Single Use and Shiny Stuff

Combine So You Can Produce Enough!

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Process Engineer, CRB

December 7, 2017

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Cambridge, MA 02142

Single Use and Shiny Stuff

Agenda

- **Introductions**
  - Speakers
  - Quick Background on CRB
- **Deconstructing the Buzzword**
  - What does “Closed” mean to your company?
- **Good Stainless Steel Practices**
  - CIP, SIP and maintaining the processing environment.
- **Good Single Use Practices**
  - Component inspection, bag deployment, tube manifold installation and tubing management.
- **Single Use vs Stainless Steel**
  - Media and Buffer Preparation
  - Teaching and Old Facility New Tricks
- **Open Discussion**

CRB
Overview

THE RELENTLESS PURSUIT OF SUCCESS. YOURS.™

design + construction professionals nationwide **1000+**

Serving Life Sciences & Advanced Tech Markets

$485M design & build revenue

 # 2 Design

* Pharmaceuticals - ENR Magazine

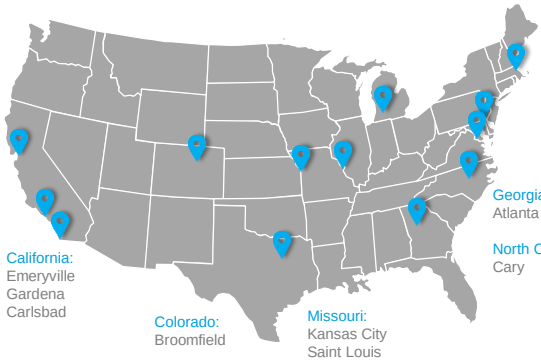
15 OFFICES

founded **1984**

10 International Society for Pharmaceutical Engineering (ISPE) Facility of the Year Awards

2017 is the 20th Anniversary of CRB's Construction Services Group

CRB
Locations



California:
Emeryville
Gardena
Carlsbad

Colorado:
Broomfield

Texas:
Irving

Missouri:
Kansas City
Saint Louis

Michigan:
Kalamazoo

Georgia:
Atlanta

North Carolina:
Cary

Massachusetts:
Medford

Pennsylvania:
Plymouth Meeting

Maryland:
Rockville

Puerto Rico:
San Juan

Switzerland:
Basel

Robert Boulanger, Ph.D., CRB Process Specialist

- 15 years experience
- Bulk of career manufacturing Vaccines
  - Lead process development, technology transfer and manufacturing operations
  - Involved in FDA licensure of two production facilities
- BPOG Viral Segregation Task Group Contributing Team Member
- ASME Bioprocessing Equipment (BPE) SIP Task Group Contributing member
- Joined CRB in April 2015, Process Specialist, Compliance, Planner
  - Based in the Rockville, MD Office

Mihir Sanghvi, CRB Process Engineer

- Joined CRB in Feb 2015
- Based in the Rockville, MD Office
- 10 years experience
  - Process Design
  - Start-Up & Commissioning
  - CIP/SIP – Cycle Development and Optimization
  - Site Construction Support
- ASME Bioprocessing Equipment (BPE) SIP Task Group Contributing member

Deconstructing the Buzzword

What do we mean by Closed?

Deconstructing the Buzzword

What does closed mean?

- Many different definitions of closed.
- Closed to/from what?
- Does closed mean clean and ready for processing?
- Why is closed processing important?
- What is the impact of closed processing on facility design?
- How do we demonstrate closed?
  - Accepted by your regulatory representatives
- Performing a closure analysis

Deconstructing the Buzzword

What does closed mean?

Aseptic connection between single-use gamma-irradiated tubing sets

Virgin Systems

Single Use Bioreactor

Aseptic tubing welder

SIPable Bioreactor

But what about?

Packed Chromatography Column

Deconstructing the Buzzword

Defining Closed

- Closed system:
  - Designed and operated such that the product is isolated and never exposed to the manufacturing environment.
  - Transfers into or from these systems must be validated as closed.
  - Risk of contamination to the product or process cannot be mitigated by addition of HEPA filtration to the immediate environment housing that operation.
  - Environment is a No Impact System
- Closed Process:
  - Process stream that is never exposed to its environment from setup through process completion

Deconstructing the Buzzword

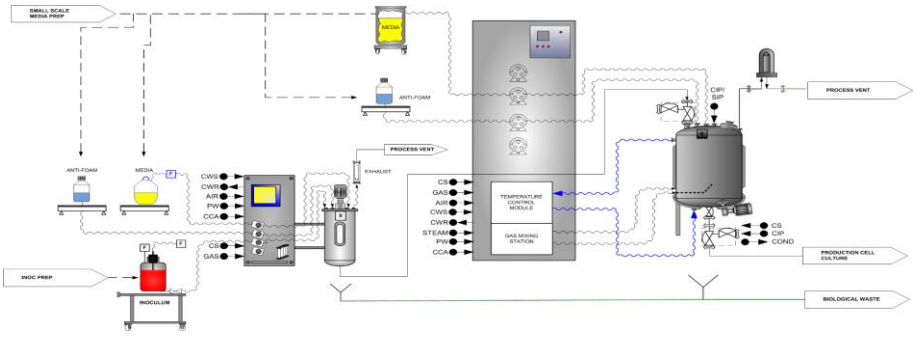
Defining Functionally Closed

- Functionally closed:
  - Closed systems that are opened between processing operations but are "rendered closed" by a cleaning, sanitization or sterilization process that is appropriate or consistent with the process requirements, whether sterile, aseptic or low bioburden.
  - Systems that remain closed during product exposure to the system but may be exposed to the environment between periods of product exposure.
- Functionally closed processing:
  - Process that is not exposed to the environment in presence product, but where system requires open setup or intervention

Deconstructing the Buzzword

Example of Functionally Closed

Cell Culture in SS Bioreactor



CIP → SIP → Inoculate → Culture → Harvest

Deconstructing the Buzzword

Defining “Open” Processes or Systems

- Open Process:
  - A process or system that is exposed to its environment before, during and/or after a process operation without appropriate measures to functionally close the process prior to use of the product stream.
  - Open operations performed in a BSC are still open operations.
- Briefly Exposed Operations:
  - Open processes containing process materials and/or product intermediates. These open processes are rendered closed by means of an appropriate closing process.
  - Definition and Validation of the “pre-closure” incubation phase is critical.

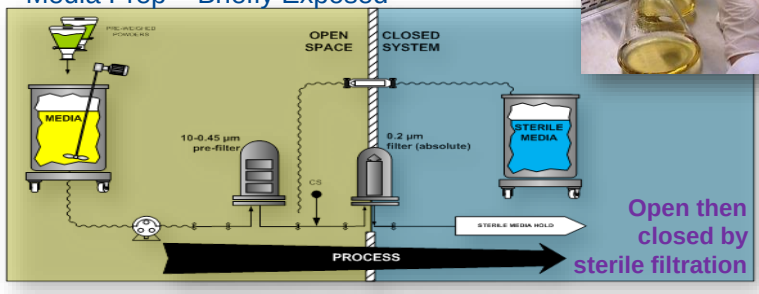
Deconstructing the Buzzword

Examples of Open and Briefly Exposed Operations

Inoculum Prep - Open

Open in BSC

Media Prep – Briefly Exposed

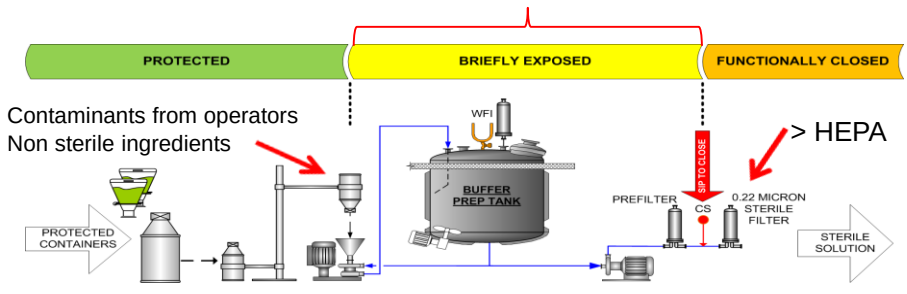


Open then closed by sterile filtration

Deconstructing the Buzzword

"Protected" vs. "Briefly Exposed" vs. "Functionally Closed"

Pre-filtration incubation periods must be carefully monitored & validated



Deconstructing the Buzzword

Formal RA Program to Assess Ability to Close Process

Our Journey begins with sound process understanding…

- Select Subject Matter Experts (SMEs):
  - Quality, Manufacturing, R&D
- The Program. Define "Closed". Set the Risk Assessment Parameters
  - Develop Impact (Severity) & Likelihood Criteria
  - Pre-Establish Risk Matrix
  - Use Tools for systematic evaluations
  - Develop Closure Philosophy and Strategy
- Understand your process & systems, PFDs, Protocols, Batch Records
- Evaluate and Confirm "Closed"
- Assess & Develop Sound Risk Mitigation Plan Based on Science & Engineering NOT on Perception of Regulatory Requirements or Legacy

Deconstructing the Buzzword

Preparing for a Closure Analysis

- Develop Closure Strategy
- All open aseptic operations shall be performed in Grade A…
- Validate closing of all systems stated as closed
- $ vs practicality vs risk vs…
- Develop a strategy to:
  - Close "closed" operations
  - House all operations appropriately
- Strategy must be approved by all stakeholders
- Initially applied to specific process then develop company-wide philosophy

Deconstructing the Buzzword

Closure Analysis: Develop and agree on the Definitions

Bioburden Control Specifications		
Control Level	**Definition**	**Example**
3 - Aseptic/Sterile	Bioburden is reduced to zero or sterile condition. Requires absolute isolation from environment. Specification is defined and typically < 1 CFU/100mL.	Cell culture or fermentation processes and final product filling where absence of microbial contamination is critical to performance of the process or quality of final product.
2 - Low bioburden	Bioburden is reduced to acceptable levels. Process solution is bacteriostatic or does not promote growth. Sanitary though non-sterile conditions required.	Chromatography processes where a sterile state is typically not required but a 0.2um filtration is in place as a measure of control.
1 - Controlled bioburden	Bioburden is controlled so as to limit microbial growth.	Media or buffer prep where raw material components are briefly exposed to the environment prior to sterile filtration.

Closure Level Specifications	
Closure Level	**Definition**
1 - closed/unexposed	Never been used such as γ-irradiated assemblies. Aseptic connections include KleenPak, Lynx, GE, SaniTek and tubing welds
2 - cleaned & SIPd	Open connection that is CIPd and SIPd prior to use or previously autoclaved and connected aseptically
3 – cleaned & sanitized	System CIPd but not SIPd prior to use, remains isolated from environment during process
4 - briefly exposed	Process exposed to environment for brief period then rendered closed by filtration or sterilization.
5 - open	Connection open to environment without subsequent cleaning or sanitization prior to use. Examples include quick connects or TC connections that have not been or CIPd or SIPd

Deconstructing the Buzzword

Closure Analysis: Set up the Closure Risk Matrix

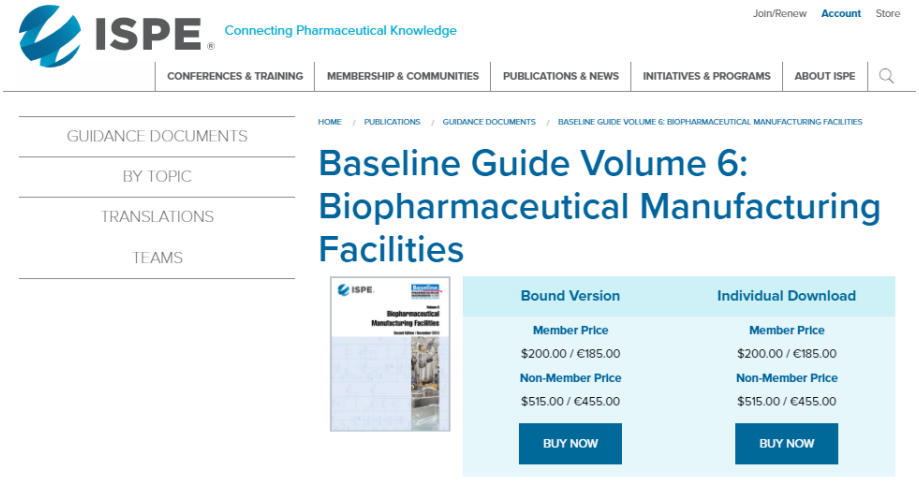
Risk Matrix

		Control Level 1 Controlled bioburden	2 Low bioburden	3 Aseptic
Closure Level	1 Closed / Unexposed	1	2	3
	2 Cleaned / SIP'ed	2	4	6
	3 Cleaned / Sanitized	3	6	9
	4 Briefly Exposed	4	8	12
	5 Open	5	10	15

Risk Ranking		
10-15	Urgent	Immediate engineering and/or managemnt control. Process must shut down until repair is implemented
8-12	High Priority	Should be addressed with engineering and/or management control with preference over medium and
5-9	Moderate Priority	Should be addressed with engineering and/or management control with preference over low priority
3-4	Low Priority	Should be addressed with engineering and/or management control without disruption to production but

Deconstructing the Buzzword

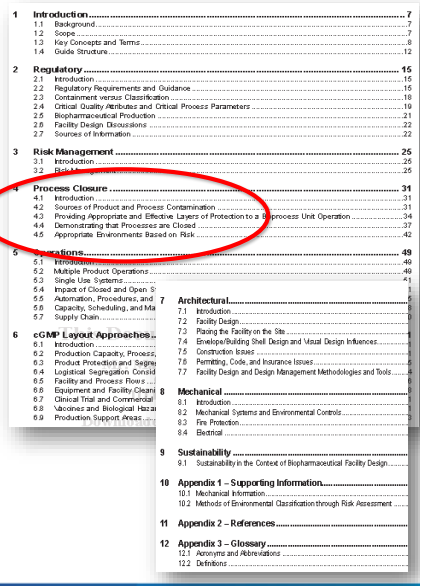
Biopharmaceutical Manufacturing Facilities Baseline Guide



Deconstructing the Buzzword

Facilities Baseline Guide TOC

- Ch 1 Introduction
- Ch 2 Regulatory
- Ch 3 Risk Management
- Ch 4 Process Closure
- Ch 5 Operations
- Ch 6 CGMP Layout Approaches
- Ch 7 Architectural
- Ch 8 Mechanical
- Ch 9 Sustainability
- Appendix

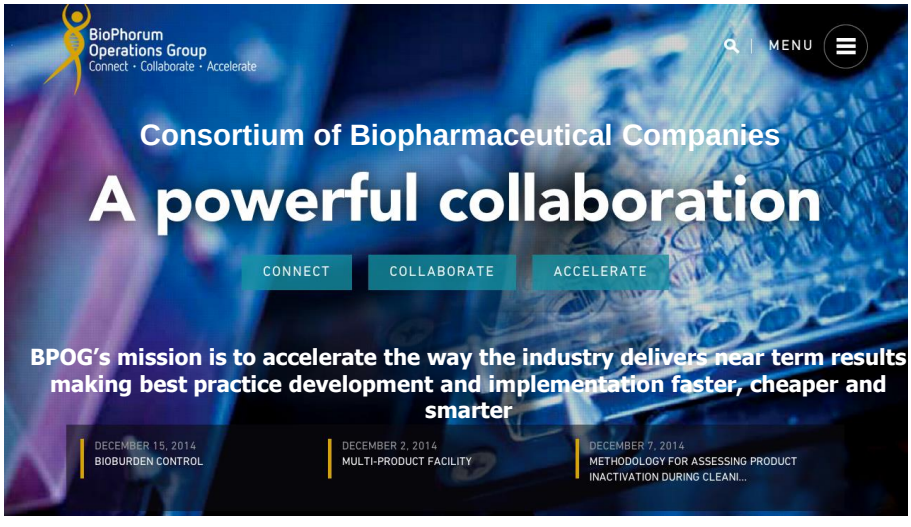


Deconstructing the Buzzword

What is a Closed System?

- **A Closed System ≠ Sterile!!!**
  - Validated to show that there are sufficient layers of protection to mitigate the risk of contamination from the environment
  - Environment housing the system is not a critical aspect of the process. Product is never exposed to outside environment.
  - Risk of contamination of closed system cannot be mitigated by housing process in bioburden-free environment
  - Contamination of a closed system represents a breach of that system.

BioPhorum Operations Group (BPOG)



BioPhorum Operations Group (BPOG)

- To create an environment where the global biopharmaceutical industry can collaborate and accelerate their rate of progress, for the benefit of all.
- Bringing leaders together to create future visions that focus the industry's energy on the key emerging opportunities;
- Mobilizing communities of the top experts around these opportunities, up and down the biopharma value chain;
- Creating partnerships that enable change and provide the quickest route to implementation and results;
- Replacing isolation with collaboration so that the industry shares, learns and builds the best solutions together;
- Making the journey better, faster, cheaper.

BioPhorum Operations Group (BPOG)

Is The "Belt And Suspenders" Approach Required?

- Many companies are implementing closed-system processing to achieve a high-level of contamination control – but within classified cleanrooms
- For concurrent multi-product manufacturing, many companies have implemented architectural segregation measures in addition to closed-system processing measures

BioPhorum Operations Group (BPOG)

Is The “Belt And Suspenders” Approach Required?

- Mission – Investigate and define how closed systems can be effectively applied to reduce the reliance on architectural, personnel, and procedural segregation for drug substance (DS) bio-manufacturing and greater use of Controlled Non-Classified (CNC) space while ensuring product quality and patient safety.
  - Risk based assessments have allowed the industry to re-think traditional approaches to area classifications
  - Focus on integrity of closed systems rather than reliance on secondary containment and environmental controls
  - Newer technology and improved engineering & procedural controls, facility designs, bioprocess equipment and analytical test methods enables greater use of closed and functionally closed systems

BPOG Workstream”

Closed Systems in CNC “Room classification

Mission

- Simplify & harmonise selection of room classification across the industry
- Consistent implementation by regulators and industry
- Common risk based approach
- Endorsement by internal and external stakeholders

Benefits

- Improving product quality and patient access to products
- Simplified design
- Reduction of capital and operational costs
- Reduced Cost of Quality
- Flexibility in facility operation
- Consistent regulatory feedback

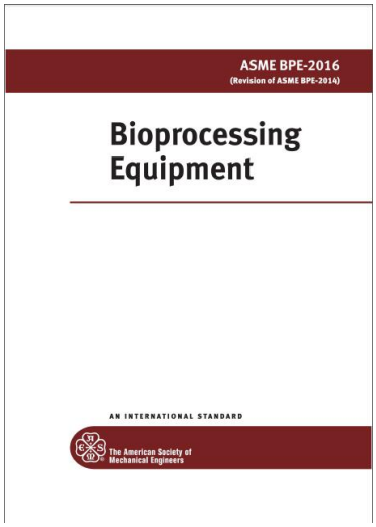
Good Stainless Steel Practices

CIP, SIP and Maintaining the Processing Environment.

27

Not so Quickly Cowboys!

SIP & CIP Principles

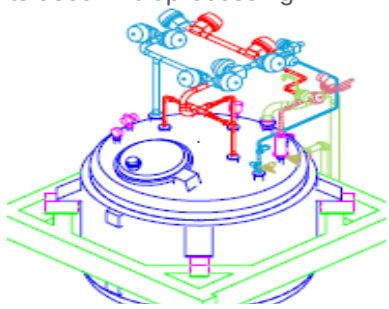


ASME CODES & STANDARDS 125

ASME BPE

Scope

- provides requirements for systems and components that:
  - are subject to **cleaning** and **sanitization** (incl **sterilization**)
    - **cleaned in place (CIP'd)**
    - **steamed in place (SIP'd)**
    - **other processes used in the manufacturing of biopharmaceuticals.**
- provides requirements for
  - **single-use systems** and components used in bioprocessing.
- This standard may be used, in whole or in part, for other systems and components where **bioburden risk** is a concern.

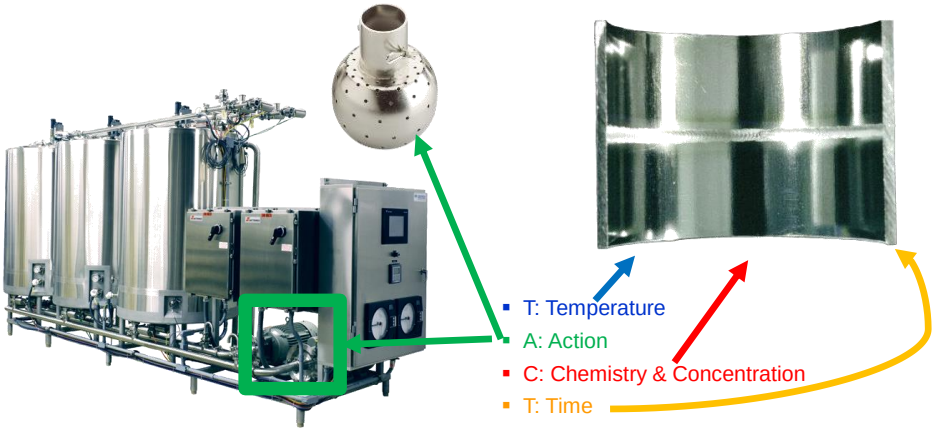


ASME BPE

Voluntary Consensus Standard

- Balanced group of experts:
  - Engineers, Consultants and Specialists
  - Component manufacturers
  - Drug manufacturers
- Updated every two years to meet industry accepted best practices
- Corrections & clarifications requested by anyone
- Single Use and Stainless Steel Equipment
- How to Close Bioprocess Systems

ASME BPE
Keys to Successful CIP



ASME BPE
Keys to Successful CIP - **Action**

Flow: turbulent, drainable, branches

Fig. SD-3.9.2.1-2 Flow Rate Guideline for Vertical Cylindrical Vessels

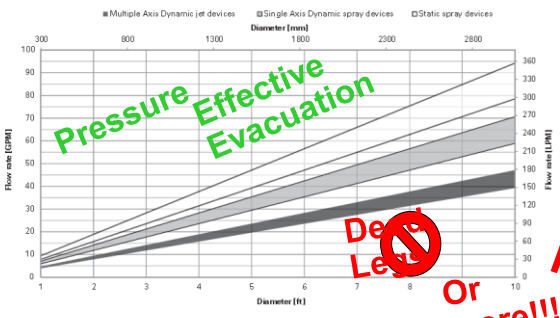


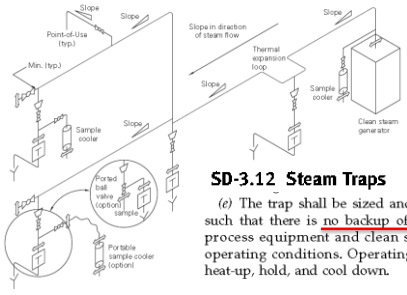
Table SD-5.3.3.3-1 Flow Rates to Achieve 5 ft/sec (1.52 m/s)

Sanitary Tube Size					
O.D.		I.D.		Flow Rate	
in.	mm	in.	mm	gpm	Lpm
0.5	12	0.37	9.4	1.7	6.3
0.75	19.1	0.62	15.7	4.7	18
1.0	25.4	0.87	22.1	9.3	35
1.5	38.1	1.37	34.8	23	87
2	50.8	1.87	47.5	42.8	162

ASME BPE
Keys to Successful SIP – **Saturated Steam**

Air is your enemy!
Water is your enemy!
ASME BPE-2016 is your friend!

Fig. SD-6.2.2-1 Typical Clean Steam System Isometric



SD-3.12 Steam Traps

(*e*) The trap shall be sized and installed to operate such that there is no backup of condensate into the process equipment and clean steam system under operating conditions. Operating conditions include heat-up, hold, and cool down.

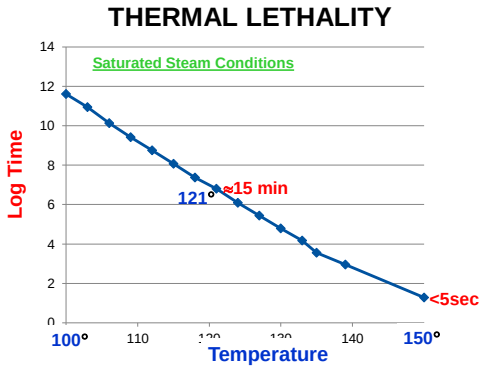
SD-2.3.1.1 Steam in Place. Equipment parts and components subjected to SIP should withstand continuous flow of saturated steam at a minimum temperature of 266°F (130°C) for duration of 100 hr minimum under continuous steady-state conditions. However, at the discretion of the owner/user, conditions that are more stringent may be imposed. The use of elastomers/fluoroelastomers (within a piece of equipment or certain process instrumentation) that may thermally degrade during SIP will need to be thoroughly evaluated by the owner/user or manufacturer. The overall life of the equipment may be shortened significantly if the correct elastomer or process instrument is not selected.

ASME BPE
Keys to Successful SIP

- Temperature
- Time

Pressure

Air is your enemy!

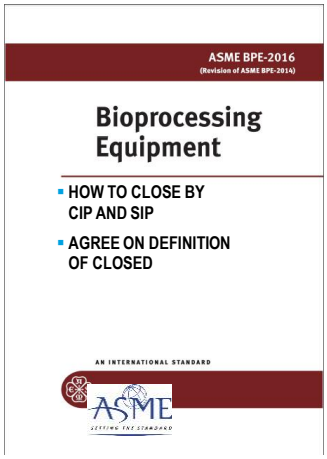


Note: approx. **250°C required for dry heat sanitization**

Saturated Steam Table

Pressure (PSIG)	Temperature (°F)	Temperature (°C)
0	212	100
1	216	102
2	219	104
3	222	106
4	224	107
5	227	108
6	230	110
7	232	111
8	235	113
9	237	114
10	240	116
15	250	121
20	259	126
24	266	130
25	267	131
30	274	134
35	281	138
40	287	142
45	292	144
50	298	148

Key to Success is Harmonization

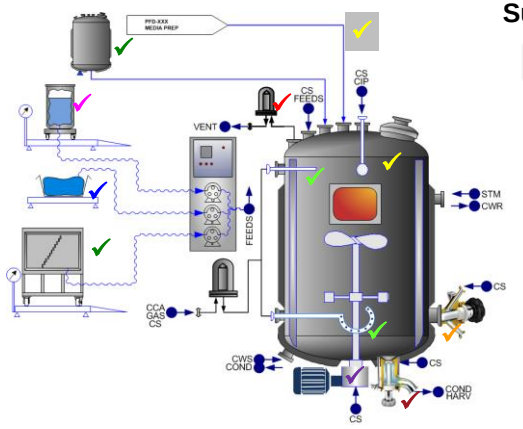


ASME BPE
Defining System Boundary

Process Contact: a surface under design operating conditions that is in contact with, or has the potential to be in contact with, raw materials, in-process materials, APIs, clean utilities (e.g., WFI, CIP, pure steam, process gases), or components and where there is a potential for the surface to affect product safety, quality, identity, strength, or purity.

Product Contact: a process contact surface that is in contact with, or has the potential to be in contact with, a product where product is defined by the owner.

ASME BPE
Defining System Boundary



Successful Cell Culture:

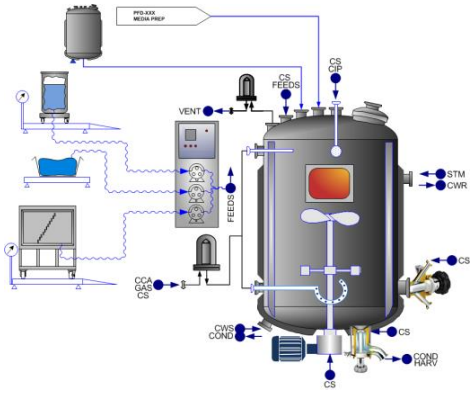
- ✓ Axenic culture (14-90 days)
- ✓ Aseptic aeration
- ✓ Aseptic venting with no fouling
- ✓ Aseptic foam control
- ✓ Aseptic pH control
- ✓ Aseptic nutrient feeds
- ✓ Aseptic mixing
- ✓ Aseptic sampling
- ✓ Aseptic harvest

ASME BPE
Recall Keys to Successful CIP

TACT
- Temperature
- Action
- [Chemistry]
- Time

Therefore monitor:
- Temperature
- Flow, Pressure, Rotation, Inspect
- Conductivity, pH
- Time

Ensure appropriate hydraulics

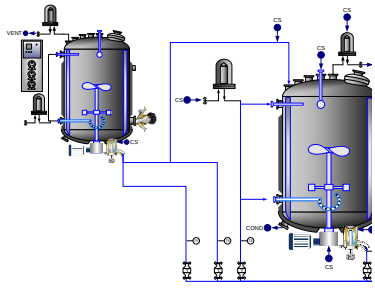


ASME BPE
Appropriate Conditions for Successful CIP

- 5 ft/sec velocity in tubing
  - No dead legs
  - Hydraulic balance
  - Pump capacity
- 3 gpm / ft of circumference
- Efficient evacuation
  - Tank bottom valve
  - Ring seal vacuum pump
  - Low points
- Monitor flow & press
  - Plugged spray devices
  - Leaks

ASME BPE
Appropriate Conditions for Successful SIP

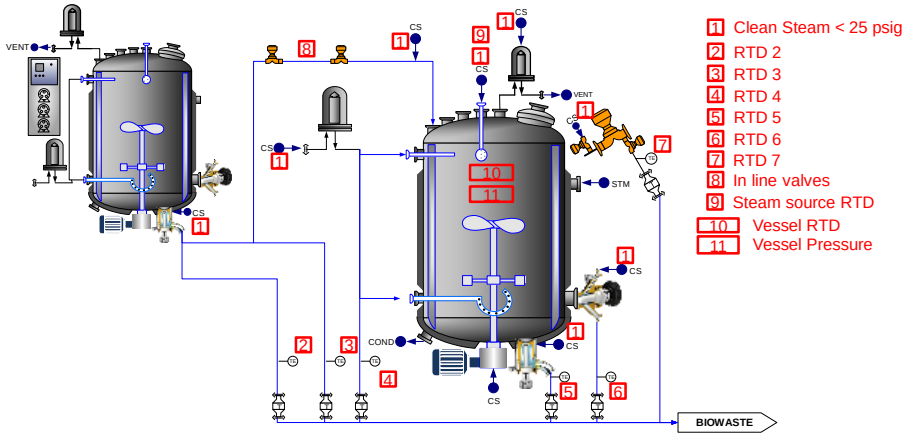
- Monitor temperature & pressure
  - All RTDs +/- 1°C
  - > 130°C requires special materials
  - >24 psig requires special materials
- Effective draining
  - Trap lines
  - Low point drains, no dead legs
- Overlap of steaming on separate SIPs
- No vacuum
- Maintain integrity/closure



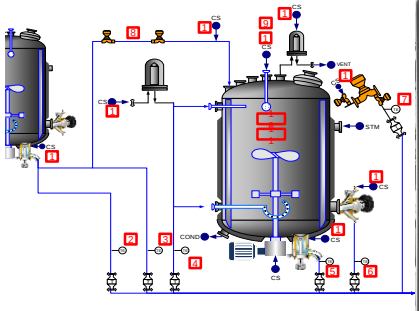
Field Observations
Disclaimer

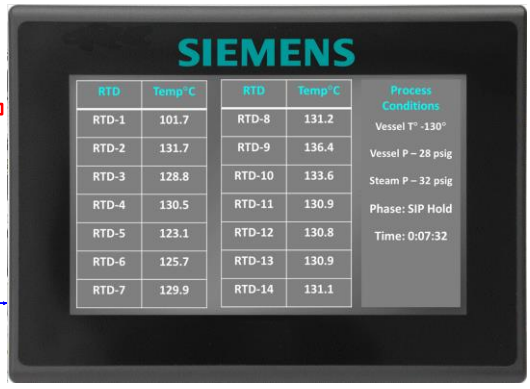
- The following slides represent a compilation of field observations from various biotech facilities. These photographs were provided with permission to present to biopharmaceutical society conferences in an effort to help improve designs and installations. Every effort has been made to hide the identity of the sources of these images. If you recognize any settings, we ask that you not disclose the sources.

Field Observations
SIP Monitoring Points

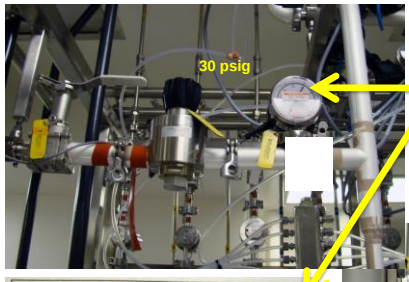


Field Observations
Initial SIP Conditions



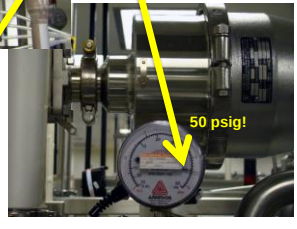


Field Observations
SIP conditions lead to damage

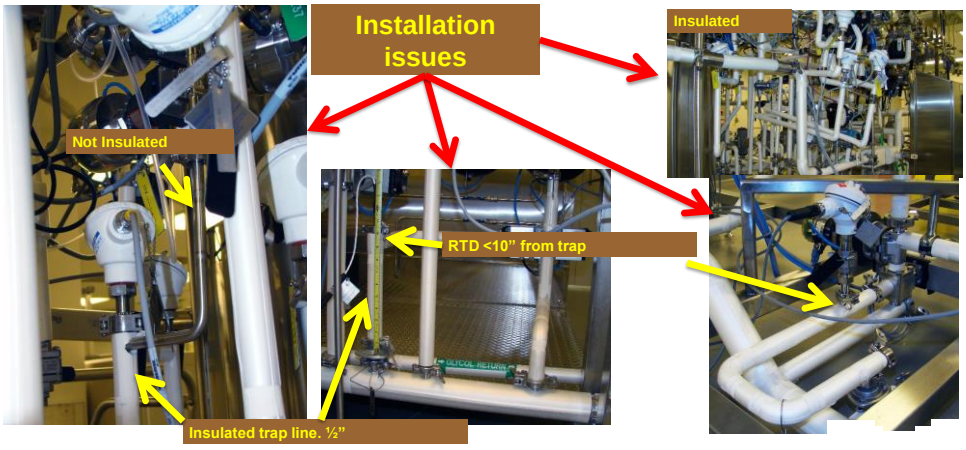


SIP Pressures above 24 psig

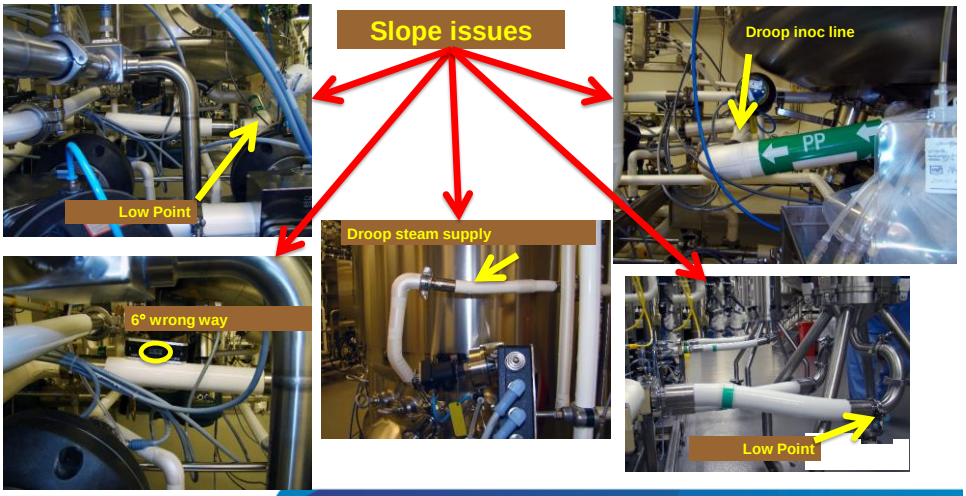
Utility	Pressure (psig)
Pure Steam	30 ± 5
OFCA	35 ± 5
Nitrogen	35 ± 5
Oxygen	35 ± 5
Carbon Dioxide	35 ± 5
Helium	35 ± 5



Field Observations
Installation affects SIP Performance

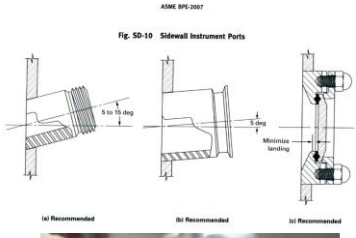


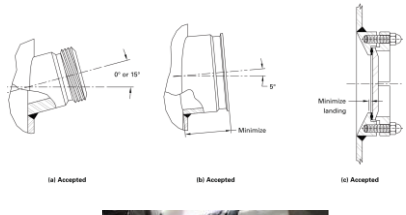
Field Observations
Installation affects CIP and SIP Performance



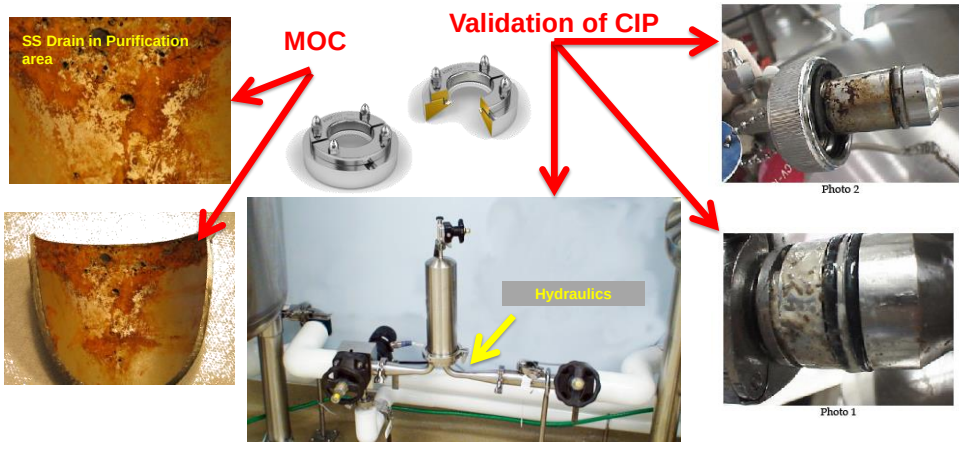
Field Observations
Ingold™ fittings: Different shapes and sizes

Field Observations
Recommended and Accepted Sidewall Designs

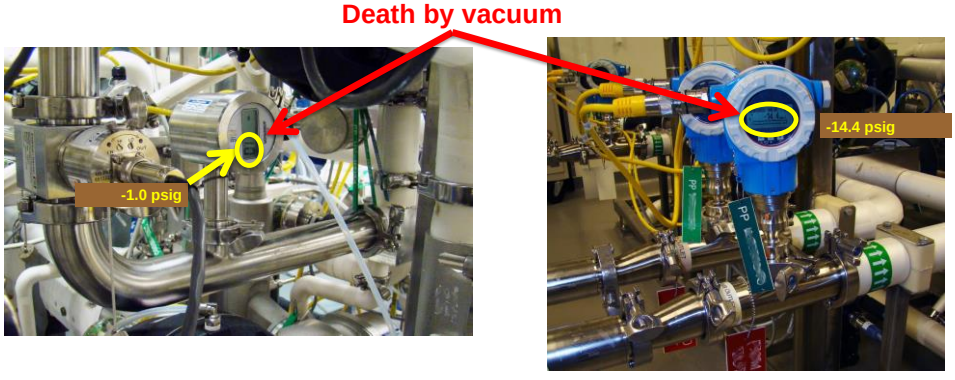




Field Observations
Design affects CIP and SIP Performance



Field Observations
Post SIP Vacuum Condition



Good Single Use Practices

Inspection, Deployment, and Management.

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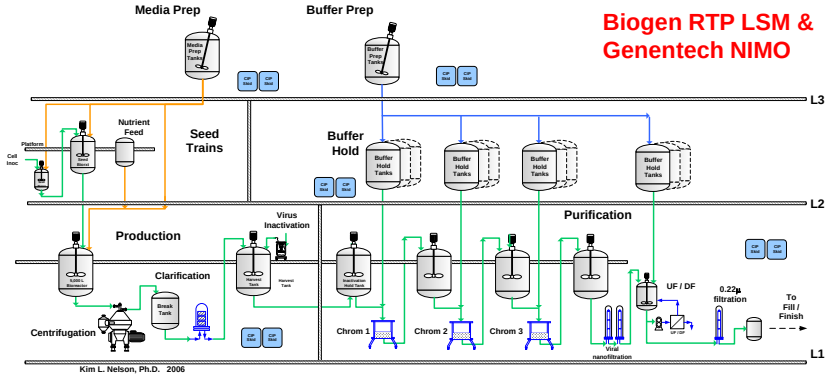
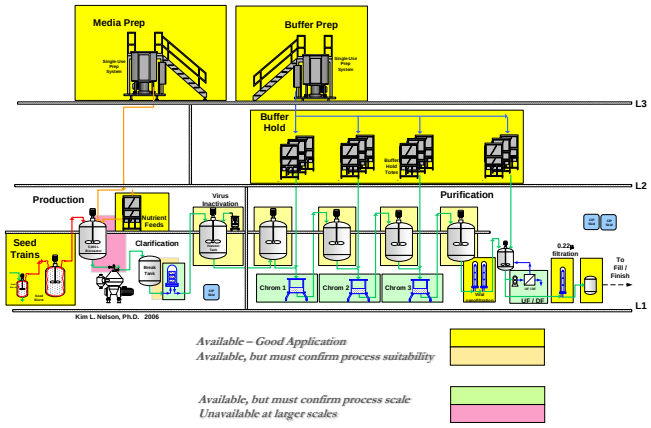
Stainless Steel Biomanufacturing



Single Use Technology Implementation



Single Use Technology Implementation
Well Suited For

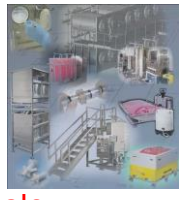
- Retrofits accommodating new products
- Process change – adding new addition buffer/media
- Clinical manufacturing
- Applications where SIP or CIP capability is limited
- Small volume applications (2000L or less)
  - At beginning (small volume Cell Culture) and end (sterile bulk fill)
- New, uncharacterized processes
- Products requiring frequent changeovers
- Cost of Goods driven???
- Ultra fast-track projects with low capital budget
  - (BUT customization adds time & $)
- Key Features:
  - Vendor supplied pre-sterilized and as a ‘closed’ system
  - No CIP or SIP required

Single Use versus Stainless Steel

- Single Use-based facility requires:
  - Less Capital
    - **No Integrated CIP & SIP**
    - **Reduced HVAC for Clean Space**
    - **Reduced Clean Space Footprint**
  - Potentially Less Expense
    - **Minimal separation of personnel (ballroom & closed processing)**
    - **More flexible**
    - **Lean process - Less idle equipment time**
  - Less construction time
    - **Allows more time between clinical results and commercial launch**

Single Use versus Stainless Steel
Issues to Consider

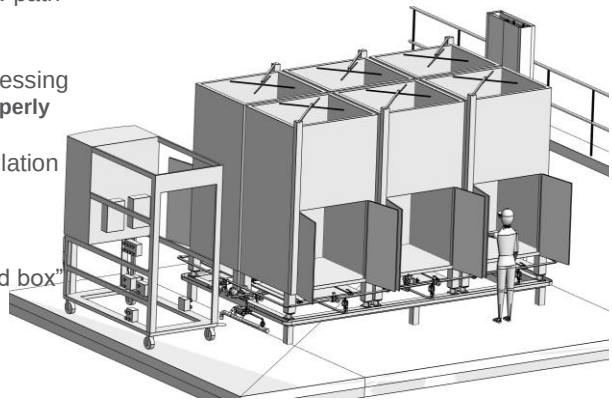
- Single use systems are scale limited:
  - Bioreactors = 2,000L (ABEC just introduced 4,000L unit)
  - Tubing = ~1 inch (weldable, dry), <1" (weldable, wet), general use 1-1/2"
  - Solution Prep (mixing) = 5,000L (only one vendor (ThermoFisher)
  - Storage Bags = 5,000L (fixed) and 1,000L (movable)
  - Chromatography systems & columns (60 cm max)
  - Depth Filtration, Viral Filtration & Ultrafiltration (may need multiple units)
- Heat Transfer
- Weight of high density solution hold
- Warehousing & Disposal
- Lead times due to customization
- Sole sourcing can be considered to increase scale, however, **supply chain risk** must be assessed



Single Use Best Practices

Operator Training: Set up, Deployment and Operation of SUS

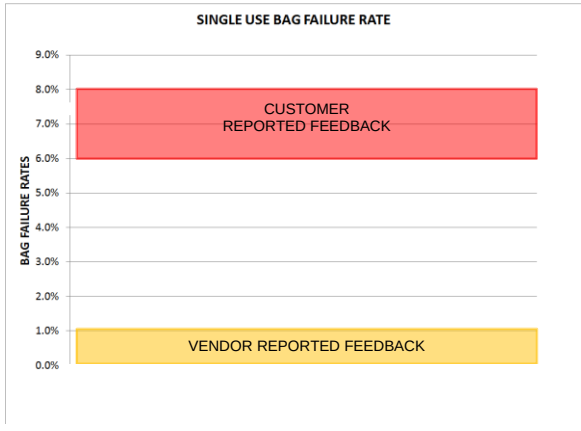
- **Tubing**
  - Organized management systems
  - Connectors fasten properly
  - Unobstructed tubing flow path
- **Ergonomics**
  - Bag set up
  - Observation during processing
    - **Confirm bag deploy properly**
- **Probe set up**
  - Training on proper installation
  - Properly supported and protected
- **Hands-on experience**
  - Visit vendor site or "Sand box"



Single Use Best Practices

Initial Learning Curve with SUS

Failure Risk

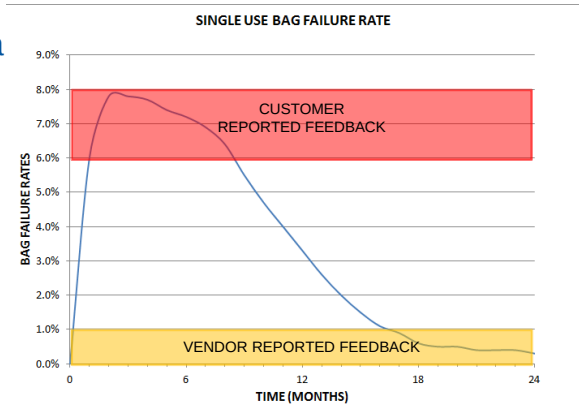


Single Use Best Practices

Initial Learning Curve with SUS

Failure Risk

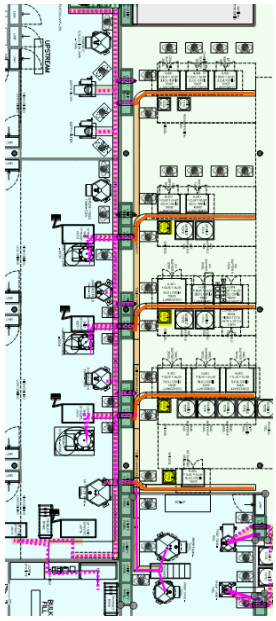
- Failures derive from a variety of causes:
  - Packaging / Unpacking
  - Poor Shipping / Handling
  - Storage conditions
  - Improper Installation
  - Legitimate failures from manual construction:
    - **Attachment of clamps / fittings**
    - **Weld seems**
    - **Zip tied hose barb connections**
  - Manufacturer's defect



Single Use Best Practices

Tube Set Design Considerations

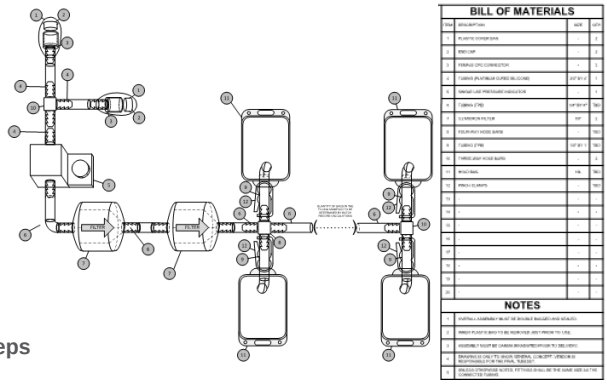
- **Standardize Tube sets**
  - Reduce number of unique tube sets
  - I.D.,O.D., and Length
  - Aseptic and quick connect (briefly exposed)
- **Risk based approach**
  - Determine appropriate connector requirements
    - **Don't use aseptic connectors unless required.**
  - Are pre-sterilized components/bags required?
  - Identify proper room classification
- **Walk through deployment and setup strategy**
  - Ergonimics



Single Use Best Practices

Tube Set “PIDs”

- **Approach like SS piping**
- **What flushes are required?**
- **Sampling?**
- **Bag Fill/Dispense**
  - Bottom/top
  - Multiple additions
    - **Liquid**
    - **Solids**
  - Open
  - Closed
  - Briefly Exposed
  - Dispense
    - **Samples**
    - **Multiple batches**
    - **Multiple process steps**



Single Use Best Practices

Tubing Management

- **Transfers over long distances can require work at elevation which pose safety concerns.**
- **Overhead tubing transfers should be avoided.**
- **Stainless steel transfer lines are easy to clean so hybrid considerations should be evaluated where practical.**

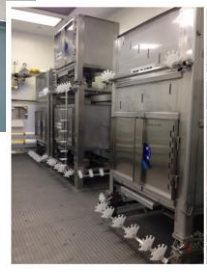
Low Run

Overhead Run

Single Use Best Practices

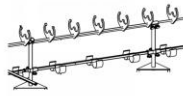
Tubing Management

- SS Cable Tray
- Tubing Support
- Custom Tubing Management system
- Tubing Trench



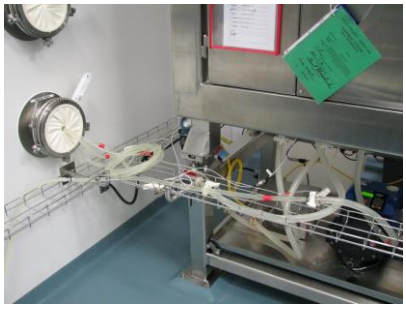
cone craft

Tube Management System



Single Use Best Practices

Tubing Pass-through



Iris Valve

AdvantaPass

Capped ports with inserts around tubing

Single Use Best Practices

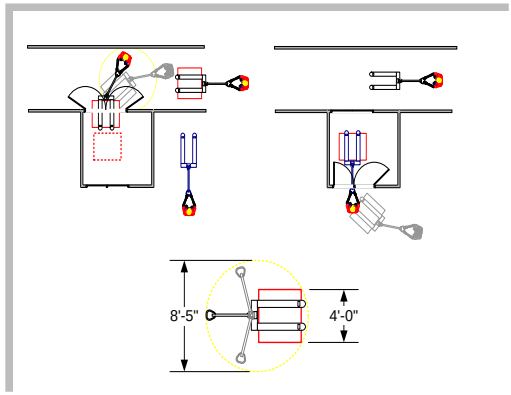
Tubing Identification

- **Intelligent Couplings can help to confirm proper connections are made to prevent mix-ups**
- **Color coding of tubing or zip ties are another option to prevent confusion**

Single Use Best Practices

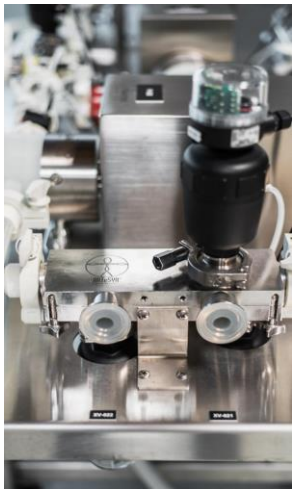
Tubing Identification

- **Airlocks must be sized to accommodate movement of media / buffer bags**
- **Must account for**
- **Door Swings**
- **Wipe Down Space**
- **Turning radius of palette jacks / movers**
- **Material movement / traffic studies are a good idea**



Single Use Best Practices

Skided Solutions (Custom, Automated and or Manual solutions)



PROCESS MODULES

Single Use Vs. Stainless Steel

Media and Buffer Preparation

Media & Buffer Preparation

Process Assumptions

- Scale comparable to small and medium size biotech GMP facility.
- 2000L Biotech Facility
- Mab Model with Titer around 1.5 - 3 g/l

Media Preparation

Single Use Mixers

Based on the process model and media requirements following sizes were selected (SUMs with jacket, load cells and temperature transmitters)-

1. 2000L
2. 500L
3. 200L

Media Preparation

Stainless Steel Vessel

Following vessel sizes are comparable to the single use equipment selected -

1. 2000L
2. 650L
3. 200L

Stainless Steel Vessels are assumed as skid with capability to perform rinses, CIP and SIP.

For Media vessel the skids will be CIP'ed at end of each run. SIP will be on as needed basis.

Media Preparation

3-D Model of Media Skid

Media Preparation

Single Use Bag Consumption and Unit Cost

Media Bag Consumption per Batch

- 2000L SUM – 1 bag
- 500L SUM – 2 bags
- 200L SUM – 4 bags

Bag Unit Cost

- 2000L Bag - $2,000
- 500L Bag - $1,200
- 200L Bag - $1,000

Media Preparation

WFI Cost and Estimation for CIP

WFI Cost = $ 0.07 per liter.

This cost does not include maintenance cost, inflation and equipment depreciation. It is assumed that site has WFI producing capability.

CIP Cost Estimate per Vessel –

- 2000L Vessel CIP (2800L) – $200
- 500L Vessel CIP (1700L) – $120
- 200L Vessel CIP (1000L) - $70

Media Preparation

Equipment Cost Comparison

Single Use Mixer	Stainless Steel Vessel
2000L SUM - $160,000	2000L Vessel* - $300,000
500L SUM - $110,000	650L Vessel* - $260,000
200L SUM - $80,000	200L Vessel - $150,000
Total Capital Cost - **$350,000**	Equipment Cost - **$710,000**
* SUM Cost Includes SUM+TCU+Instruments+Load Cell	* Vessel Cost Includes Tanks+Valves+ Platform
Negligible	Automation Cost – $100,000
Annual Maintenance - $10,000	Annual Maintenance - $60,000
Installation + Eng. - $70,000	Installation + Eng. - $140,000
Validation - $50,000	Validation - $140,000
No CIP Skid	CIP Skid (Equip+Instal) - $400,000
Total - **$480,000**	Total - **$1,550,000**

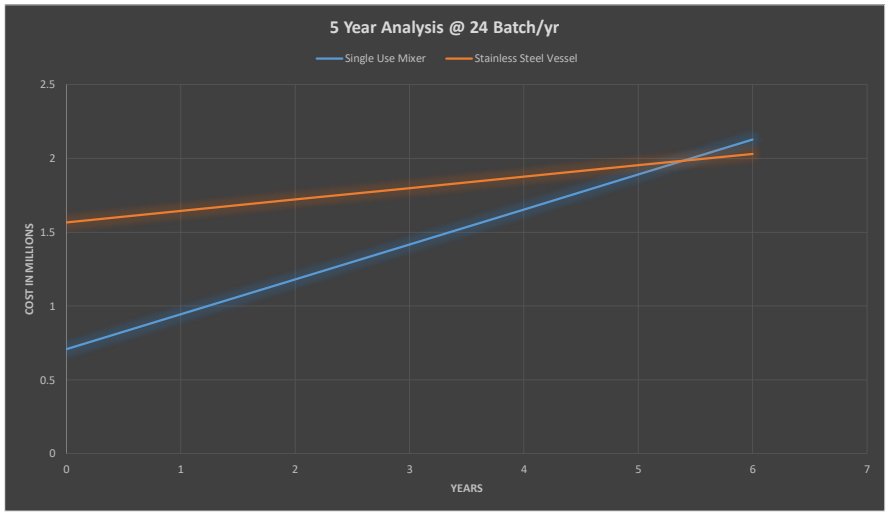
Media Preparation

Operation Cost Comparison

Single Use Mixer	Stainless Steel Vessel
2000L Bags - $2,000	2000L Vessel CIP - $200
500L Bags - $2,400	500L Vessel CIP - $240
200L Bags - $4,000	200L Vessel CIP - $280
Total Bag Cost per Batch - **$8,400**	WFI Cost per Batch- **$720**
24 Batch Annually - **$226,800**	24 Batch Annually - **$17,280**
48 Batch Annually - **$403,200**	48 Batch Annually - **$34,560**

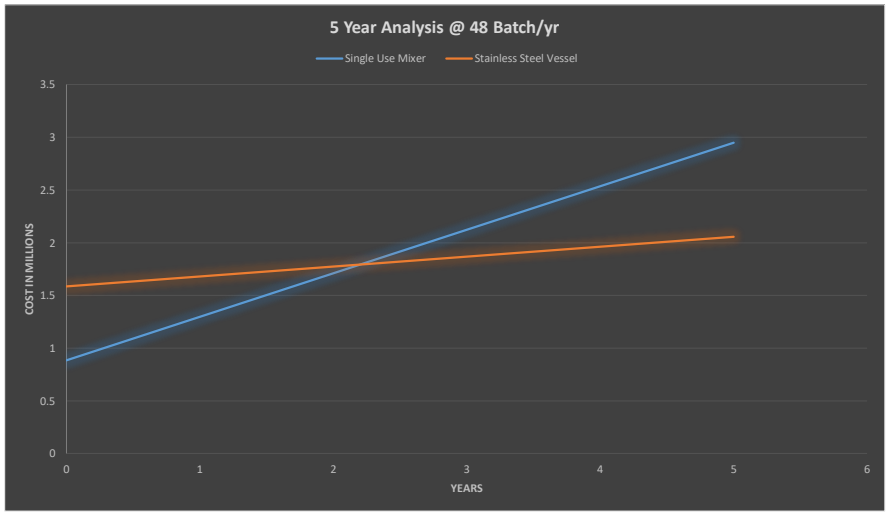
Media Preparation

5 Year Analysis at 24 Batch/yr



Media Preparation

5 Year Analysis at 48 Batch/yr



Media Preparation

Additional Key Decision Points

If Stainless Steel Option than –

- Additional Pre-Treatment capacity
- Bigger WFI Generation and Storage
- Clean Steam Generator

All this can significantly add to the capital cost.

Buffer Preparation

SUMs and Bag Consumption per Batch

Single Use Mixer (Size)	Number of Bags per Batch @1.5 g/l Titer	Number of Bags per Batch at 3 g/l Titer
2000L	2	4
1000L	5	6
500L	2	2
200L	1	4

SUMs are non-jacketed with load cells and have temperature transmitters.

Buffer Preparation

Stainless Steel Vessel

Following vessel sizes are comparable to the single use equipment selected -

1. 2000L
2. 650L
3. 200L

Stainless Steel Vessels are assumed as skid with capability to perform rinses, CIP and SIP. Buffer Prep vessels during everyday operations will be rinsed with WFI.CIP and SIP on as needed basis and accounted in Annual Maintenance.

WFI Rinse Cost Estimate per Vessel –

- 2000L Vessel Rinse (10 mins*175 l/m*0.07) – $125
- 500L Vessel Rinse (10 mins*110 l/m*0.07) – $80
- 200L Vessel Rinse (10 mins*60 l/m*0.07) - $50

Buffer Preparation

Equipment Cost Comparison

Single Use Mixer	Stainless Steel Vessel
2000L SUM - $125,000	2000L Vessel* - $300,000
1000L SUM - $90,000	650L Vessel* - $260,000
500L SUM - $75,000	200L Vessel - $150,000
200L SUM - $60,000	
Total Capital Cost - **$350,000**	Equipment Cost - **$710,000**
* SUM Cost Includes SUM+Instruments+Load Cell	* Vessel Cost Includes Tanks+Valves+ Platform
Negligible	Automation Cost – $100,000
Annual Maintenance - $10,000	Annual Maintenance - $60,000
Installation + Eng. - $70,000	Installation + Eng. - $140,000
Validation - $50,000	Validation - $140,000
No CIP Skid	CIP Skid (Equip + Instal) - $400,000
Total - **$480,000**	Total - **$1,550,000**

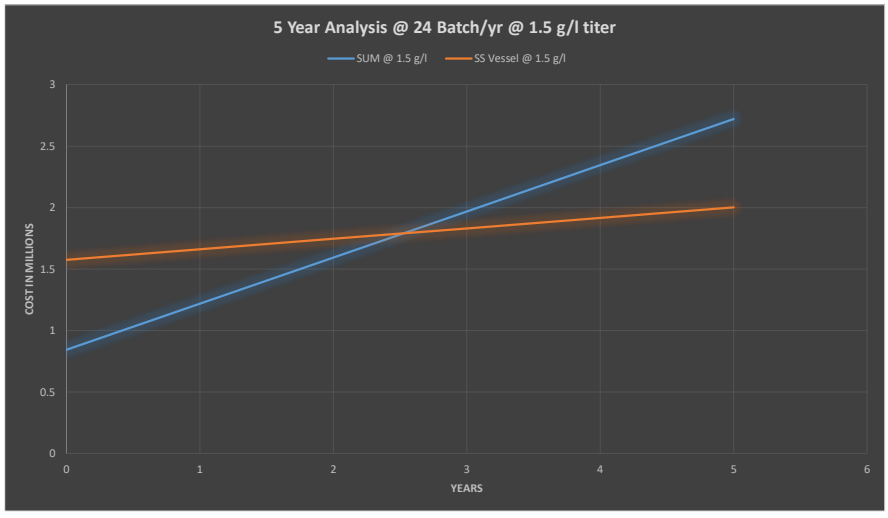
Buffer Preparation

Operation Cost Comparison

Single Use Mixer	Bag Cost per Batch	Stainless Steel Vessel	Cost Per Batch
@ 1.5 g/l			
2000L = 2*2000	$4000	2000L Vessel= 7 Rinses*125	875
1000L = 5*1600	$8000	650L Vessel = 1 Rinse*80	80
500L = 1*1200	$1200	200L Vessel = 2 Rinses*50	100
200L = 2*1000	$2000		
Total Cost/Batch	**$15,200**	Total Cost per Batch	**$1055**
@ 24 Batches/Year	**$364,800**	@ 24 Batches/Year	**$25,320**
@ 48 Batches/Year	**$729,600**	@ 48 Batches/Year	**$50,640**

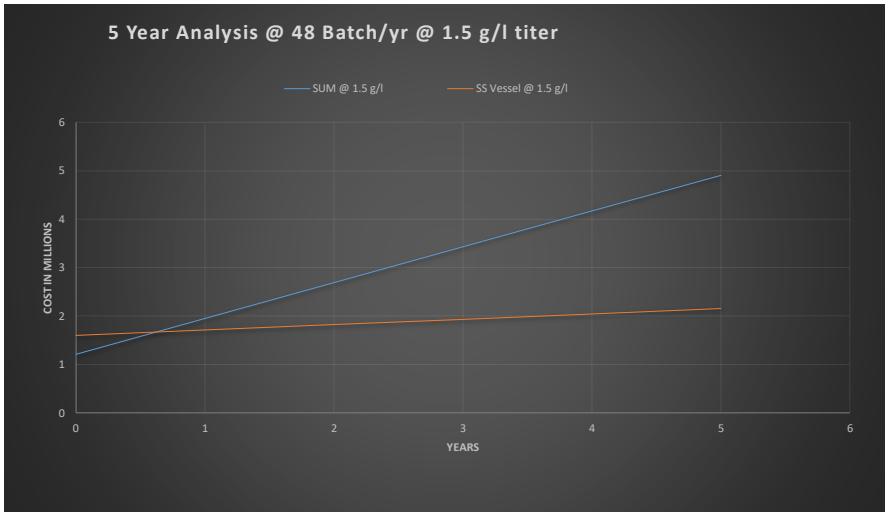
Buffer Preparation

5 Year Analysis at 24 Batch/yr



Buffer Preparation

5 Year Analysis at 48 Batch/yr



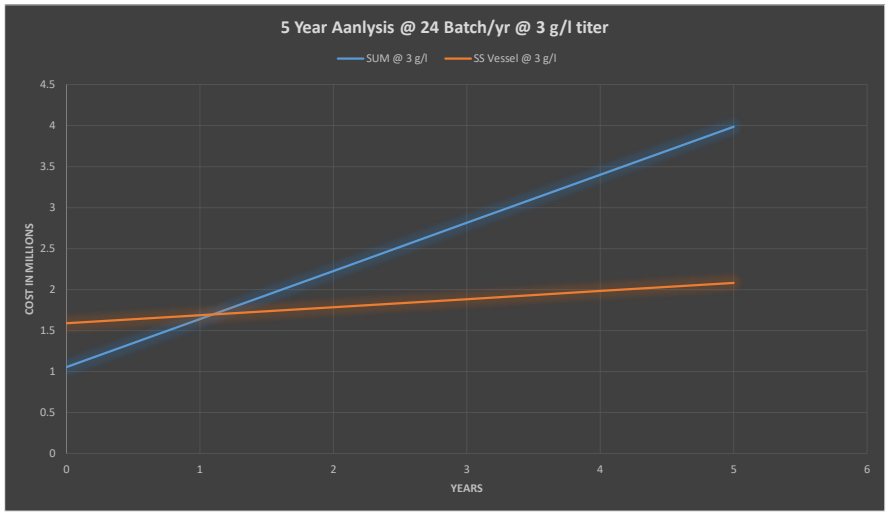
Buffer Preparation

Operation Cost

Single Use Mixer	Bag Cost per Batch	Stainless Steel Vessel	Cost Per Batch
@ 3 g/l			
2000L = 4 * 2000	$8000	2000L Vessel = 10 Rinses * 125	$1250
1000L = 6 * 1600	$9600	650L Vessel = 2 Rinse * 80	$160
500L = 2 * 1200	$2400	200L Vessel = 4 Rinses * 50	$200
200L = 4 * 1000	$4000		
Total Cost/Batch	**$24,000**	Total Cost/Batch	**$1610**
@ 24 Batches/Yr	**$576,000**	@ 24 Batches/Yr	**$38,640**
@ 48 Batches/Yr	**$1,152,000**	@ 48 Batches/Yr	**$77,280**

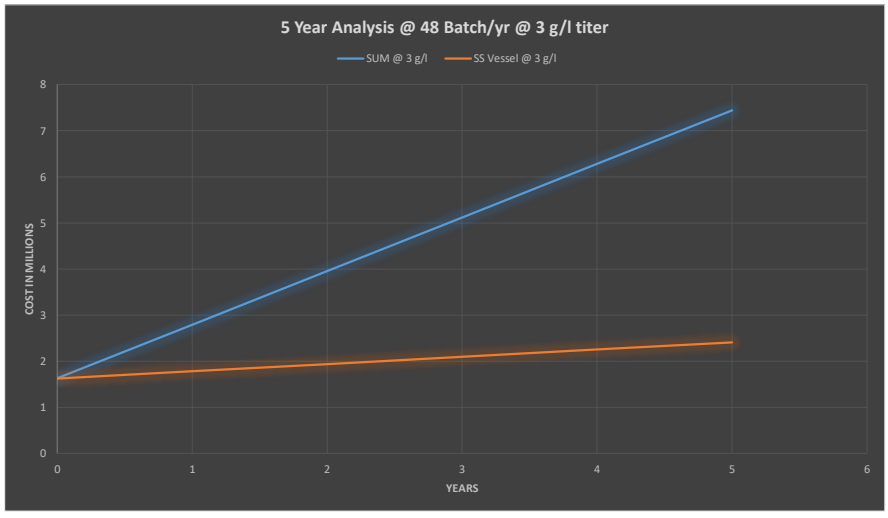
Buffer Preparation

5 Year Analysis at 24 Batch/yr



Buffer Preparation

5 Year Analysis at 48 Batch/yr



Media and Buffer Preparation

Additional Considerations

	Single Use Mixer	Stainless Steel
1	Bag Storage - Additional load on warehouse storage. Supply chain management. Dependency of vendor.	If existing facility than bigger WFI storage tank needed. If new facility than it could mean bigger pre-treatment skid, WFI generation and storage vessel.
2	Bag Disposal Cost – incineration, landfill (environmental impact)	Increased Process Waste Treatment
3	No cleaning validation needed.	Cleaning Validation & Requalification
4	Low annual maintenance	High annual maintenance (gasket replacement, agitator maintenance, re-passivation, tank cleaning and finishing)
5	No CIP Skid	CIP skid needed
6	Low flowrates	Can be pressure transferred, no flowrate restrictions

Buffer Preparation

Higher Titers

- With increasing titers its becoming difficult sizing buffer vessels.
- Leads to high investment costs.
- Substantial space requirements.
- Higher running costs and time consuming

Buffer Preparation

IN-LINE DILUTION SKIDS are key to managing variability.

- One size can fit all.
- Helps eliminating large buffer hold vessels
- Improved buffer consumption efficiency
- Better process floor space utilization
- Increases flexibility and throughput
- Optimize or eliminates CIP

Key points to consider
- Ability to concentrate (5x, 10x…)
- Expiration Date Strategy

Media and Buffer Preparation

Conclusion

- Both Single Use Technology and Stainless Steel options have their own unique sets of challenges and advantages.
- Identifying site key requirements (multiple products, space, number of batches, cadence….), conditions (new facility vs renovation) will help narrow down choice.
- **HYBRID** option would help in extracting the best of both worlds. This gives you increased flexibility, can significantly reduce operational cost and also offset high capital cost.

Single Use ~~Vs.~~ AND! Stainless Steel

Teaching an Old Facility New Tricks

Teaching an Old Facility New Tricks

"Protein Sciences lauds benefits of Pfizer refit in commercial scale-up"

- **Background – Licensed at 500L scale and wanted to go larger!**

In January 2013, the US Food and Drug Administration (FDA) approved Protein Sciences Corporation's trivalent influenza vaccine Flublok, made using the firm's Baculovirus Expression Vector System (BEVS) rather than the influenza virus itself or eggs, as in other flu vaccines.

Up-scaling to commercial production was therefore needed, VP of Manufacturing Operations Mireli Fino said at last month's Biological Production Forum in Dublin, Ireland, especially in light of the firm's $147,000 contract with the Biomedical Advanced Research and Development Authority (BARDA) to produce up to 50 million doses of Flublok within six months of a potential flu pandemic.

- **Greenfield Option – too much time and too much money**

"The first thing we looked at was a greenfield facility, but the primary concerns with constructing a new facility is time and money," she continued, adding construction and validation would have taken up to five years, rather than the one-two years' timeline the company was looking for.

- **CMO Option – path forward until…**

"The focus was then on contract manufacturing," she said, saying a CMO is the most expedient way to launch a commercial product. *"We looked at companies in the US primarily and we narrowed it down from 30 companies to just two,"* with contract negotiations beginning in July/August 2012.

- **Pfizer lease was available**

"[The Pfizer site] was a licensed manufacturing facility since 2000, just recently decommissioned and it had been fully qualified… There were relative minor expenses expected for equipment and it was ready for capacity expansion… and Pfizer offered very attractive lease terms," so the firm moved in in December 2012.

https://www.biopharma-reporter.com/Article/2014/06/13/Protein-Sciences-lauds-benefits-of-Pfizer-refit-in-commercial-scale-up

Teaching an Old Facility New Tricks

Technology transfer and scale-up of the Flublok® recombinant hemagglutinin (HA) influenza vaccine manufacturing process

Barry Buckland[a,c,*], Robert Boulanger[a], Mireli Fino[a], Indresh Srivastava[a], Kathy Holtz[a], Nikolai Khramtsov[a], Clifton McPherson[a], Jamal Meghrous[a], Paul Kubera[b], Manon M.J. Cox[a]

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Pandemic
H7N9

ABSTRACT

Multiple different hemagglutinin (HA) protein antigens have been reproducibly manufactured at the 650 L scale by Protein Sciences Corporation (PSC) based on an insect cell culture with baculovirus infection. Significantly, these HA protein antigens were produced by the same Universal Manufacturing process as described in the biological license application (BLA) for the first recombinant influenza vaccine approved by the FDA (Flublok®). The technology is uniquely designed so that a change in vaccine composition can be readily accommodated from one HA protein antigen to another one. Here we present a vaccine candidate to combat the recently emerged H7N9 virus as an example starting with the genetic sequence for the required HA, creation of the baculovirus and ending with purified protein antigen (or vaccine component) at the 10 L scale accomplished within 38 days under GMP conditions.

The same process performance is being achieved at the 2 L, 10 L, 100 L, 650 L and 2500 L scale. An illustration is given of how the technology was transferred from the benchmark 650 L scale facility to a retrofitted microbial facility at the 2500 L scale within 100 days which includes the time for facility engineering changes.

The successful development, technology transfer and scale-up of the Flublok® process has major implications for being ready to make vaccine rapidly on a worldwide scale as a defense against pandemic influenza. The technology described does not have the same vulnerability to mutations in the egg adapted strain, and resulting loss in vaccine efficacy, faced by egg based manufacture.

- **Lease approval to facility conversion and process demonstration batch completed in <100 days!**

Buckland B, et al. Vaccine (2014), Volume 32, Issue 42, 22 September 2014, Pages 5496-5502 (http://dx.doi.org/10.1016/j.vaccine.2014.07.074)

Teaching an Old Facility New Tricks

The Pfizer manufacturing facility was a traditional stainless steel design for a vaccine produced using a microbial system

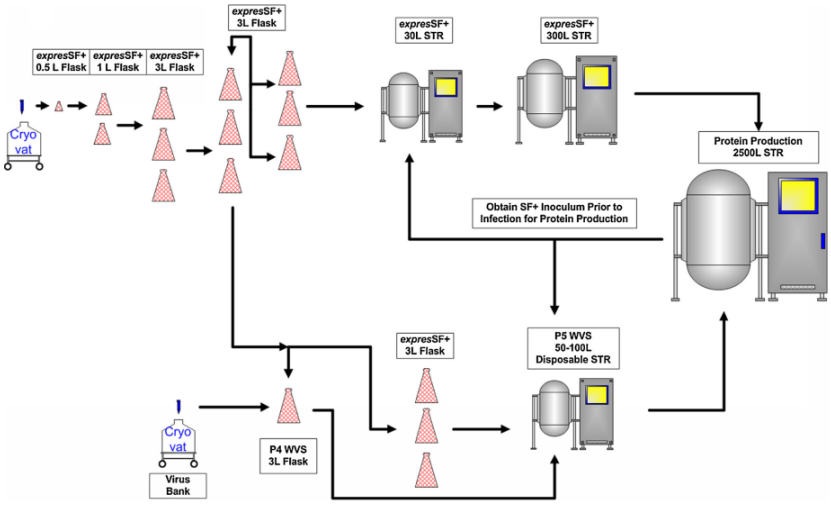
So would a Baculovirus/insect cell system FIT?:

- **Fermenters/bioreactors were designed for a microbial system**
  - Ruston impellers
  - No sparge capability
  - Media was sterilized in situ
  - Short incubation times in each vessel
- **SS media and buffer systems**
  - SS buffer/media preparation tanks
  - SS buffer hold tanks
- **Continuous centrifuge**
  - Supernatant contains product - solids discarded
- **Available downstream equipment**
  - Large scale chromatography, filtration housings and TTF equipment available.
- **Large autoclaves are available**
  - Single use assemblies can be sterilized in house
  - Manual ported valves allow SU integration with stainless steel

Buckland B, et al. Vaccine (2014), Volume 32, Issue 42, 22 September 2014, Pages 5496-5502 (http://dx.doi.org/10.1016/j.vaccine.2014.07.074)

Teaching an Old Facility New Tricks

The Upstream HA process:



Buckland B, et al. Vaccine (2014), Volume 32, Issue 42, 22 September 2014, Pages 5496-5502 (http://dx.doi.org/10.1016/j.vaccine.2014.07.074)

Teaching an Old Facility New Tricks

The Upstream Retrofit

- **SF+ Inoculum and P4 Virus stock preparation**
  - Ability to store frozen virus stock and cell stock
  - Disposable shake flasks
  - Addition BSC required for required aseptic operations
  - Segregate into separate rooms to separate Baculovirus +/-

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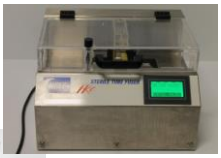
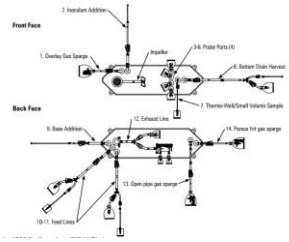
Teaching an Old Facility New Tricks

The Upstream Retrofit (con't)

- **SF+ Inoculum Transfer and P5 Virus stock preparation (and transfer)**
  - 50-100L of P5 WVS required
  - Closed system for transfer of inoculum to the R1 Fermentor
  - Aseptic connectors vs tube welders
  - Segregate into separate rooms to separate Baculovirus +/-





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Teaching an Old Facility New Tricks

The HA Recovery process:

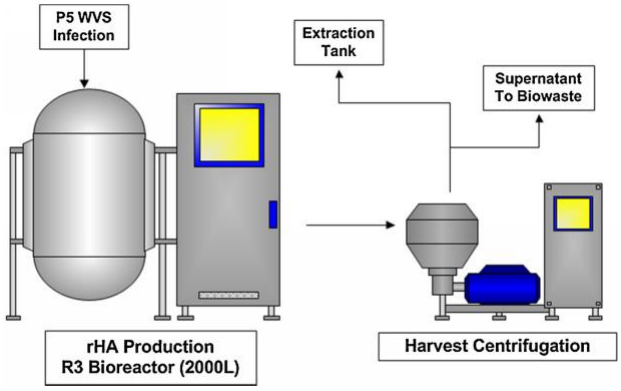
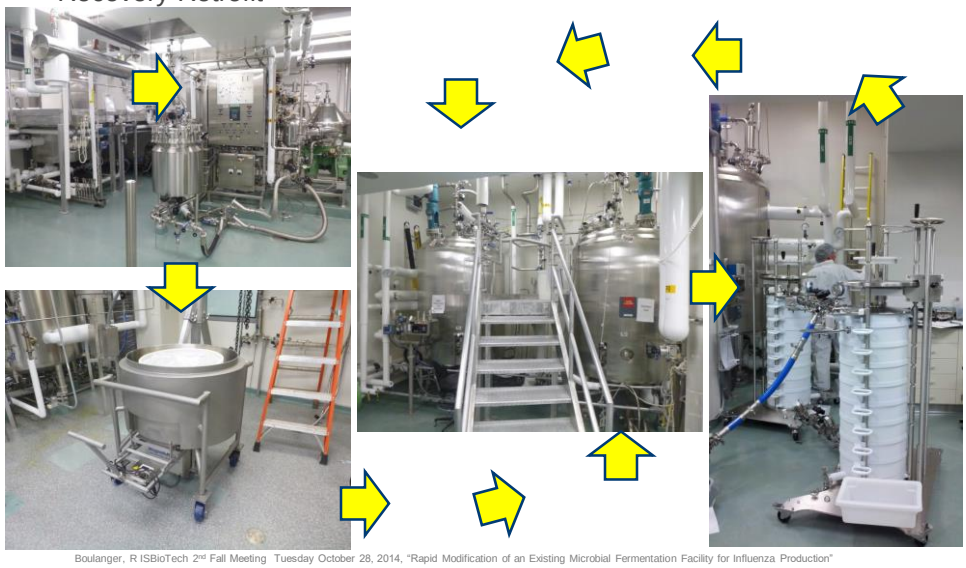


Fig. 3. Harvest of the suspension culture insect cell pellet by disk stack centrifuge at the 2500 L scale.

Buckland B, et al. Vaccine (2014), Volume 32, Issue 42, 22 September 2014, Pages 5496-5502 (http://dx.doi.org/10.1016/j.vaccine.2014.07.074)

Teaching an Old Facility New Tricks

Recovery Retrofit



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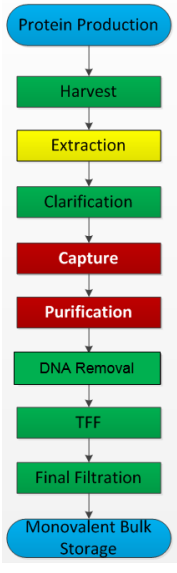
Teaching an Old Facility New Tricks

The Downstream HA process:



www.who.int/immunization/research/meetings_workshops/Manon_Cox.pdf; Meghrous, et al. Vaccine (2009) Volume 28, Issue 2, 11 December 2009, Pages 309-316

Questions?

Please use the microphone indicated so our recording includes audio of your question