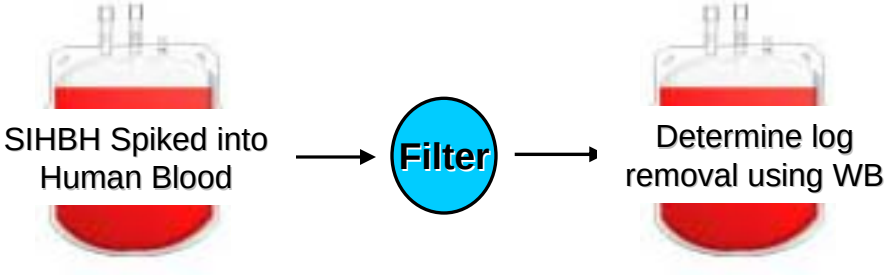
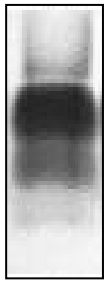
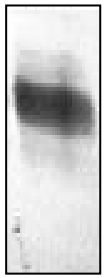
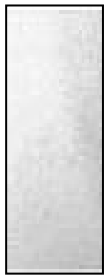
Exogenous Western Blot Studies



Leukocyte Filtration	
Pre Filtration	Post Filtration
	
Associated with 42% reduced infectivity†	

†Gregori *et al.* Lancet 2004 Aug 7;364(9433):529-31

Prototype Design	
Pre Filtration	Post Filtration
	
Removal > 2 Log standard Western blot	

Final Filter Design	
Pre Filtration	Post Filtration
Removal = 2.9±0.7 log reduction (N=48)	

*PTA ppt & densitometry

Bioassay In Hamsters

SIHBH Spiked into Hamster Blood

Filter

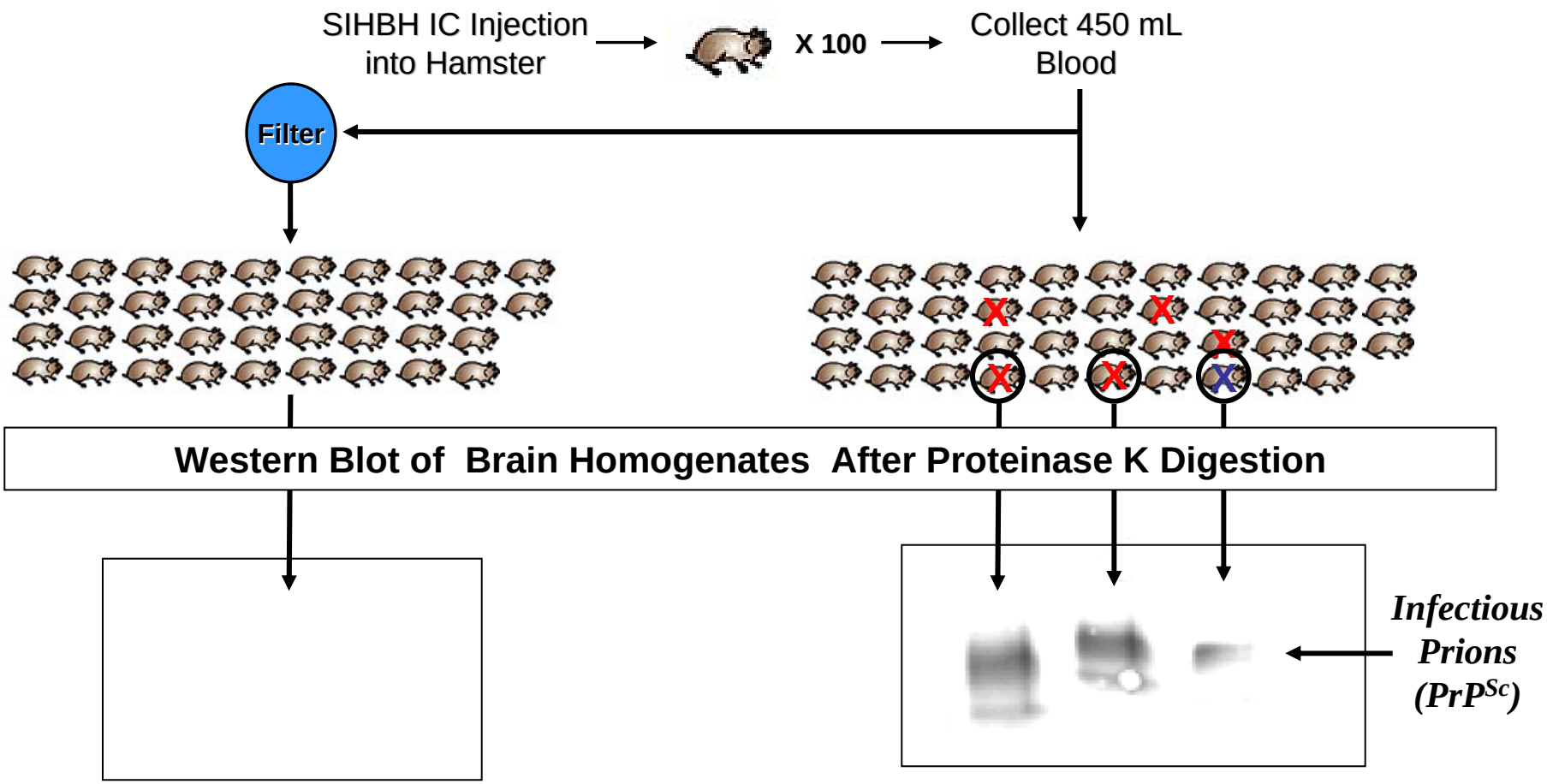
Dilution	$1:10^{-5}$	$1:10^{-6}$	$1:10^{-7}$	$1:10^{-8}$	$1:10^{-9}$
# Hamsters Infected	6/6	6/6	6/6	4/6	0/6
Incubation period	95-100d	108-122d	100-130d	110-160d	>250d

Dilution	$1:10^{-2}$	$1:10^{-3}$	$1:10^{-4}$	$1:10^{-5}$
# Hamsters Infected	6/6	4/6	5/6	0/6
Incubation period	108-122d	115-130d	110-180d	>250d

Total Infectious Prions Removed = 3.7 Log*

* Calculated using the Karber method to account for differences in volume

Infectivity Study In Hamsters

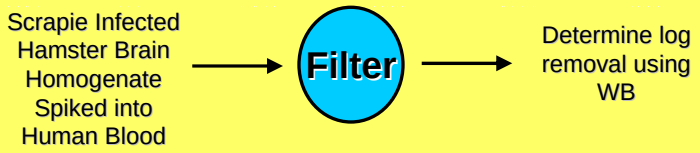
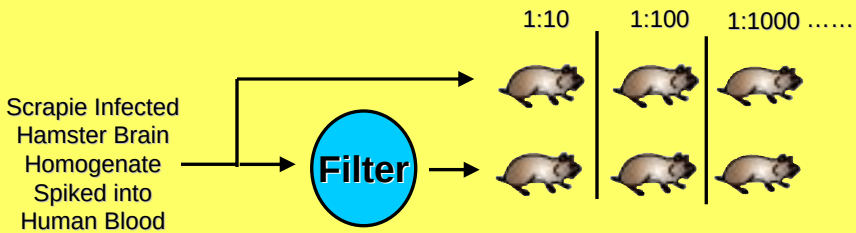
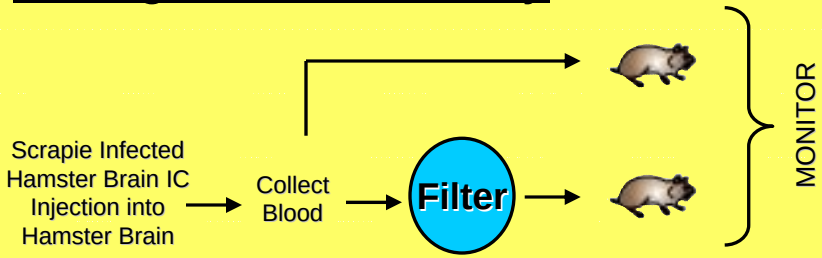


0 out of 38

6 out of 43 hamsters were infected
p=0.0384 (Fisher exact test)

X = Clinical signs of scrapie X = No clinical signs, pathogenic prion found in Western blot only

Studies to Validate Filter Performance

		RESULTS
<u>Exogenous Western Blot</u>  	Screening method used as a first order look to determine prion removal capability	**Prototype**: >2 Log Removal **Final Design**: 2.9±0.7 log reduction
<u>Exogenous Bioassay</u>  	Quantitative method to determine log removal of the infectious prion in an animal model	**Prototype**: 3.7 Log Reduction (N=1) **Final Design**: Ongoing (end '05)
<u>Endogenous Infectivity</u>  	Animal study to determine clinical impact of filter removal performance	**Prototype**: 0/35 versus 6/43 (p=0.0384 Fisher exact) **Final Design**: Ongoing (Spring '06)