The Upstream/ Downstream Process Balancing Act

Early collaboration and open communication between upstream and downstream are crucial to ensure a consistent end-to-end bioprocess.

By Serena Fries Smith and Hunter Malanson

Traditionally, bioprocess development is split into "upstream" and "downstream" functional groups. There are individual challenges associated with the development of both pieces of the process, with upstream focused on producing quality product, and downstream on purifying it. While these two functions rely on each other to be fully successful, they frequently work in parallel – yet separately – to meet the aggressive timelines of the program. This approach may allow for some efficiencies during development, but if the two teams are not working together, it can also create problems.

Developing a robust process

During downstream development, scientists are focused on a number of unit operations. For a therapeutic protein, these processes would start with a primary recovery step – where the cells and debris are removed. The clarified material would then move through a variety of subsequent processing steps including buffer exchange, material hold, viral inactivation/filtration, and chromatography – which are required to remove process impurities and isolate the protein of interest. Each individual step has critical process parameters (CPPs) that need to be monitored and controlled to ensure the critical quality attributes (CQAs) of the intermediates and purified protein will be achieved at the end. But the biggest factor to ensure a consistent and successful downstream process is not even one that the downstream team can control: it is the consistency of the upstream harvest material.

"If you think of a manufacturing process as a chain of inter-connected blocks, with each block representing a specific unit operation, changes to any block or series of blocks can have lasting and unpredictable consequences to blocks further down the chain. In that sense, upstream processes have a profound impact on the reproducibility and performance of downstream processes." – Pratik Jaluria, Executive Director of Process Development and Manufacturing at Adverum Biotechnologies.

Development of a robust and consistent downstream process must include the ability to understand and balance the output from the interconnected upstream process. During upstream production, we tend to overly focus on product titers, but other factors such as cell concentration, cell viability, and various product quality characteristics may be impacted to achieve those high titers. And these upstream factors will most likely impact the subsequent recovery and purification process steps.

As an example, one NS0 process developed was initially harvested at a viability of 30 percent to maximize the antibody titer. Unfortunately, this low harvest viability resulted in significant problems downstream and caused very low cumulative process yields. Through discussions with the purification team, it was decided that a new harvest viability specification of greater than 50 percent would be used for this process. Upstream, there was a 20 percent loss in productivity, but the downstream process yields were much higher than before and the overall amount of purified protein increased. This example highlights the importance of cross-functional collaboration to ensure the entire process, and not just one discrete area, is successful.

"During the process development phase, it's important that the upstream process delivers "representative" material that has a varied level of process impurities to ensure the downstream process will consistently remove these to acceptable levels. For example, a lower cell viability at harvest typically generates a greater release of host cell protein and DNA impurities. The higher impurity load can lead to diminished product recovery, or overwhelmed chromatography processes leading to a failed batch." – Ben Hughes, Director of Global Tech Transfer Biologics at Patheon.

Evaluating process variations

Collaborating to establish upstream harvest parameters is crucial to the overall downstream success, but there is also the added challenge of accommodating unexpected and unknown variations in the upstream process. Some variations can be measured, while with others the true impact may not be known until a problem emerges. When there are process challenges during purification, reviewing the following with the upstream team can help to identify the root cause of the problem:

- Has the harvest viability or titer changed?
  A change in harvest viability or titer, can disrupt the approved downstream process by fouling filters or falling outside qualified column loading ranges. Working with the upstream team to understand the expected variation and define acceptable limits will increase success. Additionally, the upstream team should immediately communicate when there are deviations in expected growth and production profiles, so that the downstream team can assess the deviation and plan accordingly.
- Were new raw materials used?
  Raw materials used in the upstream process have the potential to impact a variety of elements, including cell

growth, protein production, product quality and process impurities. As an example during the manufacturing of a recombinant protein used an animal-derived component upstream. Due to increasing regulatory requirements on animal-derived material, the team was forced to identify and qualify a new source from a different country. While in theory this was a "like for like" material change the newly sourced material resulted in an unexpected 50 percent increase in titer. Unfortunately, there was not enough capacity in the downstream process at the existing facility to handle the unforeseen increase and some material needed to be discarded. This example demonstrates that any upstream variability, even increases in titer, can be a problem when the downstream process isn't designed for it. It also highlights the importance of identifying critical raw materials, and closely monitoring any changes in lots or suppliers that could result in upstream variability.

- Is the quality profile different?
Some variability can be identified through rigorous measurements and tracking of the upstream process (e.g., titer, viability, cell growth) and some can be identified due to supply chain changes of critical raw materials. But there are other changes, that are completely unexpected, and do not become readily visible until something goes wrong downstream. Reviewing and understanding the characteristics of the product quality profiles (such as the glycosylation profile and charge distribution for antibodies) in the production bioreactor, and leveraging qualified small-scale models to troubleshoot variability during manufacturing can provide key insights when the process isn't performing as expected.

Collaborating for success

The process development teams need to openly interact with one another from the very beginning, verifying that changes made to improve or further control the upstream process will not have a negative effect downstream. The open communication and cross-pollination of ideas will also improve the coordination of project timelines and minimize material waste. Additionally, linking upstream and downstream experimental studies could provide benefits to the analytical, product characterization and formulation teams by providing them with material for their studies earlier.

Conclusion

It's important to frequently communicate and collaborate. Upstream constantly needs to be thinking about what materials they're using in their processes and what this means for downstream. Can they clear it? Will it cause interference? Variability should be minimized – a robust and consistent upstream process is key to a robust and consistent downstream process.

In short, by working together, we won't just have a successful upstream process or a successful downstream process. We can ensure that we have a robust end-to-end manufacturing process.

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