Technical and Safety Considerations for TB Challenges in Mice

Hardy Kornfeld, M.D.
Division of Pulmonary and Critical Care Medicine
UMass Medical School
Worcester, MA

Modeling Pulmonary Infection

North RJ. Mycobacterium tuberculosis is strikingly more virulent for mice when given via the respiratory than via the intravenous route. **J. Infect. Dis.** 1995; 172:1550

Cardona PJ, Cooper A, Luquin M et al. The intravenous model of murine tuberculosis is less pathogenic than the aerogenic model owing to a more rapid induction of systemic immunity. **Scan J. Immunol.** 1999; 49:362

Leemans JC, Juffermans NP, Florquin S, et al. Depletion of alveolar macrophages exerts protective effects in pulmonary tuberculosis in mice. **J. Immunol.** 2001; 166:2604.

Modeling Pulmonary Infection

- IV injection
- Nasal instillation
- Tracheal instillation
- Aerosol systems

IV Injection

- Typically < 0.1% seeds the lung
- Systemic immunity is rapidly induced
- Low contamination risk
- Minimal cost and complexity
- Sharps are required

Nasal Instillation

- Typically < 5% delivery to the lung
- Low contamination risk
- Minimal cost and complexity
- Requires anesthesia
- Labor intensive

Tracheal Instillation

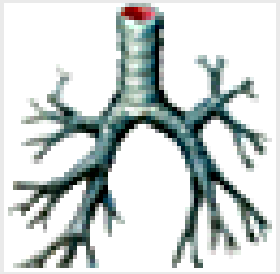
- Indirect or direct approaches
- Requires anesthesia/surgery
- Minimal cost, moderate complexity
- Delivery of bolus to one lung
- Minimal contamination risk, but difficult to operate in a biosafety cabinet
- Labor intensive

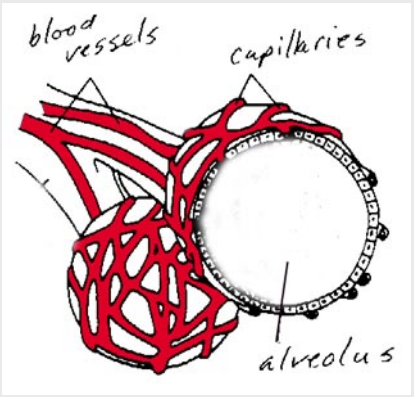
Aerosol Delivery

- Infection is deep and bilateral
- Minimal variation in delivered dose
- High throughput
- Moderate to high contamination risk
- Poor efficiency for large organisms
- High initial cost

Particle Deposition in the Lungs

1. Impaction, sedimentation, diffusion, and dispersion
2. Effects of relative humidity
3. Particle size distributions





> 10 um

5 - 0.1 um

< 0.1 um

Some Aerosol Systems

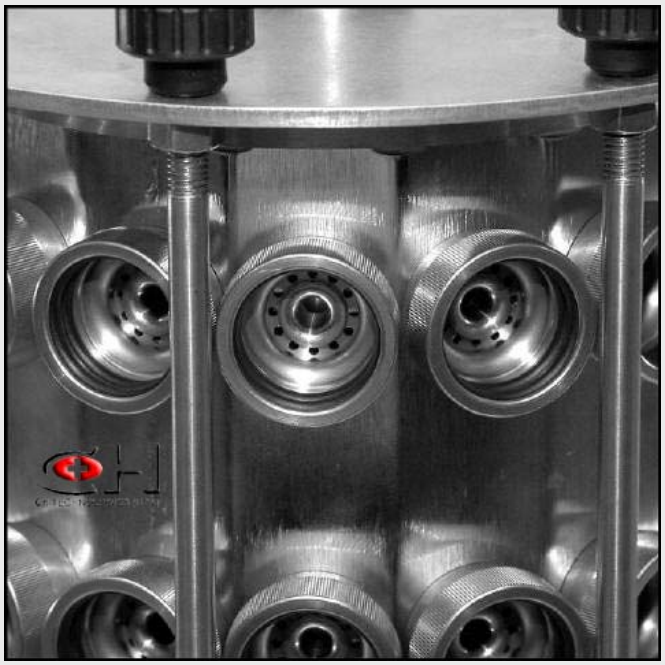
- CH Technologies
- InTox
- Glas-Col
- Madison

CH Technologies

Modular platform
16 ports per row

"Nose-only" exposure

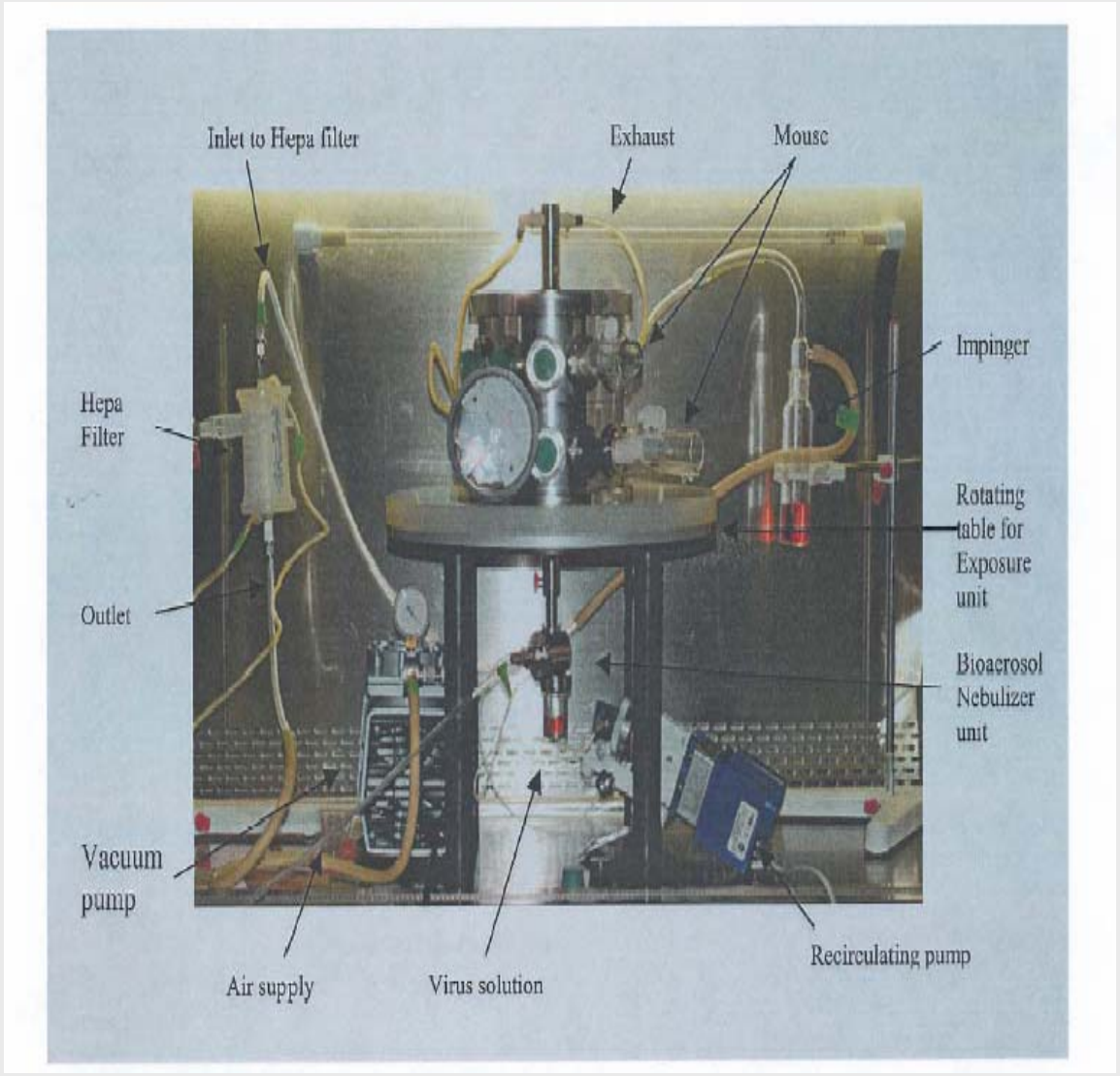
CH Technologies



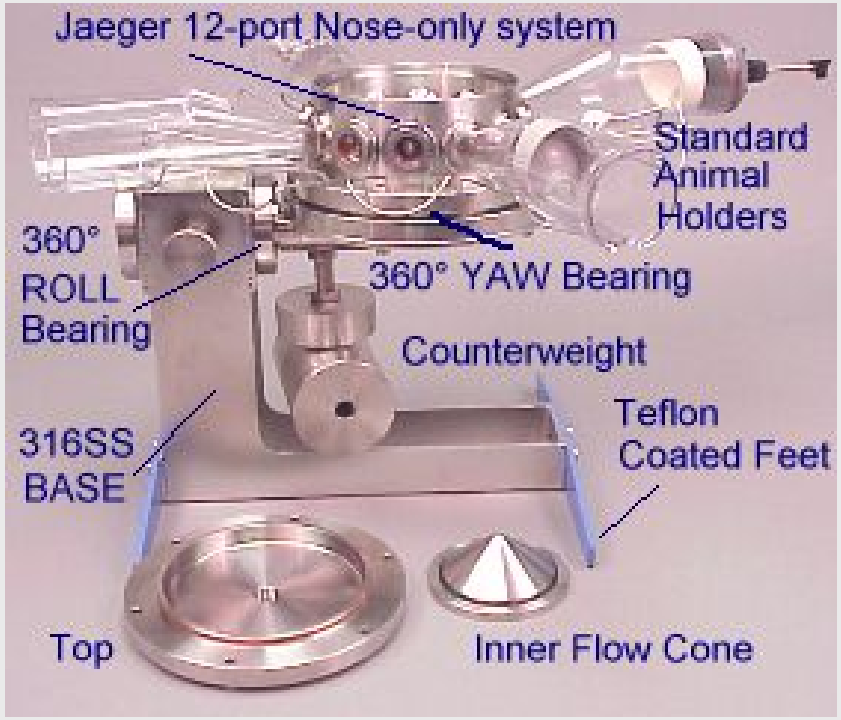
Mouse tube connection

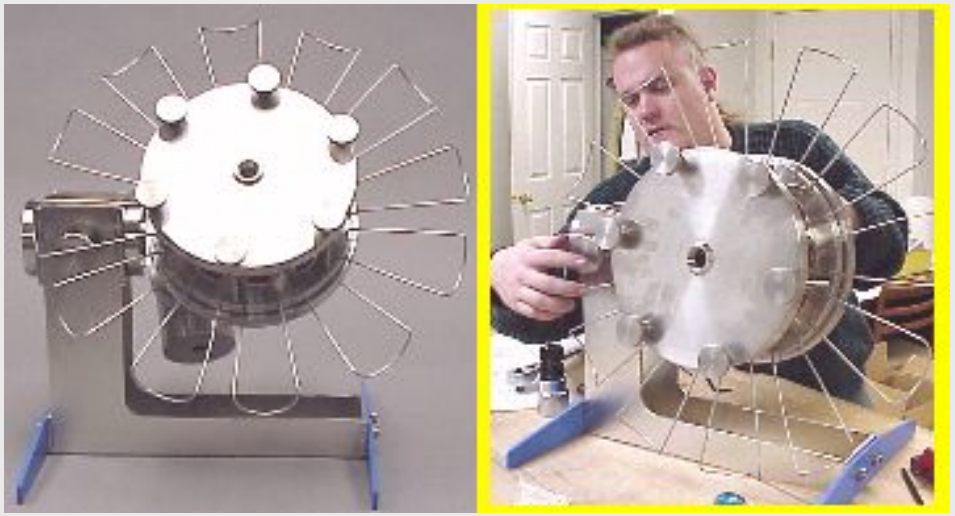
Nebulizer connection

CH Technologies



CH Technologies





High Toxicity Agent System

CH Technologies

Pros

- Modular format
- Custom design
- Nose-only exposure
- Brief run time
- Breaks down for cleaning

Cons

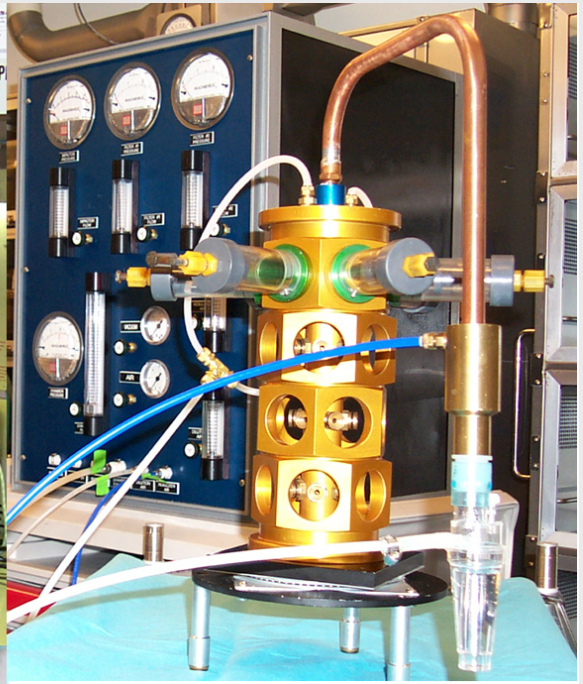
- Not plug-and-play
- Complex assembly
- Requires secondary containment
- May be time consuming to load and operate

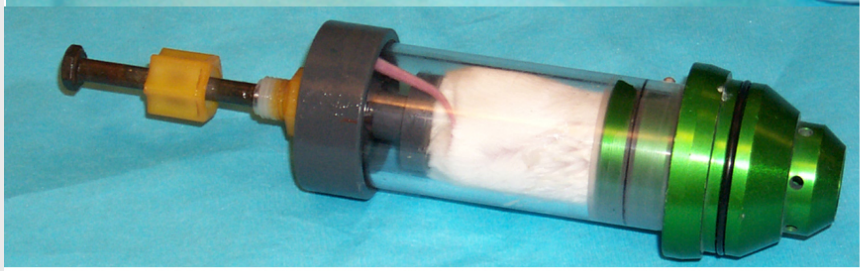
InTox

- Nose-only system
- Requires compressor and vacuum pump
- Requires secondary containment
- In-line HEPA filter
- 24 ports (scalable)

InTox

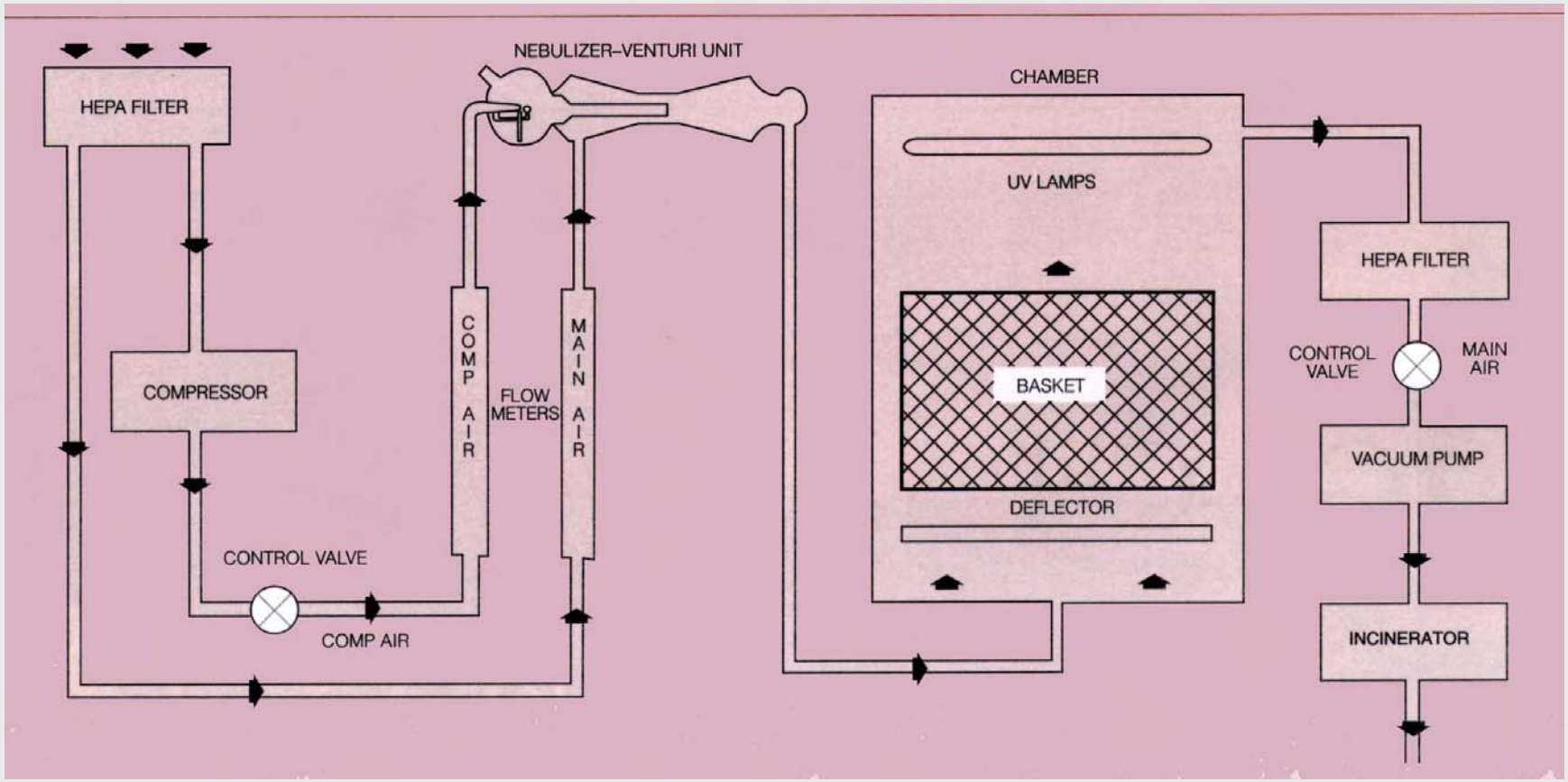
InTox





Glas-Col

Glas-Col


HEPA FILTER
COMPRESSOR
CONTROL VALVE
COMP AIR
NEBULIZER–VENTURI UNIT
COMP AIR
FLOW METERS
MAIN AIR
CHAMBER
UV LAMPS
BASKET
DEFLECTOR
HEPA FILTER
CONTROL VALVE
MAIN AIR
VACUUM PUMP
INCINERATOR

Glas-Col

- Internal basket holds up to 100 mice, 20 rats, or 5 rabbits
- Programmable controller sets pre-heat, nebulizer, cloud decay, and decon cycle length
- 110 or 220 V

Glas-Col

Expt	1	2	3
Mean ± SD	16.8 ± 8.3	87.7 ± 7.1	675.3 ± 76.1
Range	9 - 27	76 - 95	605 - 781
Median	15	85	675

Glas-Col

Pros

- Plug-and-play
- No external connections besides outlet
- Autonomous operation
- High throughput

Cons

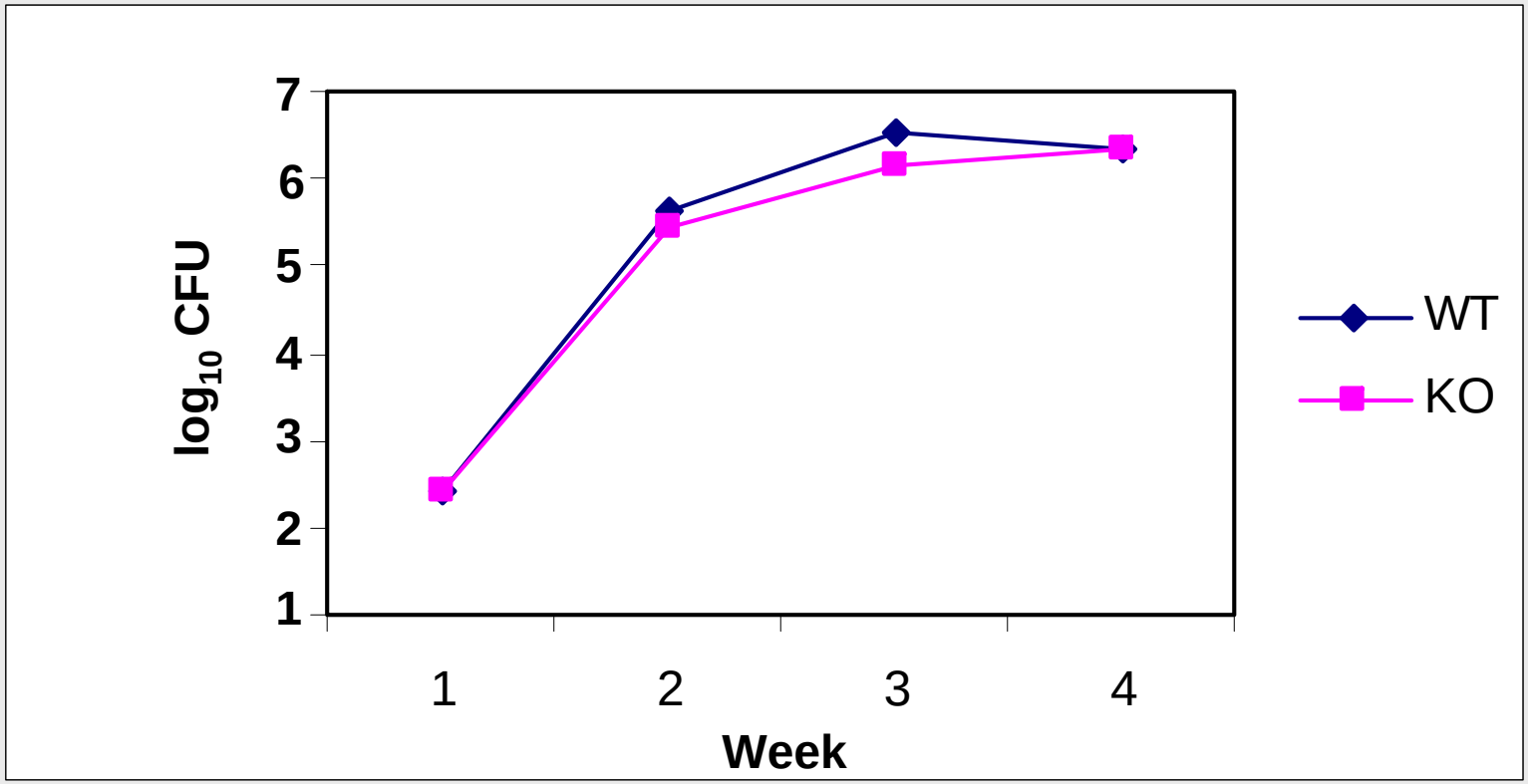
- Whole body exposure
- Exposed glass nebulizer
- Decontamination inconvenient

www.glascol.com

Getting What You Need

- What are the safety issues for your agent?
- What is the physical size of the agent?
- What species do you want to infect, now and in the future?
- What are the biological parameters of your model?
- What is your budget?

Mtb Growth Curve



4 time points, 6 mice per group per time point
Total 48 experimental mice plus 2 dose controls

Setting Up The Lab

- Renovations for ABSL-2/3
- Caging options
- Waste handling
- Proximity to in vitro BL-3 lab
- Animal care personnel
- SOP and surveillance