Microbial Control During Drug Substance Manufacturing

Juhong Liu, Ph.D.
Principal Consultant
GlobalSubmissions Consulting, LLC

PDA Aseptic Processing of Biopharmaceuticals Conference
Seoul, 2024

Disclaimer

The views and opinions expressed in the following slides are purely my personal opinion.

They should not be attributed to positions from any Health Authorities or pharmaceutical companies.

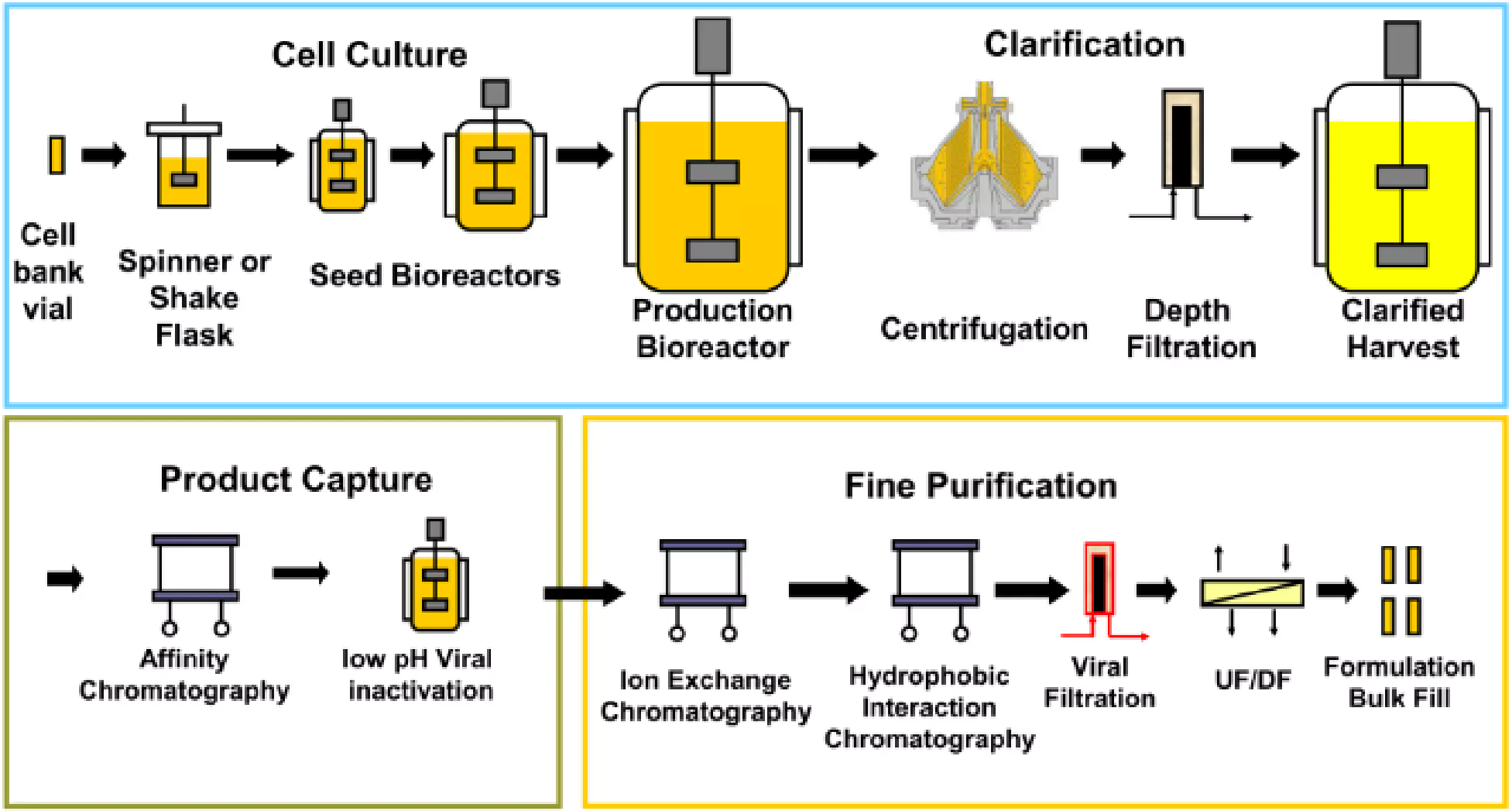
DS Production – A Bioburden Controlled Process

Biologics DS Processes: environments accommodate microbe growth

- Cell culture / fermentation media
- Cell culture /fermentation time
- Protein rich intermediates
- Buffers
- Chromatography resins
- Open operations

A Bioburden Controlled Process

Typical Mab DS Production



Adopted from: Kevin Lauziere

Risk Assessment

- Microbial growth during the process is inevitable
- Risk evaluation:
  - Unit operations susceptible to microbial contamination
  - Distance to final bulk DS
  - Unit operation Clearance capability
  - Placement of bioburden reduction filters
  - Impact on product quality and safety

Microbial Control – Process Specific Risks

Bacterial/yeast expression (Insulin)

- Fermentation – contaminations are difficult to identify or quantify
- Resins are pH and salt resistant
- Many unit operations performed at low (3.0) or high (10.0) pH
- Intermediates stored at high/low pH
- Final steps uses organic solvents
- Bulk DS storage form and temperature

CHO expression (MAbs)

- Cell Culture – contaminations are easy to identify
- Pro-A resin is susceptible for contamination
- Unit operations performed at mild pH
- Intermediates stored at mild pH
- Bulk DS in liquid form, some stored at 4°C

Mitigating Microbial Contamination

Maintain an appropriate clean environment:

- Environmental monitoring
- Class A, C, D rooms
- EM frequency
- Static vs. production EM

Case Study 1: Inadequate Routine EM + Insufficient Monitoring During Production

- Routine EM performed every two months
- Due to COVID, no production for some time
- Insufficient microbial and particle testing during production
- Bulk DS fill – Class A:
  - All routine EM were performed without production
  - No particle or viable data at the time of fill

FDA 483:

- EM program is inadequate
- No batch-related dynamic EM is performed during production

Mitigating Microbial Contamination

Cleaning

Facility:

- Cleaning agent validation
- Cleaning frequency
- Sporicidal agent use
- Cleaning when yeast or mold were identified

Purification column/resins:

- Cleaning effectiveness – blank runs
- Post-cleaning storage time
- Cleaning procedures after a contamination event

Case Study 2: Cleaning after Mold Contamination

- Mold was identified during a routine EM
- Facility was cleaned using routine cleaning agent
- No sufficient batch-related EM to rule out mold contamination
- Impact by mold on product is uncertain

FDA 483

Mitigating Microbial Contamination - Process

1. Column and Filter cleaning
2. Bioburden reduction filtration
3. Storage of intermediates
4. Limit of intermediate hold time

Mitigating Microbial Contamination

Bioburden Reduction Filtration and Intermediates Storage

- Purification columns are susceptible to microbial growth
- Intermediates are stored as liquid form at room temperature
- Most intermediates are in buffers that promote microbe growth
- Including a bioburden reduction filtration step is critical to mitigate carryover of contaminant to the next step
- Filtered intermediates should be stored in clean tanks or pre-sterilized bioprocessing bags

Bioburden Sampling and Filter Placement

- Bioburden and endotoxin of intermediates should be tested to demonstrate microbial contamination is under control
- Intermediates should be sampled prior to filtration
- Bioburden reduction filtrate should preferably be transferred to storage containers through aseptic connections

Case Study 3: Sampling after Filtration

- PPQ campaign – Planned for 3 production runs
- Runs 1 & 2: bioburden and endotoxin sampled post-filtration, no excursion found
- Sampling moved to pre-filtration per industry standard for the third run – TNTC for Pro-A column, excursions for others
- 4 months to identify problems
- Re-initiate 3 PPQ production run
- 3 batches wasted, 7 months delay

Intermediate Hold Time Validation

Microbial testing must sample from intermediate stored in commercial production containers under commercial production conditions

Case Study 4: Validation of hold time study during PPQ

- Chemical stability and microbial samples stored in small storage bags with the same construction
- The intermediates are chemically stable within the proposed hold time at the proposed temperature
- Microbial samples are not representative of the at-scale production condition, results can not be used to demonstrate control of microbial growth
- Microbial tests repeated in the following 3 commercial production runs
- The three PPQ batches were not released for commercial distribution

Summary

- Biologics DS manufacturing process is a bioburden-controlled process
- Demonstration of microbial control is critical through the entire process
- Can be achieved by:
  - Facility EM: routine and batch-related monitoring
  - Cleaning: appropriate cleaning agents, frequency, and for-cause cleaning
  - Filtration: at appropriate points
  - Adequate sampling
  - Validation of intermediate hold time