NEPDA meeting
11Sep2013

Presented by:
Dawn Tavalsky
Sr. Director QA, genzyme, a Sanofi Company
Dawn.tavalsky@genzyme.com
Outline:

- Scope of a cleaning validation program
- Where do I start
  - How to collect the data I need
  - Understand your soils
  - Understand your equipment
- Grouping of Soils
  - Worst Case Soil Studies
- Grouping of Equipment
Definition of Cleaning Validation

The purpose of cleaning validation is to establish documented evidence with a high degree of assurance that the cleaning process will consistently yield results that meet predetermined specifications and quality characteristics.

The action of proving in accordance to principles of GMP that the cleaning procedure actually leads to expected results.

Validation helps to know the process capability and create an avenue for process improvement.
Definition of Cleaning Validation (Cliff Notes)

- Removing:
  - all components of the previous product (antigen, Hg, Al, egg proteins, etc)
  - Bioburden
  - Endotoxin
  - Detergents
- Down to levels that are “safe”
  - have no impact on the next product or the patient with a large safety factor built in
- Prior to using the equipment for the next product
Why do we need Cleaning Validation?

Sources of Regulatory Guidance for Cleaning Validation

- Canadian HPFB, Cleaning Validation Guidelines (2001)
- Pharmaceutical Inspection Convention (PIC), Recommendations on...Cleaning Validation (2004)
Why do we need Cleaning Validation?

BUT – more than the reason “because we have to,” we need cleaning validation to ensure that our products remain safe and effective for our patients – let’s move our industry culture from one of “compliance” to one of “quality”
Scope of Cleaning Validation Program

- It is important to define the scope of your cleaning validation program.
  - The cleaning must be validated for all “product contact parts”
  - There is a difference between parts that come into contact with product and parts that are in the process flow path
Product Contact Equipment

VS

Equipment that is Contacted by Product
Scope of Cleaning Validation Program (cont)

- Biologically Active Material
  - (BAM)
- Non-Biologically Active Material
  - (non-BAM)

- BAM and non-BAM contact parts in the process flow path
  - Interior of tanks, pipes, etc
- BAM and non-BAM parts NOT in the process flow path that material may see spillage or splashing
  - Tools, tables, floors, walls, cleaning equipment (parts washer, sonicator)
The Cleaning Validation Team Mascot
The Manufacturing Process from a Cleaning Validation Perspective

The closer you get to the patient, the more stringent your cleaning validation criteria become –

Sanofi Example

**Processing Stage 1** - those phases of a manufacturing process that precede the purification steps. Stage 1 process equipment includes the preparation of raw materials, the fermentation or culture process and primary filtration steps.

**Processing Stage 2** - those phases of a manufacturing process involving product purification and conjugation up to but not including final formulation

**Processing Stage 3** - those phases of a manufacturing process involving final formulation and filling
Scope of Cleaning Validation Program (cont)

The remainder of this training will be with respect to equipment that is in the process flow path and comes into contact with BAM and non-BAM materials.
Where do I begin?

Start with a document Blueprint for your cleaning validation program
Where do I begin?

Next Make a Project Validation Master Plan that describes how the cleaning validation program will be established and implemented – include phasing in of new SOP requirements
Validation Master Plan:

The establishment of a dynamic written plan that defines the overall approach to a validation discipline or project. It will define the terminology to be used in all subsequent documentation, and outline descriptions of the facility site, the manufacturing processes, and the scope and implementation of the validation sequence.
Where do I begin?

- Understand your processes
  - Collect Process Flow Diagrams (PFD’s) and operating instructions (batch records, SOP’s, etc) for the processes onsite that will be included in the cleaning validation program

- Understand your soils
  - Make a list of all soils that must be cleaned from product contact equipment (including intermediate soils)
  - Make a list of all detergents/cleaning agents approved for use on site

- Understand your Equipment
  - Make an inventory list of all equipment that requires cleaning
  - Make an inventory list of all equipment USED for cleaning

- Understand the Legacy Cleaning onsite
  - Make a trace matrix for all existing cleaning validations

- Understand the Analytical Methods available for use
Where do I begin?

From all of the data you have gathered, develop a trace matrix “snapshot” for the processes that require cleaning validation.
<table>
<thead>
<tr>
<th>Step Description</th>
<th>Step Reference</th>
<th>Room</th>
<th>Equipment</th>
<th>Material(s) of Construction</th>
<th>Equipment ID Number</th>
<th>Soil</th>
<th>Product Contact (D, I, N)</th>
<th>Cleaning SWI(s)</th>
<th>Cleaning SWI Notes</th>
<th>Cleaning Validation Report(s)</th>
<th>Soils Encountered</th>
</tr>
</thead>
</table>
| Inoculum
Preparation | Batch Production Record #3030413, section 2; PFD3030044FA | 109  | 20L Inoculum Bottle | Glass or Stainless Steel | MR0023, Flu Seed Culture | 1970 | D | J000722 (sec 8 & 11) | To Service Area | C000540 (BSA WSH)*if GLASS ● C002004 & C002011 (BSA WSH)*if GLASS C002077 & C002078 (BSA WSH)*if GLASS | Thimerosal ● N. meningitides Polysaccharide Depolymerized Concentrate ● TET Toxin |
|                  |                |      | Magnetic Stirring Bar | Plastic | MR0023, Flu Seed Culture |        | D | J000722 (sec 8 & 11), J000491, J000823, & J000575 | To Service Area | C001815 (BSA ULT) ● C000540 (BSA ULT) | Meninge Conjugate ● Thimerosal |
|                  |                |      | Inoculum Bottle Stopper | Silicone | MR0023, Flu Seed Culture |        | D | J000722 (sec 8 & 11), J000491, J000823, & J000575 | To Service Area | C000540 (BSA ULT) ● C000540 (BSA WSH) | Thimerosal ● Thimerosal |
|                  |                |      | Automatic Inoculum Bottle Siphon | Stainless Steel | MR0023, Flu Seed Culture |        | D | J000722 (sec 8 & 11), J000536 (sec 5), J000491 (sec 6), J000823, & J000575 | To Service Area | C001815 (BSA ULT) ● C0011159 (B37 ULT) ● C000540 (BSA ULT) | Meninge Conjugate ● Harvest Fluids ● Thimerosal |
|                  |                |      | Bottle Stopper Clamp | Unknown | MR0023, Flu Seed Culture |        | I | Not Found | N/a | C000540 (BSA ULT) ● C000540 (BSA WSH) | Thimerosal ● Thimerosal |
| Egg Unloading, Storage, and Pre-Inoculation | Batch Production Record #3030413, section 3; SWI #3030413; PFD3030044FA | 144  | Punch/Needle Assembly | Stainless Steel | In-Process Monovalent Vaccine | N/a | D | J000722 (sec 8 & 11), J000823, & J000575 | To Service Area | C001159 (B37 ULT) | Harvest Fluids |
|                  |                |      | Inoc. Manifold | Stainless Steel | In-Process Monovalent Vaccine | N/a | D | J000722 (sec 8 & 11), J000525 (sec 5), J000491, J000823, & J000575 | To Service Area | C006108 (B37 ULT) | Influenza Inoculum |
|                  |                |      | Needle Rinsing Manifold | Stainless Steel | In-Process Monovalent Vaccine | N/a | D | J000491, J000823, & J000575 | To Service Area | C006108 (B37 ULT) | Influenza Inoculum |
|                  |                |      | Rinsing Manifold Tubing | Unknown | In-Process Monovalent Vaccine | N/a | D | Not Found | N/a | N/a | N/a |
|                  |                |      | Forces | Stainless Steel | In-Process Monovalent Vaccine | N/a | D | Not Found | N/a | C001815 (BSA ULT) ● C001159 (B37 ULT) | Meninge Conjugate ● Harvest Fluids & Virus Subunit Fluids |
|                  |                |      | Replacement Punch | Stainless Steel | In-Process Monovalent Vaccine | N/a | D | J000491, J000823, & J000575 | To Service Area | C001159 (B37 ULT) | Harvest Fluids |
|                  |                |      | Replacement Needle | Stainless Steel | In-Process Monovalent Vaccine | N/a | D | J000491, J000823, & J000575 | To Service Area | C001159 (B37 ULT) | Harvest Fluids |
|                  |                |      | 4L Collection Bottle | Glass | In-Process Monovalent Vaccine | N/a | D | J000491 (sec 6) & J000569 | To Service Area | C002077 & C002078 (BSA WSH)*if GLASS ● C006476 (BSA WSH) | TET Toxin ● Tetanus Toxoid (NT0151) |
|                  |                |      | Inoculum Volume Cup Tray | Stainless Steel or Plastic | In-Process Monovalent Vaccine | N/a | D | Not Found | N/a | C006108 (B37 ULT) ● C001159 (B37 ULT) | Influenza Inoculum ● Harvest Fluids |
|                  |                |      | Inoculator | Stainless Steel | In-Process Monovalent Vaccine | 7491 & 335 | D | J000722 (sec 8 & 11) | Manual Cleaning | Not Found | N/a |
|                  |                |      | 20L Inoculum Bottle (not previous step) | Unknown | In-Process Monovalent Vaccine | 1970 | D | J000722 (sec 8 & 11) | To Service Area | C002004 & C002011 (BSA WSH)*if GLASS ● C002077 & C002078 (BSA WSH)*if GLASS | N. meningitides Polysaccharide Depolymerized Concentrate ● TET Toxin |
|                  |                |      | 10mL Graduated Cylinder | Plastic or Glass | In-Process Monovalent Vaccine | N/a | D | Not Found | N/a | C00540 (BSA ULT) | Thimerosal |

Notes:
- "Warm Room" is applicable
- "Soil" refers to types of soil encountered during the cleaning process
- "Equipment ID Number" is applicable for certain equipment records
- "Cleaning SWI" refers to cleaning step identification numbers
- "Cleaning SWI Notes" provide additional notes on the cleaning process
First:

Know and Understand the Soils and Residues You Must Clean
The residues remaining on process equipment surfaces contain proteins, lipids, sugars and salts. These residues result from

1) the cellular growth process, with protein and lipid components comprising the primary constituents

2) the media preparation process, with amino acids, vitamins, sugars, lipids and salts or

3) the buffer preparation process with salts, sugars and organics (alcohol, glycol).

4) Other (e.g., microcarriers, etc)
This refers to the degradation and dispersion of proteinaceous residues.

Hydrolysis typically involves the cleavage of proteins into smaller peptide chains that are soluble in aqueous solutions. Hydrolysis can be accomplished, depending upon the physical and chemical properties of the specific protein, by alkaline or acidic cleaning solutions.
Dissolving

This refers to the direct solubility of residues in aqueous cleaning solution.

Readily dissolved residues include non-heat treated proteins, short chain alcohols, excipients, and monovalent salts (dissolution of polyvalent salts typically occurs in acidic solutions).

Dissolving is often the mechanism of choice as it is the simplest of the commonly used cleaning mechanisms, and works well in conjunction with degradation mechanisms that render residues soluble.
Saponifying

This mechanism is used for the chemical cleavage and degradation of lipids, in the form of tri-glycerides, which are not freely soluble in aqueous solutions.

Saponification involves a hydration reaction where free hydroxide breaks the ester bonds between fatty acids and glycerol of tri-glycerides, resulting in free fatty acids and glycerol.

The resultant materials are both soluble in aqueous solutions.

As lipids are nearly always present in some stage of a process, especially those involving cell growth and separation, saponification is an important cleaning mechanism for residue removal from process equipment surfaces.
De-Mineralizing

This mechanism involves the removal of insoluble salts on process equipment surfaces.

The formation of insoluble salts is caused by the exposure of polyvalent cations to the high pH of alkaline-based cleaning solutions.

These residues can be effectively removed through the use of an acidic cleaning agent, which will dissolve the mineral salts.

FIGURE 5. Octahedral sheet with aluminum cations. Oxygen ions are balanced with the addition of hydrogen protons (H⁺).
Neutralizing

Alkaline cleaning agents often do not rinse very freely, and complete removal by rinsing would require relatively large amounts of water.

These alkaline residues can be effectively removed through the use of an acidic cleaning agent.
Chelating agents bind or capture trace amounts of iron, copper, manganese, calcium and other metals that occur naturally in many materials.
Raw Meat and Prions
Formalin

Sucrose

Sodium Citrate

Triton X-100

Aluminum

Mercury
Allantoic Fluid

Figure 1. A diagrammatic representation of the extra-embryonic membranes and fluid compartments for the chick embryo around a third of the way through incubation. Note that the sizes of these structures, and their development relative to each other, have been modified to clarify types of cells present in each of the membranes.
Your Soils may Change with Heat/pH/Time
Gelatin
Gram-positive bacteria

Gram-negative bacteria are pathogenic, meaning they can cause disease in a host organism. This pathogenic capability is usually associated with certain components of Gram-negative cell walls, in particular the lipopolysaccharide (also known as LPS or endotoxin).
Endotoxin
Viral Material after Cleaning and/or Disinfection

Resulting Vaccine - Split Virus

Resulting Material after cleaning with cleaning detergents or disinfectants (to be demonstrated through degradation studies)
Protein Folding / Protein Surface Bonding

Primary structure
Secondary structure
Tertiary structure
Quaternary structure

Amino acid residues
α Helix
Polypeptide chain
Assembled subunits

PEG
γ-APS
γ-APS

(a)
(b)
(c)
Evaporative Dispersion

The coffee-stain effect: solute goes to edge of evaporating drop
Soil Lists from in-process Streams

Use the PFD and list all of the components that are present on the equipment just prior to cleaning – and then assign “that” soil a name

Example: Final soil on the 3000L fermentor:

Name - Fermentation Soil

- Bacteria/product
- Soy Media
- HCl
- NaOH
- Antifoam
- Phenol
# Obtain Subjective Data

**Bldg-37: Sonicator Cleaning Development Questionnaire**

<table>
<thead>
<tr>
<th>Name:</th>
<th>Employee Position:</th>
</tr>
</thead>
<tbody>
<tr>
<td>Reporting Supervisor:</td>
<td>Date:</td>
</tr>
</tbody>
</table>

1) Please fill out the following table:

<table>
<thead>
<tr>
<th>Soil Category</th>
<th>Sub Category</th>
<th>Cleanability</th>
<th>Viscosity</th>
<th>Solubility</th>
</tr>
</thead>
<tbody>
<tr>
<td>ACP</td>
<td>1 Dose</td>
<td>Rank from 1 - 5 with 1 being easiest to clean and 5 being hardest to clean</td>
<td>Rank from 1 - 5 with 1 being the least viscous and 5 being the most viscous</td>
<td>Rank from 1 - 5 with 1 being the least soluble in water and 5 being the most soluble in water</td>
</tr>
<tr>
<td>Flu PBS</td>
<td>(none)</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Flu</td>
<td>Adult Stake Syr-Mex</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td></td>
<td>Adult Syringe</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td></td>
<td>Ped Syringe (Dedicated)</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Fluzone</td>
<td>10 Dose</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Diluent for Meninge</td>
<td>1 Dose</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td></td>
<td>Bact Dil 10 Dose</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Meninge</td>
<td>1 Dose</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td></td>
<td>10 Dose</td>
<td></td>
<td></td>
<td></td>
</tr>
</tbody>
</table>
### Obtain Subjective Data

<table>
<thead>
<tr>
<th>Soil Category</th>
<th>Sub Category</th>
<th>Cleanability</th>
<th>Ivy</th>
<th>Mitchell</th>
<th>Paul</th>
<th>Gladys</th>
<th>Mercedes</th>
<th>Avg.</th>
<th>STDEV.</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td></td>
<td>Rank from 1 - 5 with 1 being easiest to clean and 5 being hardest to clean</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Fluzone, SV, 2007</td>
<td>NP Unit DS Vial</td>
<td></td>
<td>2</td>
<td>2</td>
<td>2</td>
<td>2</td>
<td>1</td>
<td>1.8</td>
<td>0.4472136</td>
</tr>
<tr>
<td></td>
<td>10 Dose</td>
<td></td>
<td>3</td>
<td>2</td>
<td>2</td>
<td>2</td>
<td>1</td>
<td>2</td>
<td>0.7071068</td>
</tr>
<tr>
<td></td>
<td>NP Unit DS</td>
<td></td>
<td>1</td>
<td>2</td>
<td>2</td>
<td>2</td>
<td>1</td>
<td>1.6</td>
<td>0.5477226</td>
</tr>
<tr>
<td></td>
<td>NP Unit DS Syr .25ml</td>
<td></td>
<td>1</td>
<td>2</td>
<td>2</td>
<td>2</td>
<td>1</td>
<td>1.6</td>
<td>0.5477226</td>
</tr>
<tr>
<td>Tet-Meninge</td>
<td>Tetanus/Diptheria, AD. Unit Dose PF</td>
<td></td>
<td>4</td>
<td>4</td>
<td>5</td>
<td>4</td>
<td>4</td>
<td>4.2</td>
<td>0.4472136</td>
</tr>
<tr>
<td></td>
<td>Diphtheria/Tetanus, Absorbed</td>
<td></td>
<td>4</td>
<td>5</td>
<td>5</td>
<td>4</td>
<td>4</td>
<td>4.4</td>
<td>0.5477226</td>
</tr>
<tr>
<td></td>
<td>Syrinbge, Tet-Diph Toxoid, Abso</td>
<td></td>
<td>4</td>
<td>5</td>
<td>5</td>
<td>4</td>
<td>4</td>
<td>4.4</td>
<td>0.5477226</td>
</tr>
<tr>
<td></td>
<td>Tetanus Toxoid, AD. 1 DS, Prese</td>
<td></td>
<td>2</td>
<td>2</td>
<td>5</td>
<td>4</td>
<td>4</td>
<td>3.4</td>
<td>1.3416408</td>
</tr>
<tr>
<td></td>
<td>DtabP, Absorbed (biken)-1 DS Pres</td>
<td></td>
<td>3</td>
<td>3</td>
<td>3</td>
<td>3</td>
<td>3</td>
<td>3</td>
<td>0</td>
</tr>
</tbody>
</table>
# Product Information

<table>
<thead>
<tr>
<th>Reagent</th>
<th>Solubility</th>
<th>Toxicity</th>
<th>Product A</th>
<th>Product B</th>
<th>Product C</th>
<th>Product D</th>
<th>Unit/dose</th>
</tr>
</thead>
<tbody>
<tr>
<td>Formalin</td>
<td>Freely</td>
<td>Moderately Toxic</td>
<td>0.00504826</td>
<td>0.00504826</td>
<td>0.00504826</td>
<td>0.005107597</td>
<td>ml</td>
</tr>
<tr>
<td>Thimerosal</td>
<td>Soluble</td>
<td>Moderately Toxic</td>
<td>0</td>
<td>0</td>
<td>0</td>
<td>0</td>
<td>0.00456592 ml</td>
</tr>
<tr>
<td>NaOH</td>
<td>soluble</td>
<td>Slightly Toxic</td>
<td>0.002581532</td>
<td>0.002581532</td>
<td>0.002581532</td>
<td>0.003286401</td>
<td>g</td>
</tr>
<tr>
<td>Sodium Citrate</td>
<td>Soluble</td>
<td>Slightly Toxic</td>
<td>0.025890179</td>
<td>0.025890179</td>
<td>0.025890179</td>
<td>0.025219891</td>
<td>g</td>
</tr>
<tr>
<td>P.B.S 0.006M</td>
<td>Freely</td>
<td>Slightly Toxic</td>
<td>52.55405564</td>
<td>52.55405564</td>
<td>52.55405564</td>
<td>50.46327387</td>
<td>ml</td>
</tr>
<tr>
<td>60/40 Isopropanol</td>
<td>soluble</td>
<td>Slightly Toxic</td>
<td>1.943832433</td>
<td>1.943832433</td>
<td>1.943832433</td>
<td>0.050560013</td>
<td>ml</td>
</tr>
</tbody>
</table>
Now – Onto Soil Grouping

Working smarter not harder means determining how to do fewer total validation executions while making the validation executions you do perform more valuable and defendable.
One Example of Soil Grouping

After you have a COMPLETE list of Soil – put the soils into categories

Figure 1: Soil Grouping Strategy
Example Continued

- **Group 1 Soil:**
  Soil containing proteins, nucleic acids and other components of viral origin. Group 1 soils may also contain cellular debris from viral culture, and components derived from in-process activities (e.g., inorganic salts, formaldehyde).

- **Group 2 Soil:**
  Soil containing proteins, nucleic acids and other components of bacterial origin. Group 2 soils may also contain components derived from in-process activities (e.g., inorganic salts, phenol).

- **Group 3 Soil:**
  Soil containing proteins for use as bacterial growth media or in-process reagents. Group 3 soils contain no components derived from viral or bacterial culture.

- **Group 4 Soil:**
  Soil containing small molecule organic compounds and/or inorganic salts. Group 4 soils are typically reagent solutions, inactivation solutions and buffers.
Now Choose a Worst Case Soil in each group

A worst-case process soil may be selected within each grouping. Worst-case soil selection is based on the difficulty to clean and, where applicable, the risk that any carryover would present to a subsequently manufactured product.
11.2.1 Non-Specific Soils:

The worst-case non-specific soil is selected on the sole criterion of “difficulty to clean”:

11.2.1.1 The soil with the greatest score for “difficulty to clean” is selected for cleaning validation. If two soils have the same score, the soil resulting from the product most frequently manufactured is selected.

Table 1: Risk Scores for Non-specific Soils

<table>
<thead>
<tr>
<th>Parameter</th>
<th>Risk Level = 0</th>
<th>Risk Level = 1</th>
<th>Risk Level = 2</th>
<th>Risk Level = 3</th>
<th>Risk Level = 4</th>
<th>Risk Level = 5</th>
</tr>
</thead>
<tbody>
<tr>
<td>Product Difficulty to Clean</td>
<td>Very easy to clean</td>
<td>Easy to clean</td>
<td>Moderately easy to clean</td>
<td>Moderately hard to clean</td>
<td>Hard to clean</td>
<td>Very hard to clean</td>
</tr>
<tr>
<td>(Subjective)</td>
<td>Free flowing, mobile liquid</td>
<td>Viscous liquid</td>
<td>Viscous liquid</td>
<td>Viscous, sticky liquid or gel</td>
<td>Viscous oily product or denatured protein</td>
<td>Very viscous oily product or highly denatured protein</td>
</tr>
</tbody>
</table>
11.2.2 Specific Soils:

Worst-case specific soil is selected on the combined criteria of “difficulty to clean” of the process soil, and the toxicity and solubility of the selected tracer compound:

11.2.2.1 The strategy for grouping begins by evaluating the composition of each process soil, and identifying potential tracer compounds.

11.2.2.2 After evaluation of the soils and identification of potential tracer compounds, each soil/tracer combination is scored for soil “difficulty to clean”, tracer toxicity, and tracer solubility.

11.2.2.3 The soil/tracer combination with the greatest cumulative score is selected for cleaning validation.

11.2.2.4 If two soils have the same cumulative score, the soil containing the tracer with the higher toxicity is selected.
Table 2: Risk Scores for Specific Soils

<table>
<thead>
<tr>
<th>Parameter</th>
<th>Risk Level = 0</th>
<th>Risk Level = 1</th>
<th>Risk Level = 2</th>
<th>Risk Level = 3</th>
<th>Risk Level = 4</th>
<th>Risk Level = 5</th>
</tr>
</thead>
<tbody>
<tr>
<td>Product Difficulty to Clean</td>
<td>Very easy to clean</td>
<td>Easy to clean</td>
<td>Moderately easy to clean</td>
<td>Moderately hard to clean</td>
<td>Hard to clean</td>
<td>Very hard to clean</td>
</tr>
<tr>
<td>(Subjective)</td>
<td>(Free flowing, mobile liquid)</td>
<td>(Viscous liquid)</td>
<td>(Viscous, sticky liquid)</td>
<td>(Oily product or denatured protein)</td>
<td>(Very oily product or highly denatured protein)</td>
<td></td>
</tr>
<tr>
<td>Tracer Toxicity LD$_{50}$(oral/rat)</td>
<td>2500mg/kg $\leq$ LD$_{50}$</td>
<td>1000mg/kg $\leq$ LD$_{50}$ $&lt; 2500mg/kg$</td>
<td>250mg/kg $\leq$ LD$_{50}$ $&lt; 1000mg/kg$</td>
<td>50mg/kg $\leq$ LD$_{50}$ $&lt; 250mg/kg$</td>
<td>10mg/kg $\leq$ LD$_{50}$ $&lt; 50mg/kg$</td>
<td>LD$_{50}$ $&lt; 10mg/kg$</td>
</tr>
<tr>
<td>Tracer Solubility (g/100mL in water)</td>
<td>Very soluble (100 $\leq$ sol)</td>
<td>Freely Soluble (10 $\leq$ sol $&lt; 100$)</td>
<td>Soluble (1 $\leq$ sol $&lt; 10$)</td>
<td>Slightly Soluble (0.1 $\leq$ sol $&lt; 1$)</td>
<td>Very Slightly Soluble (0.01 $\leq$ sol $&lt; 0.1$)</td>
<td>Practically Insoluble (sol $&lt; 0.01$)</td>
</tr>
</tbody>
</table>
11.3 Cleaning Agents:

Cleaning agents are not part of the manufacturing process and are only added to facilitate soil removal during the cleaning process.

11.3.1 Cleaning agents are evaluated for composition, and potential tracer compounds are identified.

11.3.2 A worst-case tracer is selected on the combined criteria of toxicity and solubility of the selected tracer compound.

11.3.2.1 The soil/tracer combination with the greatest cumulative score is selected for cleaning validation.

11.3.3 Cleaning agent tracer selection may be restricted by limitations of the analytical procedures available. If no suitable analytical procedure is available for the worst-case tracer, then the worst-case tracer for which an acceptable analytical procedure exists will be selected.
Evaluate Cleanability – but how?

1 option - Perform Laboratory Coupon Studies

- If you use an analytical method (i.e., TOC) – it would have to be validated for every soil you are testing.

- And if it is validated for every soil…..how do you apply the soil to the coupon when different soils have varying concentrations of the components (i.e., varying levels of TOC content)

Weight turns out to be a very good option for coupon studies
Evaluate Cleanability – but how?

There are several articles available on different laboratory method study designs that have been used for worst case soil studies. They all try to address the issue of “how do you expose several coupons to exactly the same cleaning conditions at the same time?”

- Recirculatory / flow over bath that holds coupons in place
- Controlled temperature water bath with coupon holder that is attached to a shaker
- Use of a dissolution bath
Gravimetric Last to Leave Determination

Coupon Dissolution
Gravimetric Last to Leave Determination

- Coupon Preparation
- Soil Preparation
- Dissolution
- Evaluation
- Worst Case Soil Determination
Gravimetric Last to Leave Determination

**Coupon Preparation**
- Select MOC that matches your process
- Properly Label Coupons
- Thoroughly Clean Coupons Prior to Analysis
- Drying, Desiccation, and Storage of Coupons
- Tare Weight
Gravimetric Last to Leave Determination

Coupon Selection
- MOC
- In-Process Soils
- Temperatures
- Detergents
- Size/weight
- DOE: Analysis Desired

We Used 316SS #8 2”x2”

<table>
<thead>
<tr>
<th></th>
<th>Teflon</th>
<th>316SS #8</th>
<th>316SS #8</th>
<th>Viton</th>
<th>Noryl</th>
</tr>
</thead>
<tbody>
<tr>
<td>Polypropylene</td>
<td>316SS #7</td>
<td>H2O2 Cured Silicone</td>
<td>Valox</td>
<td>Glass</td>
<td></td>
</tr>
<tr>
<td>Aluminum</td>
<td>316SS #4</td>
<td>Titanium</td>
<td>Pt-Cured Silicone</td>
<td>Buna-N Rubber</td>
<td></td>
</tr>
</tbody>
</table>
Labeling

- Should Identify MOC
- Should Reference Certificate of Manufacturing
- Should have a unique Identifier

We use Marking Methods Inc. Electro-Chemical Marking Equipment
Gravimetric Last to Leave Determination

Cleaning: Clean with a method and detergent compatible with the MOC

Coupon for Current Gravimetric Dissolution Studies are being:
- Sonicated for 20min in 3%CP310
- Rinsed 3X with Milli-Q H2O
- Dried under a Biosafety Hood
- Visually inspected
- Smudges are removed with Alcohol
- Stains are passivated with 5% H2SO4
- Dried at 125C for >20 minutes
- Desiccated for >30 minutes
Gravimetric Last to Leave Determination

- ** Coupon Cleaning  
  - Sonication in 3% CP310  

- ** Rinsed with Milli-Q H20  

- ** Dried under a Biosafety Hood
Gravimetric Last to Leave Determination

- Coupon Cleaning
  - Drying
- Desiccation
- Storage
Gravimetric Last to Leave Determination

Weighing
- Tare Weight of the MOC Coupon is the Most Important Measurement
  - Make sure balance is level
  - Wear gloves or handle coupons with forceps
  - Make sure Coupon is dry and free from debris
  - Accurately record coupon ID
  - Wait for mass measurement to stabilize before recording weight
  - Record out as far as possible
Gravimetric Last to Leave Determination

Soil Preparation

- Accurately measured (Volume, Mass, Dose)
- Reproducible Application
  - Pipette
  - Dry/Desiccate/Store
  - Proper Handling

Document Visual Observations
Gravimetric Last to Leave Determination

- **Dissolution (standardization)**
  - Volume and Concentration of Detergent

- Temperature

- Turbulence

- Time
Gravimetric Last to Leave Determination

- Distek Model 2100C Water Bath Dissolution Unit
  - 200RPM
  - 500ml Detergent
  - 50°C
  - Time (+~30sec)
Gravimetric Last to Leave Determination

Distek 2100C Controls

- Hydraulic Lift (Time)
- Set Temperatures
- Set RPMs
- Set Paddle Height
Gravimetric Last to Leave Determination

- Evaluation of Data
- Dry Soil Weight Pre-Cleaning
- Dry Soil Weight Post Cleaning
- Percent Soil Loss
Second:

Know and Understand the Equipment you Must Clean
Surrogate Soil

Consider using a surrogate soil for your cleaning validations!
Product Contact Equipment

vs

Equipment that is Contacted by Product
Biologically Active Material – BAM – live or inactivated vaccines (intermediates, bulk and final product) that have a potential to engender an immune response.

**BAM Contact Surfaces** –

**Condition 1:** Equipment surfaces that are in direct contact with biologically active material (BAM) during manufacturing activities. During the production and cleaning processes, these surfaces are soiled with BAM, product components (e.g., buffers, media, etc.) and cleaning agents. The cleaning must be validated to show removal of soils.

**Condition 2:** Equipment surfaces that are not contacted by BAM by design but may contact BAM in practice (due to splashing or spillage) will be individually assessed to determine the risk involved should the surfaces not be sufficiently cleaned and/or sterilized before contact is made.
Non-BAM Contact Surfaces –

Condition 1: Equipment surfaces used for preparing and/or transferring reagents, medias, buffers, components (e.g., stoppers) etc. that are incorporated into the product. During the production and cleaning processes, these surfaces are soiled with product components and/or cleaning agents but are not soiled with BAM. The cleaning must be validated to show removal of these soils.

Condition 2: Equipment surfaces that are not contacted by reagents, medias, buffers, components by design but may contact reagents, medias, buffers, components in practice (due to splashing or spillage) will be individually assessed to determine the risk involved should the surfaces not be sufficiently cleaned and/or sterilized before contact is made.
Ensure your Trace Matrix contains all equipment

You must be able to ASSURE the auditors that you have captured ALL product contact equipment in your cleaning validation
Third:

Know and Understand the Cleaning Processes you must Validate
Understand the Manufacturing Process

Processing Stage 1 - those phases of a manufacturing process that precede the purification steps. Stage 1 process equipment includes the preparation of raw materials, the fermentation or culture process and primary filtration steps.

Processing Stage 2 - those phases of a manufacturing process involving product purification and conjugation up to but not including final formulation

Processing Stage 3 - those phases of a manufacturing process involving final formulation and filling
Understand the Equipment Flow
Know the Cleaning Systems Utilized

**Clean-in-Place (CIP):** The process of cleaning a piece of equipment, piping, or device without taking it out of line; cleaning performed without the need to relocate the equipment being cleaned (may be manual or automated.)

**Clean-Out-of-Place (COP):** The process of cleaning a piece of equipment, piping, or device after it has been taken out of line (may be manual or automated.)

**Cabinet Washer** – alternately described as a glass or bottle washer - is a compartment type pressure washer with doors that is designed to utilize racks, spray nozzles, cleaning solutions and air to clean and dry the interior and exterior of items placed within its chamber.

**Manual Cleaning** - Manual cleaning is performed by trained personnel who follow detailed procedures pertaining to preparation and use of cleaning solutions, disassembling, scrubbing using non-abrasive and non-shedding brushes or pads, soaking, rinsing and drying.
Sonicators
Sonicators
Parts Washer
Parts Washer
Parts Washer
Vial Washer
Vial Washer
Clean In Place Skid (CIP)
CIP Flow Example

- CIP Storage
- CIP Target
- Heat Exchanger
- Balance tank
- CIP Centrifugal Supply pump
- Pressure indicator
- Temperature transmitter
- Flow Control Valve
- Temperature indicator
- CIP Rinse Sample Point
- Rinse Conductivity Sensor (select transfer lines)
- CIP Rinse Conductivity Sensor
- CIP Liquid Ring Return Pump
- Flow Switch
- Rinse Conductivity Sensor

Connections:
- To drain
- Recirculation or Balance Tank Fill
- WFI Final Rinse once thru
- Temperature switch
- Flow switch
- Clean Compressed Air
Manual Clean
Manual Clean
Manual Clean
Fourth:

Know and Understand the Acceptable Philosophy at your Site
Your Quality Counterpart
Your Quality Counterpart
Method Approach - Direct – cleaning validation execution is performed on subject equipment and residues.

Method Approach – Matrix - cleaning validation execution is performed on equipment and/or residue that is grouped or bracketed to minimize the amount of runs while still providing evidence that all specified equipment cleaned will be in a validated state. If the matrix method is utilized the grouping strategy and the rationale for the grouping must be justified in the project plan and/or the protocol.

**Parent** - when using the matrix approach the item chosen to represent the group or bracket is termed the parent (typically the “worst case” or most challenging item).

**Child/children** – when using the matrix approach the item(s) identified as belonging to a group.
Confirmation – when applying the matrix approach, the act of executing one (1) successful cleaning validation run on equipment categorized as a child. When applying confirmation runs to the family approach, the cleaning process is considered validated after the parent cleaning validations are executed. Rationale must be included in the project plan and/or the protocol justifying which children will require a confirmation run and which will not require a confirmation run.

Verification – A one-time testing of equipment for cleaning efficacy. Verifications may be performed during Demonstration, Definition and/or Consistency runs, or after process changes/deviations where the validated cleaning no longer applies (eg, violation of dirty hold time) prior to performing cleaning validation.

Validation - Validation can be described as establishing documented evidence that provides a high degree of assurance that a specific process/equipment will consistently produce a product meeting its pre-determined specifications and quality attributes.
Acceptance Criteria

Required Testing

Definition of Failure

Degradation Studies

Analytical Methods and Recoveries

Filing Strategy

Company 5 year and 10 year plan for the site

Routine Monitoring

Revalidation
Degradation Studies

Building 37 Experience – Full Scale

Cleaning Validation Program Prerequisites

Full Scale Cleaning Development Studies in B59

Risk Assessment for equipment and Soils

Detergent and Cleaning equipment Experience

Worst Case Soil Studies

Cleaning Monitoring Program

Lab Scale Studies
Now – Onto Equipment Grouping

Working smarter not harder means determining how to do fewer total validation executions while making the validation executions you do perform more valuable and defendable.
One Example of how to Group Equipment

12.1.2 Equipment may be grouped if it meets the following criteria:

- The grouped equipment is of the same type;
- In all cases, equipment cannot be grouped unless the equipment items are cleaned using the same cleaning procedure and see the same residues.

12.1.3 Two equipment items are considered "similar" if they are constructed with the same material and functionality, but have different physical attributes (i.e., working volume, geometry).

Example: Two stainless steel tanks of 100L and 500L, fitted with mechanical stirrers would be considered similar and grouped into one family.

12.1.4 Two equipment items are considered "identical" if they meet the following qualifications:

12.1.4.1 they are constructed of the same material
12.1.4.2 they are constructed using the same design drawing
12.1.4.3 they have the same physical attributes (i.e., working volume, geometry, surface finish, top ports)
12.1.4.4 they encounter the same residues
12.1.4.5 the Installation and Operational Qualification Reports verify that the vessels meet the same specifications

Example: Two 500L stainless steel tanks used to transport flu vaccine, made from drawing DWG001, with a surface finish of 25 RA, with 2 – 2" ports and 1 – 6" port, without stirrers would be considered identical and grouped within one family as a "type".
12.2 Scoring Procedure:

12.2.1 The strategy for grouping begins by identifying and scoring each critical element or item on the equipment (e.g., ports, dip tubes, etc). When all critical elements/items have been identified and scored, a global risk score can be calculated for that equipment.

12.2.2 The risk scoring is performed in three steps with input from the operators:

- Classification of the equipment by material and attributes;
- Identification of the equipment attributes and design features;
- Assignment of individual risk scores and calculation of the Global Risk Score.

12.2.3 The Global Risk Score is calculated as follows:

\[ GRS = (Ic_1 \times Ne) + (Ic_2 \times Ne) + \ldots + (Ic_n \times Ne) \]

Where:
- \( GRS = \) Global Risk Score;
- \( Ic_1 \) \ldots \( Ic_n = \) Risk score for each critical element or item;
- \( Ne = \) Number of each critical element present on the equipment.
One Example of how to Group Equipment

Table 3: Risk Scores for Equipment Attributes

<table>
<thead>
<tr>
<th>Risk or Criticality of the Attribute</th>
<th>Risk Score</th>
</tr>
</thead>
<tbody>
<tr>
<td>Very low risk or non-critical</td>
<td>1</td>
</tr>
<tr>
<td>Low risk or low impact</td>
<td>2</td>
</tr>
<tr>
<td>Moderate risk or moderate impact</td>
<td>3</td>
</tr>
<tr>
<td>High risk or major impact</td>
<td>4</td>
</tr>
</tbody>
</table>
One Example of how to Group Equipment

Table 4: Risk Attributes for Tank Accessories

<table>
<thead>
<tr>
<th>Design Feature</th>
<th>Risk Score</th>
</tr>
</thead>
<tbody>
<tr>
<td>Removable Elbow</td>
<td>1</td>
</tr>
<tr>
<td>Removable Dip Tube</td>
<td>2</td>
</tr>
<tr>
<td>Port</td>
<td>2</td>
</tr>
<tr>
<td>Baffles</td>
<td>2</td>
</tr>
<tr>
<td>Penetration (probe, rupture disk, vent filter, etc)</td>
<td>2</td>
</tr>
<tr>
<td>Vortex Breaker</td>
<td>3</td>
</tr>
<tr>
<td>Fixed Elbow</td>
<td>3</td>
</tr>
<tr>
<td>Spray Ball Shaft</td>
<td>3</td>
</tr>
<tr>
<td>Fixed Dip Tube</td>
<td>3</td>
</tr>
<tr>
<td>Mixer Impeller</td>
<td>4</td>
</tr>
<tr>
<td>Impeller Shaft Bushing</td>
<td>4</td>
</tr>
<tr>
<td>Septum</td>
<td>4</td>
</tr>
</tbody>
</table>
One Example of how to Group Equipment

Table 5: Risk Attributes for Small Equipment Parts

<table>
<thead>
<tr>
<th>Design Feature</th>
<th>Risk Score</th>
</tr>
</thead>
<tbody>
<tr>
<td>Flask or Bottle</td>
<td>1</td>
</tr>
<tr>
<td>Filling Pump Body</td>
<td>2</td>
</tr>
<tr>
<td>Filter Housing</td>
<td>2</td>
</tr>
<tr>
<td>Filling Pump Inlet/Outlet</td>
<td>3</td>
</tr>
<tr>
<td>Filling Pump Piston</td>
<td>4</td>
</tr>
<tr>
<td>Filling Needle Assembly</td>
<td>4</td>
</tr>
</tbody>
</table>
Consider Using Surrogate Small Parts

- Identify worst case / grouped parts list and then purchase a complete validation loads worth of the parts that will belong just to the cleaning validation group – the cost is fairly easy to justify (dirty hold time and clean hold time requirements during validation can shut down production for weeks during validation execution)
Finally:

Execute!