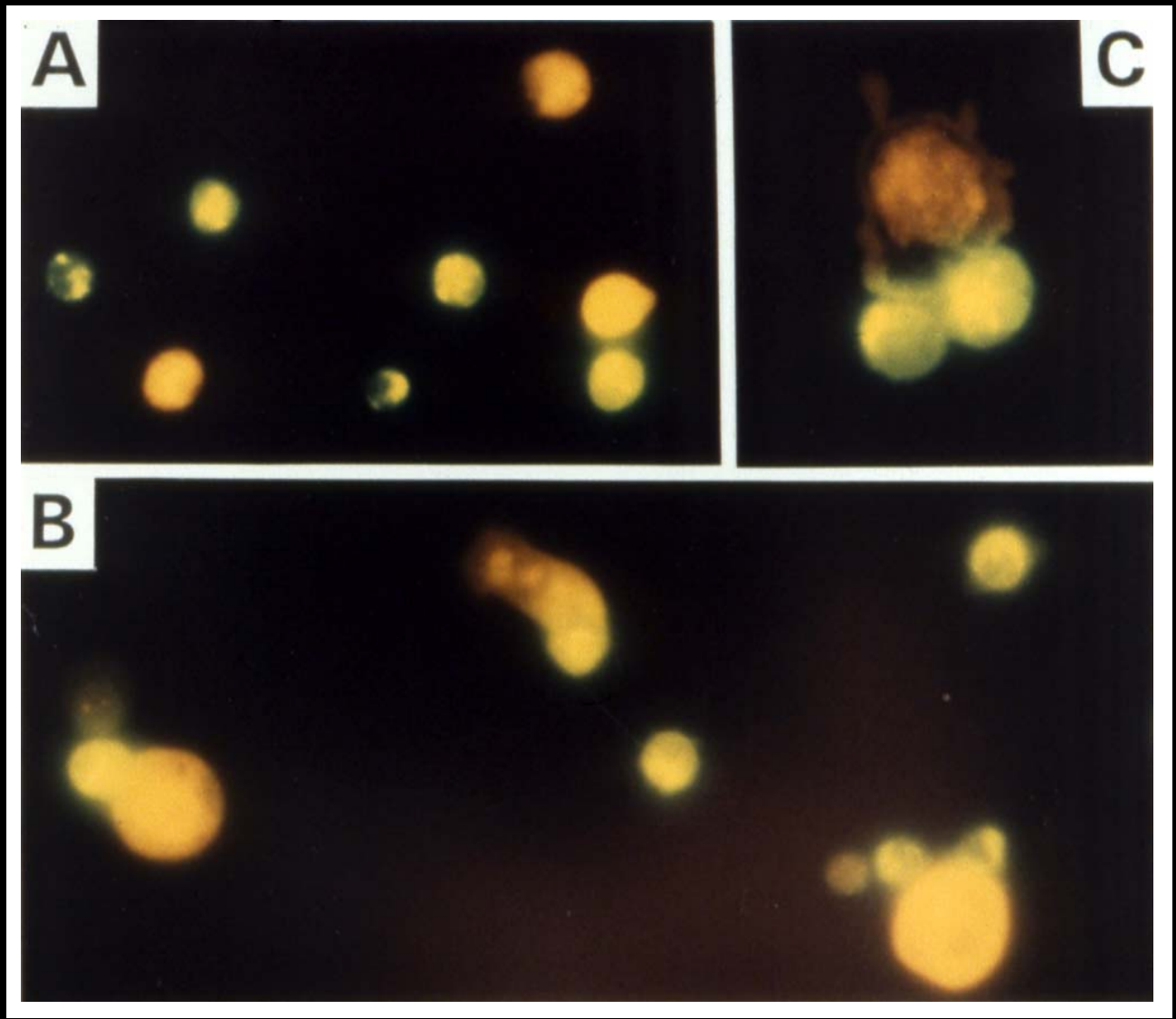
- Single gene sFv-CD8α hinge-TCRζ construct
- No prokaryotic selection marker
- No internal regulatory elements

Expression

DESIGNER T CELLS ARE EFFICIENTLY GENERATED AND EXPRESS HIGH LEVELS OF IgTCR

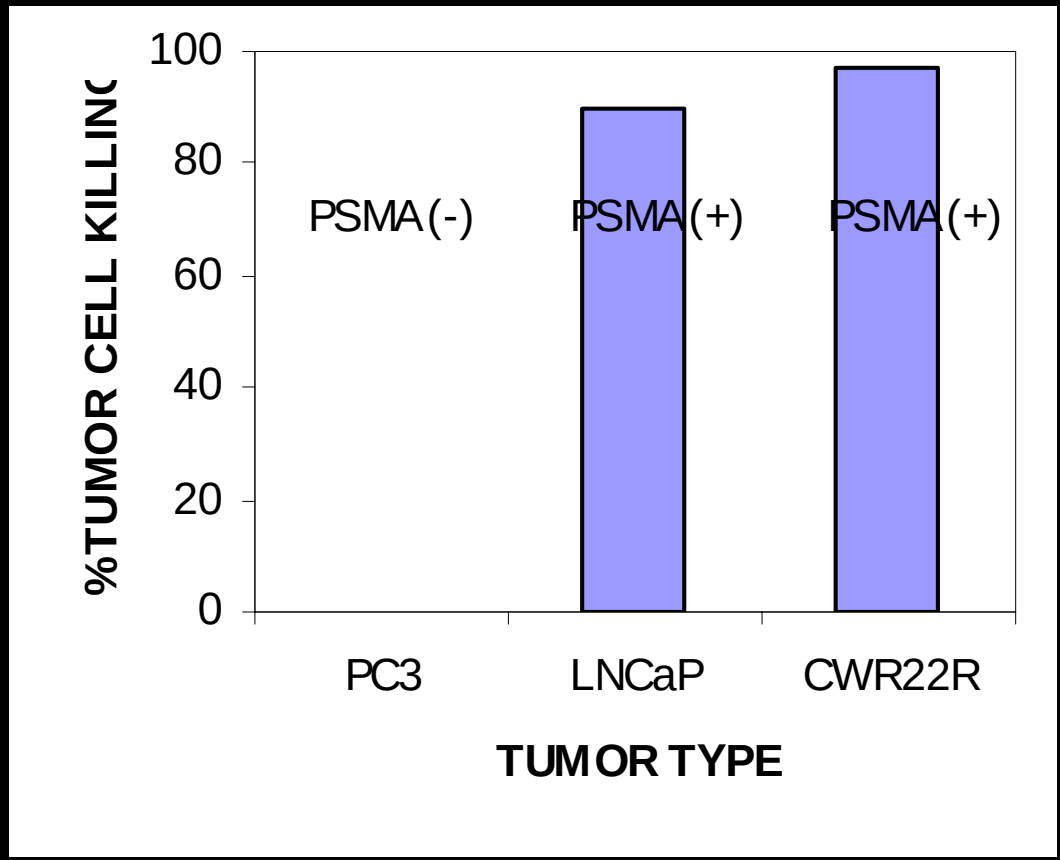


Tumor Cell Killing by T Cells



Activation: Cytotoxicity

ANTI-PSMA DESIGNER T CELLS KILL PSMA+ TUMOR CELLS



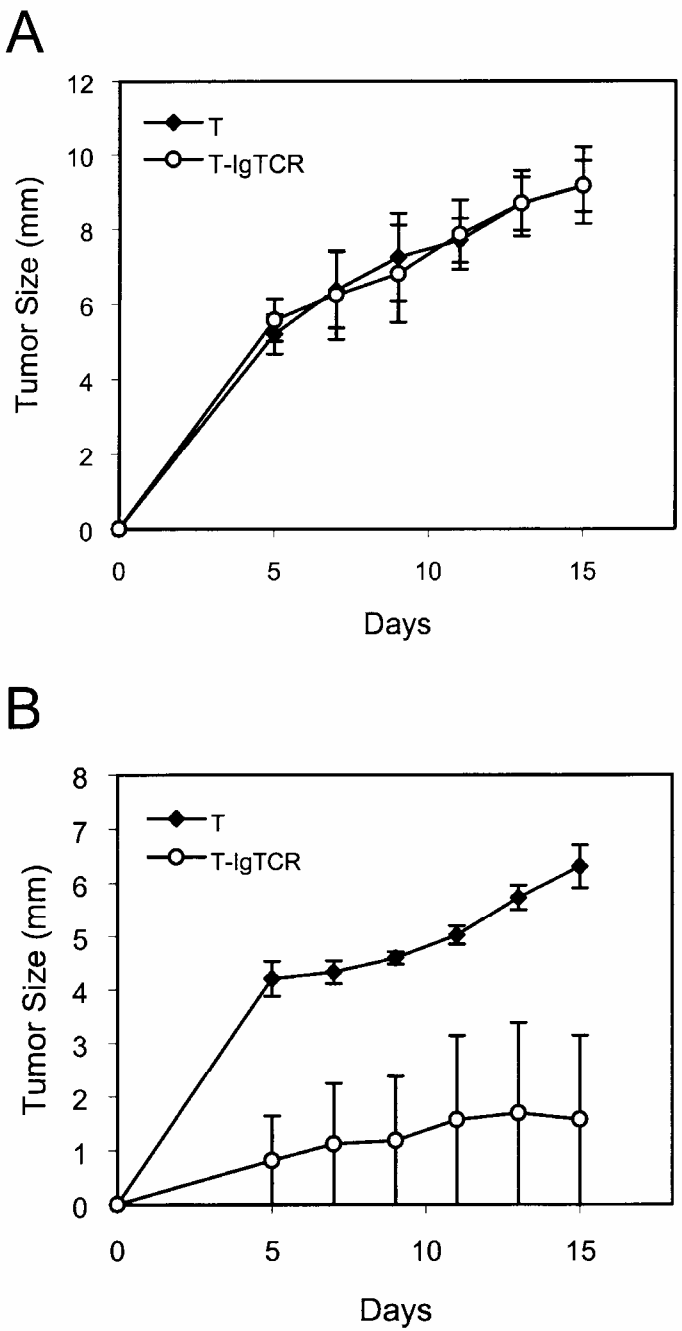
Animal model:

PSMA(-)

Selective *in vivo* Tumor Suppression by Anti-PSMA Designer T Cells

PSMA(+)

55% (5/9) tumor free



Conclusion

- Anti-PSMA designer T cells prepared
- Ready for use in prostate cancer

- *But……*

Clinical Data: 1st Generation
(heterologous system)

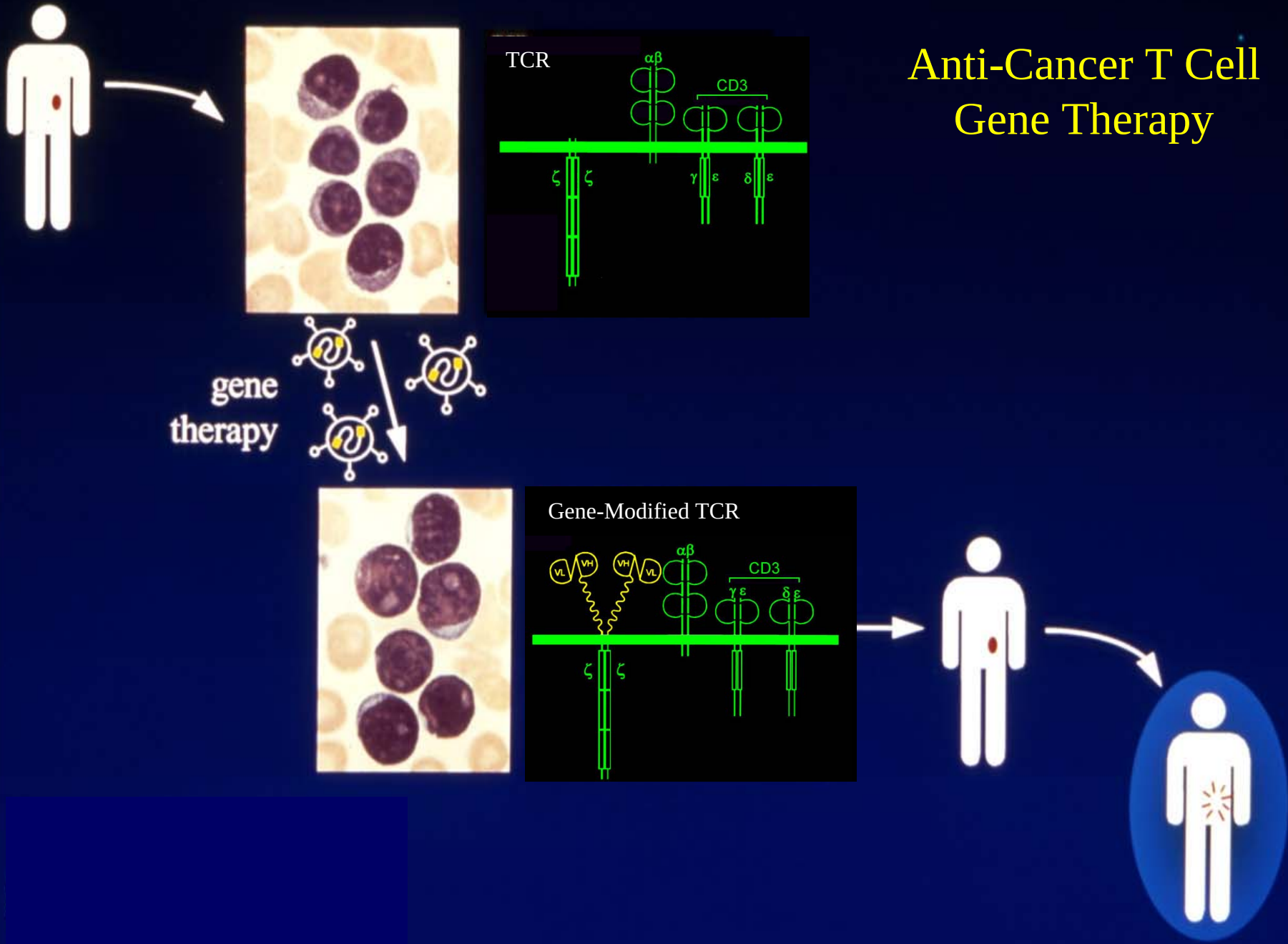
Phase I Study of Anti-CEA Designer T Cells in Adenocarcinoma ("1st generation")

FDA BB IND
7301

Hypotheses

- IgTCR will redirect modified T cells to recognize CEA+ tumor in an MHC-independent manner.
- Recognition of tumor cells will induce proliferation, IL2 secretion (CD4+) and cytotoxicity (CD8+) by “designer T cells” *in vivo*.
- A self-sustaining immune response will cause tumor regression *in vivo*.

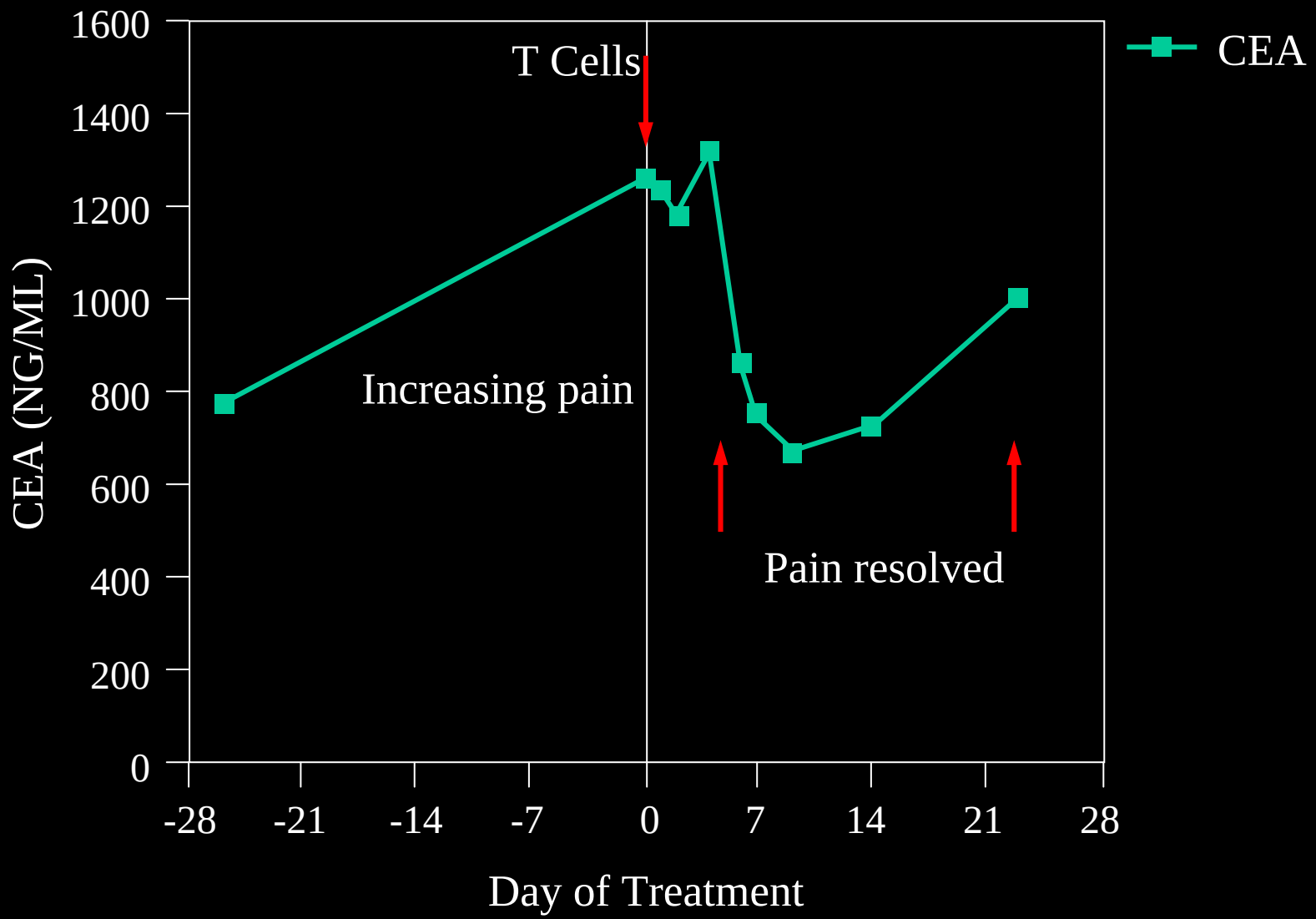
Anti-Cancer T Cell Gene Therapy



Clinical Summary

- Number of doses administered (24): 17 -IL2, 7 +IL2
- Patients treated (7): 5 colorectal, 2 breast; 5 -IL2, 2 +IL2
- Doses sizes administered
  - 1×10^9, 3×10^9, 1×10^{10}, 3×10^{10}, 1×10^{11} cells
- Drug Toxicity (“Probably related” or “Definitely related”)
  - No grade III toxicity, one grade IV toxicity (grade II fever >> grade IV SVT)
  - No delayed grade III, IV toxicity
  - Positive for low grade fevers, mild GI symptoms (<grade III)
- Response
  - Partial, transient (1 patient -IL2)
  - Minor, transient (1 patient +IL2)

CEA Profile on Patient GT



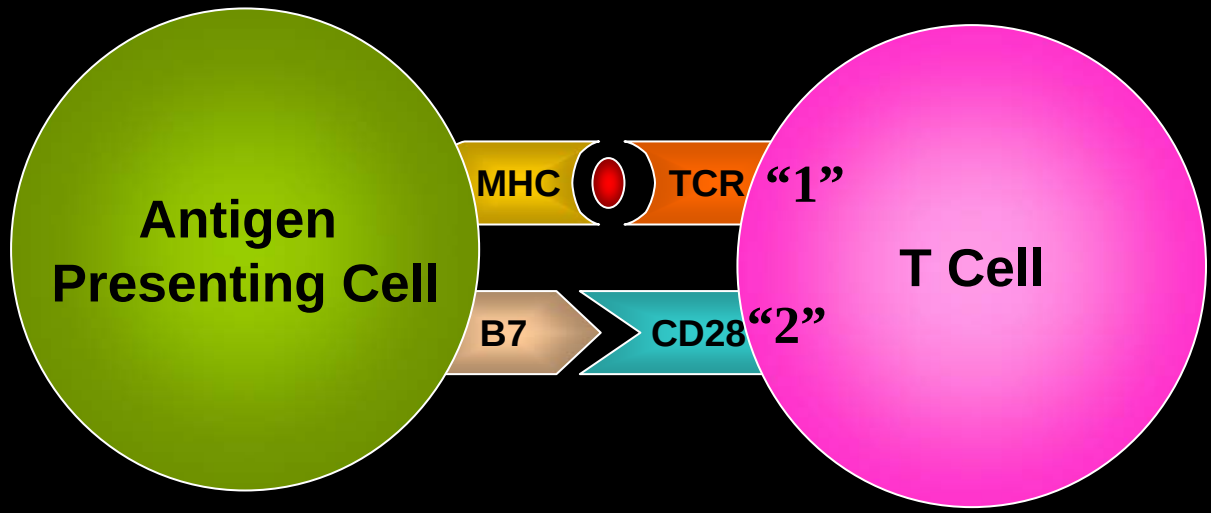
Conclusions:
1st Gen Phase I Study

- Adequate safety
- Proof-of-principle biologic response
- *But…* Time-limited efficacy profile
  - Don’t want PSMA targeting to have same fate…

- WHY DIDN’T IT *CURE*???
  - Laboratory correlate studies >>>>>>>>>

Immunology 101

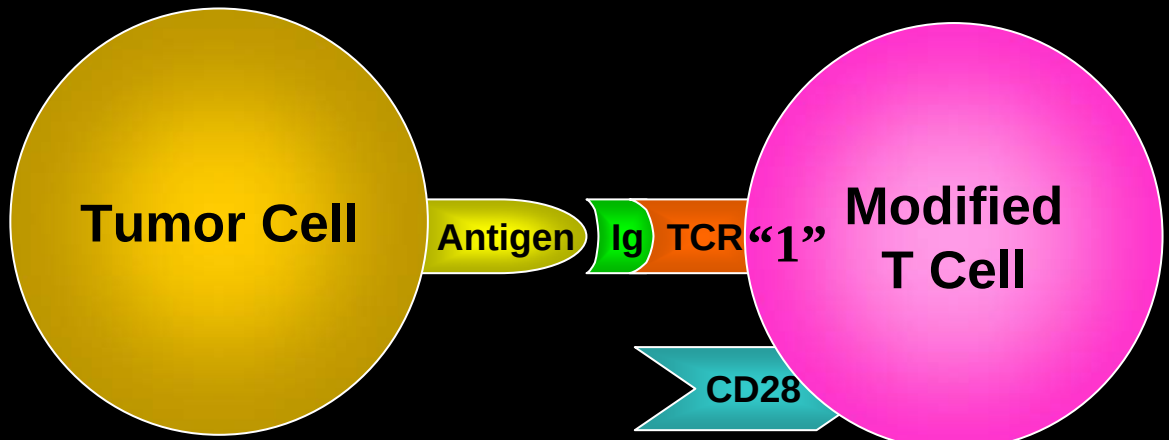
T Cell Activation



- Gene expression
  - Cytokines (IL-2, 4, IFN-γ, etc)
  - Surface molecules (CD25, CD40L, etc)
- Cytotoxicity
- Proliferation

Designer T Cells – First Generation

- **IgTCR** – *chimeric immunoglobulin – T cell receptor*

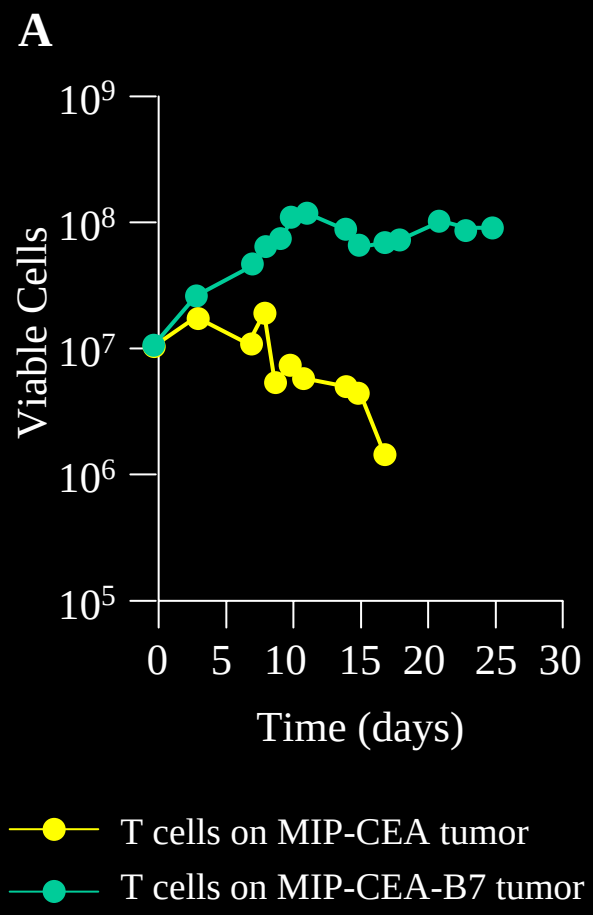


MIPCEA

Advantage: IgTCR provides Signal 1: adequate T cell cytotoxicity.

***Disadvantage*:** Lacking Signal 2, undergoes Activation-Induced Cell Death (AICD) after killing target cells. [HYPOTHESIS]

Comparing Signal 1 with Signal 1+2



Signal 1-only = AICD Signal 1+2 = Proliferation

Proliferation = Increased tumor cell killing

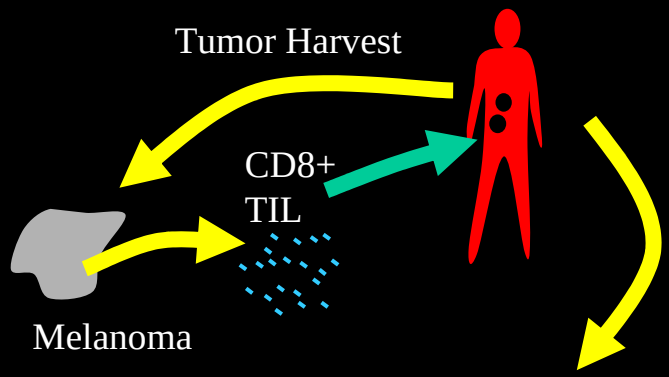
--> EFFICACY HAMPERED BY PROLIFERATION DEFECT

Strategy

Bypass co-stimulation:

Auto-Transplant: Engraft designer T cells via lympho-expansive capacities of the body after lympho-depletion treatments

TIL -- Melanoma



11/20 objective tumor responses

But:

Responses not durable

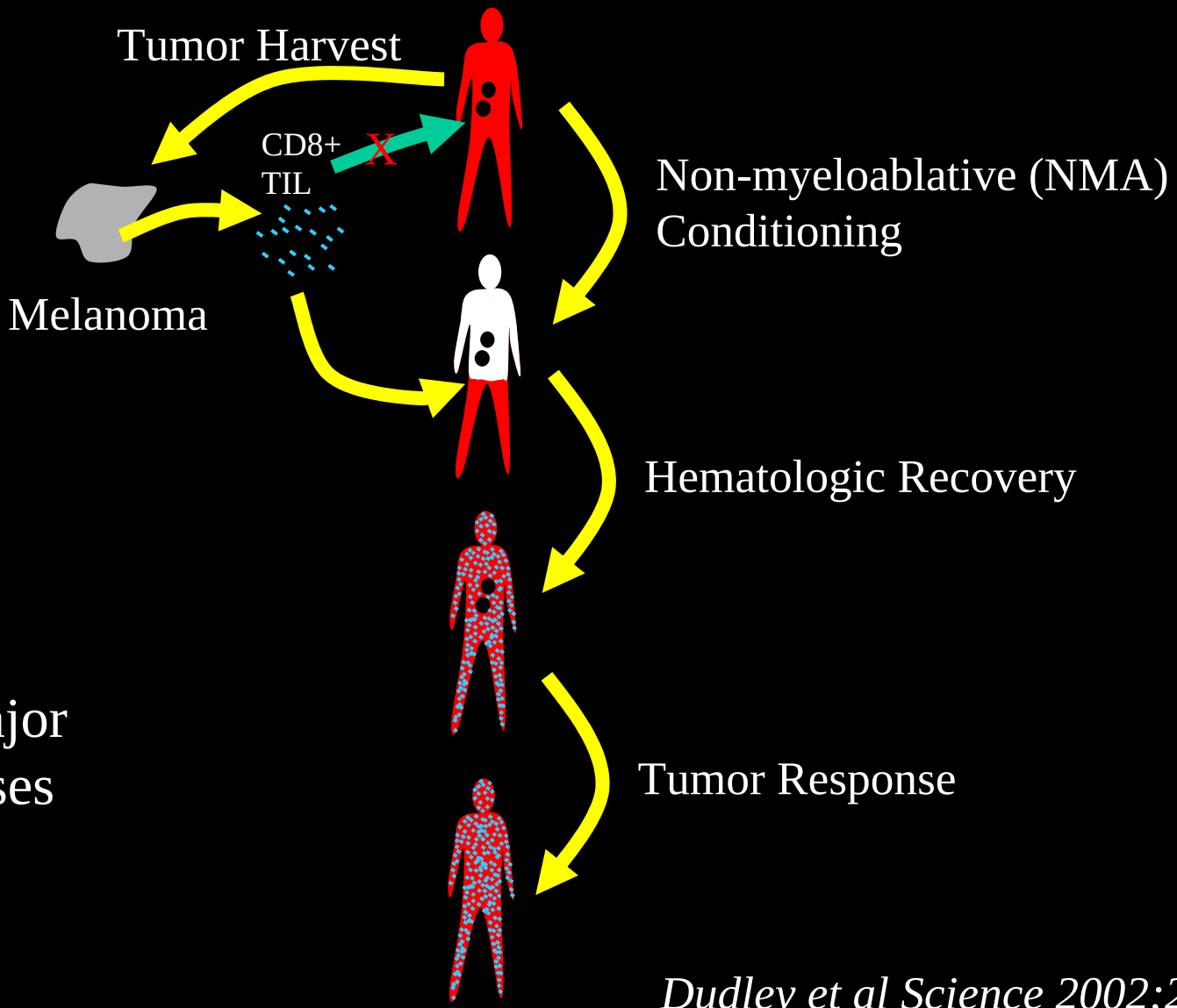
Only melanoma, limited numbers

Technically challenging, antigen(s) unknown

Not reproducible in other studies

Rosenberg et al NEJM 1988;319:1676.

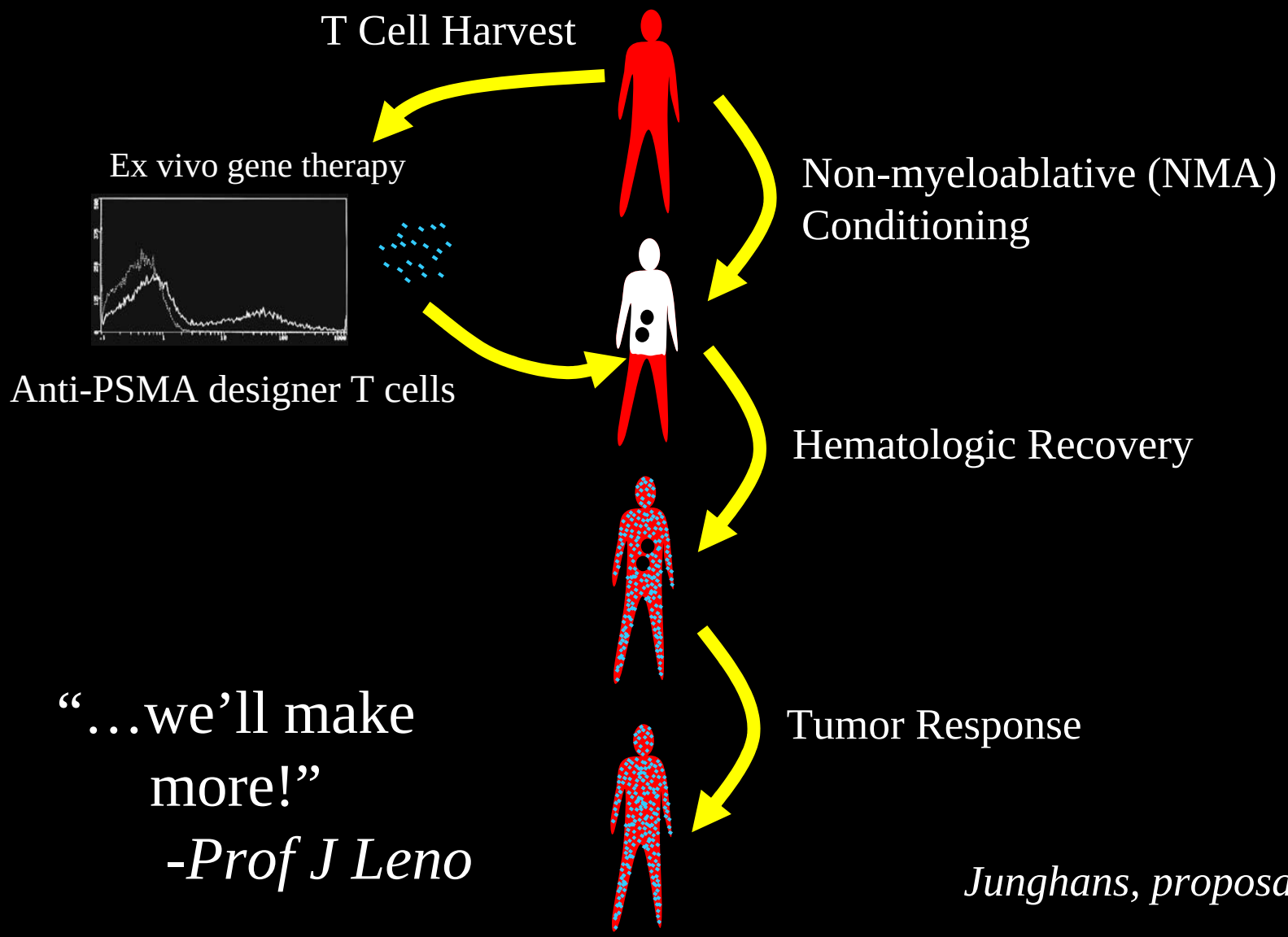
NMA – Melanoma TILs



6/13 major responses

Dudley et al Science 2002;298:850

Prostate Cancer

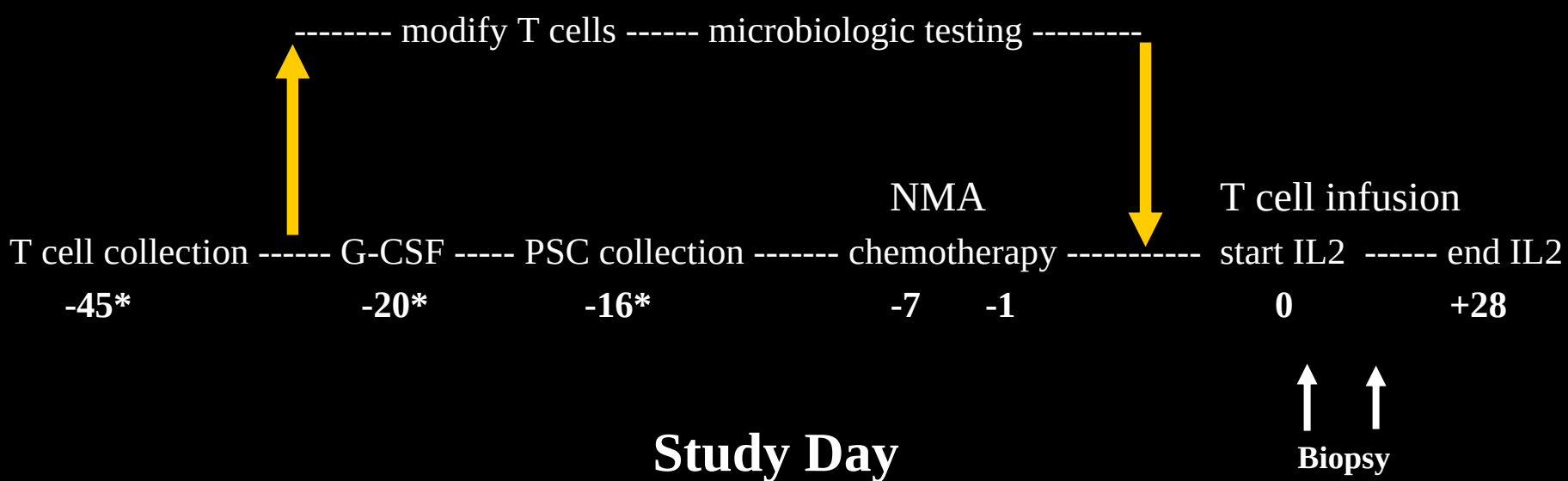


"…we'll make more!"
-Prof J Leno

Junghans, proposal

Phase I Study of NMA Autologous Transplantation with Anti-PSMA Designer T Cells in Prostate Cancer

Treatment Schema



CTX 30 or 60 mg/kg d-7, d-6
Fludarabine 25 mg/m2 d-5 to d-1

Phase I Study Enrollment Plan

Pt #	Cohort	T Cell Dose, Number of Cells 10.9	10.10	10.11
#1		X		
#2	I	X		
#3		X		
#4		X		
#5		X		
#6		X		
#7			X	
#8	II		X	
#9			X	
#10			X	
#11			X	
#12			X	
				(Bx)
#13				X
#14	III			X
#15				X
#16				X
#17				X
#18				X

- Monitoring
  - –Safety
  - –Designer T cell persistence/expansion
    - –in blood
    - –In tumor
  - –Tumor response

Summary

- Uses in vivo expansion capabilities *independent* of costimulation
  - requires chemotherapy, but should be well tolerated
- All technologies available now
- FDA IND filing (11/09/04)
  - no clinical hold issues
  - minor product hold issues
- IRB/IBC approvals
- NGVL vector production pending
- Funding for trial pending
- Potential clinical start date 1/06

Regulatory Implications

- Safety of retroviral gene therapy
  - Replication competent retrovirus
  - Oncogenesis (insertional mutagenesis)
- Occurrence of T cell leukemias
  - 3/12 children treated for X-SCID
  - Autologous stem cells, RV vector, gamma-c gene
  - Mean 3 years after treatment/engraftment
  - 2 leuks with activation of LMO2; 1 with something else (TBD, but not LMO2
- Uncertain relevance to other studies

Probability of Hit #1: LMO2 targeting in X-scid

LMO2 targeting suggests either that there is a “physical hotspot” of integration at this locus, or more likely, that random, activating, *LMO2* integrants are selected simply by the growth advantage conferred on them. The chance of integration of any active gene is assumed to be 1/10.5 (a rough estimate of a random hit within 10 kbp among the estimated transcriptionally active 1 x 10.9 base pairs). It is likely that each patient received at least 1 to 10 *LMO2*-targeted cells, because the patients received 1 x 10.6 or more transduced T lymphocyte precursors (estimating that at least 1% of the total number of injected transduced cells—92 x 10.6 and 133 x 10.6 for patients P4 and P5, respectively—could give rise to T cells).

Hacein-Bey-Abina et al, 2003

More Than 1 Hit

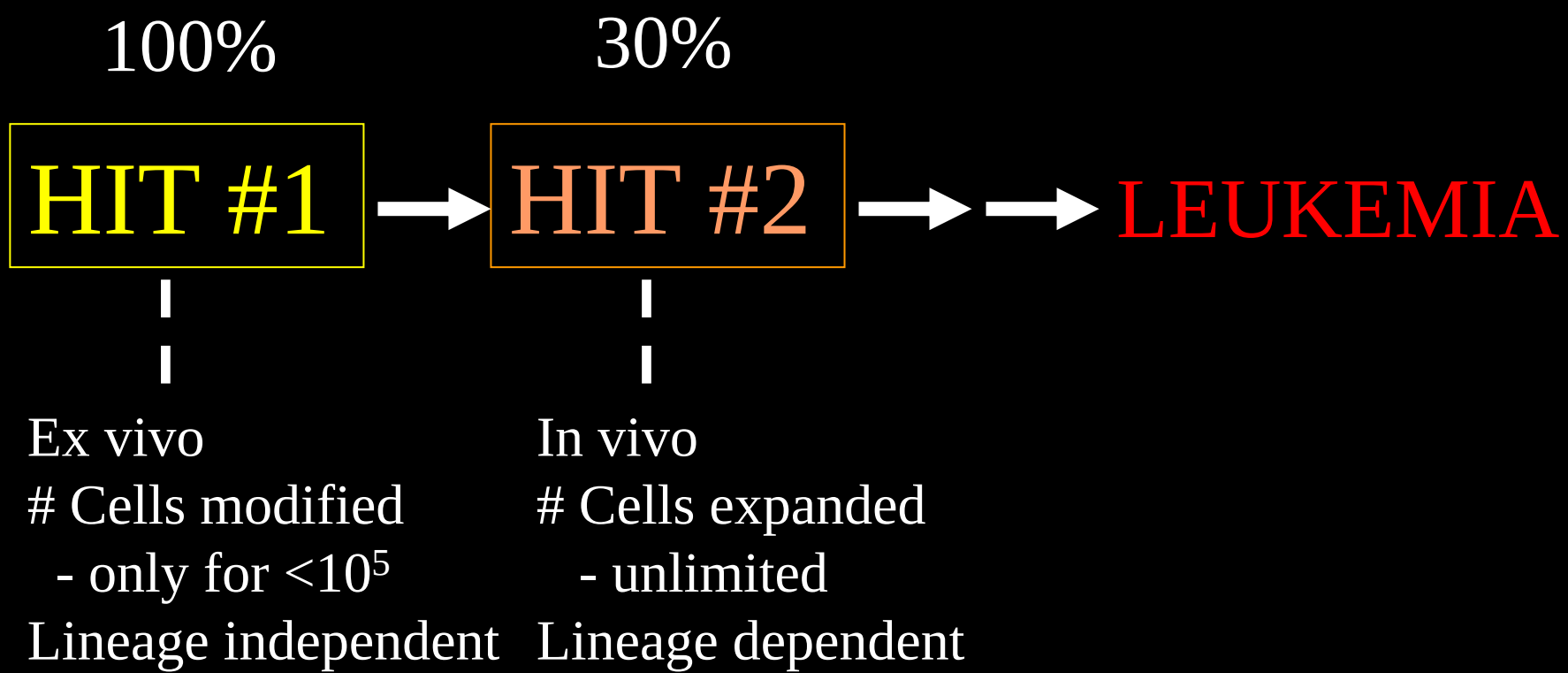
LMO2++ mice, 100% of stem cells express only 10-70% --> leukemia (T-ALL), clonal lag time of 1 year.

Neale et al., Blood 86:3060;
Larson et al., Oncogene 9:3675.

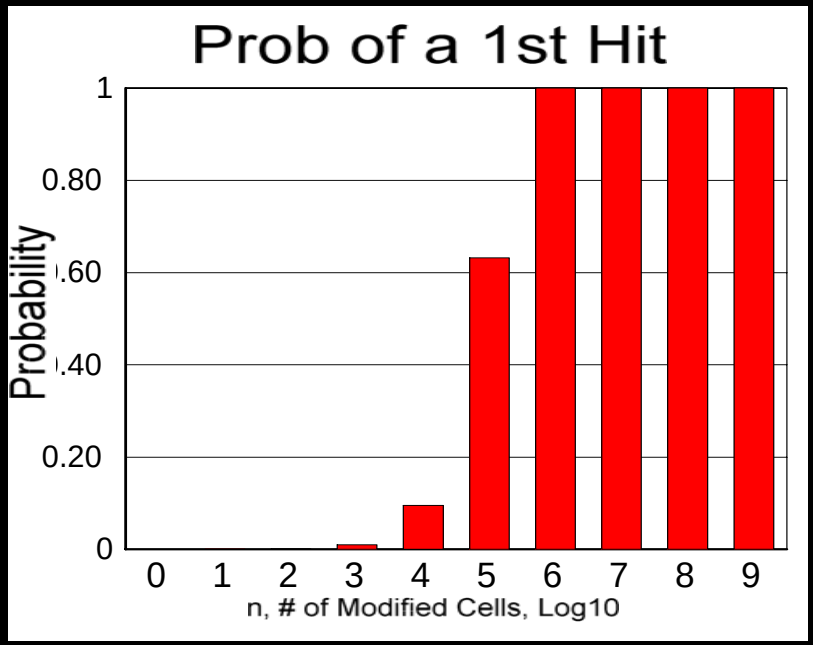
This strongly suggests that additional factors leading to secondary genomic alterations were required for the development of the leukemia-like stage of lymphoproliferation in these patients.

Hacein-Bey-Abina et al, 2003.

Progression to Leukemia



Effect of Cell Dose on 1st Hit



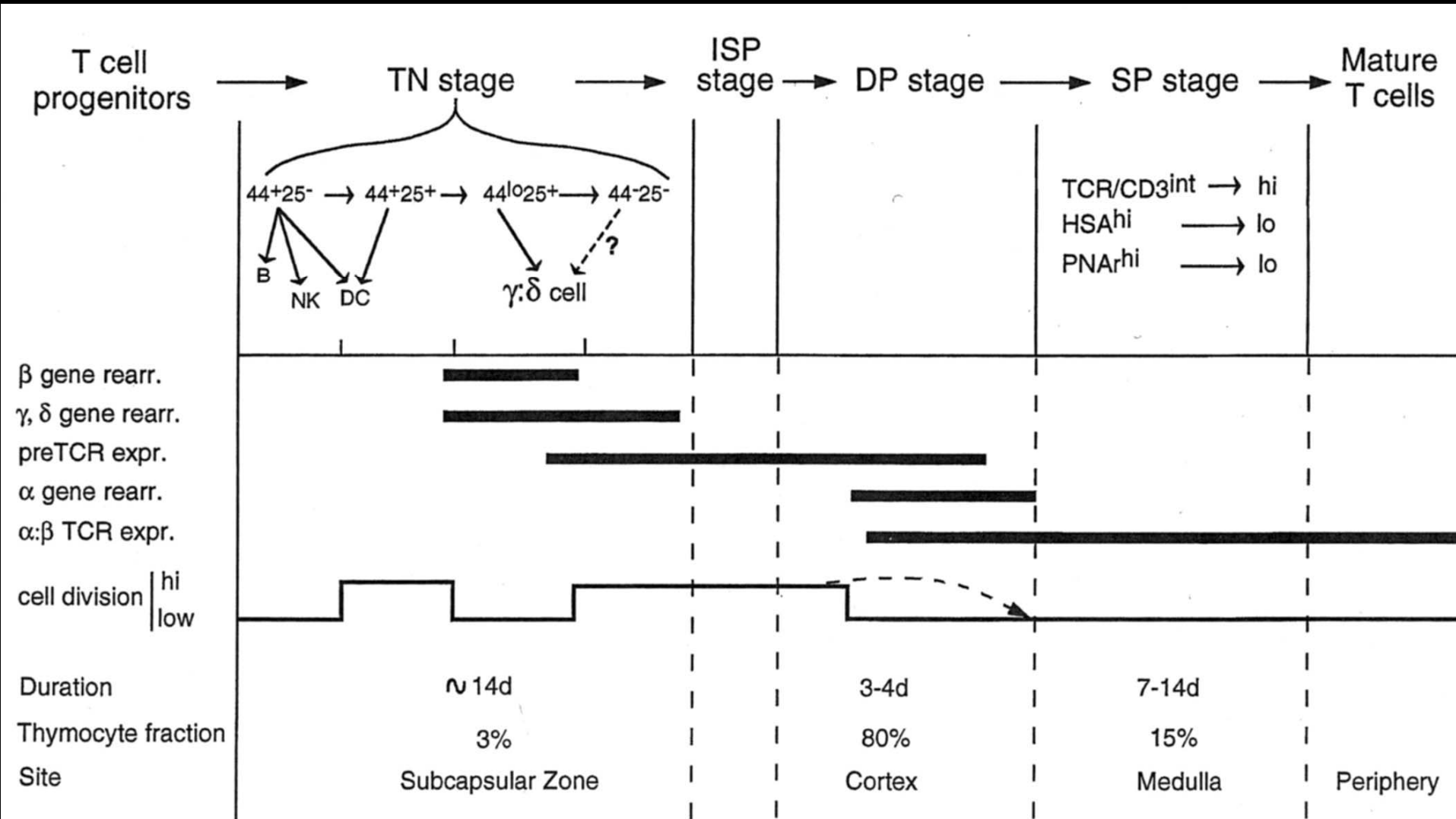
$P(1^{st}) = 1 - (1-p)^n$
p = 10^-5 of hitting a site

- Only chance to make real difference is n < 0.1*(1/p)
- Above this, all preps have same fraction of cells with first hit (= p).
- Final number at risk for 2nd hit = pN, where N is final # of expanded cells.
  - Child 10^-5 x 10^11 = 10^6 (4)
  - Adult 10^-5 x 10^12 = 10^7 (5)

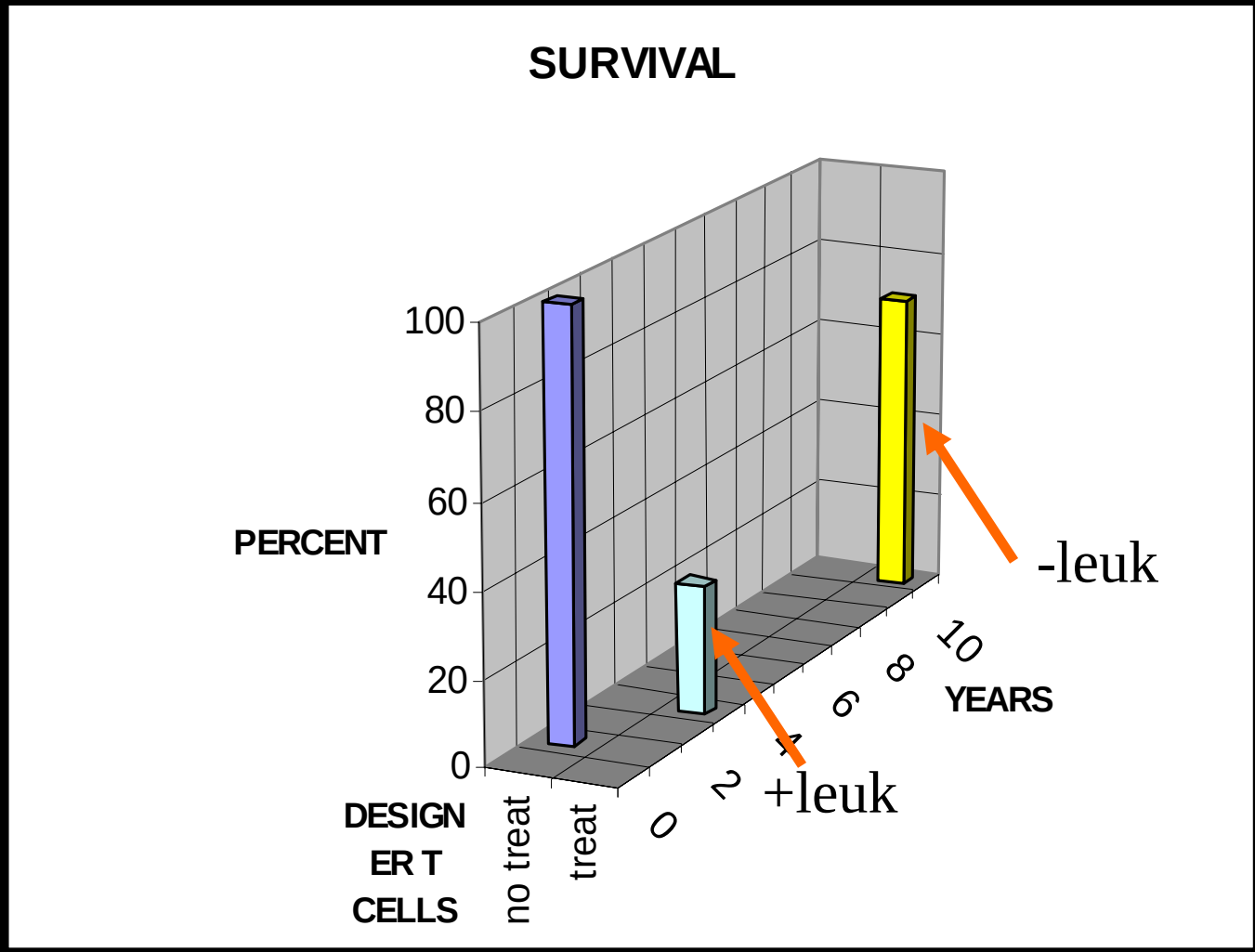
Comparing Gene Therapy Settings 2nd Hit

- X-scid
  - Insertional events saturating
  - Stem cells
    - Recombination may be part of risk
  - Infant/child
    - T-ALL childhood disease
- Cancer
  - Insertional events saturating
  - Mature T cells
    - Recombination complete
  - Adult
    - T-ALL rare

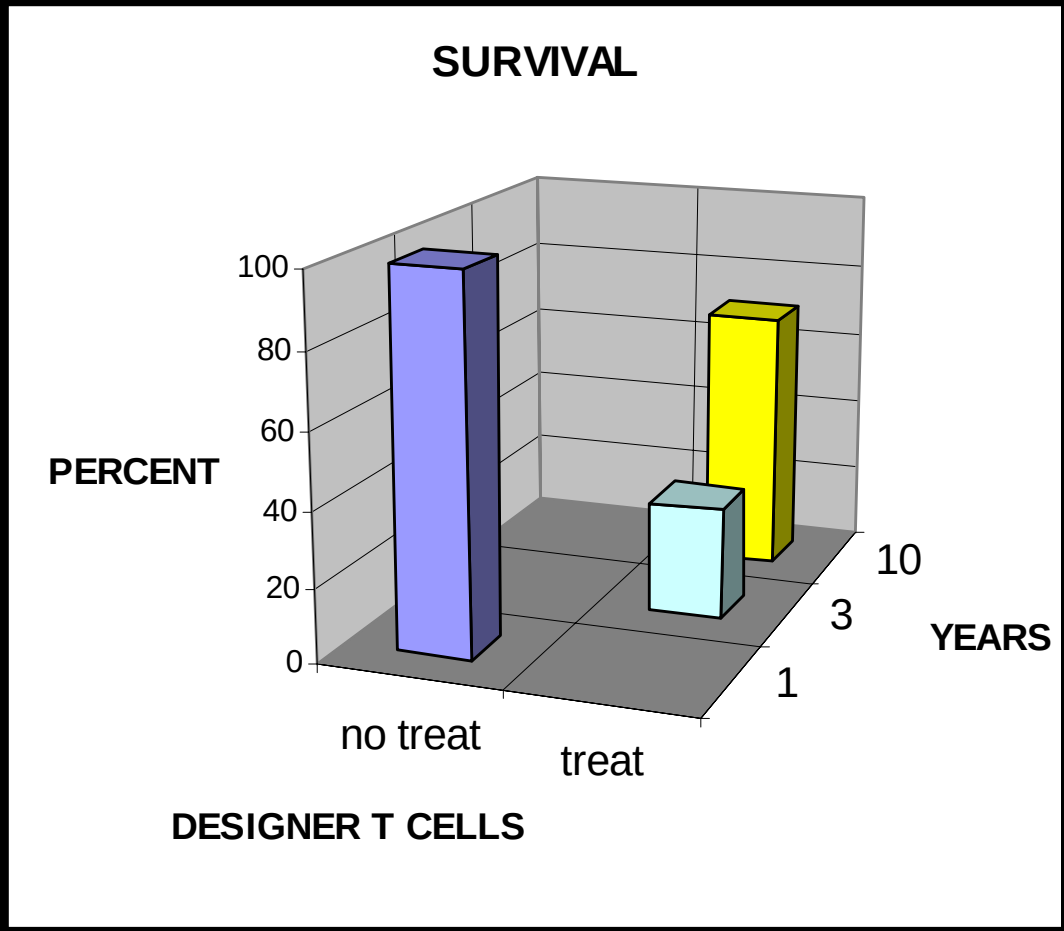
T Cell Ontogeny



Patient Survival with Treatment



Patient Survival with Treatment



Suicide Gene

- Suicide gene drawbacks
  - No data to support need
    - 100 adults treated with modified T cells, no leukemias
    - If cures obtained with T cell engraftment, can readdress risk
  - Immunogenic
  - Efficacy is major hurdle presently
  - Occupies important site for gene co-expression
  - Complicates vector
    - Difficult to omit “safety feature” once introduced
    - Increases delay to test efficacy
  - May not work

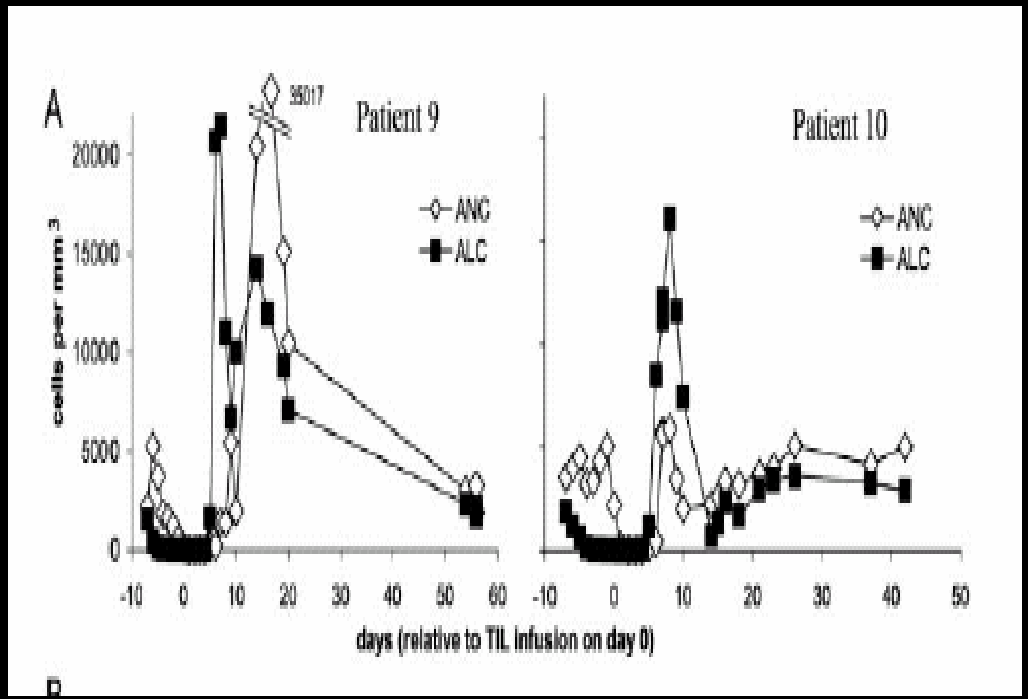
Solutions

- Potential for leukemia in cancer protocol unknown
  - Data needed
  - Some cured patients *may* die from leukemia
    - Acceptable if treatment prolongs life for most (e.g., allo-BMT)
    - Focus on means
      - To prevent leukemia
      - To treat leukemia
- Cost to delay treatment while awaiting solutions
  - Each month delay = cost of life of 3000 patient deaths from prostate ca
- Best option: treat now
  - obtain data
  - solutions applied only if needed

THE

END

IL2 (High Dose) and T Cell Numbers



- IL2 d1-d5
- T cells rebound in periphery after IL2 stop
- Gradually reduce in number to baseline
- Engraftment = stable

Tumor Infiltrating Lymphocytes (TIL)

- N Engl J Med. 1988 Dec 22; 319(25): 1676-80.
-
- **Use of tumor-infiltrating lymphocytes and interleukin-2 in the immunotherapy of patients with metastatic melanoma. A preliminary report.**
-
  Rosenberg SA, Packard BS, Aebersold PM, Solomon D, Topalian SL, Toy ST, Simon P, Lotze MT, Yang JC, Seipp CA, et al.
-
  Surgery Branch, National Cancer Institute, Bethesda, MD 20892.
-
  Lymphocytes extracted from freshly resected melanomas can be expanded in vitro and can often mediate specific lysis of autologous tumor cells but not allogeneic tumor or autologous normal cells. We treated 20 patients with metastatic melanoma by means of adoptive transfer of these tumor-infiltrating lymphocytes and interleukin-2, after the patients had received a single intravenous dose of cyclophosphamide. Objective regression of the cancer was observed in 9 of 15 patients (60 percent) who had not previously been treated with interleukin-2 and in 2 of 5 patients (40 percent) in whom previous therapy with interleukin-2 had failed. Regression of cancer occurred in the lungs, liver, bone, skin, and subcutaneous sites and lasted from 2 to more than 13 months. Toxic effects of interleukin-2 occurred, although the treatment course was short (five days); these side effects were reversible. It appears that in patients with metastatic melanoma, this experimental treatment regimen can produce higher response rates than those achieved with interleukin-2 administered alone or with lymphokine-activated killer cells. It is too early to determine whether this new form of immunotherapy can improve survival, but further trials seem warranted.

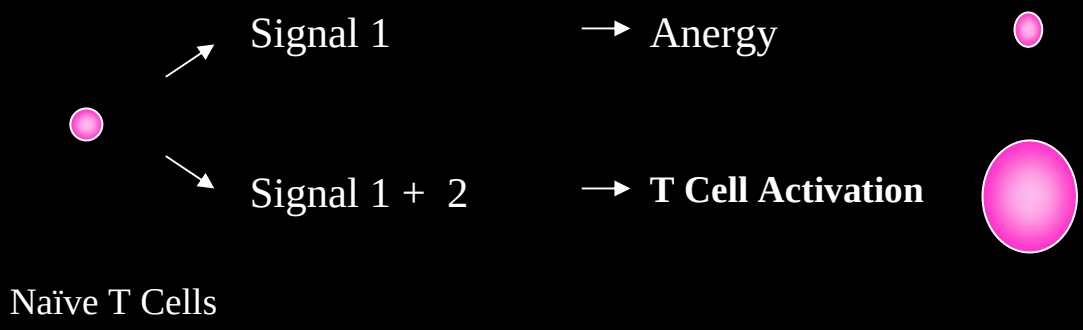
- TIL (11/20) = 55%

Interventions

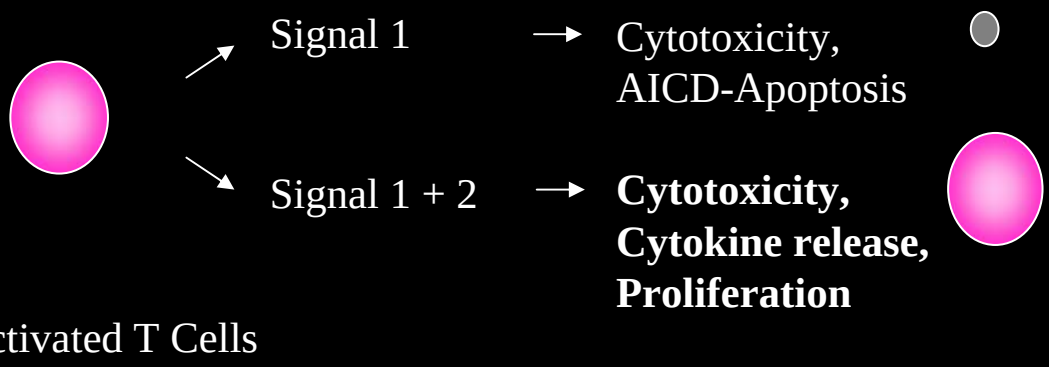
- Phlebotomy/Apheresis
- Isolate patient's peripheral blood mononuclear cells (PBMC)
- Activate/transduce with IgTCR
- Expand in IL2
- Harvest cells equal to dose; Infuse
- Monitor for Toxicity/Response

Costimulation For T-Cell Activation

T Cell Activation



T Cell Re-activation



- Gene expression
  - Cytokines (IL-2, 4, IFN-γ, etc)
  - Surface molecules (CD25, CD69, CD40L, etc)
- Effector function (T help, Cytotoxicity)
- Proliferation

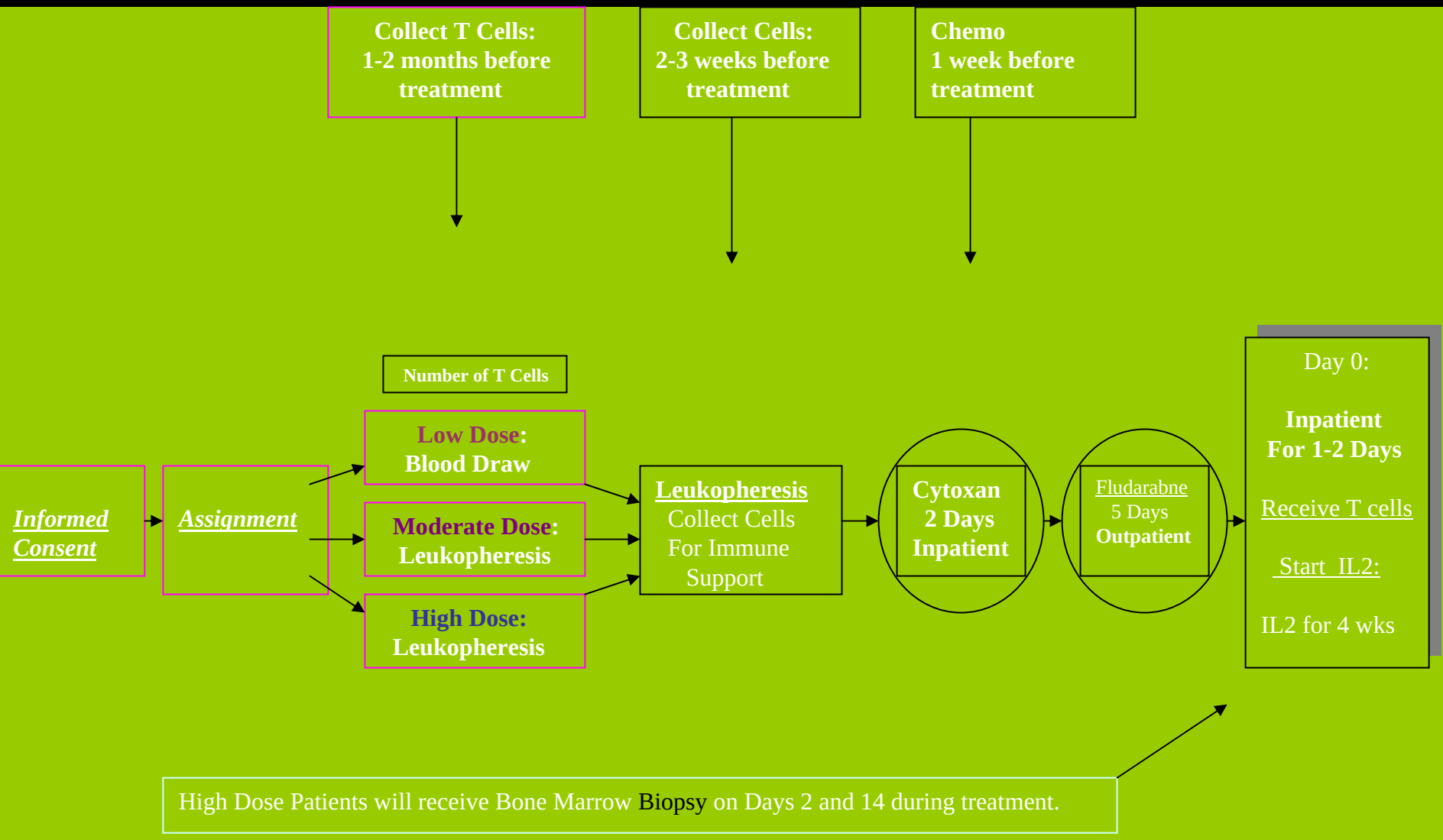
- Apoptosis (AICD – Activation induced cell death)

25 OCTOBER 2002 VOL 298 SCIENCE

Cancer Regression and Autoimmunity in Patients After Clonal Repopulation with Antitumor Lymphocytes

Mark E. Dudley,[1] John R. Wunderlich,[1] Paul F. Robbins,[1] James C. Yang,[1] Patrick Hwu,[1] Douglas J. Schwartzentruber,[1] Suzanne L. Topalian,[1] Richard Sherry,[1] Nicholas P. Restifo,[1] Amy M. Hubicki,[1] Michael R. Robinson,[2] Mark Raffeld,[3] Paul Duray,[3] Claudia A. Seipp,[1] Linda Rogers-Freezer,[1] Kathleen E. Morton,[1] Sharon A. Mavroukakis,[1] Donald E. White,[1] Steven A. Rosenberg[1]*

Schema


Collect T Cells:
1-2 months before
treatment
Collect Cells:
2-3 weeks before
treatment
Chemo
1 week before
treatment
Number of T Cells
Informed Consent
Assignment
Low Dose:
Blood Draw
Moderate Dose:
Leukopheresis
High Dose:
Leukopheresis
Leukopheresis
Collect Cells
For Immune
Support
Cytoxan
2 Days
Inpatient
Fludarabne
5 Days
Outpatient
Day 0:
Inpatient
For 1-2 Days
Receive T cells
Start IL2:
IL2 for 4 wks
High Dose Patients will receive Bone Marrow Biopsy on Days 2 and 14 during treatment.

Animal model:

Tumor Free Mice after Anti-PSMA Designer T Cell Therapy

	PSMA(-)	PSMA(+)
	PC3	CWR22R
T	0/8	0/8
T-IgTCR	0/9	5/9 (55%)

Treatment Plan

- T cell harvest, Designer T cell preparation
- NMA conditioning (CTX, Fludarabine)
- Designer T cell infusion (x1)
  - interleukin 2 co-administration (outpatient)
- Monitoring
  - Safety
  - Designer T cell persistence/expansion in blood
  - Tumor response