Microbial monitoring of GF Isolators

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Topics to Cover

- Sterilants
- Sterility testing prior to animal entry
- QC of materials entering the isolator
- Microbial monitoring of the isolator after animal entry
- Supplies for testing and outsourcing

Chemical Sterilants- Things to keep in Mind

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- Material composition, geometry (carrier), organic load
- Clean, non-porous surface is usually assumed
- Read the label carefully for exposure times
- Validate for your purposes

Chemical Sterilants

- Peracetic acid: first germicidal agent used; high efficacy at low concentrations, active in presence of organic matter
- – Highly corrosive

- Chlorine dioxide: most commonly used; highly effective, commercially available as Exspor and Clidox

- Hydrogen peroxide and peracetic acid combination; highly effective, broad sporicidal efficacy, commercially available as Spor-Klenz, Renalin

- Chlorine dioxide gas: used in hospitals, expensive equipment to use so not practical for isolators

- Hydrogen peroxide gas: highly effective, sporicidal activity at low concentrations, expensive equipment

New GF Isolator Sterility Testing

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- Autoclaved sterile supplies are already in the isolator (caging, racks, water bottles etc.) as well as feed and bedding
- At least 3 weeks of negative sterility testing:
  - Week 1 testing
    - Mold trap swabs
    - Moisten swabs with sterile water; swab interior walls, ceiling, gloves, around filter outlets, caging, feeders and floor area
  - Week 2 testing
    - As above with addition of mold trap samples
  - Week 3 testing
    - As above with the addition of mold trap samples
- Imperative that sterilant does not get on the swabs!!

Laboratory Testing

- Vials are transported to the QC lab on the same site
- Samples are processed the same day of receipt/collection
- Cultured aerobically and anaerobically
- Wet mounts prepared
- Original vial is kept at least one week (BTC, mold, discrepancies)
- Suspect findings must be confirmed by a second sampling taken on a separate day
  - If it does not confirm, an investigation takes place
  - Must have 3 consecutive weeks of negative results to be used

QC of Materials Entering the Isolators

- Autoclaving is the safest method to sterilize materials for GF isolators
  - Biological indicators- Verify
  - Chemical indicators- Sterigage
- Validated autoclave cycles
- Validated loads
- Ethylene oxide (Verify and Sterigages for chemical sterilization)
- Irradiated materials (feed particular concern)

Self-Contained Biological Indicators

Biological Indicators

- STERIS' Verify® Self-Contained Biological Indicators (SCBIs) are available as either single or dual species formats (Bacillus atrophaeus (BA) for ethylene oxide and dry heat sterilization and/or Geobacillus stearothermophilus (GS) for saturated steam sterilization).

Negative and Positive Verify Indicators

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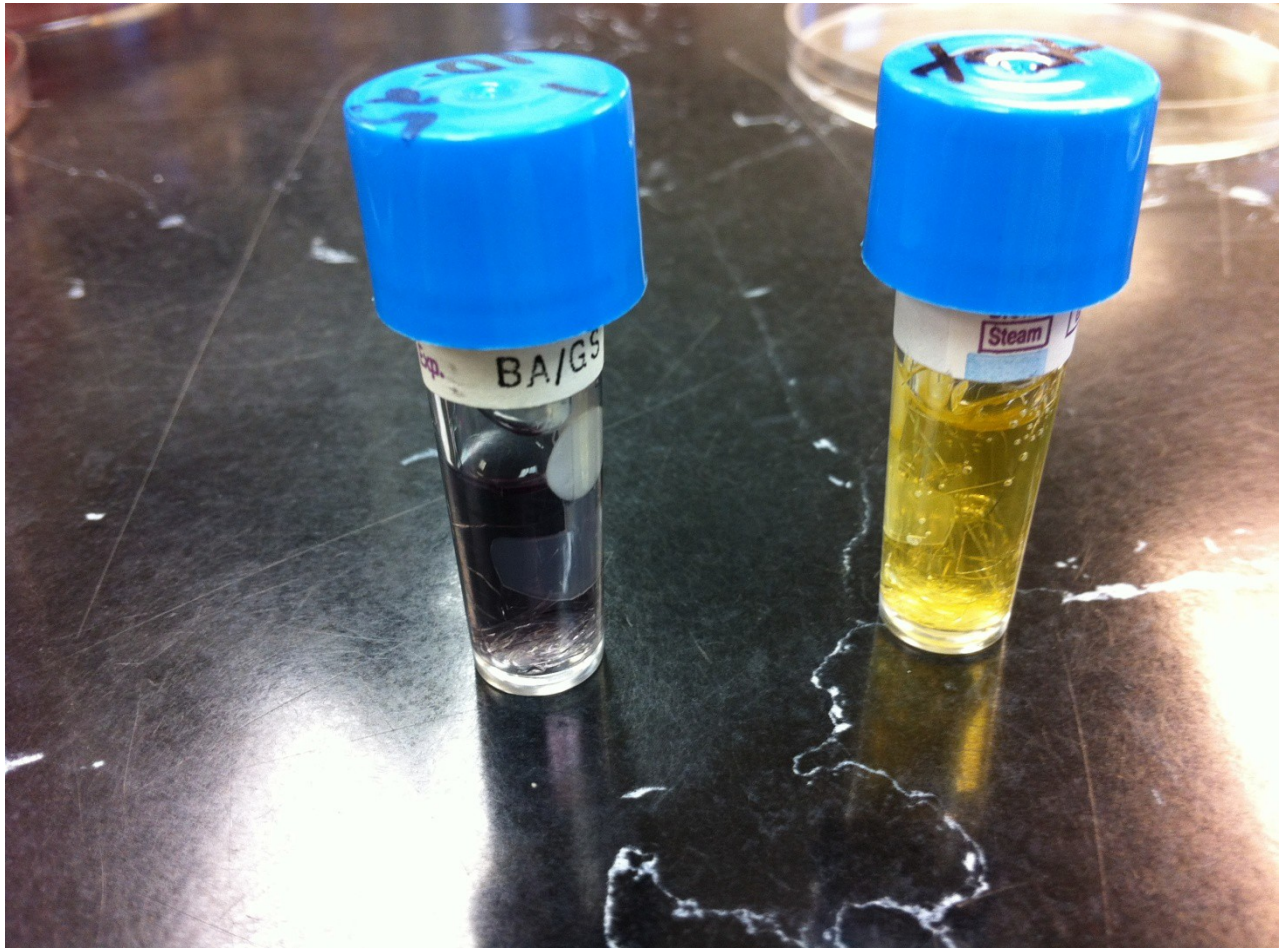
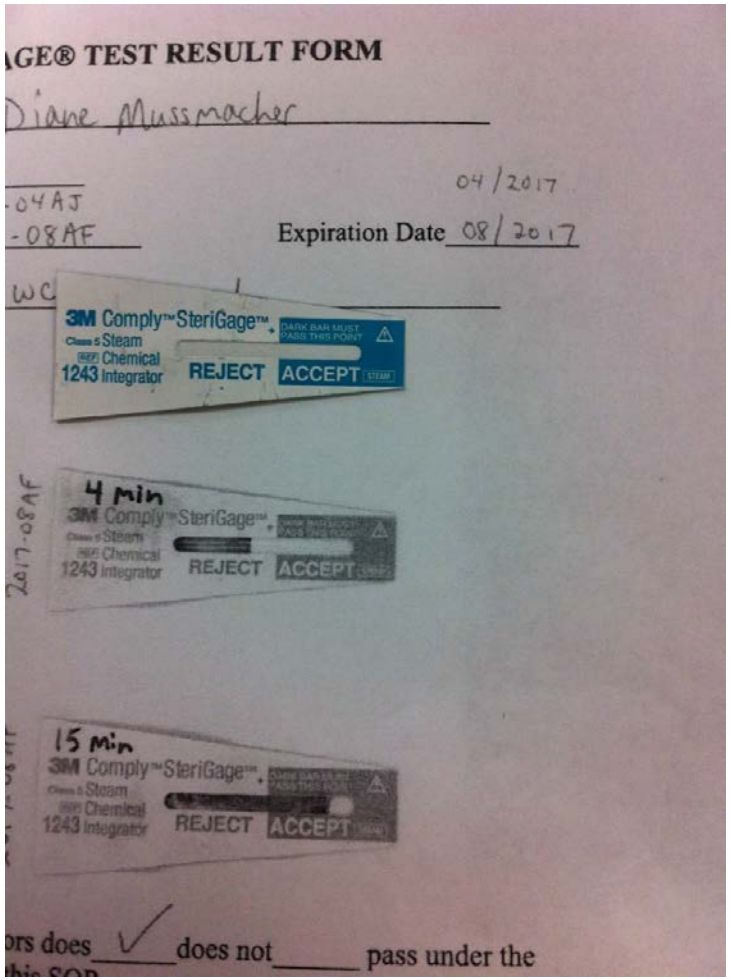


Spore Strips

Chemical Indicators

- 3M Comply SteriGage
- These chemical integrators are used for pack control as a method for monitoring sterilization process conditions inside each pack. The 3M™ Comply™ Steam Chemical Integrators can also be used inside a process challenge device (PCD) to release processed items, (load control). Use for this purpose does not replace the use of biological indicators.
- Steam enters the permeable topside of the device - the chemical pellet melts and migrates as a color along the paper wick; the distance or extent of migration depends on exposure to steam, time, and temperature.



Microbial Monitoring After Animal Entry

- Weekly vial testing
  - Swab of isolator interior surfaces (moisten with “dirty” water), swab caging, feed, feeders, floor area
  - Randomly sample 10-12 cages-collect fresh feces and soiled bedding
  - Add water from water bottles (several)
  - Transport to laboratory

- Culture: aerobic, anaerobic, mold
  - Trypticase soy agar with 5% sheep blood (37 degrees C)
  - Nutrient agar with dextrose (22 degrees C)
  - Brucella agar with 5% horse blood (anaerobic 37 degrees C)
  - Wet mount
  - Store original sample

Culture

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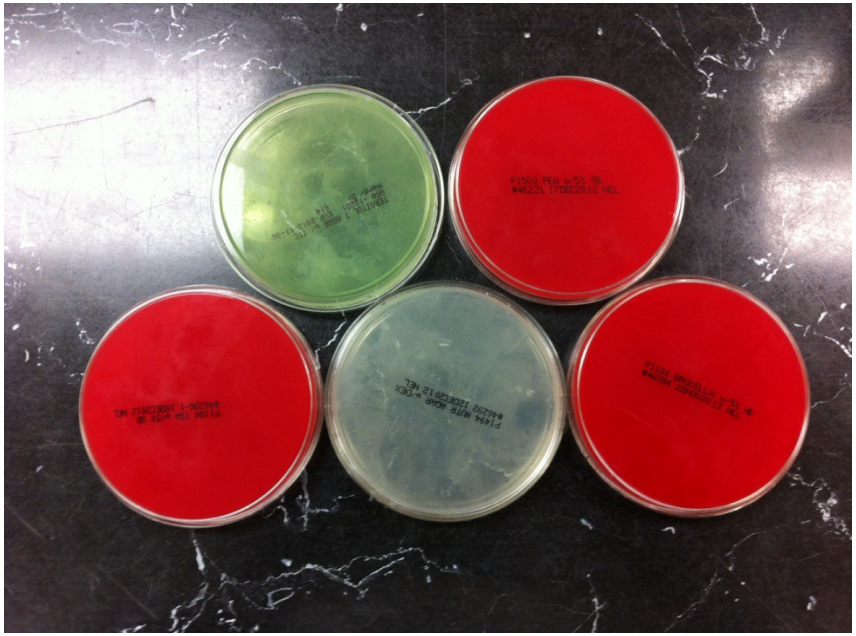

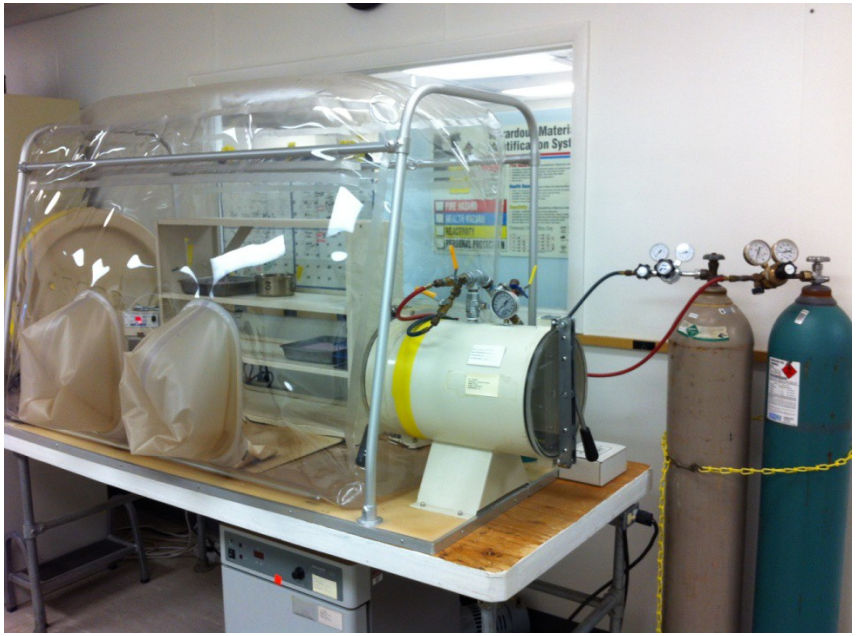

Isolator Samples

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Common Contaminants

Transport Media

- •Viability of a wide variety of strict anaerobic, aerobic and microaerophilic organisms is maintained for up to 72 hours at 20 to 25 degrees C.
- •Tubes
- •Jars
- •Vials
- •BBL™ Port-A-Cul™ Transport Systems
- •Validate the transport system works for your particular testing

Summary

- Microbial monitoring of new GF isolators
  - At least 3 weeks of negative results prior to animal entry
- QC/monitoring of supplies
  - Autoclaving is recommended
  - Validation of autoclave, runs, loads
  - Use of biological indicators/chemical indicators
- Microbial monitoring after animal entry
  - Weekly microbial monitoring

Independent Testing Laboratories

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- Enviro-Tech Laboratories- Mail in laboratory testing service http://www.sporestriptesting.com/services.htm

- Accugen Laboratories (independent contract microbiology laboratory) http://www.accugenlabs.com/

- Microbac Laboratories, Inc. http://www.microbac.com/index.php