

ABSA

Anaheim, California

54th Annual Biological Safety Conference
Session XIV: Roundtable-Animal Biosafety

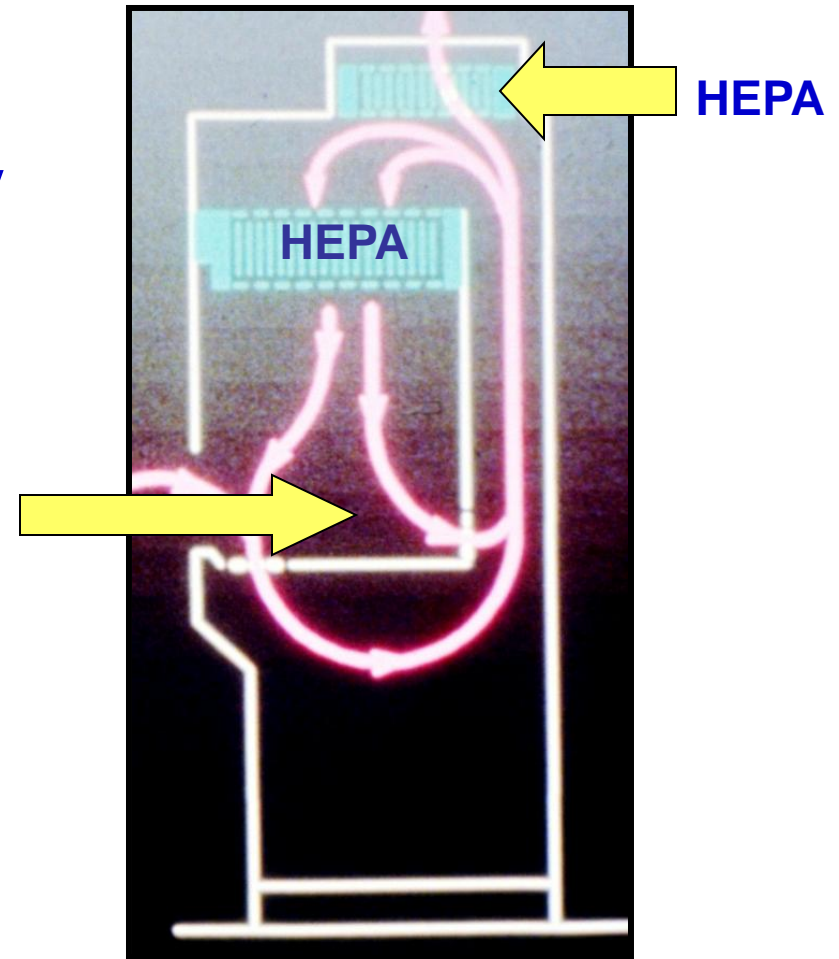
**Prep for Animal Handling, Biocontainment
Systems, & Decontamination Options**

November 2, 2011

Michael G. Sidelsky, Sr., RLATG
Rodent Facility & Housing Specialist
Allentown, Inc.
Allentown, New Jersey
msidelskysr@allentowninc.com

Class II Biosafety Cabinet (BSC)

Personnel, Environment and Product protection provided by HEPA filtration which bathes work area in sterile air



Vertical Flow Biosafety Cabinet

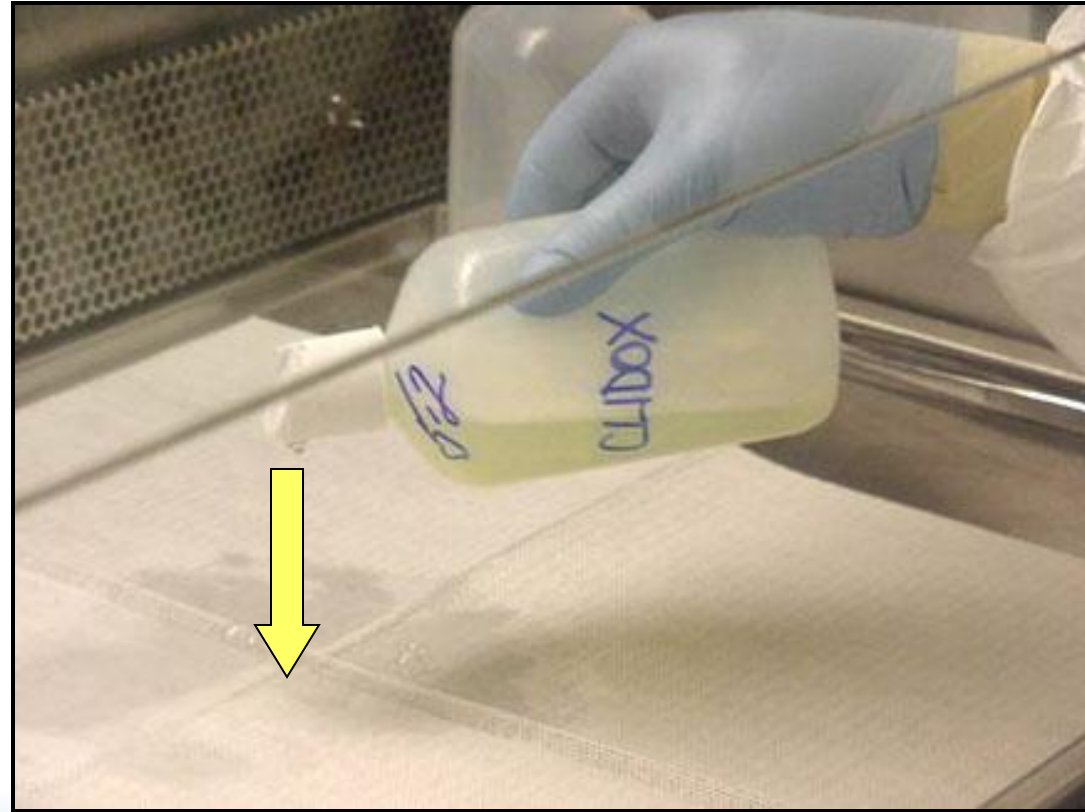
Materials and Methods

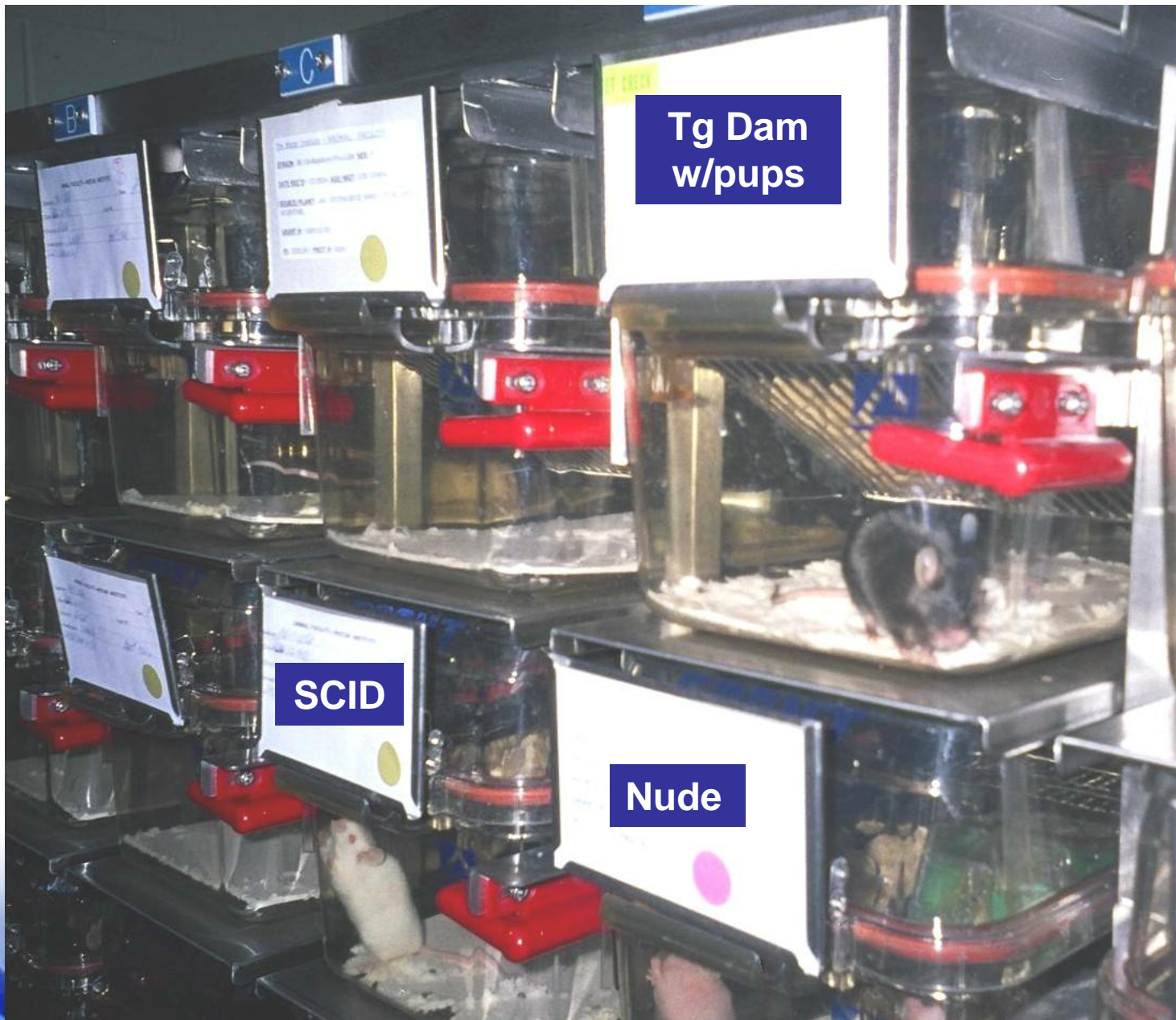
- Sterile or Irradiated cages, water, water bottles, sipper tube/stoppers, micro-isolation tops, wire bar lids, feed, and bedding
- Aseptic technique using Chlorine Dioxide solution, fiber cloth towels, Class II Biosafety Cabinet
- ONE cage at a time for changing or mouse manipulation

First & Second Fiber Towels onto BSC Work Surface



- Preparation of an aseptic field
- Application of Chlorine Dioxide to sport towel
- Use a fresh solution daily

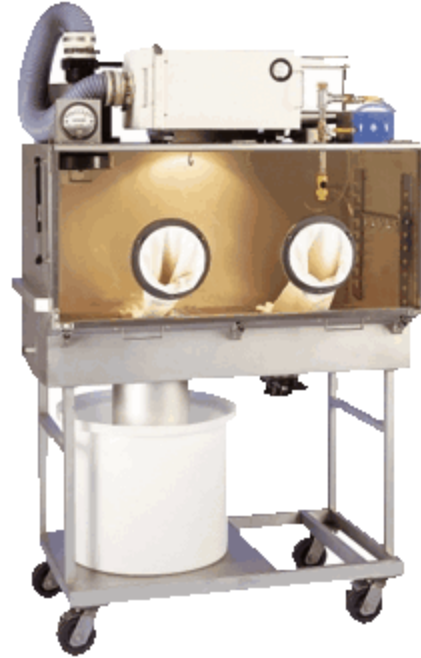




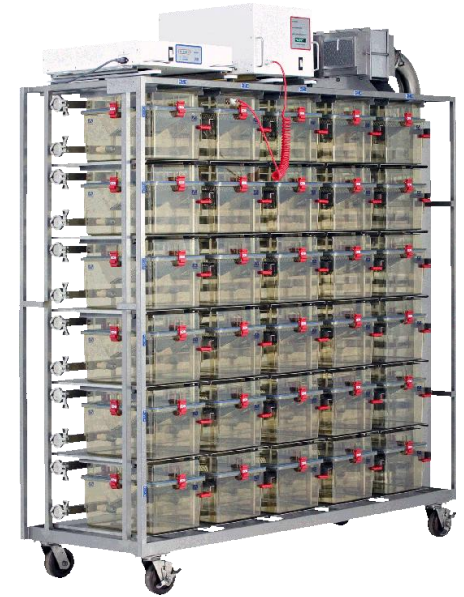
Tg Dam
w/pups

SCID

Nude



Biocontainment Systems

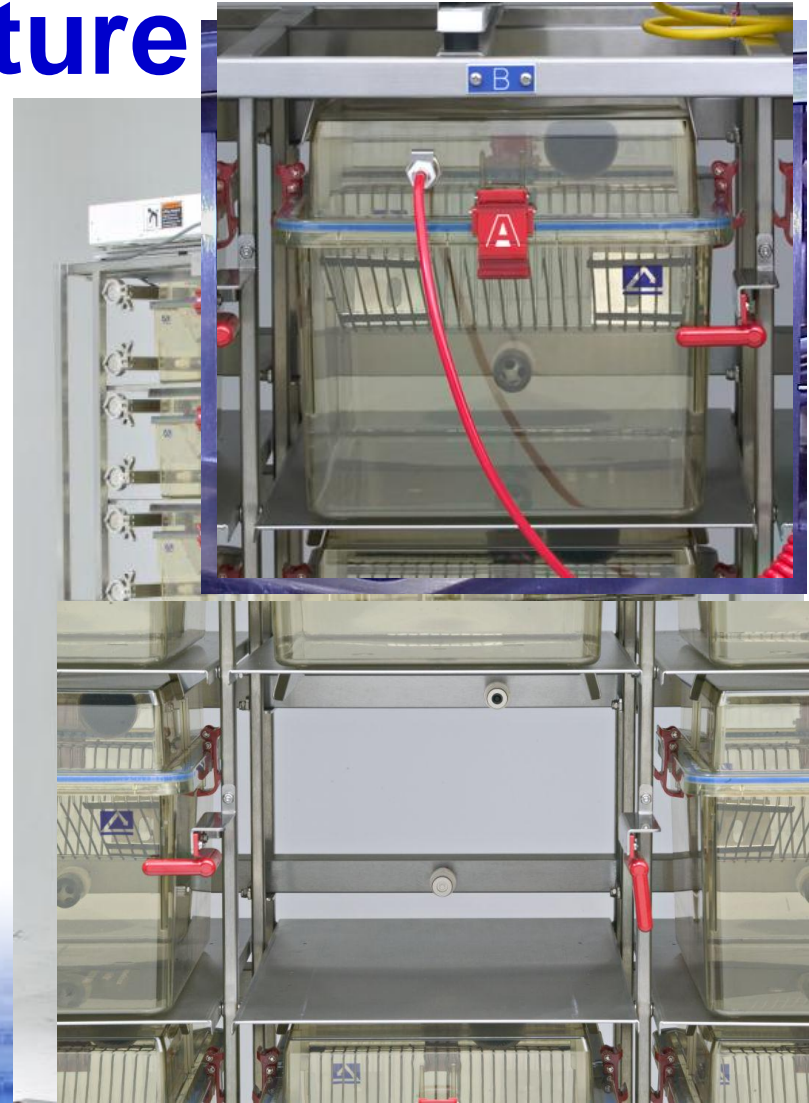


Biocontainment Animal Transfer Unit



Biocontainment Systems Should Feature

- Containment or Quarantine Use
- Rack or wall mounted HEPA assembly must have hard connections
- Single-pass negative only airflow
- Pressurized seals on all manifold connections
- Auto-sealing ports on supply & exhaust
- Cage level lock system
- In-room decontamination and HEPA challenge capability



Sealed Cage Should Feature

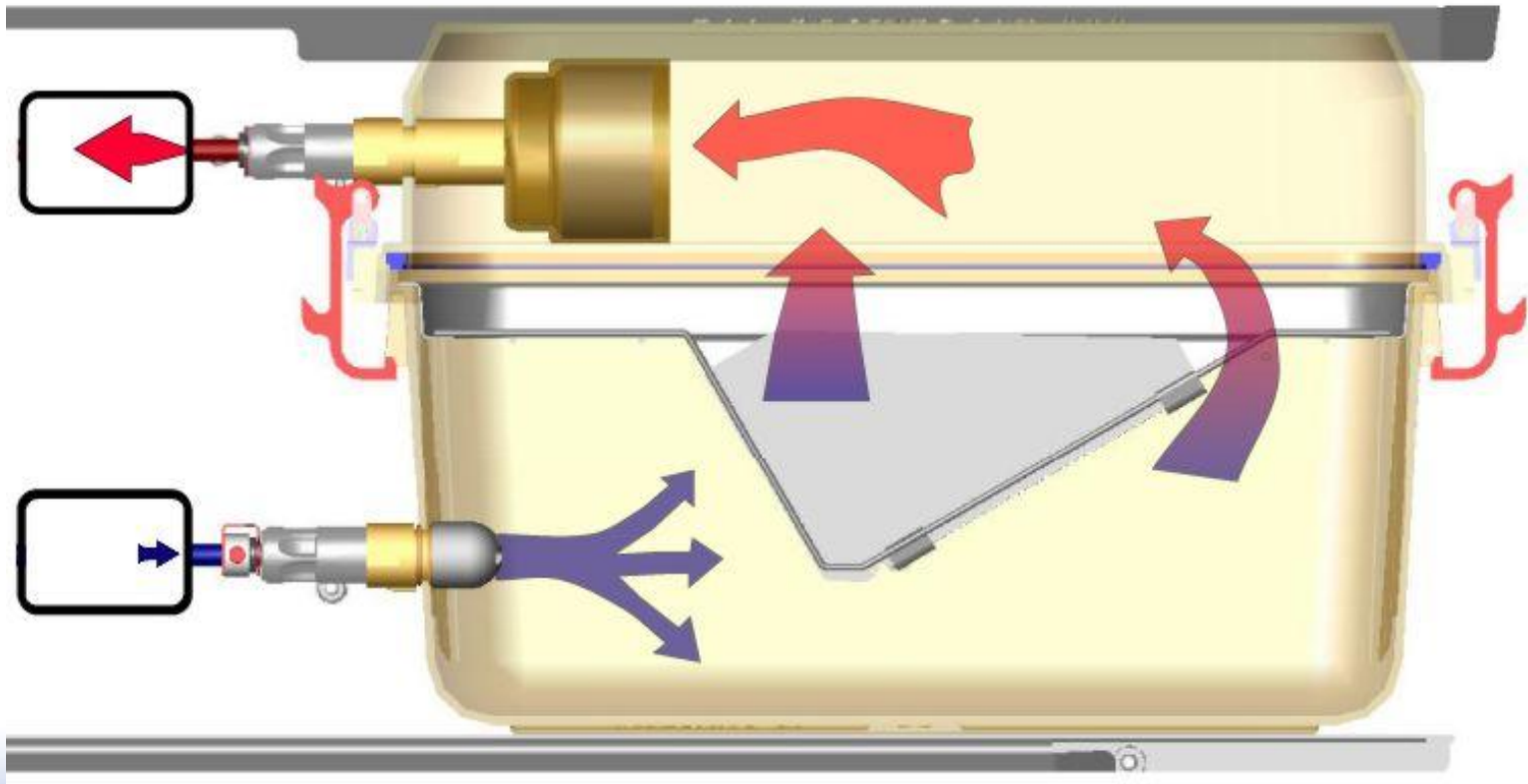
- Auto sealing supply & exhaust ports
- Secure locking mechanisms
- Air tight silicone seal
- Debris Filter
- Autoclavable
- Tunnel washer friendly
- 7-10 day change out
- Bag watering option



Supply Airflow Path



Airflow Design Path



Exhaust Airflow Path

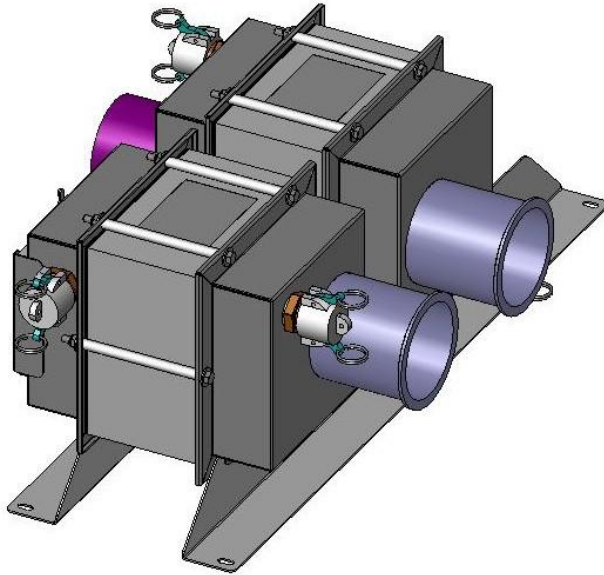


Decontamination Methods

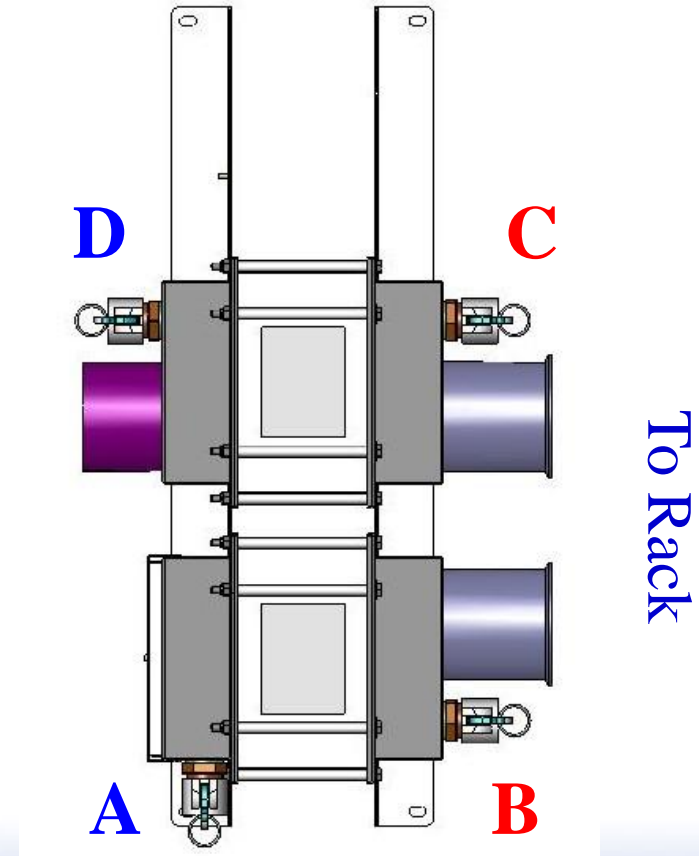
- Vaporized Hydrogen Peroxide *
- Chlorine Dioxide Gas*
- Steam Sterilization *
- Dry Heat Sterilization

* Method demonstrated for following slides

Decontamination & Challenge Ports



| | In | Out |
|-------------------|----------|--------|
| Supply Challenge | Port A | Port B |
| Exhaust Challenge | Port C | Port D |
| Decontamination | Port A/B | Port D |



IVC Biocontainment cage/rack

- Cage level
 - Supply
 - Exhaust
- Rack level
 - Supply
 - Exhaust

VHP or ClO₂ must flow through cage and rack apertures



HEPA Contamination/Decon

- HEPA banjo decon ports
- Decontamination through decon ports and through rack

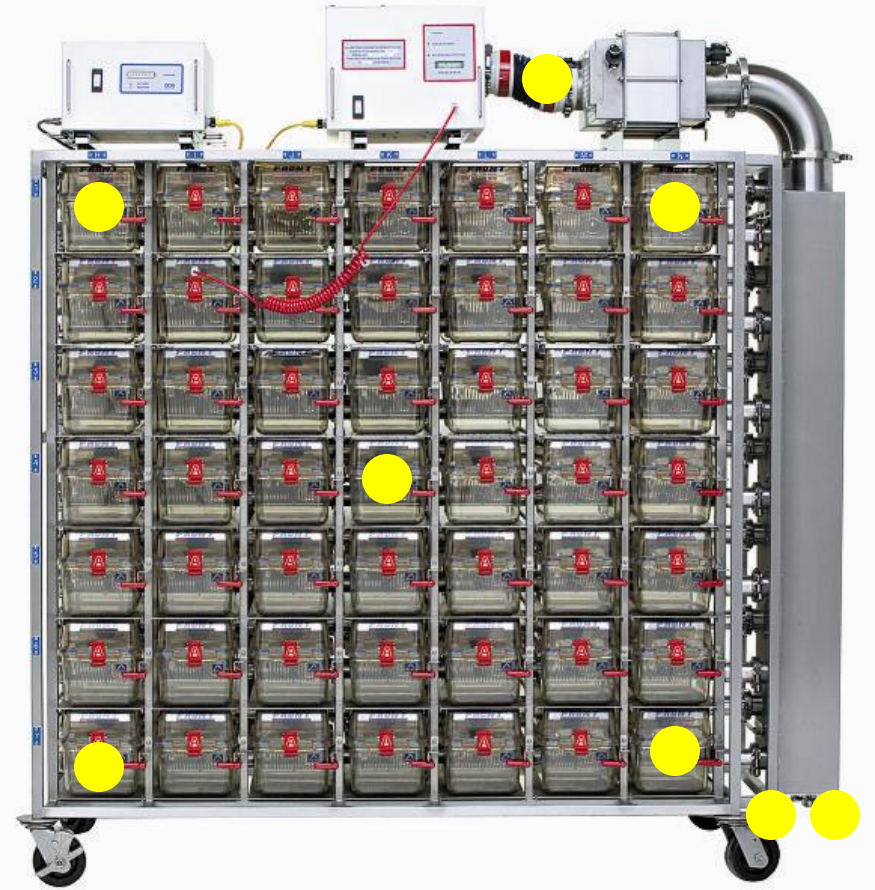


VHP Decontamination



VHP – Validation

- Biological indicators (BIs) located at strategic positions and the use of controls (Geobacillus Stearothermophilus HMV-091) must be incorporated for validation
- After a 7-day incubation, all cultured BIs must be negative except for control (positive for turbidity and precipitate)



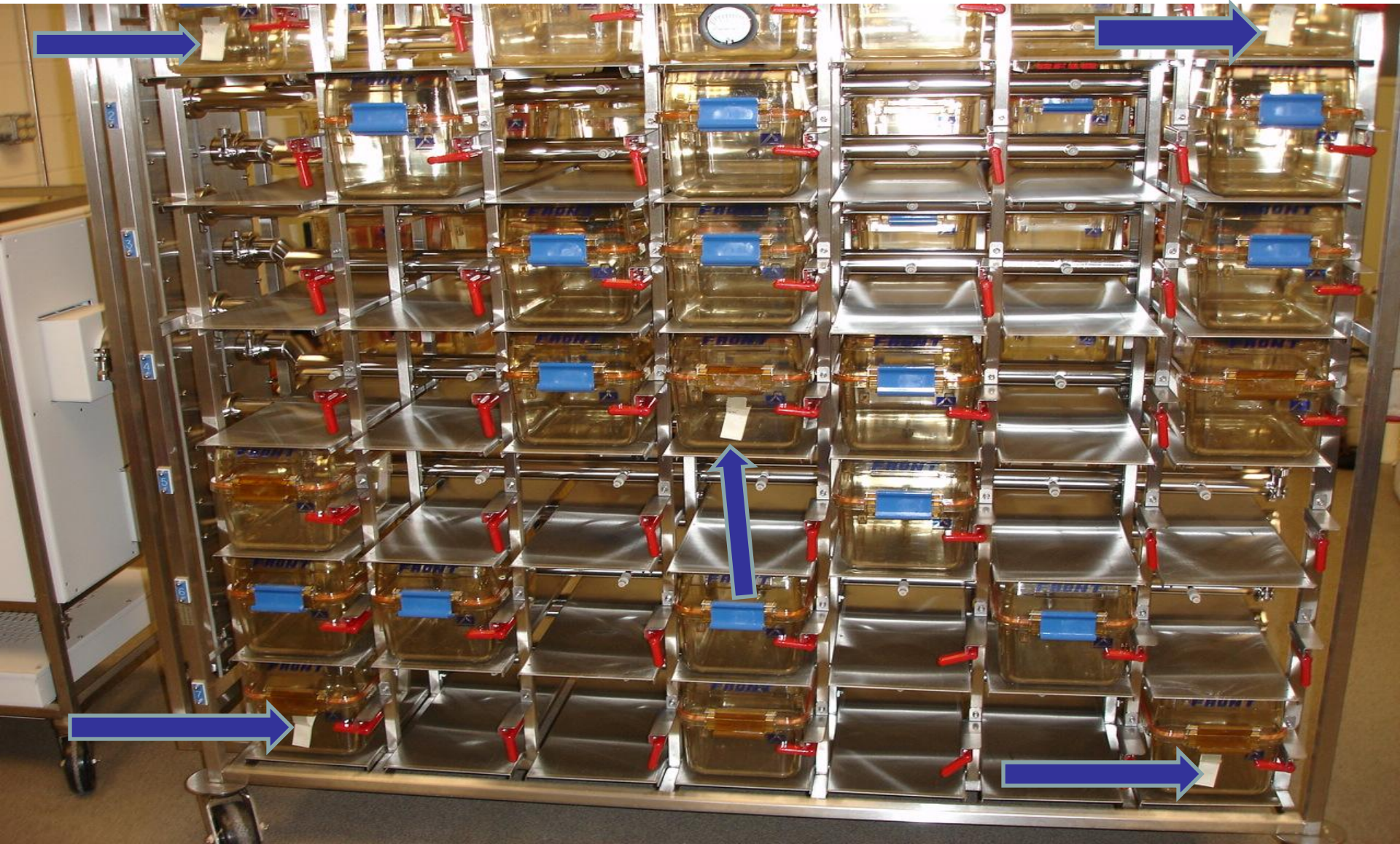


Second Type Of VHP Method For Rack/Cage Level Decontamination

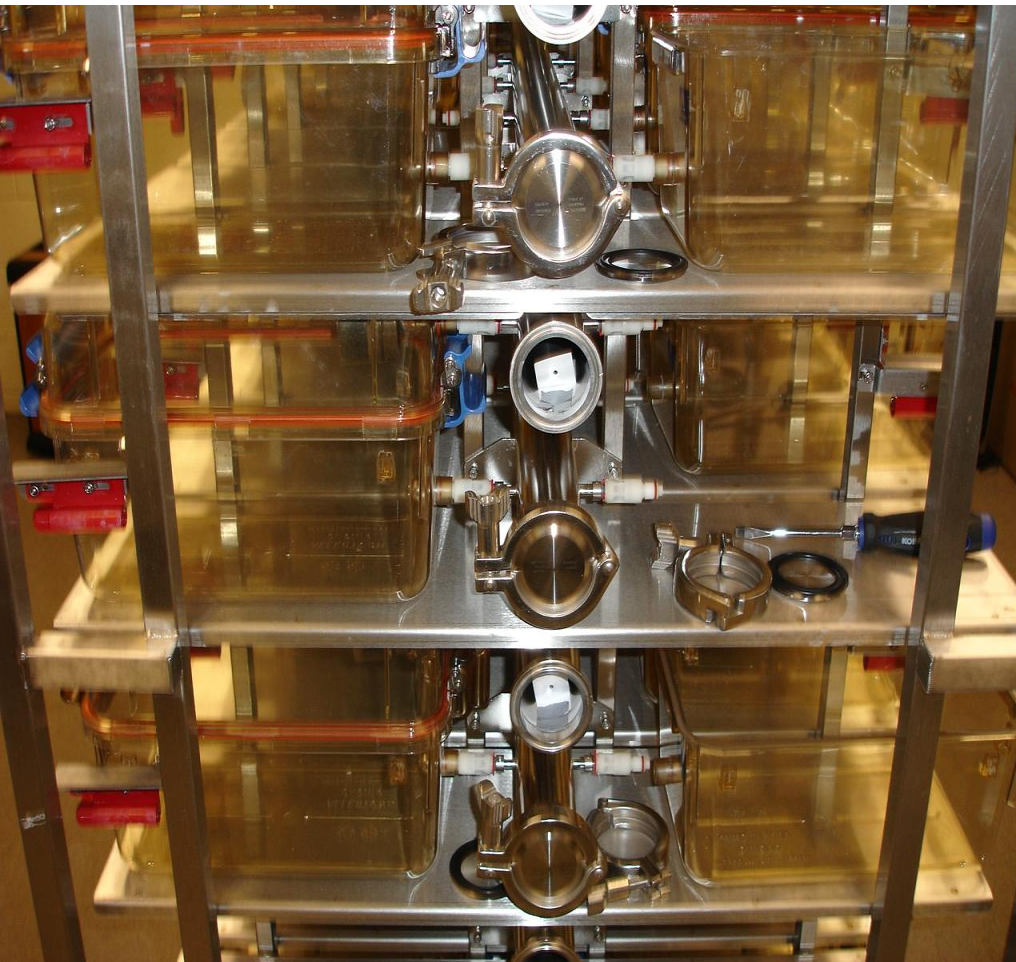
Biological Indicator Placement



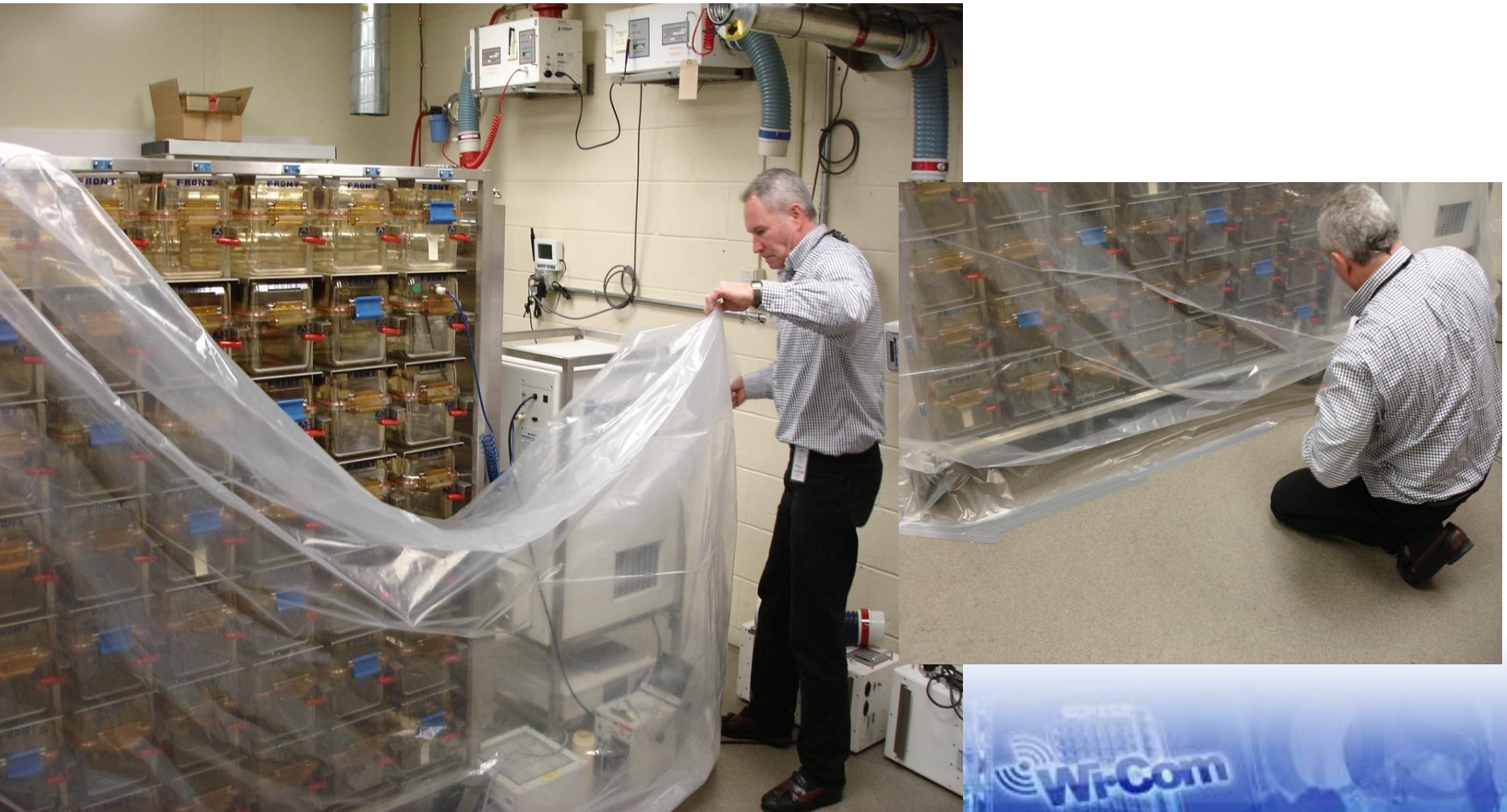
BIs Placed in X Pattern



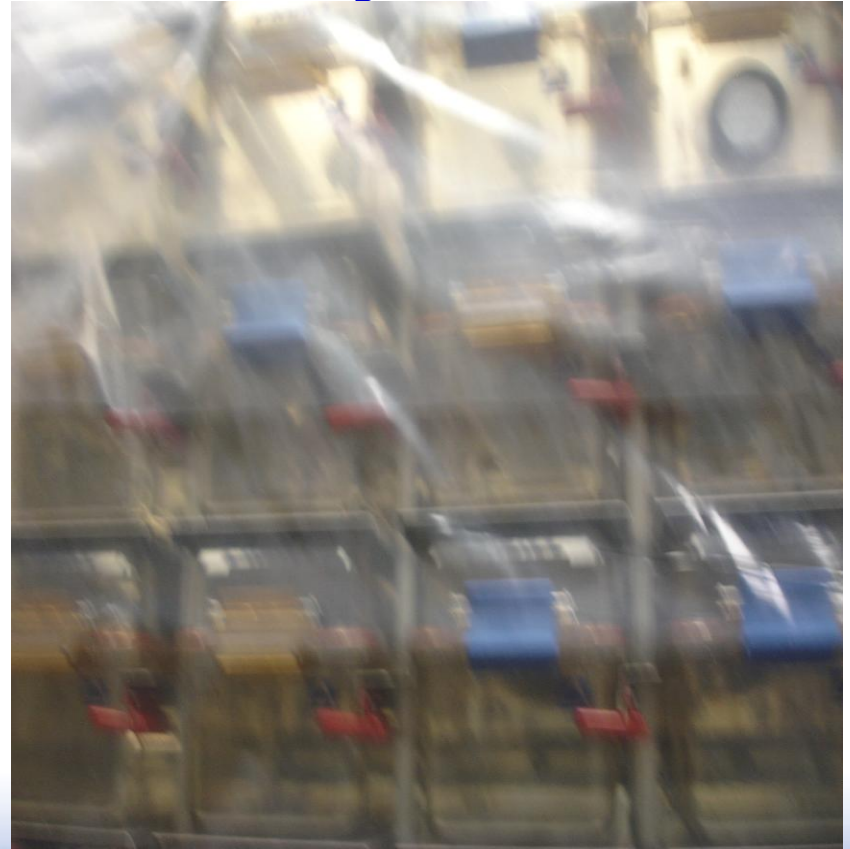
Plenum BI Preparation



Drape & Tape Preparation



Taping & Draping of Entire IVC/Biocontainment System



Monitoring Device for VHP



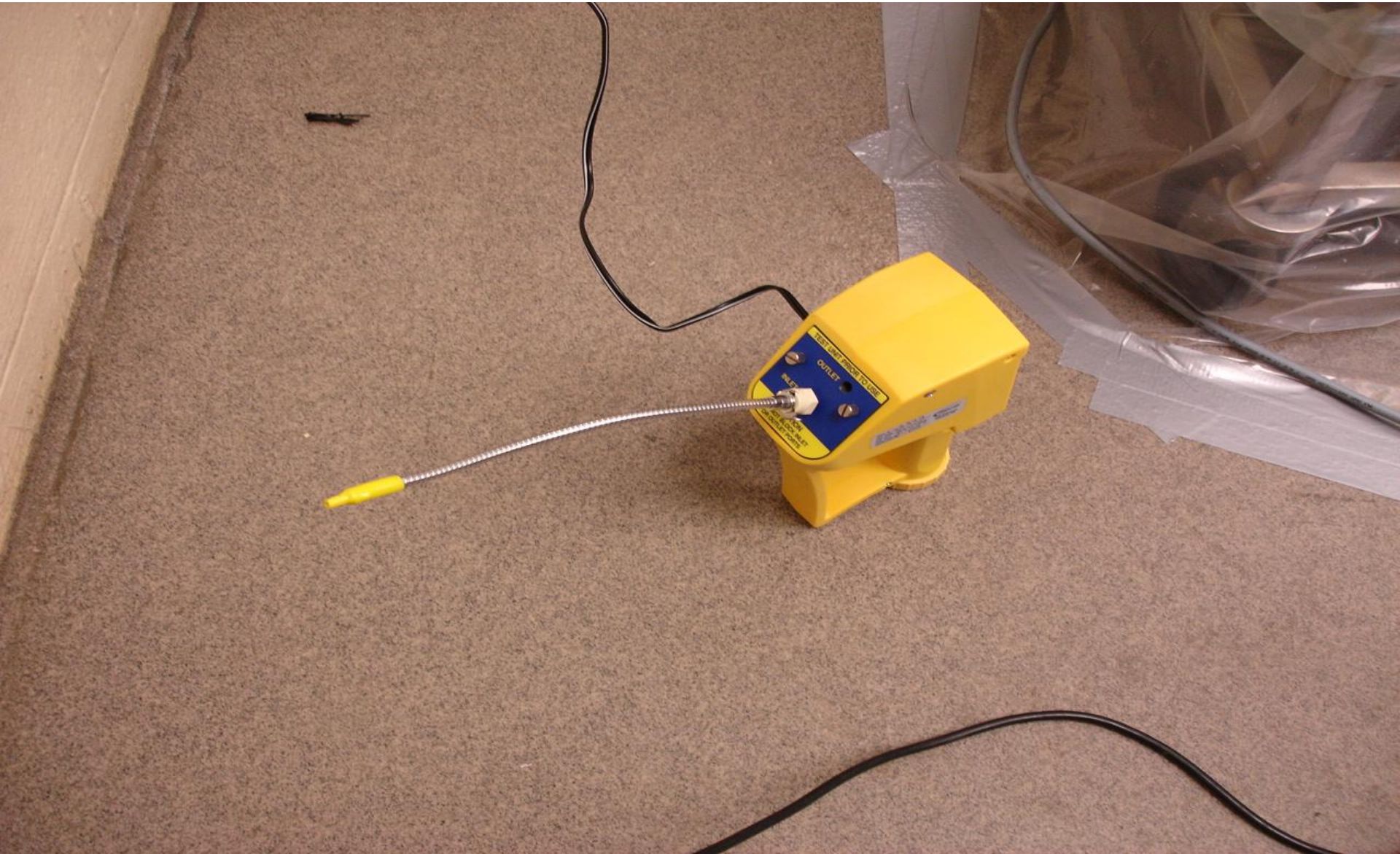
VHP Delivery & Monitoring System



Flex Hose Assembly for Exhausting Vaporized Hydrogen Peroxide



Room Air Monitoring During and After VHP Application



20 TSB VIALS with BI's in Vial
Before Incubation
05-Apr-2007



**20 Biological Indicators
placed in TSB plus one(1)
control for incubation**

After Incubation TSB Vials Showing no
Growth. Control Vial in Front Shows Growth
11-Apr-2007



**After seven(7)days no growth
in all 20 TSB vials that had BIs
from VHP treated cages, rack,
and blower. Only control was
POSITIVE for growth.**

TSB Vial on Left Showing no Growth.
TSB Vial on Right (Control) Showing
Growth
9-Apr-2007

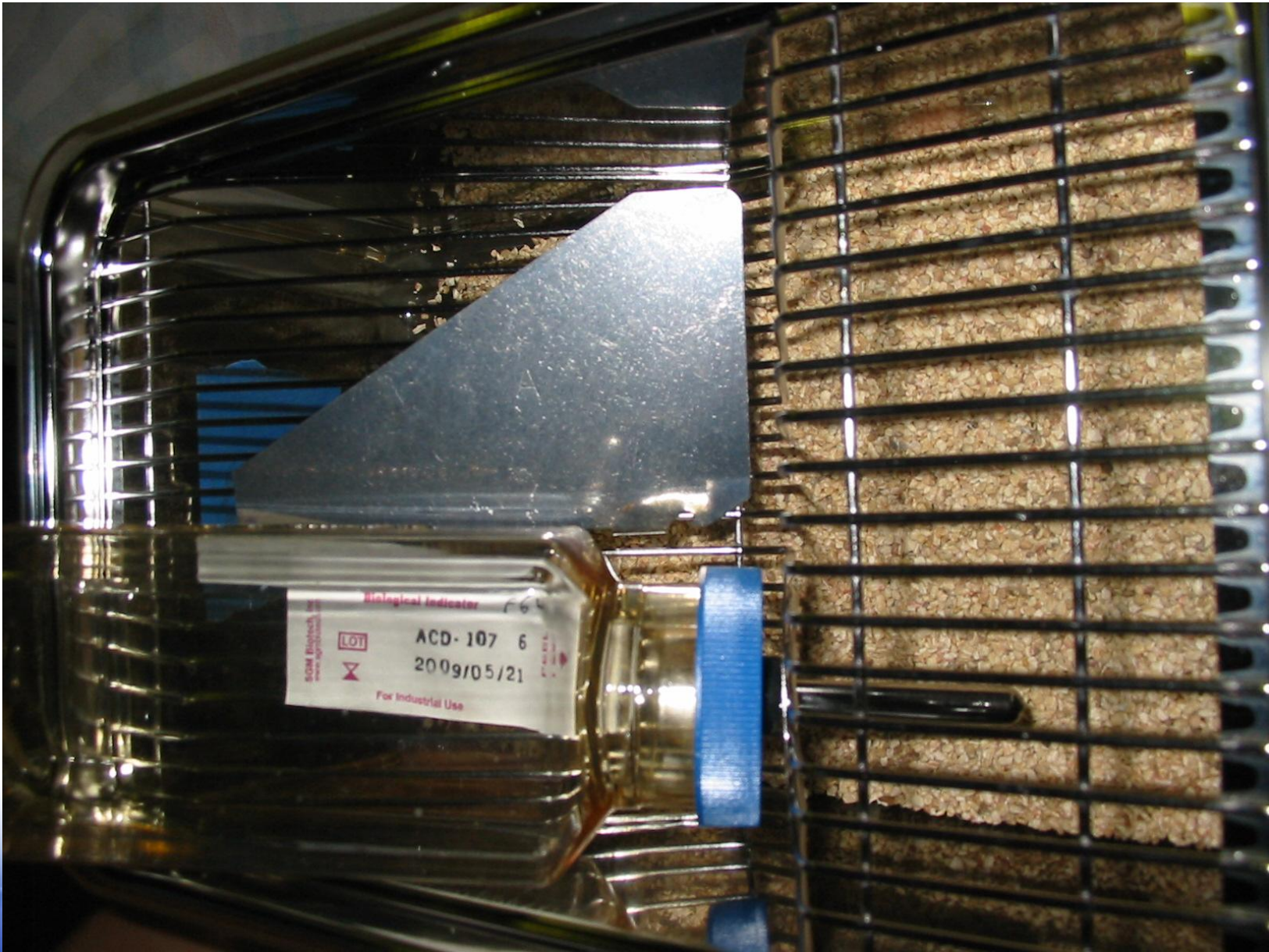


**TSB vial on left
with NO signs of
growth or spore
kill.**

**Vial of TSB on
right with control
BI shows growth
or spores not
killed.**

Chlorine Dioxide Gas Decon

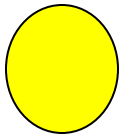
- Preliminary testing showed no growth of *Bacillus Atrophaeus* biological indicators (BIs) in TSB after 7 days
- Over one hundred (100) *B. Atrophaeus* BIs were placed in both the sealed 42 cages and biocontainment system itself, including under the bedding material of each cage, in water bottles, and in horizontal and vertical supply and exhaust plenums



Bacillus Atrophaeus Biological Indicator being placed under bedding material



SEALED ROOM LEVEL CLO2 GAS APPLICATION REMOTE ACTIVATION



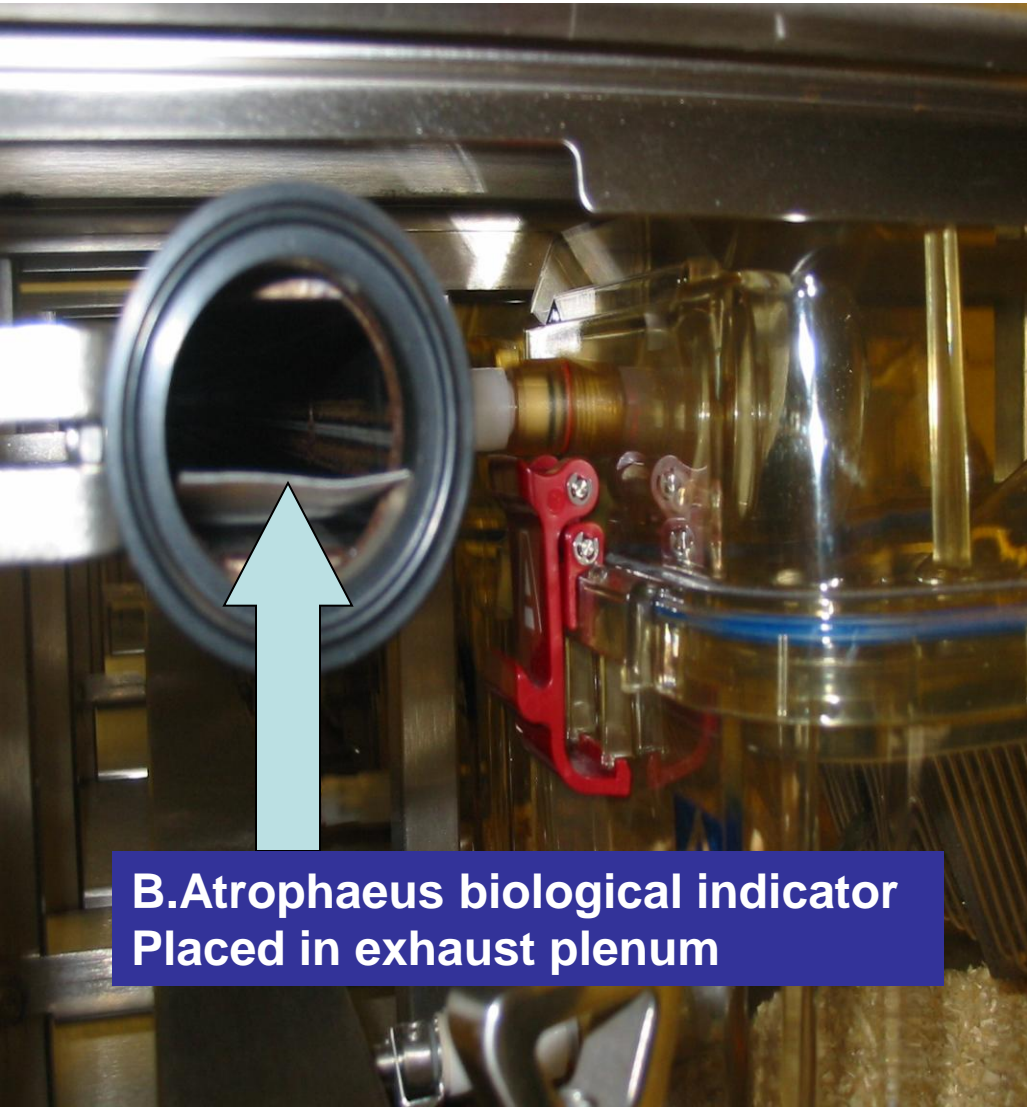
Biological indicator placed inside both the battery box and blower box



BIs Inside Bottles & Under Bedding



BIs Placed inside IVC Supply & Exhaust Plenums



**B. Atrophaeus biological indicator
Placed in exhaust plenum**



Exhaust Plenum BI

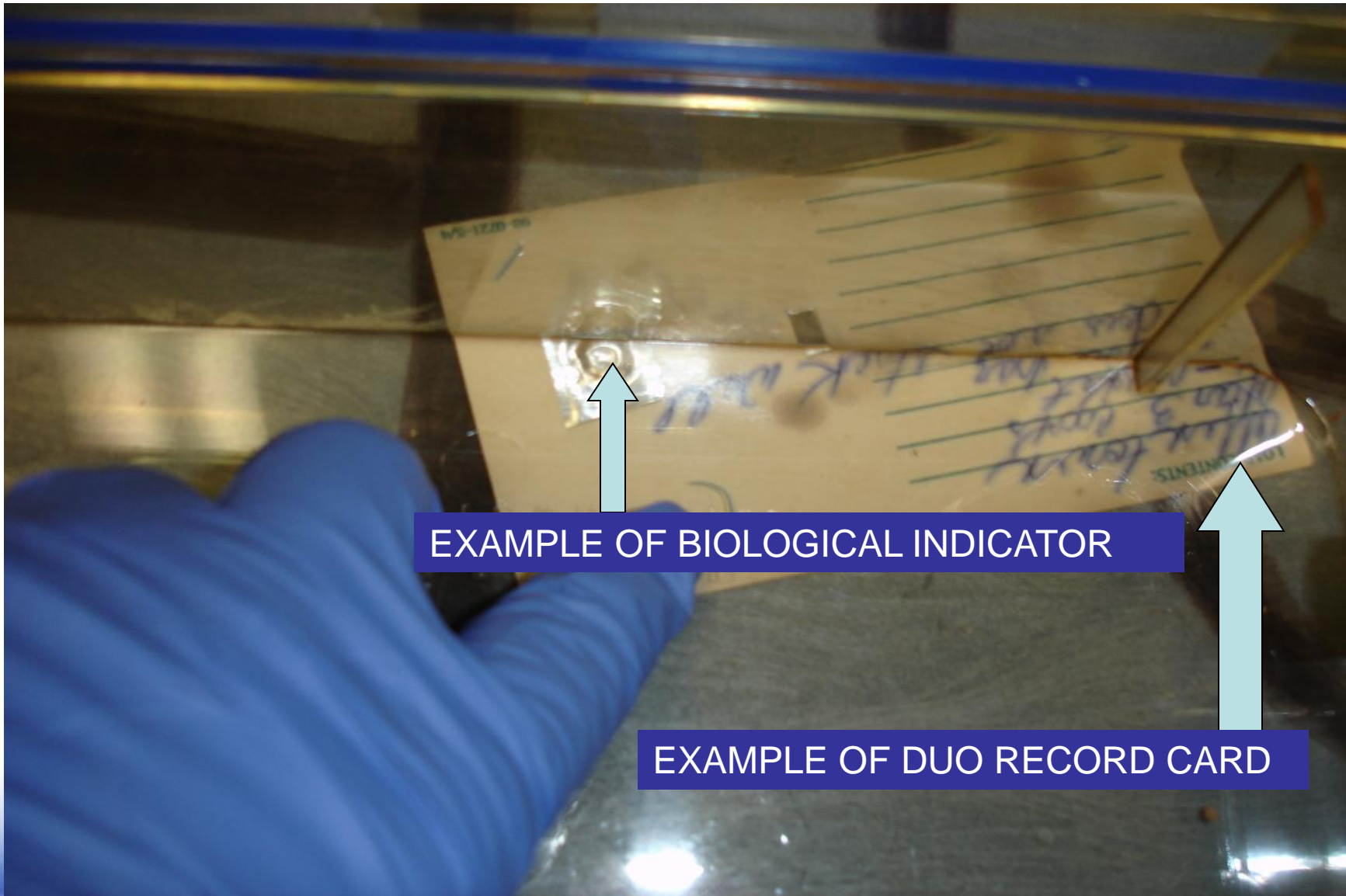
Supply Plenum BI

ClO2 Gas Potential

- Cages, water bottles and rack, as well as the room could be decontamination/sterilized in place via remote activation (plug cord into power source from external area) or provide a quick disconnect for attaching gas dispensing machine
- Sensitive equipment such as computers and other electronic equipment could also be decontaminated in the same room. However, be certain to check with the manufacturer and/or applicator for the correct suggested time, temperature , and humidity settings.

Monitoring Supplies For Steam Autoclaving Validation

- Biological indicator- *Geobacillus Stearothermophilus*
- Autoclave tape
- Duo Record Card- color change shows steam penetration
- Hard copy of cycles from Autoclave printer
 - Documents Pre-Vac (removes air from chamber)
 - sterilization temperature (250 F or 121 C), PSI,
 - exhaust (slow or fast), cool down/dry time, total run time



EXAMPLE OF BIOLOGICAL INDICATOR

EXAMPLE OF DUO RECORD CARD

PACKAGING

- Examples of autoclave packaging

Opaque or Clear Autoclave bag*

Fabric cover

Polyspun cover

Tyvek (use caution due to possible shrinkage)

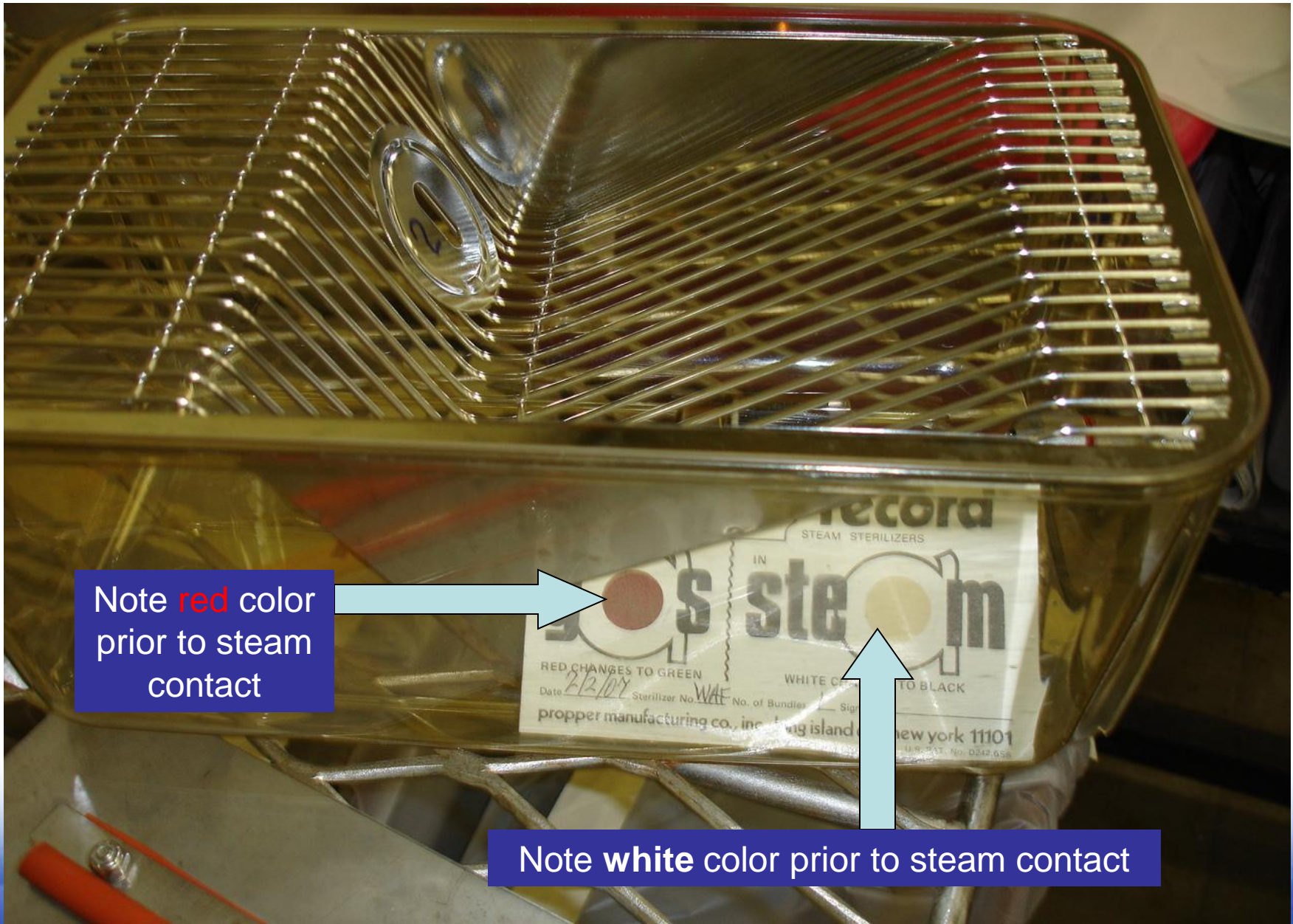
- * BE SURE TO KNOW THE HIGHEST POSSIBLE TEMPERATURE TOLERANCE FOR THE BAG YOU SELECT



OPAQUE STEAM AUTOCLAVE BAG

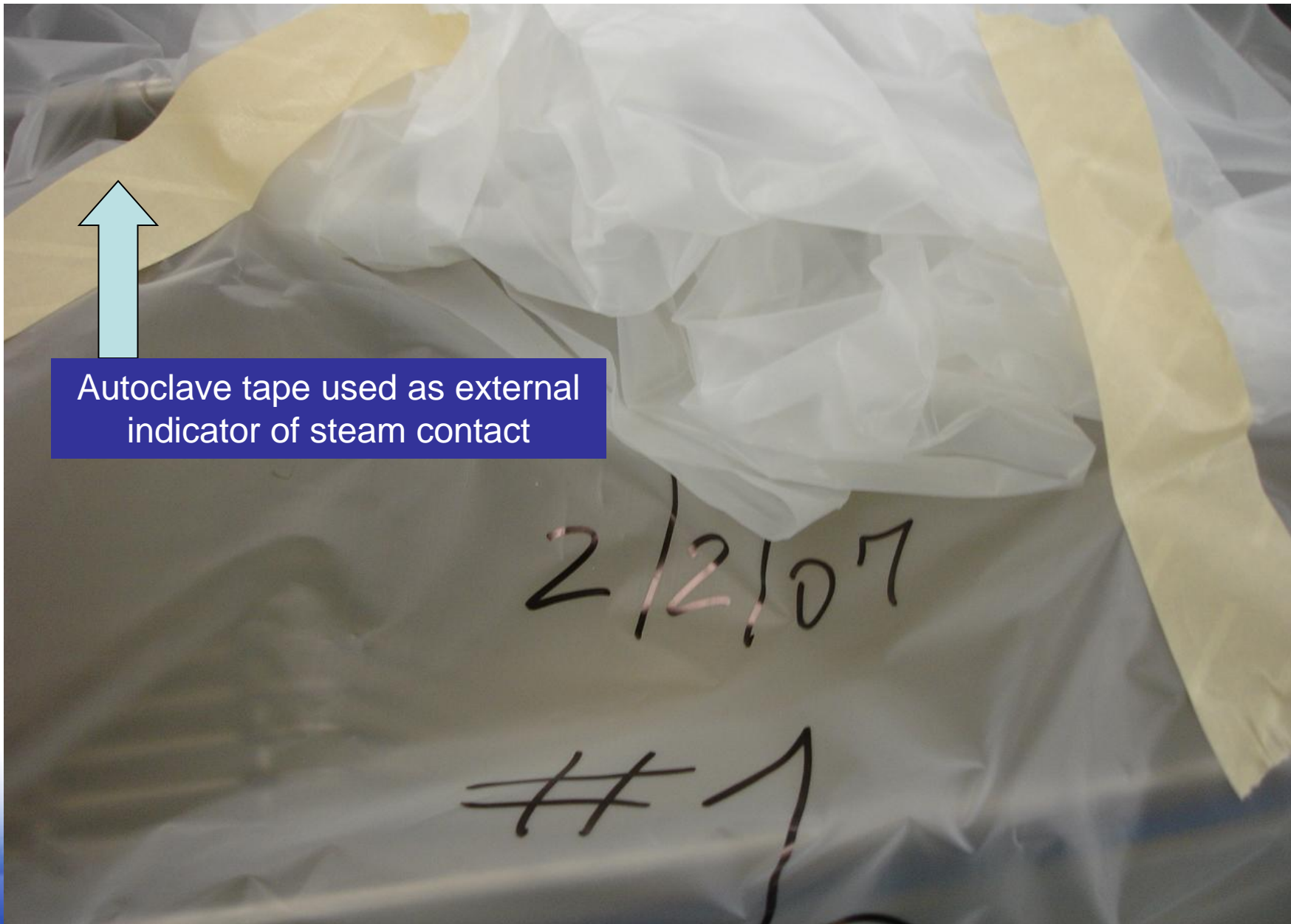
PREPARATION OF ITEMS TO BE AUTOCLAVED

- Bagged items with duo card
- Opaque autoclave bag marked w/sharpie
- Autoclave tape use as indicator & closure



Note red color prior to steam contact

Note white color prior to steam contact



Autoclave tape used as external indicator of steam contact

2/2/07

#1

Five items post autoclaving all with external steam contact tape indicators
Changed from clear to **BLACK**



Note color change to
BLACK
After autoclaving

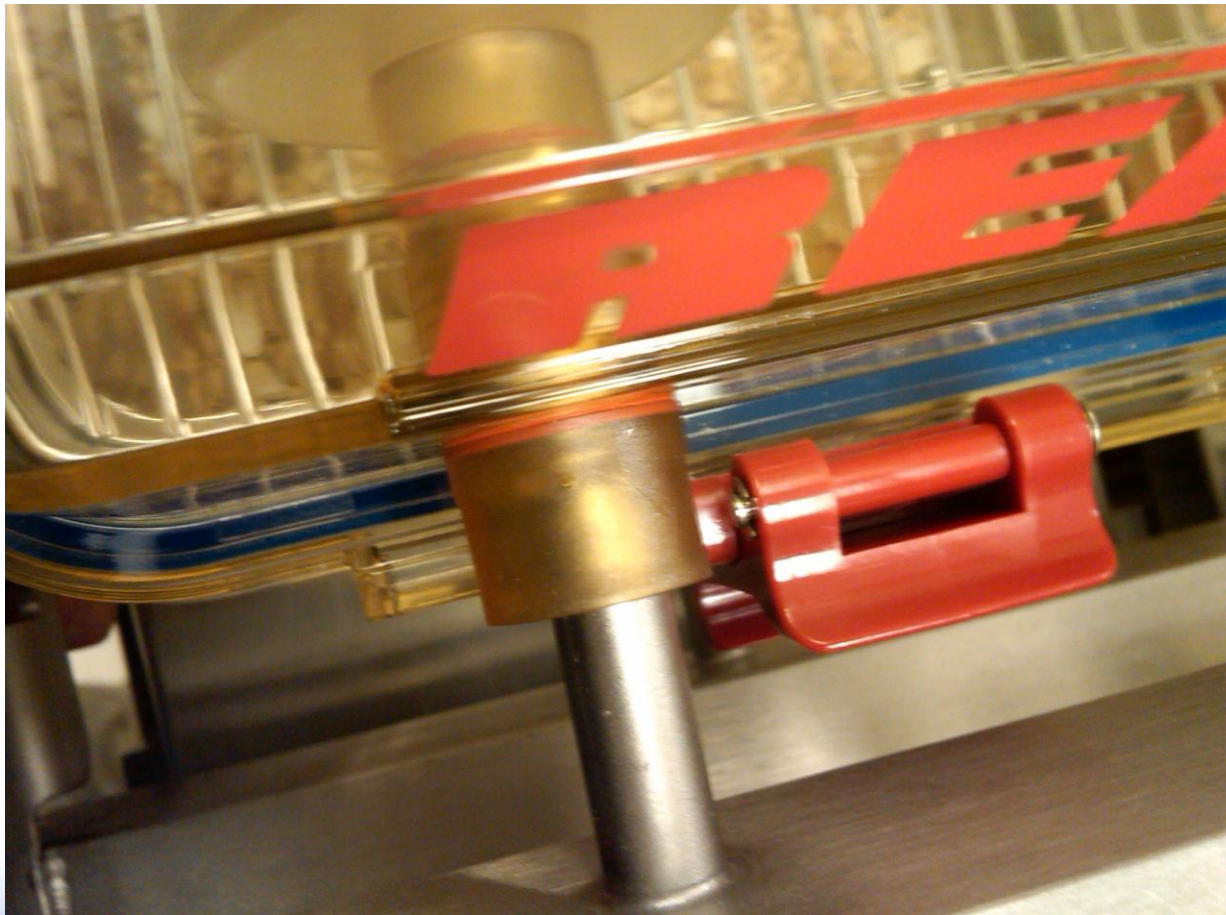
PREPARATION OF IVC COMPONENTS TO BE AUTOCLAVE



RACK OF COMPLETE IVC UNITS TO BE AUTOCLAVED



CONNECT RACK TO IVC UNITS BY SOLID TUBE TO ALLOW AIR TO BE REMOVED AND STEAM IN





COMPLETION OF GRAVITY FLOW AUTOCALVE CYCLE OF IVC UNITS @ 131C



COMPLETION OF GRAVITY FLOW AUTOCALVE CYCLE OF IVC UNITS @ 131C



PREP FOR PRE-VAC AUTOCLAVE CYCLE OF IVC UNITS @ 121 C



3/31/2010 11:17:28
USER: admin

CYCLE STATUS

NO ALARMS

CYCLE: 9 PREVAC

BATCH: 3

CYCLE PHASE: PRECONDITION 1

CYCLE STEP: VACUUM LEVEL

PULSE No: 3

REMAINING TIME: min sec

CHAMBER PRESSURE: 15.5 Psia

CHAMBER TEMP: 101.7 °C

PRESSURE SETPOINT: 6.0 Psia

JACKET TEMP: 115.8 °C

TEMPERATURE SETPOINT:

LOAD 1 TEMP: 101.8 °C

STEAM VALVE (PID):

DECONT. TEMP: 42.2 °C

MENU

SELECT
CYCLE

ABORT

ALARMS

PREVIOUS

40

COMPLETION OF PRE-VAC AUTOCLAVE CYCLE OF IVC UNITS @ 121 C







THANK YOU