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Custom Development & Manufacturing Organization (CDMO)

Analytical and microbiological methods in industrial biotechnology: Indispensable attribute of bioprocess development and commercial biomanufacturing

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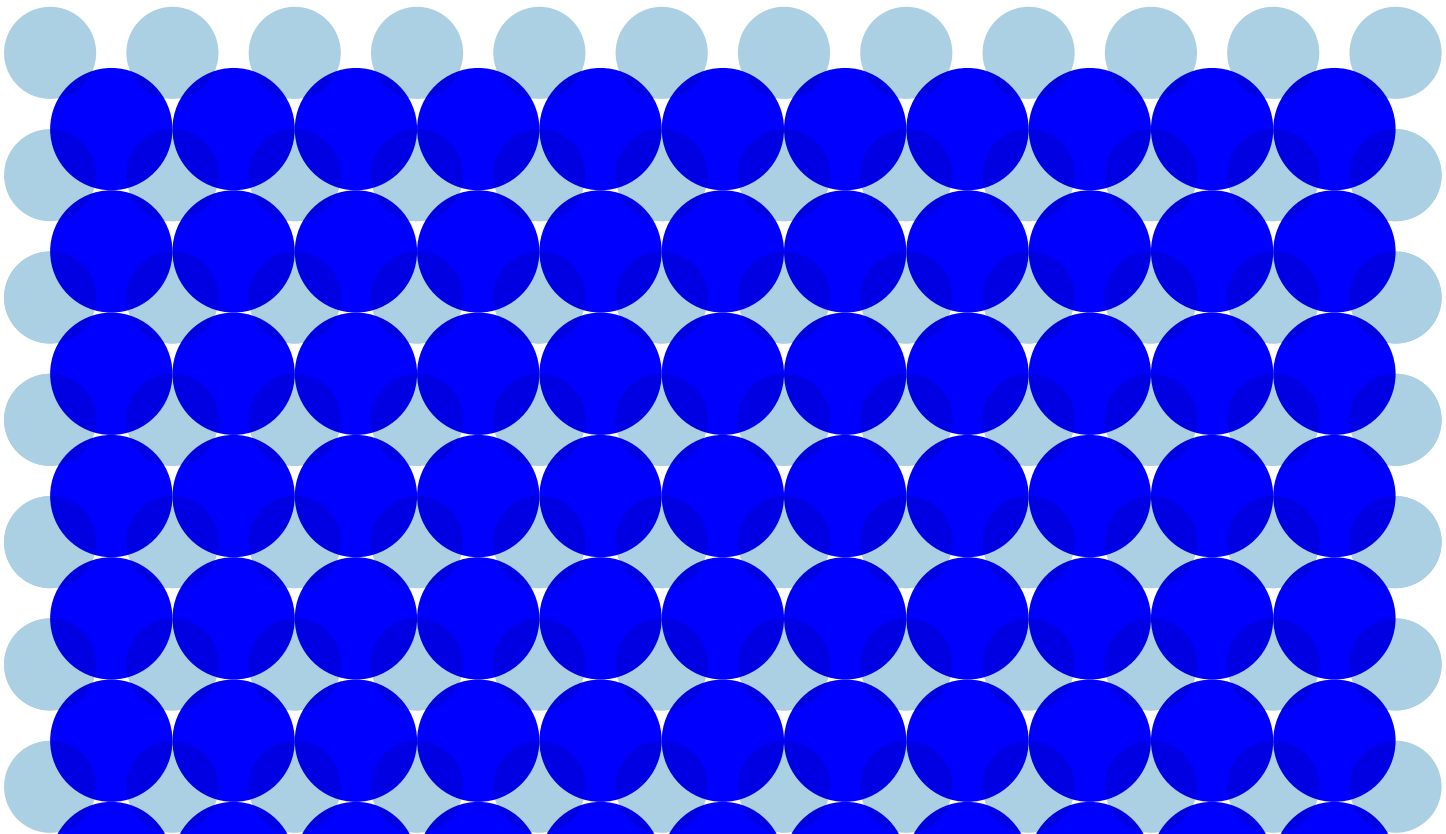
The development and manufacturing of biotechnological products involve multiple integrated technologies and distinct stages such as microbial strain development, fermentation, and downstream processing. Throughout each of these stages, robust analytical methods are required to assess and ensure the bioprocess's efficiency, yield, and final product quality. For contract development and manufacturing organizations (CDMOs) such as Arxada, state-of-the-art analytical capabilities and certified quality control laboratories are essential assets. These facilities allow CDMOs to maintain product integrity, uphold regulatory standards, and deliver high-quality solutions for customers.



Analytical and microbiological methods in industrial biotechnology: Indispensable attribute of bioprocess development and commercial biomanufacturing

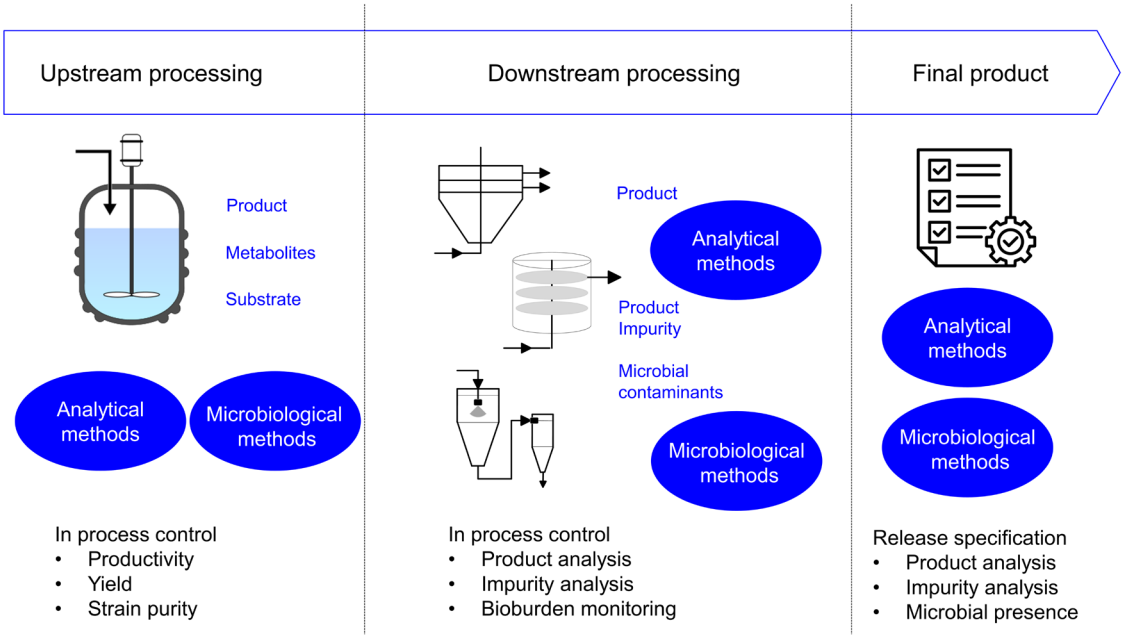
In the complex landscape of biotechnological product development and manufacturing, the integration of analytics plays a pivotal role across the product development flow. Analytical methods are not only important for confirming product quantity, quality, and regulatory compliance but also crucial for monitoring and optimizing bioprocess parameters. Thus, the transfer of analytical methods is essential part of the entire technology transfer. Such methodologies are usually transferred and implemented first, even for not fully mature industrial processes. This is a prerequisite for setting up first laboratory experiments following successful technology transfer. Let us dive deeper into the challenges and best practices associated with analytical and microbiological method transfer and their implementation in the commercial manufacturing environment.

A wide variety of analytical technologies from classical to instrumental have been used to assess product's physicochemical and functional characteristics in biomanufacturing. Analytical methods based on various physical, biochemical or biological underlying principles and advanced instrumentation are pivotal not only for the product itself, but also for quantification of other parameters directly or indirectly linked to the product or process attributes.

Some of the common techniques used in biomanufacturing nowadays are:

- Spectroscopy methods employed for e.g. measurements of metabolites, optical density, enzyme activity assays or assessment of raw materials,
- Analytical chemistry techniques such as high-performance liquid chromatography (HPLC) and gas chromatography (GC) are standard methods for accurately quantifying products, metabolites or substrates in fermentation samples,
- Molecular biology methods such PCR or qPCR may be used to determine genetic stability of the production strain, presence of the residual DNA in the sample if required in the product specification or presumable pathogen identification. Other techniques such as SDS-PAGE and Western blotting may be used to identify protein products and assess their quality,
- Other modern methods such as flow cytometry might be helpful to monitor cell physiology (viability and vitality) throughout the course of fermentation,
- Various microbiological methods are used to detect, identify, and quantify microorganisms in biotechnological products, for monitoring of bioburden during the product processing and for validation of cleaning protocols. For the latter other methods such as total carbon content analysis or conductivity measurements are used as well (Figure 1).

Figure 1: Schematic illustration of analytical and microbiological method application throughout the product manufacturing flow.



No matter which methods are used for the product development and manufacturing, the key objectives of analytics throughout the product development are always followed:

- **Continuous process monitoring and optimization**: To ensure stability, efficiency, and yield at each stage of the manufacturing process. Analytical methods used for in-process testing include both online (e.g. pH, oxygen, temperature) and offline measurements (e.g. product quality and quantity, by-product, substrate and biomass quantity, or cell physiology), providing actionable insights for bioengineers to monitor the manufacturing process. As some methods such as biomass quantity monitoring (optical density, wet cell weight, dry cell weight, packed cell volume, pH) are highly standardized, some other need adjustments or new method development to adapt to increasing demand for quantification of novel products of the expanding biotechnology market.
- **Quality control and compliance**: To meet stringent and often different regulatory requirements for biotechnological products, given by the product intended application in various regulated market segments such as pharma, medical device, food, or cosmetics. Product specification defines analytical methodologies and their associated limits, as these ensure the product quality attributes are met.

Analytical methods frame a biotechnology process from early stages of development to a routine commercial production. To maintain analytical integrity and process scalability across different manufacturing environments, efficient and flawless analytical and microbiological method transfer needs to be executed. Such transfer ensures that analytics (and so the process and final product) remain consistent, reliable, and compliant. There are several stages of typical method transfer:

- **Preparation stage** defines which methods will be part of the formal transfer and their validation status, the material the methods will be assessed against and assessment of differences (personnel, instrumentation) between transferring and receiving laboratories. Comprehensive documentation that includes detailed protocols, standard operating procedures (SOPs), and validation reports needs to be shared. Proper knowledge transfer and subsequent personnel training ensures that the receiving laboratory can replicate the method accurately.
- **Execution stage** may include
  - round robin testing, i.e. running the same samples in both the transferring and receiving laboratories and comparing the results to ensure they are within acceptable limits,
  - partial or full method validation (demonstration the method serves its intended purpose) is conducted e.g. in case the method is used in the receiving laboratory for the first time, before the use of the method in routine testing or if the method is part of a novel product application dossier. In case of transfer of a compendial method, its verification (simplified single-laboratory validation) in the receiving laboratory might be sufficient. Guidelines for method validation, verification and the assessed method performance parameters are well described in international pharmacopoeias,
  - transfer waiver, i.e. omitting of a method from the formal transfer if the method is the same in the transferring and receiving laboratory. This typically involves routinely used methods such as optical density measurement, dry cell weight, turbidity, density of liquids or product appearance,
- **Reporting stage** compiles a comprehensive report detailing the transfer process, describes the results obtained in relation to the acceptance criteria, and any deviations or issues encountered. The report documents that the receiving laboratory is qualified to run the transferred analytical procedures.

Transfer of analytical methods is a complex process and comes with multiple challenges (Table 1). By carefully planning and executing the stages outlined above, and addressing the key challenges, successful and efficient method transfer can be achieved, ensuring reliable and compliant analytical and microbiological testing in the biomanufacturing environment.

Table 1: The key challenges and best practice in analytical and microbiological method transfer in biotechnology.

Challenge	Description	Best Practice
Equipment, instrumentation and material differences	Variations in equipment and materials between laboratories can affect method reproducibility and performance	Ensure equipment standardization, regular calibration, detection system compatibility, use of equivalent materials and standards
Variability in personnel expertise	Differences in skill levels and experience can impact method consistency and accuracy	Provide thorough and regular trainings and standard operating procedures (SOPs)
Environmental conditions	Environmental factors such as temperature, humidity, and cleanliness may influence outcomes of particularly microbiological or enzymatic assays	Ensure appropriate environmental controls are in place
Documentation and communication	Poor documentation, communication and coordination effort can lead to unnecessary errors	Maintain detailed documentation and clear communication channels to address issues promptly and ensure a smooth transfer process
Method validation, standardization and regulatory compliance	Meeting regulatory requirements for method transfer, including documentation and validation, particularly across different regions can be complex and time-consuming	Perform rigorous validation and standardization processes. Stay updated on regulations and ensure methods comply with all relevant standards
Intellectual property and confidentiality	Protecting IP during the technology transfer may be complex, particularly when dealing with proprietary methods and technologies	Implement strict confidentiality agreements and IP protection measures
Data interpretation	Incorrect data interpretation can lead to misunderstandings and method transfer failures	Ensure consistent data interpretation and apply appropriate statistical methods to compare results

Summary

The flawless implementation, transfer and reliability of analytical and microbiological methods are critical aspects needed for the advancement of biotechnological applications. By choosing the right methods, following the best practices and proper understanding of often complex regulatory landscape, (contract) manufacturing organizations can ensure the integrity and quality of the manufactured products. Efficient technology transfer not only enhances operational efficiency but also ensures compliance with regulatory standards, ultimately contributing to the success of biotechnological innovations.

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- **Dedicated team throughout the whole project**
- **Facility registered as food manufacturing site at FDA. Holding additional certification such as cosmetic manufacturing (EFfCI cGMP), ISO 9001:2015, FSSC 22000/HACCP, FAMI QS, Halal and Kosher**
- **Long lasting experience with high quality, speed, and strong focus on continuous process improvement**
- ***Focus on what matters to you***

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