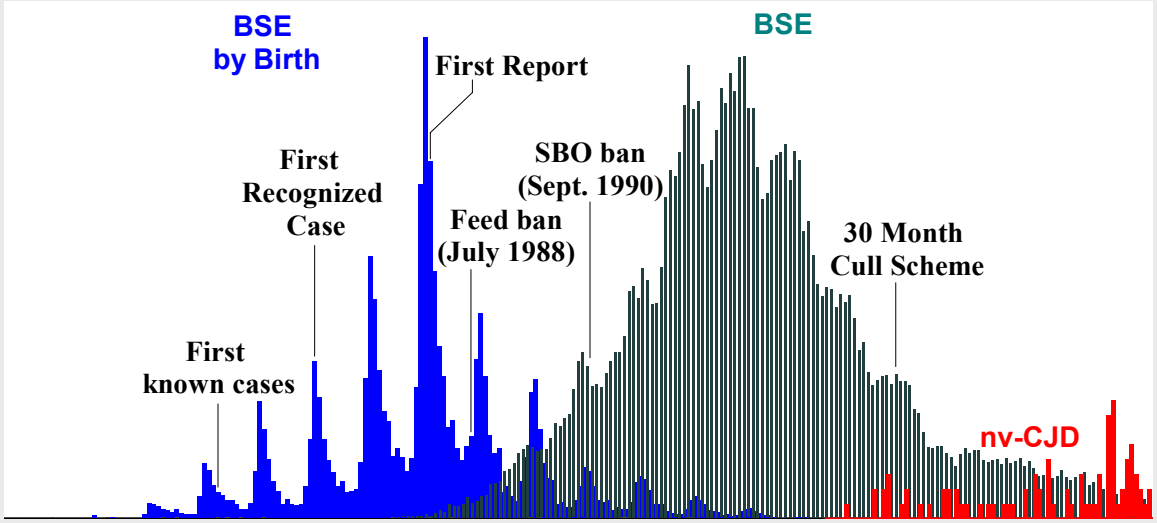
FDA TSE Advisory Committee

Bethesda, Maryland
October 31, 2005

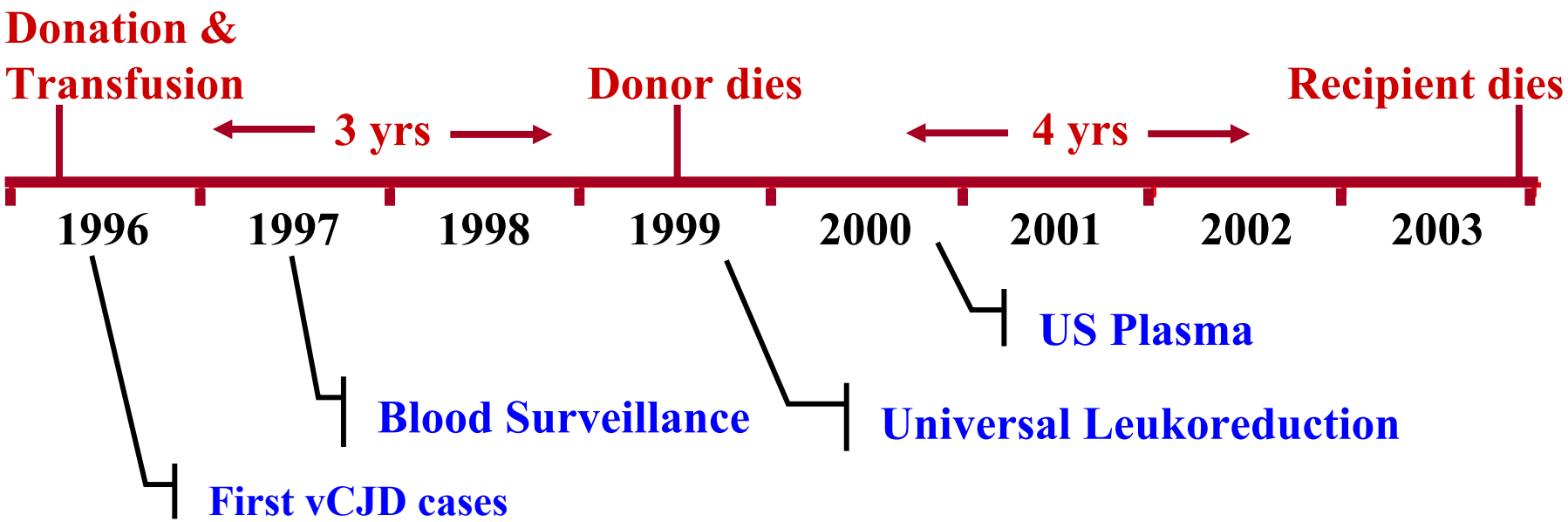


Removal of TSE Infectivity from Blood by Adsorption to Immobilized Ligands

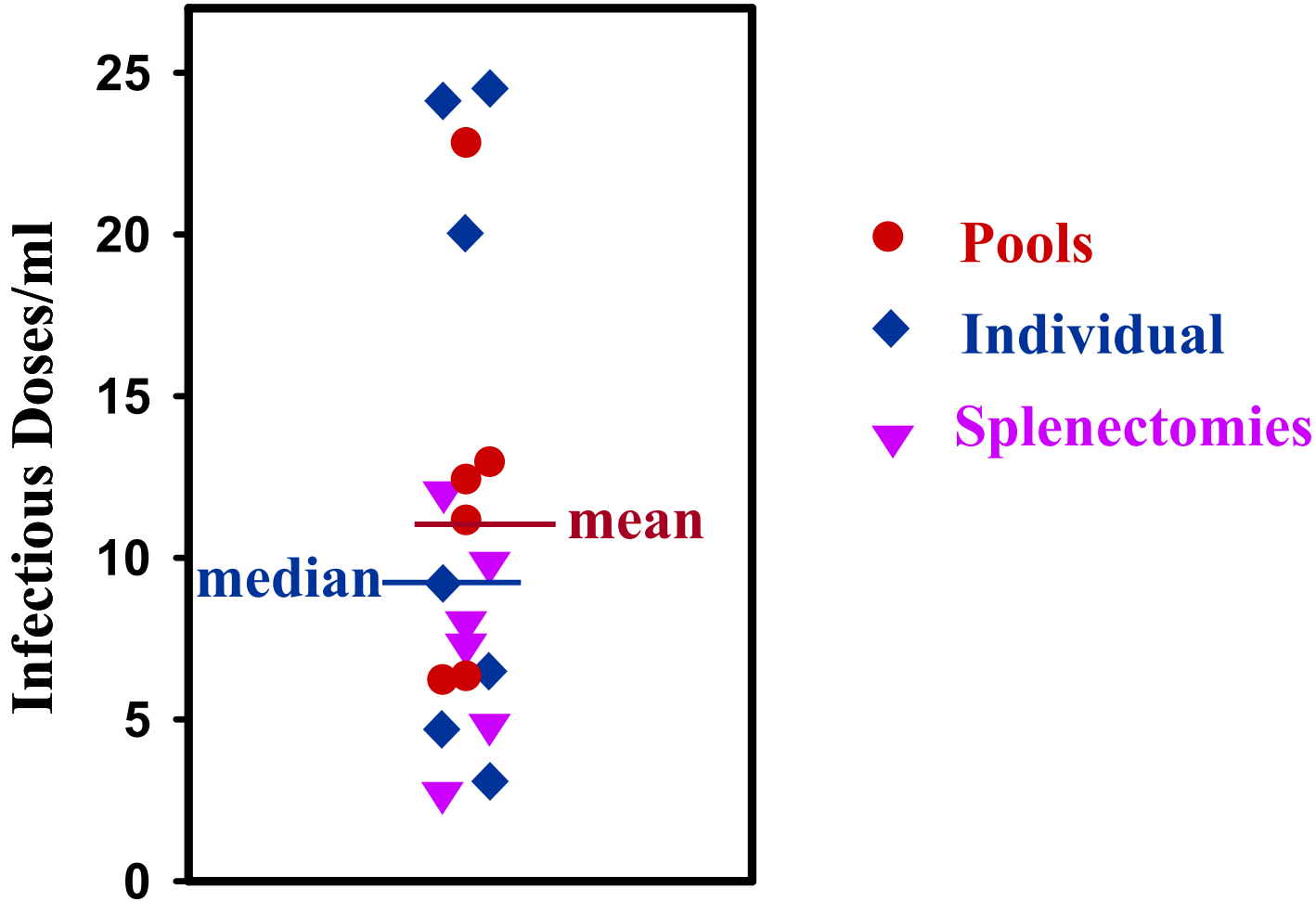
by Robert G. Rohwer, Ph.D.

VA Medical Center
University of Maryland
Baltimore, MD

Time line for vCJD transfusion transmission

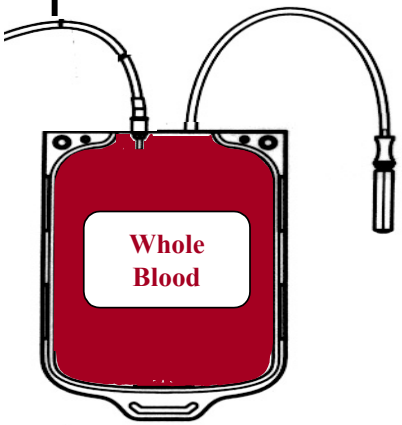


Titer of TSE Infected Blood
in Clinically Affected Animals



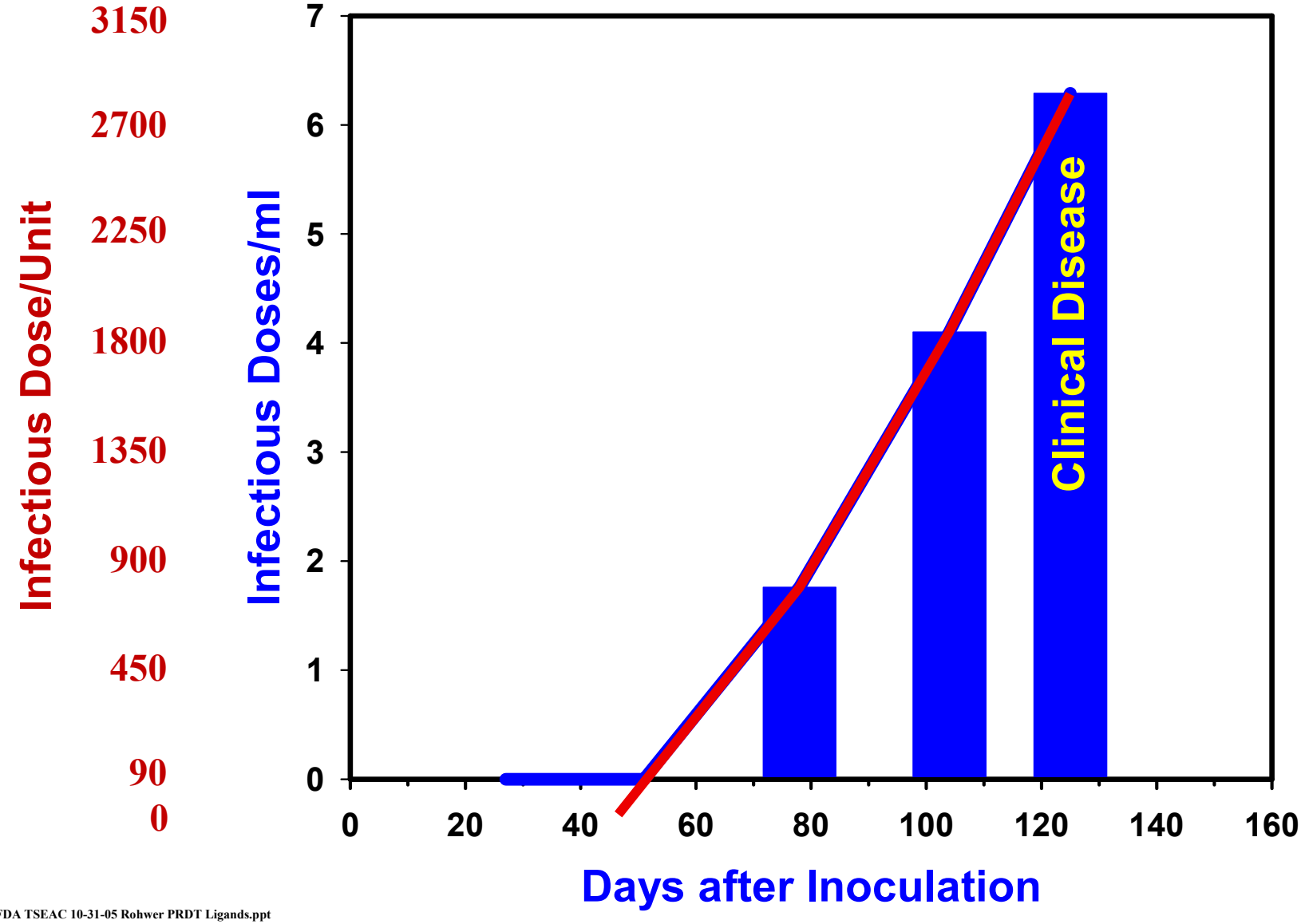
Blood vs Brain

Tissue	Infectivity Titer
Blood	10 ID/ml
Brain	10,000,000,000 ID/ml



5,000 ID/Unit

Titer in ID/ml of Preclinical Hamster Blood



Control of TSE Pathogens

- **Sourcing/Deferrals – moving target**
- **Screening – technically problematical for blood**
- **Inactivation – incompatible with product**

  or

  Risk Substitution instead of Risk Reduction

- **Removal – low risk, technically accessible**

Advantages of Adsorption for TSE Risk Reduction

- Removes infectivity that can not be detected with diagnostics
  - Clinical disease for blood
  - Preclinical disease for brain
- Discrimination of abnormal from normal PrP is not necessary or even desirable
- More comprehensive than a diagnostic
- May be less costly than diagnostics

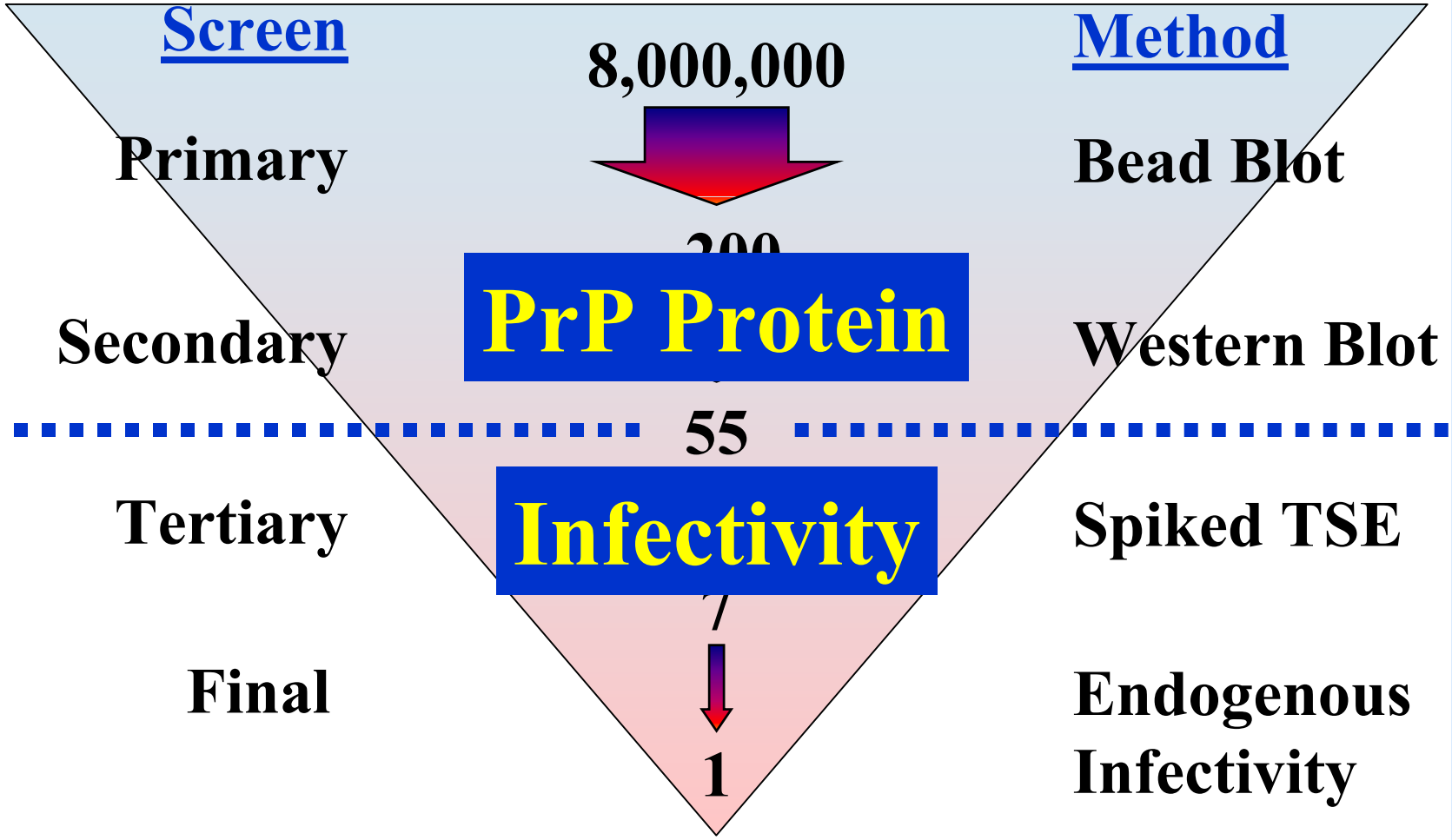
PRDT

Pathogen Removal and Diagnostics Technologies, Inc.

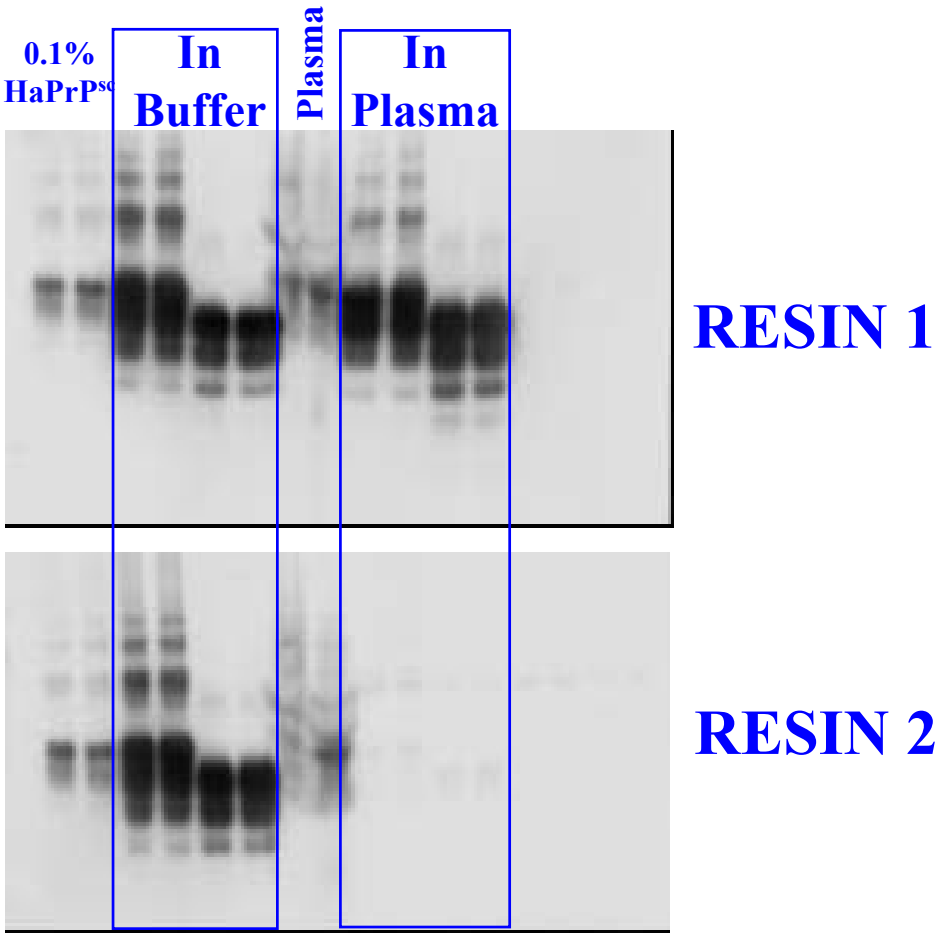
- **Founders**
  - Dave Hammond, Ph.D.
  - Ruben Carbonnel, Ph.D.
  - Robert Rohwer, Ph.D
- **Joint Venture**
  - American Red Cross
  - Prometic Corp.
- **Manufacturing and Marketing Partner**
  - MacoPharma

Selection

Library: 64,000,000 combinations



Binding in Plasma



Infectivity Experiments

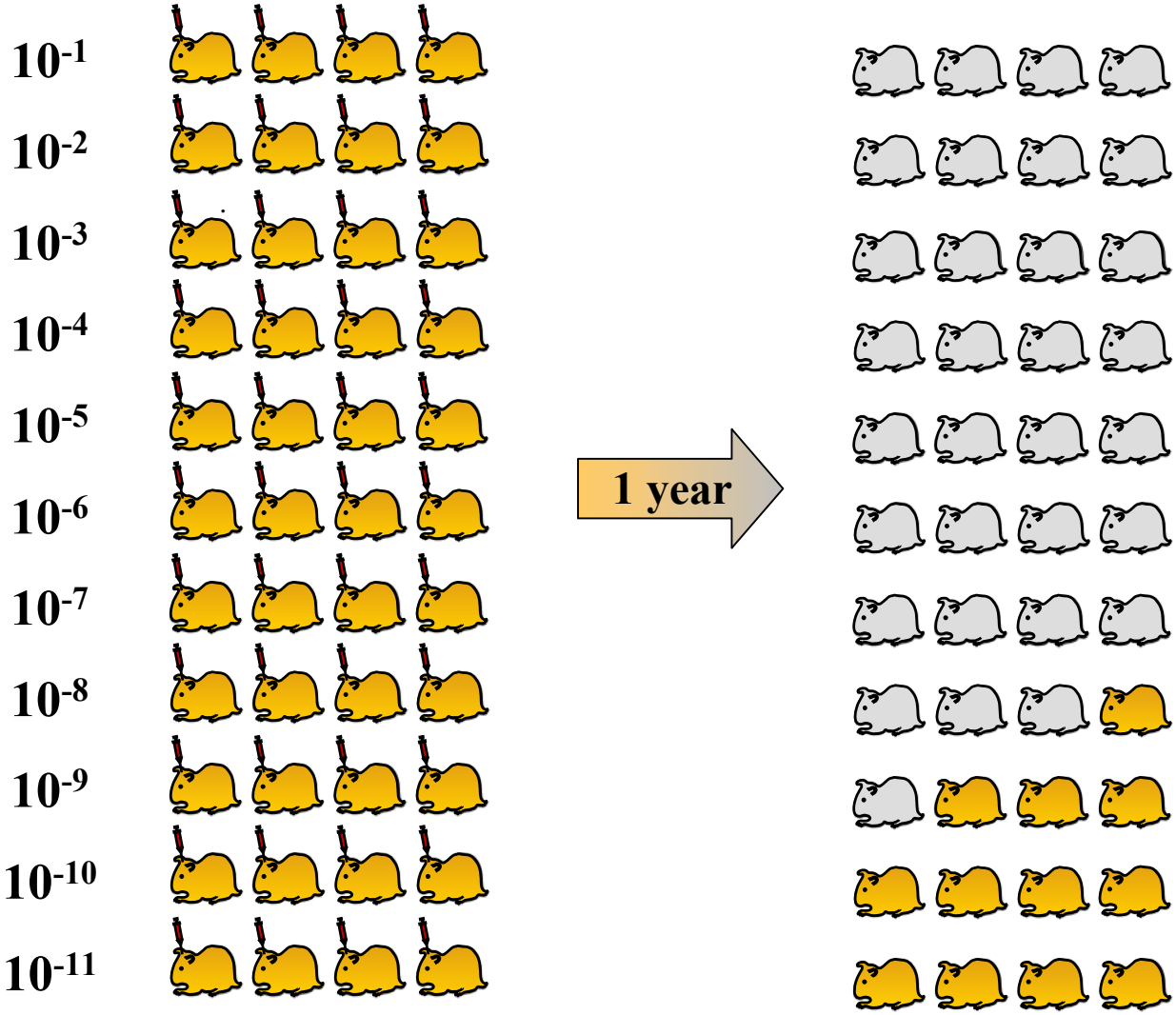
Titer vs Relevance

- **Hamster brain infectivity spiked into RBCs**
  - ***High*** **titer**
  - **High levels of removal**
  - **"Relevance" is *uncertain***
  - **Early readout**
  - **Crude measurement**
  - **Less costly**

- **Endogenous infectivity from leukoreduced WB**
  - **Very *low* titer**
  - **1.5 $Log_{10}ID_{50}$ Maximum**
  - ***Relevant***
  - **18 month experiment**
  - **Precise measurement**
  - **More Costly**

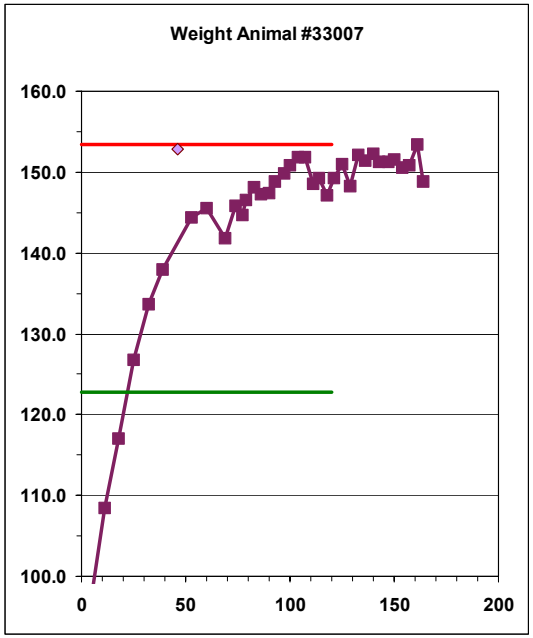
End Point Dilution Titration

Inoculate 50 µl each



Titer
by
Reed & Muench
or
Spearman & Karber

$10^{9.8\pm0.4}$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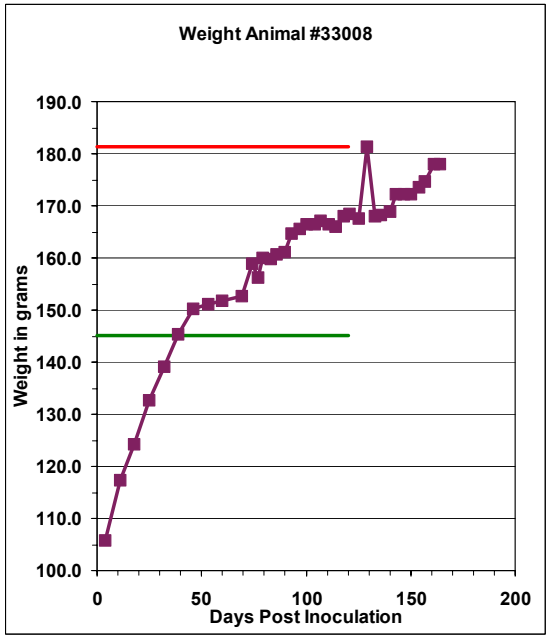
150.0
140.0
130.0
120.0
110.0
100.0
0
50
100
150
200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Weight in grams
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180.0
170.0
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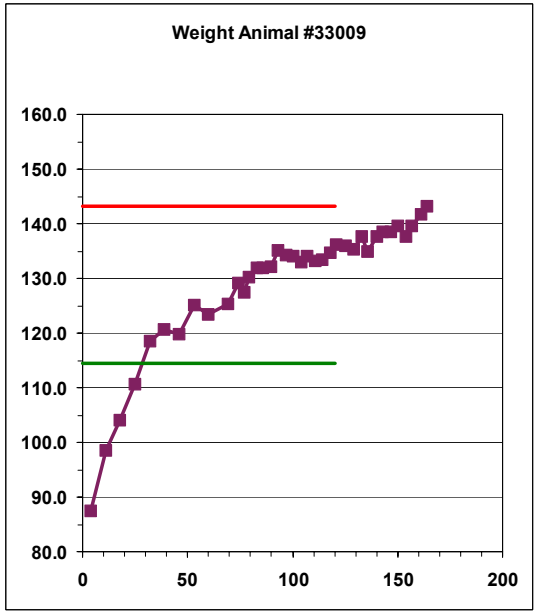
50
100
150
200
Days Post Inoculation

CAGE 10021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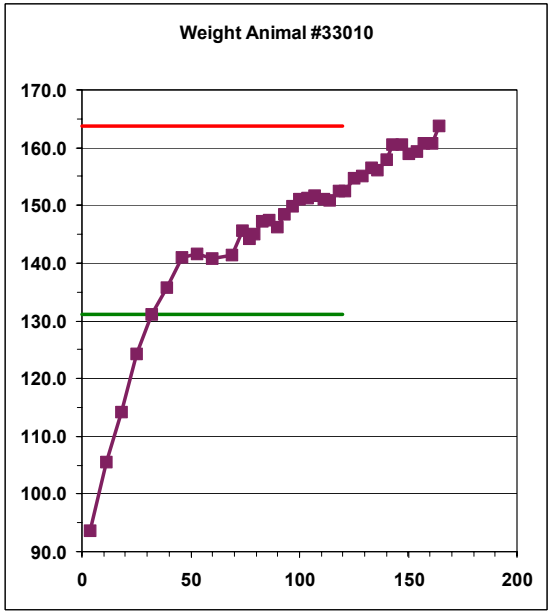
150
200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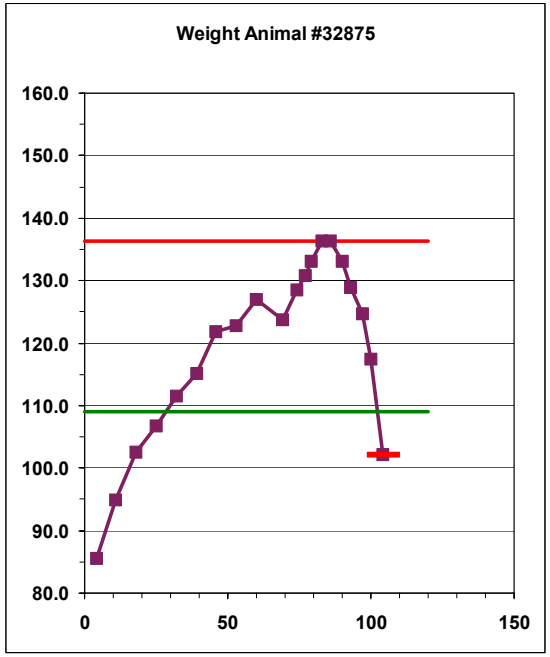
150
200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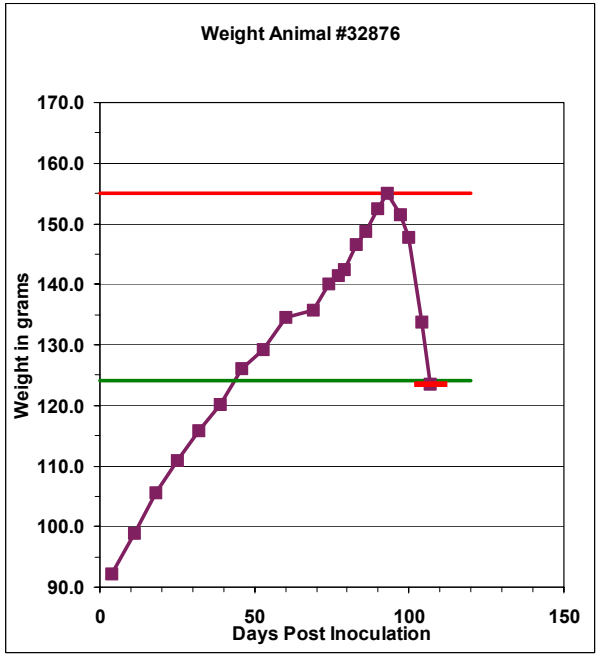
80.0
0
50
100
150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150.0
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130.0
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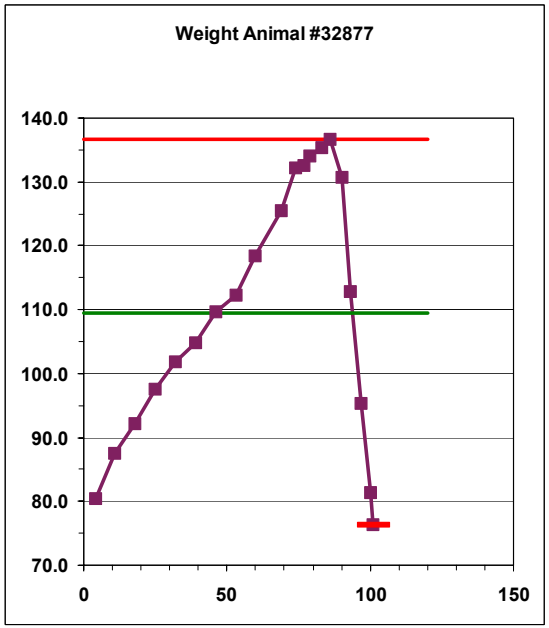
90.0
Weight in grams
0
50
100
150
Days Post Inoculation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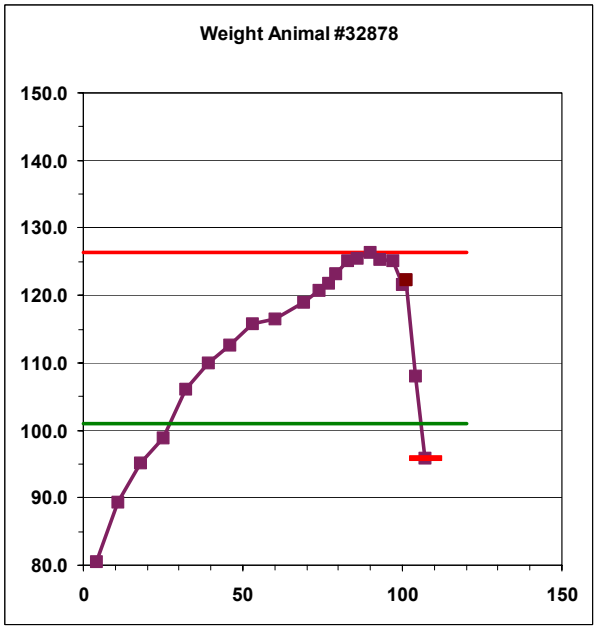
80.0
70.0
0
50
100
150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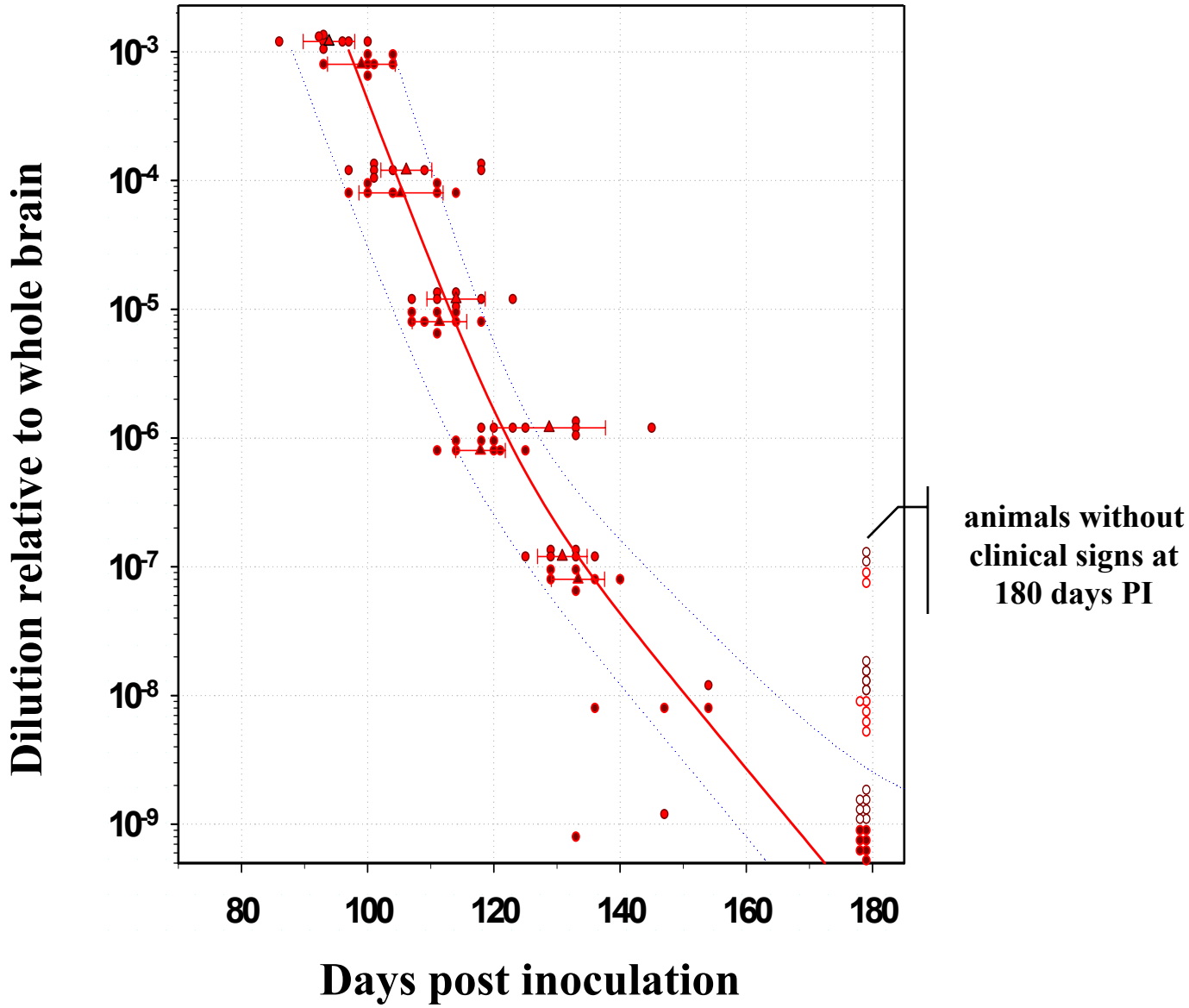
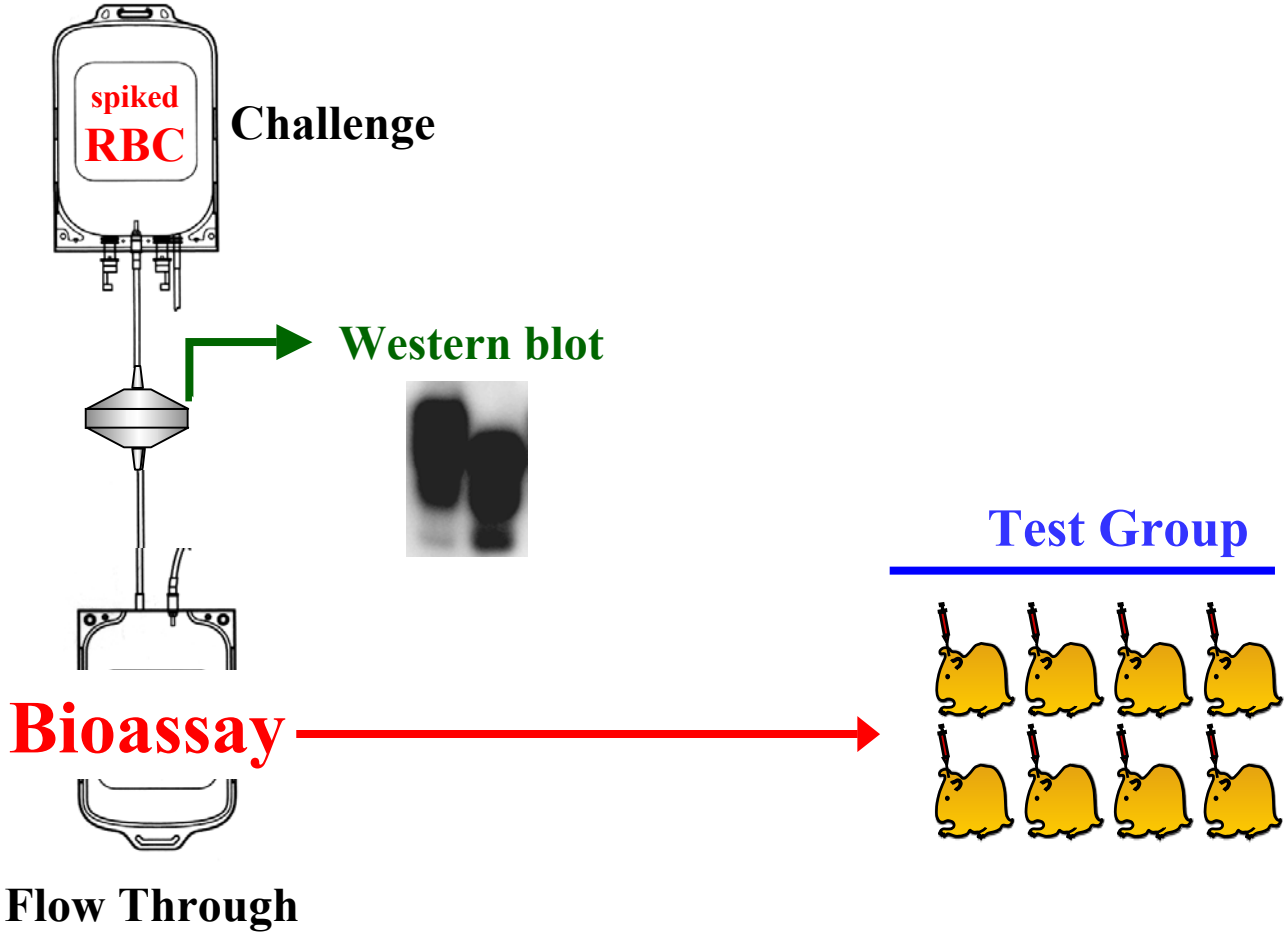
80.0
0
50
100
150

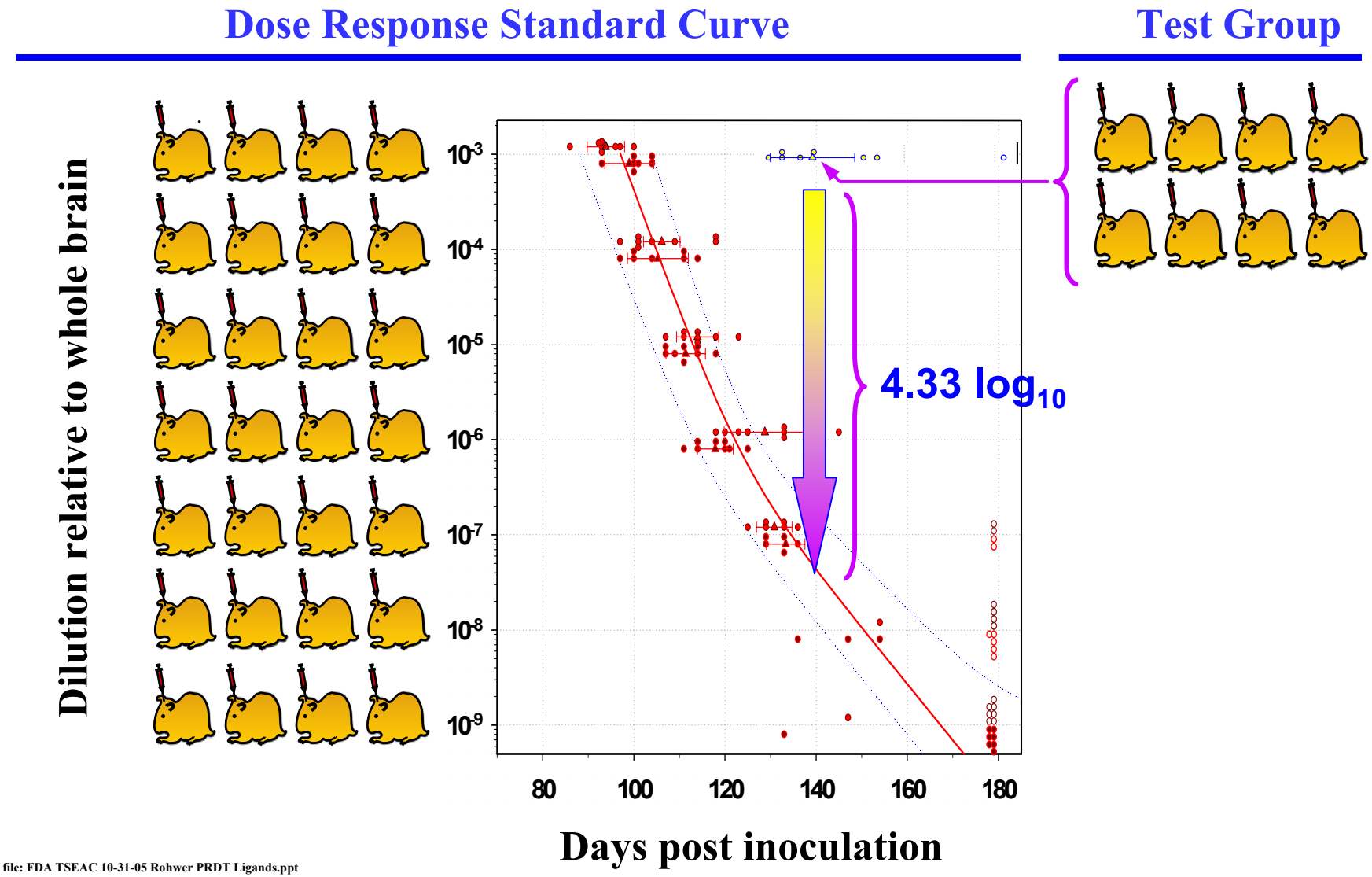
Dose response of duplicate end point dilution titrations



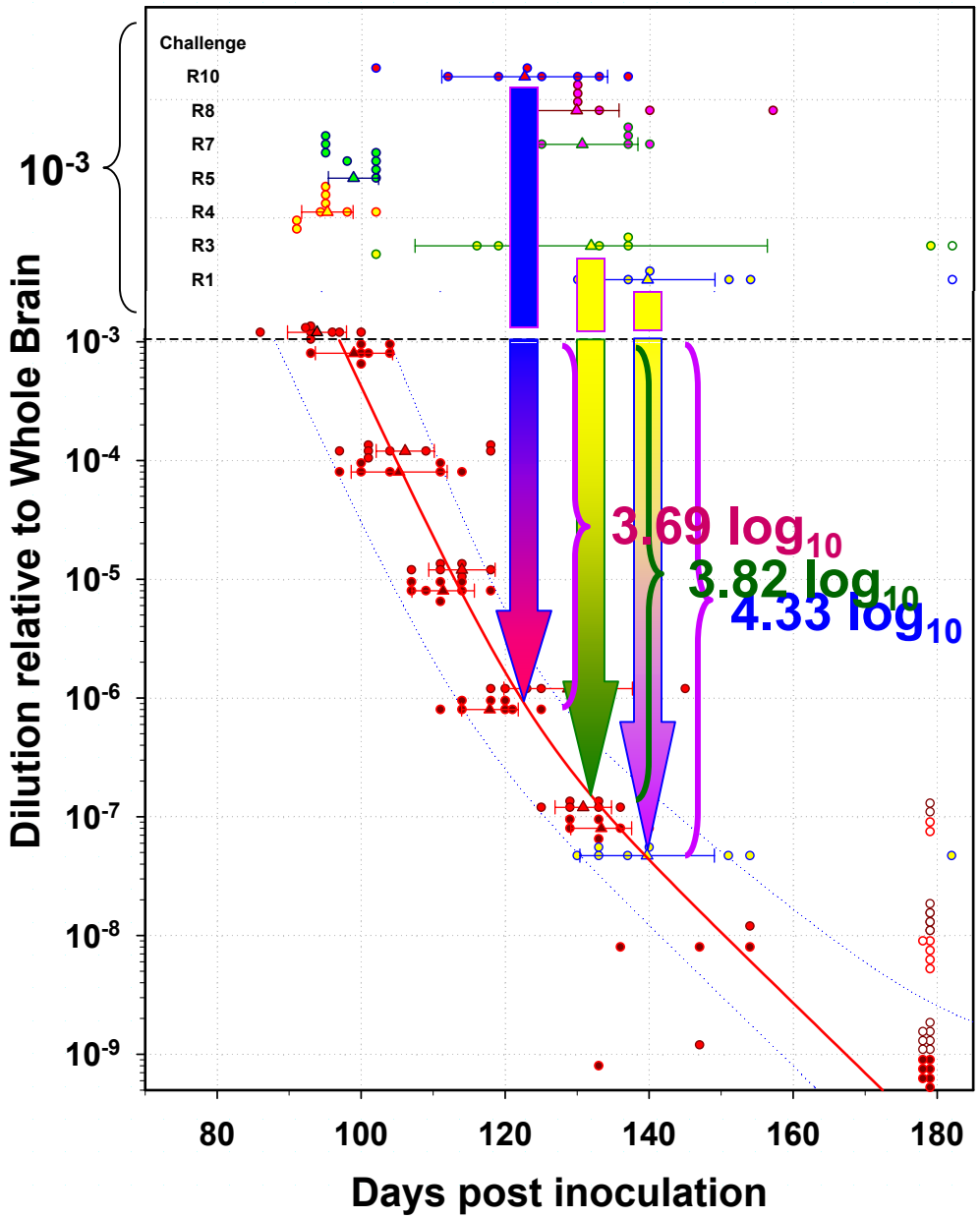
RBC spiked with Brain Homogenate

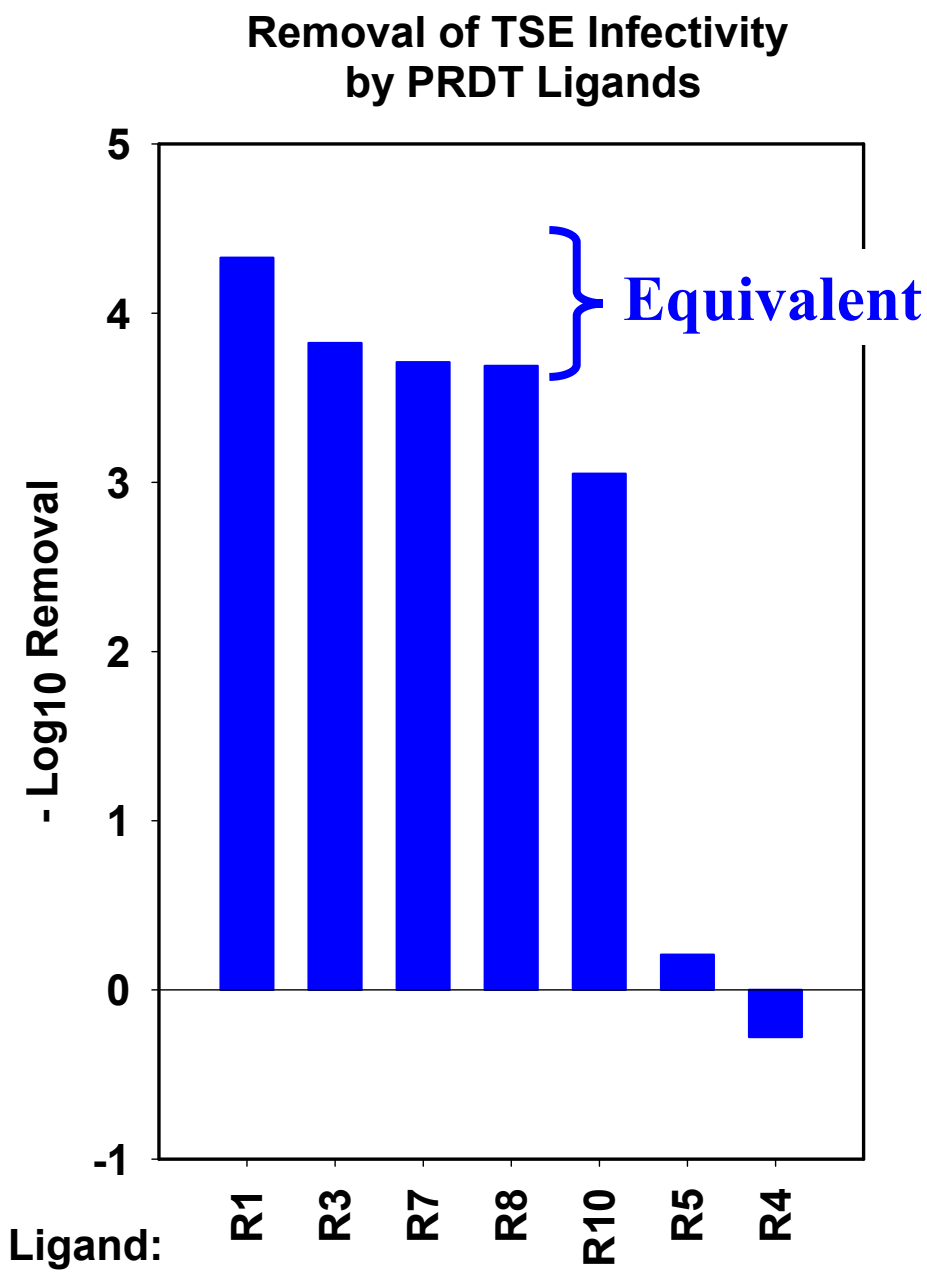


Incubation Time Bioassay



Dose response curve for SBH in RBCC





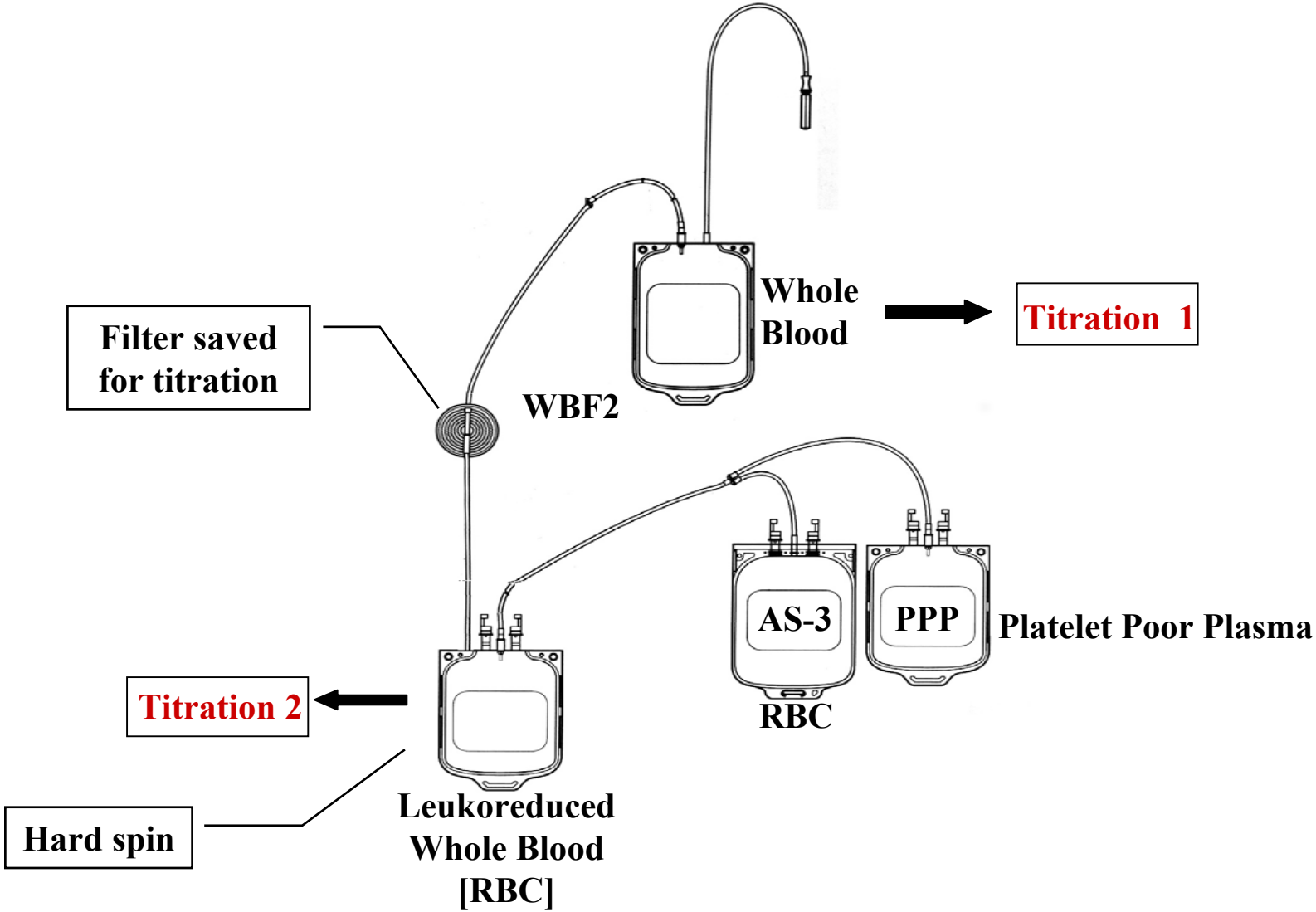
Overloaded Challenge

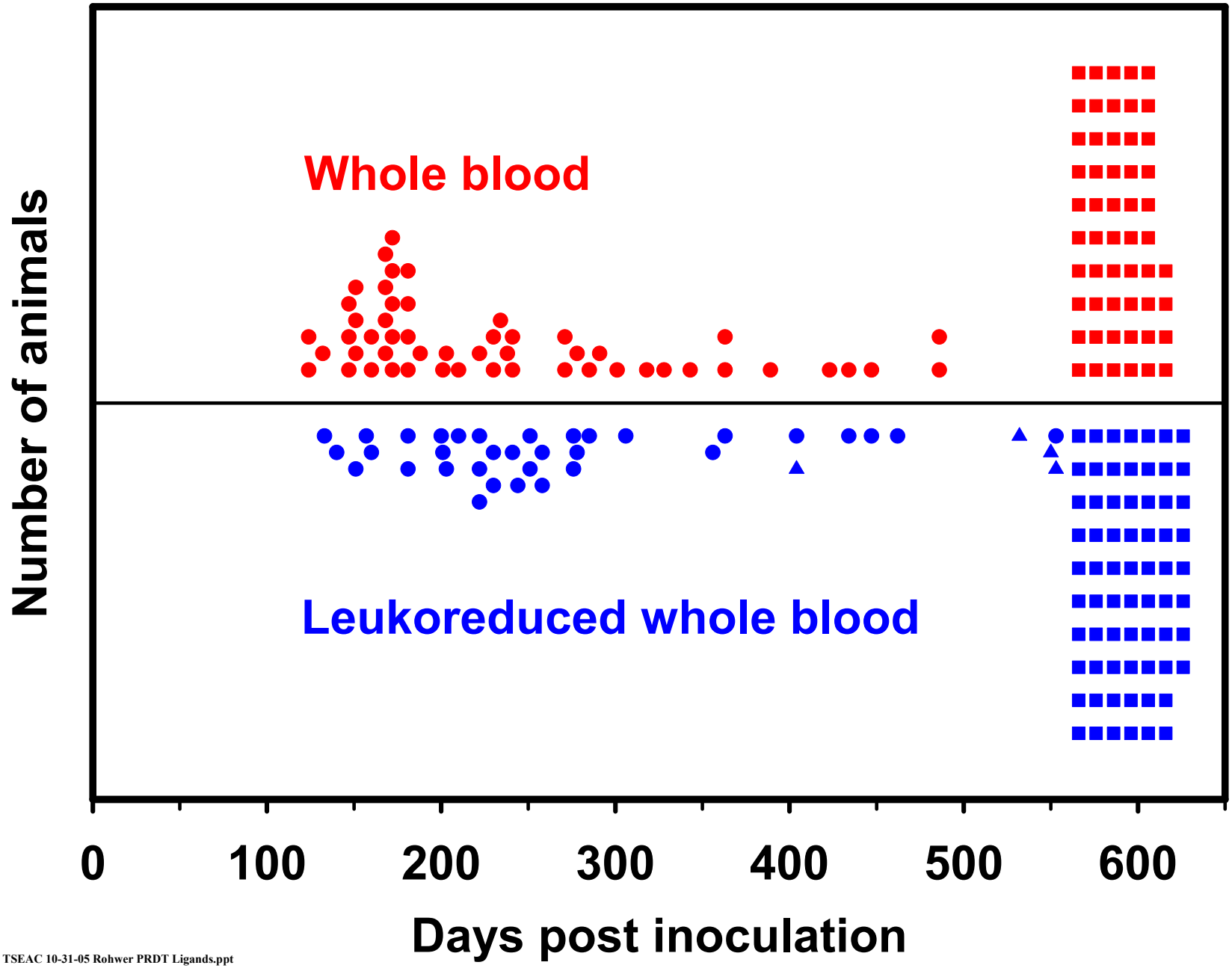
- Blood titer: 10 ID/ml
- Challenge titer: 10^6 ID/ml
- Non-removable: 20 ID/ml

Margin of Safety

Tissue	Infectivity Titer	
Brain Spike	1,000,000.	ID/ml
Residua	20.	ID/ml
Blood	10.	ID/ml
Residua	0.0002	ID/ml

Leukoreduction by Pall WBF2

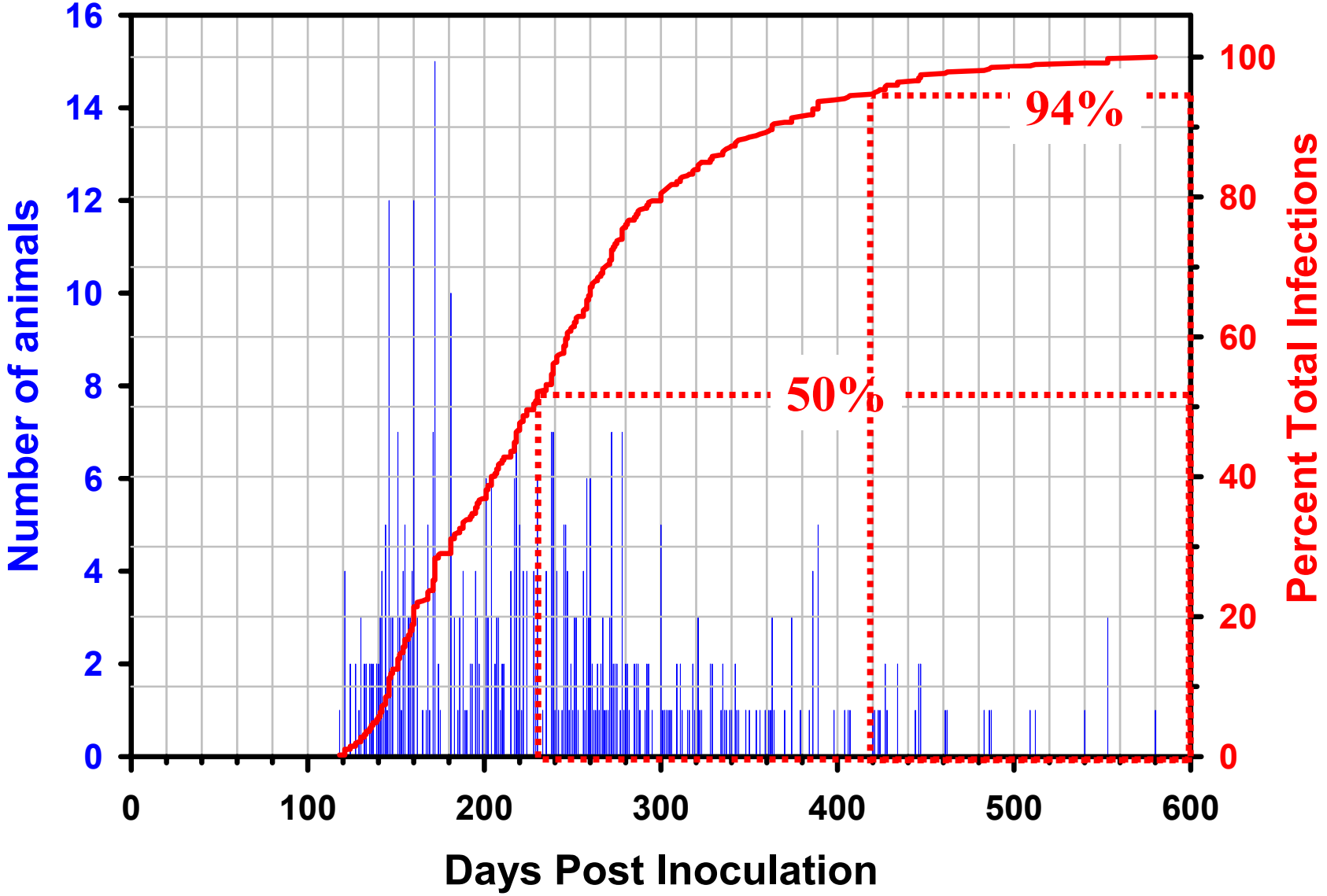



Whole blood
Leukoreduced whole blood
Number of animals
0
100
200
300
400
500
600
Days post inoculation

Endogenous Infectivity Removal

- Challenge
  - Leukofiltered Whole Blood
    - Expect ~ 6 ID/ml
  - Not Leukofiltered RBC
    - Expect ~ 0.72 ID/ml
- Sufficient titer for measurable effect
- Precedent experiment
- Current progress: 420 days / 550 days

Incubation Times of Infections at Limiting Dilution



Endogenous Infectivity Experiment

Infection Status
Sept 12, 2005
~420 days post inoculation

Whole Blood

Not Leukoreduced
414 days
SSSSSSSSSS
SSSSSSSSS●
●●●●●●●●●●
●●●●●●●●●●
●●●●●●●●●

Challenge Leukoreduced
414 days
SSSSSSSSSS
SSD●●●●●●●
●●●●●●●●●●
●●●●●●●●●●
●●●●●●●●●●
●●●●●●●●●●
●●●●●●●●●●
●●●●●●●●●●
●●●●●●●●●●
●●
392 days
●●●●●●

RESIN

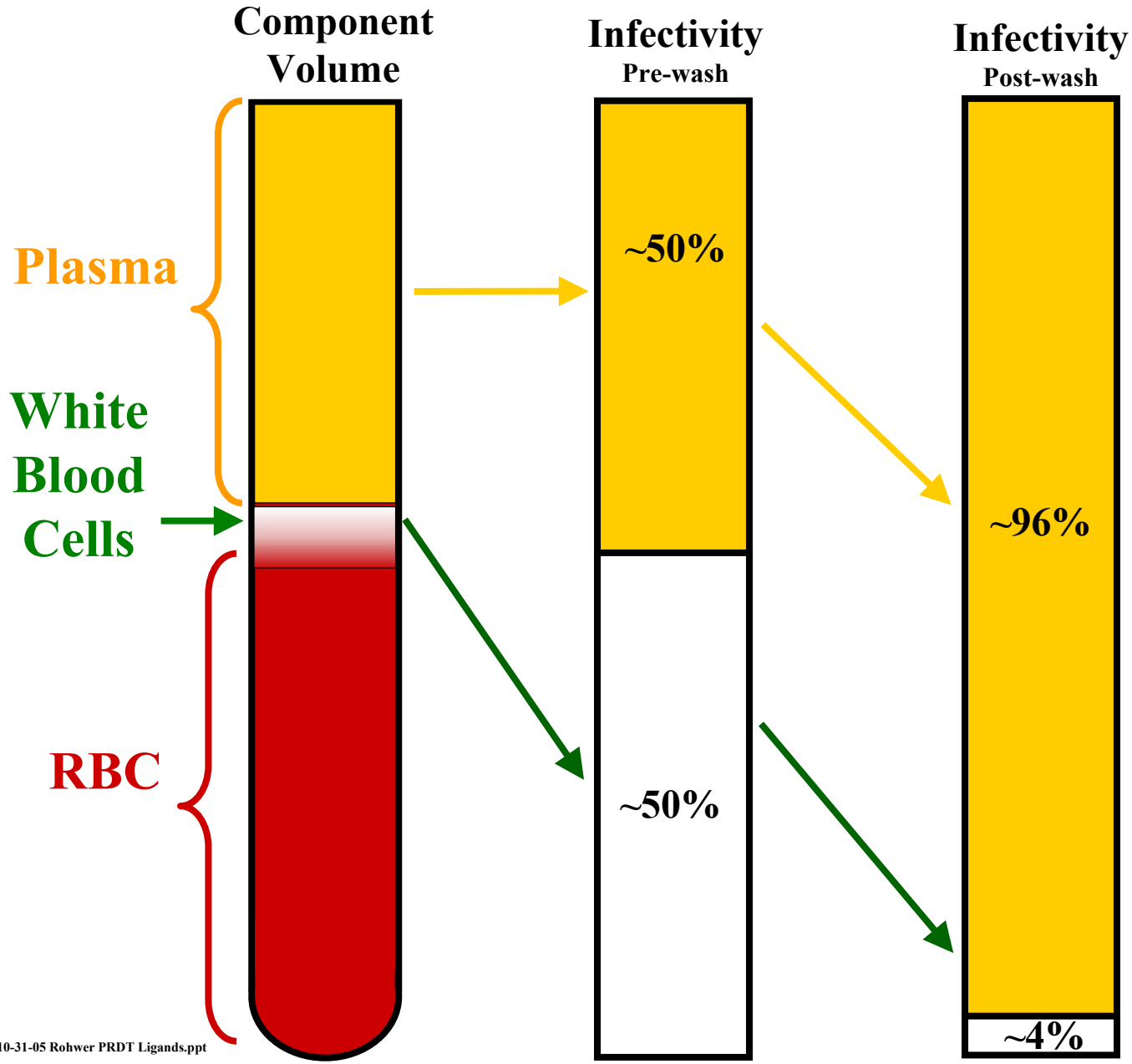
FINAL FLOW THROUGH
420 days
DD●●●●●●●●
●●●●●●●●●●
●●●●●●●●●●
●●●●●●●●●●
●●●●●●●●●●
●●●●●●●●●●
●●●●●●●●●●
●●●●●●●●●●
●●●●●●●●●
392 days
●●●●●●●●●●
●

Endogenous Infectivity Experiment

	Whole Blood		RESIN
	Not Leukoreduced 414 days SSSSSSSSSS SSSSSSSSS● ●●●●●●●●●● ●●●●●●●●●● ●●●●●●●●●	Challenge Leukoreduced 414 days SSSSSSSSSS SSD●●●●●●● ●●●●●●●●●● ●●●●●●●●●● ●●●●●●●●●● ●●●●●●●●●● ●●●●●●●●●● ●●●●●●●●●● ●●●●●●●●●● ●● 392 days ●●●●●●	FINAL FLOW THROUGH 420 days DD●●●●●●●● ●●●●●●●●●● ●●●●●●●●●● ●●●●●●●●●● ●●●●●●●●●● ●●●●●●●●●● ●●●●●●●●●● ●●●●●●●●●● ●●●●●●●●● 392 days ●●●●●●●●●● ●
94% Complete	19/50	12/99	0/100
100% Complete	20/50	13/99	0/100
Poisson Titer ID/ml	10.2 ± 1.3	2.6 ± 0.3	0
Total Challenge	13.0 ID/5ml		
Reduction	1.1 log10		
%Leukoreduction	75%		

file: FDA TSEAC 10-31-05 Rohwer PRDT Ligands.ppt

Distribution of Infectivity in Blood Components



Distribution of TSE Infectivity in Blood

- Two compartments
  - 50% Plasma
  - 50% White blood cells
- Not intrinsic to platelets or red blood cell
- White blood cell infectivity is easily washed off
  - 96% Plasma
  - 4% Washed White Blood Cells

Summary

- ✓ Lead resin defined
- ✓ High affinity for prion protein (< 10^{-9} Kd)
- ✓ Removes PrP from: rodent brain & human brain
- ✓ Removes scrapie, sCJD, fCJD and vCJD
- ✓ Works in red blood cell concentrate, whole blood and plasma (*in vitro*)
- ✓ 99.99% removal of brain derived infectivity
- ✓ ≥ 1 $\log_{10}$ removal of endogenous infectivity to date
- ✓ No impact on RBC, plasma proteins or platelet activation

Partnership

- Partnership established with MacoPharma

  - One of Europe's leading suppliers of blood collection systems
  - Will manufacture and supply the prion reduction filter

04.30.2004 09:38

The End