# Identification and Risk Mitigation of a Critical Process Parameter during Antibody Process Scale-up

Christa Short, Midge Bonzon, Jonathan Mitschelen, Upstream Process Development, R&D Services (Reference)

Cytovance Biologics

## Background

**Start-up biotechnology companies are founded on great product concepts from partners or universities**

- Early partners and investors want to see proof of concept
- Funding is often in tranches tied to development milestones

**Entrepreneurial Business Strategies**

- Rapid realization of value
- Limited financing
- Limited duration commitment-early exit preferred
- Minimal business organization or contract experience
- High risk tolerance – "Plan for Success"
- Plans change, assumptions do not

## Impact on Manufacturing

**Timeline**

- **Development of manufacturing process is rate-limiting activity for progression to clinical studies**
- Virtual or small company needs 18 – 24 months to develop process
- Investor timelines often require clinical trial initiation in 12-14 months

**Process Development Strategies**

- Process and Process Knowledge "Adequate" for Business Plan
- Graduated cGMP development and compliance
- Delay major development expenses - "Fix before Phase III"
- Rely on minimally characterized standard processes
- At risk scale up – Straight to cGMP Production for Phase II

## Company Strategies

**To meet timelines and reduce costs prior to proof of concept, companies often cut corners**

- Processes deliver safe, effective products at small scale
- Process parameters and process design space are unmapped
- Understanding likely cell culture behavior during scale-up based on scale-down models is not a priority

**Material is available for early clinical trials, but….**

- Patient safety is never at risk, nor is mode of action
- Ability to meet ongoing clinical demand IS at risk due to limited process knowledge
- Rapid requirement to scale-up to meet clinical supply can lead to unexpected surprises…even with antibodies

## Regulatory Consideration

**FDA Phase 1 Guidance**

- Phase appropriate GMPs
- Emphasis on patient safety but limited expectation for process consistency early in development
- Continuous improvement in process control throughout development
- Operation within historical ranges not considered a process change

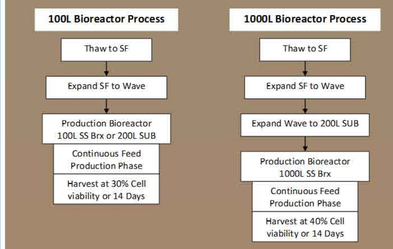
**GMPs for the 21st Century**

- Continuous process improvement
- Knowledge driving process design
- Risk-based decision making driven by data and scientific rationale

## Case Study

- Production of Phase 1 clinical trial material at 100 L scale
  - Limited process development prior to cGMP production
  - Productivity of < 1 g/L, below industry standard
- Scale-up to 1,000 L planned for Phase 2 clinical supply
  - Initial 1,000 L batch successful with similar yield to 100 L batches
  - Second 1,000 L batch failed to reach normal cell density and yield was much lower than normal
- Risk mitigation for third batch included rigorous data review and implementation of a process control strategy

## Mab Process Flowchart



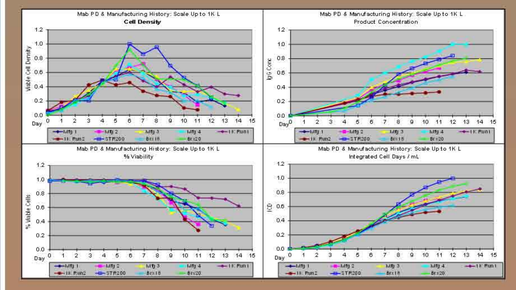




**Figure 1. PD & MFG Growth & Production.**

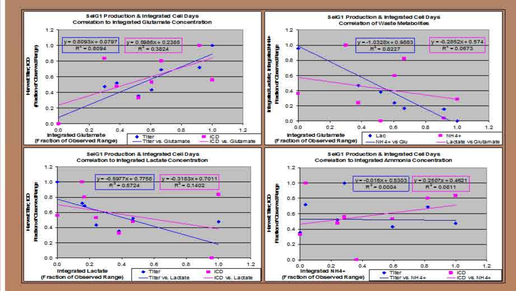



**Figure 3. Process Parameter Correlations.**

## Mab Production Process

| | |
|---|---|
| Product: | Fully human IgG1 |
| Cell Line: | CHO System |
| Growth Medium: | Gibco OptiCHO™ |
| Production Medium: | Gibco OptiCHO™<br>10% Gibco EfficientFeed™ A<br>10% Gibco EfficientFeed™ B |
| Feed Medium: | 50% EfficientFeed™ A / 50% EfficientFeed™ B |
| Feed Schedule: | 5% v/v of Initial Culture Vol/day Day 2 to Harvest<br>Glucose as required |
| Temperature: | 37°C |
| pH: | 7.05 ±0.15 |
| Harvest: | ≤ 40% Cell Viability or Day 14 |

## Process Performance History

**Process Development**

- Problematic
- Variable peak VCD
- Variable run duration
- Modest productivity
- High oxygen consumption

**Manufacturing**

- All lots met product specifications
- 100 L & 200 L Scale
- 5 runs
- Yield ≥ PD runs
- Variable duration, VCD, and yield
- 1000 L Scale Run 1
- Low vcd
- Good run duration
- Low but adequate yield
- 1000 L Scale Run 2
- Lowest vcd
- Shortest run duration
- Lowest, inadequate yield

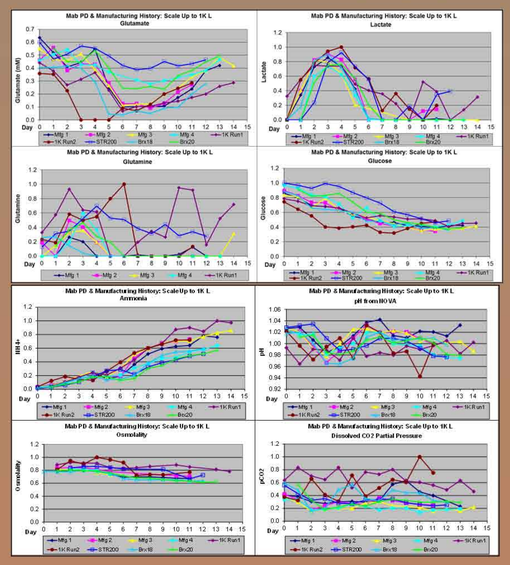



**Figure 2. PD & MFG Metabolite Profiles.**

## Glutamate Issues & Process Range

**Justification of Glutamate as a Process Issue**

- Glu is a component of both base medium and feed medium
- Glu vs Titer strongest of correlations
- Glu has roles in both protein synthesis and energy production
- Literature - Metabolomics report Asn, Asp, Glu, Pyr depletion at start of decline phase associated with interruption of TCA cycle
- Schedule did not allow time for AA depletion studies
- Schedule did not allow time to establish Asn, Asp, Pyr assays

**Process Range Considerations**

- Consistent with regulatory considerations
- Lower action level reliably distinguishable from zero – 1.5 mM
- Range wider than maximum daily consumption – 2 mM / day
- Upper level: 1.5 mM + 2 mM ≈ 3.5 mM vs historical max = 3 mM

## Process Change Options

**No Change**

- Justify as maintenance of lot to lot process consistency
- Inadequate data to support process change impact

**Supplement with Glutamate**

- Justify as continuous process improvement
- As required to maintain a standard control window
- As required to maintain to range in successful runs
- Adjust to standard concentration once at bioreactor inoculation
- Adjust to standard concentration once at N-1 bioreactor inoculation

## Final Change and Justification

**Convert Process Options to Approved Manufacturing Change**

- Agreement among Client, Regulatory & Process Consultants & CMO
- Reasonable manufacturing procedure
- Minimize potential for impact on product quality
- Minimal change to improve process control
  - Correct process deficiency: glutamate exhaustion
  - Target average Glu to historical high concentration: ~3 mM Glu

**Agreed Process Changes**

- Supplement N-1 Seed Train at inoculation only: 2.5 mM Glu
- Maintain Glu in production bioreactor: 2.5 – 4.5 mM Glu

## Results

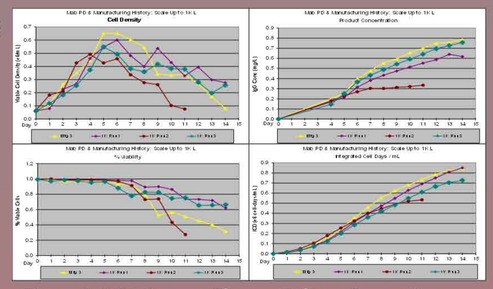



**Figure 4. 1K L Process Change Culture Properties.**

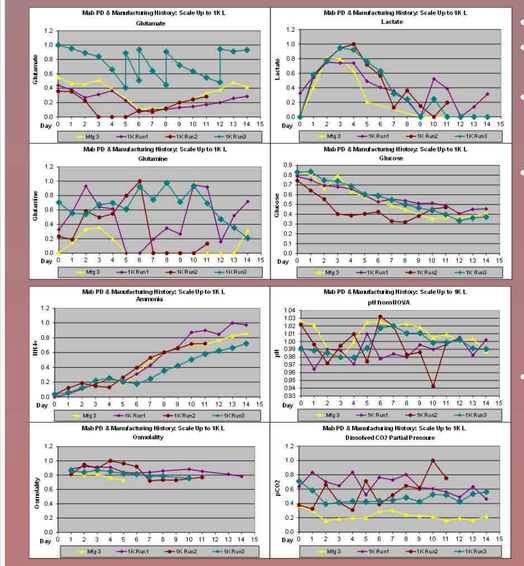



**Figure 5. 1KL Process Change Metabolites.**

- Lot met release specifications
- Yield satisfied critical clinical inventory requirements
- No process parameters significantly outside process history
- Most process parameters more favorable than historical range
  - Yield better than first 1K bioreactor run
  - Peak VCD in process history range
  - Duration to 14 days with viability >60%
- Changes implemented without introducing dramatic changes

## Conclusions

- Improved Glutamate control successfully addressed scale up issue
- Intensive, collaborative process data 28.9
- review identified process change options that satisfied client business constraints, regulatory obligations, and manufacturing facility design

## Reference

This poster is a version of slides presented at PEGS Summit in Boston, MA (May 5-9, 2014) in the session of "Scaling Up and Down Strategies for Protein Production". The presentation was a joint effort with Susan Dana Jones, Ph.D., Vice President and Senior Consultant, BioProcess Technology Consultants.

## Acknowledgements

Manufacturing, QA, QC, Project Management, BD & Client