

# PROCESS OPTIMIZATION STRATEGIES FOR BIOTECHNOLOGY PRODUCTS: FROM DISCOVERY TO PRODUCTION

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## Summary

Process optimization is an evolving mechanism of improvement; often never stopping during the life of the process. It is a complex and very costly undertaking, but with a clear economic and quality imperative, to make the most efficient use of raw materials or other resources (e.g. equipment or time) which contribute significantly to the total cost. As a competitive strategy it has its greatest impact at the design stage, which includes both discovery and development. Using *Biopharmaceuticals* as an example, it can be broken down into four phases, involving improving the chemical structure, intracellular genetics of the organism, incubation medium and operating environment. One of the most challenging aspects of process optimization is that for greatest efficiency, optimization of these four phases occurs simultaneously. There are now many tools in the process developer's toolbox to assist with process optimization. The challenge remains to use these tools to find the quickest route to climb to the top of the process performance mountain.

## 1. Introduction

Process optimization strategies are particularly appropriate for furthering the goals of sustainable development and global security. Simply put, optimized processes require fewer resources. Developing an optimized process is also an essential part of bringing new economic opportunities to fruition. Within the sphere of biotechnology, developing a new pharmaceutical drug product highlights most of the challenges involved from discovering an opportunity to coming up with an optimized, economic commercial process.

Fermentation processes are the workhorses involved in the production of many of the pharmaceuticals in use today. Optimizing fermentation processes is thus of central concern to the industry and will be used as the example for this discussion of process optimization. It also has wider application, as fermentation is used in a variety of processes from food and energy production to environmental clean up.

### 1.1. Process Development

The objective of process development is the timely production of sufficient product to meet market demand using a cost-effective and reliable process, which also meets safety and quality requirements. The broad stages of process development are product identification, process identification, product validation (i.e. approval), scale-up (turning a laboratory process into a robust practical commercial process) and process validation (for pharmaceuticals). In addition, for therapeutic products, a scale-up to at least pilot-

scale is required to produce sufficient material for clinical trials prior to product approval.

## 1.2. Process Optimization

The economic objective of process optimization is to produce the greatest quantity of saleable products at the least cost to maximize profit. To effect this optimization requires a process model spanning upstream (see Upstream Processing - Sterilization in Bioprocess Technology) and downstream processes (see Downstream Processing of Proteins Using Foam Fractionation), which can be used to define strategies for optimizing sub-processes. For example, in commodity production employing a very large-scale fermenter, increasing volumetric productivity ahead of yield may be the best strategy for maximizing total profit. However, the difficulty of global process modeling means that, in practice, process optimization is applied to the major sub-processes which have the greatest impact on the competing priorities of quality, productivity and cost (i.e. efficiency). As a general rule for commodity products, because the relative cost of raw materials is high (typically greater than 50 percent of total cost) compared to processing costs, optimizing yield is a preferred strategy to reducing operating costs.

Given the dynamic nature of markets and technology, optimized processes must be continually updated or superseded to maintain competitive position. Many fermentation processes such as those for secondary metabolites (e.g. antibiotics) operate well below theoretical maximum yields. The improvement horizon for a single product manufactured at a facility can therefore span decades with the introduction of new strains and improvements to mixing, aeration and nutrient feeding.

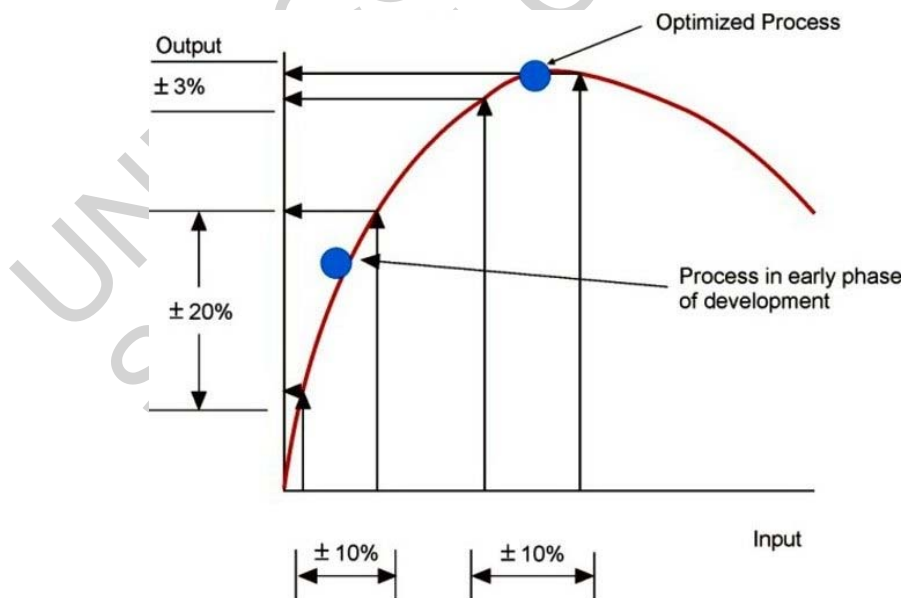


Figure 1: Uncontrolled variation in process inputs (e.g. raw materials) is transmitted to the process outputs (e.g. yield). For a given variation in inputs, an optimized process not only brings economic benefits (e.g. increased yield), but also reduces transmitted variation to downstream processes.

In contrast to an open-ended, online improvement strategy, competitive pressures and technologies focus the optimization strategy back to the design stage, for process development. The potential benefits are two-fold: the economic benefits of an optimized process are available earlier in the product's lifetime and secondly a more robust, more reliable process results (see Figure 1). Process development and process optimization are thus all part of the same process improvement continuum.

Having considered why and when to optimize, the remainder of this article considers what processes to optimize and how. It should be emphasized that process improvement (development and optimization) involves significant effort and cost.

## **2. The Drug Discovery and Development Process**

The discovery and development of a new pharmaceutical is a complex process, with many iterative loops and strategies to choose from. A generic drug development process is shown in Figures 2 and 3.

The first step in the drug development pathway is identifying a disease state that is an economically viable proposition to target. Companies are usually biased in certain directions by their prior history or areas of expertise. However, some simple generalizations apply for a target disease selection such as:

- The disease should affect a significant fraction of a population (orphan drugs are an exception to this)
- The outcome of non-treatment or using existing treatments should be severe eg. death or severe debility
- Current regimes of treatment are non-existent or work poorly

Next a discovery strategy is developed (see also *Biotechnology in Drug Discovery*). A competitive advantage could arise from a novel understanding of a disease mechanism, i.e. a novel target within a metabolic process or cascade. Another extremely valuable strategy is the re-interpretation of historical data.

This could come from testing old abandoned drugs in the light of new information, or trying old drugs used for other disease states on a new disease target. Traditional knowledge is also a valuable starting point. Libraries of chemical structures may provide building blocks for constructing new disease target ideas.

Once the disease state is identified the specific bioactivity desired must be determined; for example, if a breast cancer target is chosen, anti-angiogenesis activity may be sought. The most crucial part of this early phase of the process is developing an assay that will identify the desired bioactivity. This assay must be carefully thought out (see also *Bioassays in the Development of Biotherapeutics*). It must be able to be miniaturized, amenable to robotic execution, be quick, inexpensive, have the desired sensitivity and usually have a visual end product. Once this assay is determined, a large number of candidates can be put through the screen. This has resulted in many high-throughput screening techniques where success is a "number game" (the probability of success being proportional to the number of sources tested).

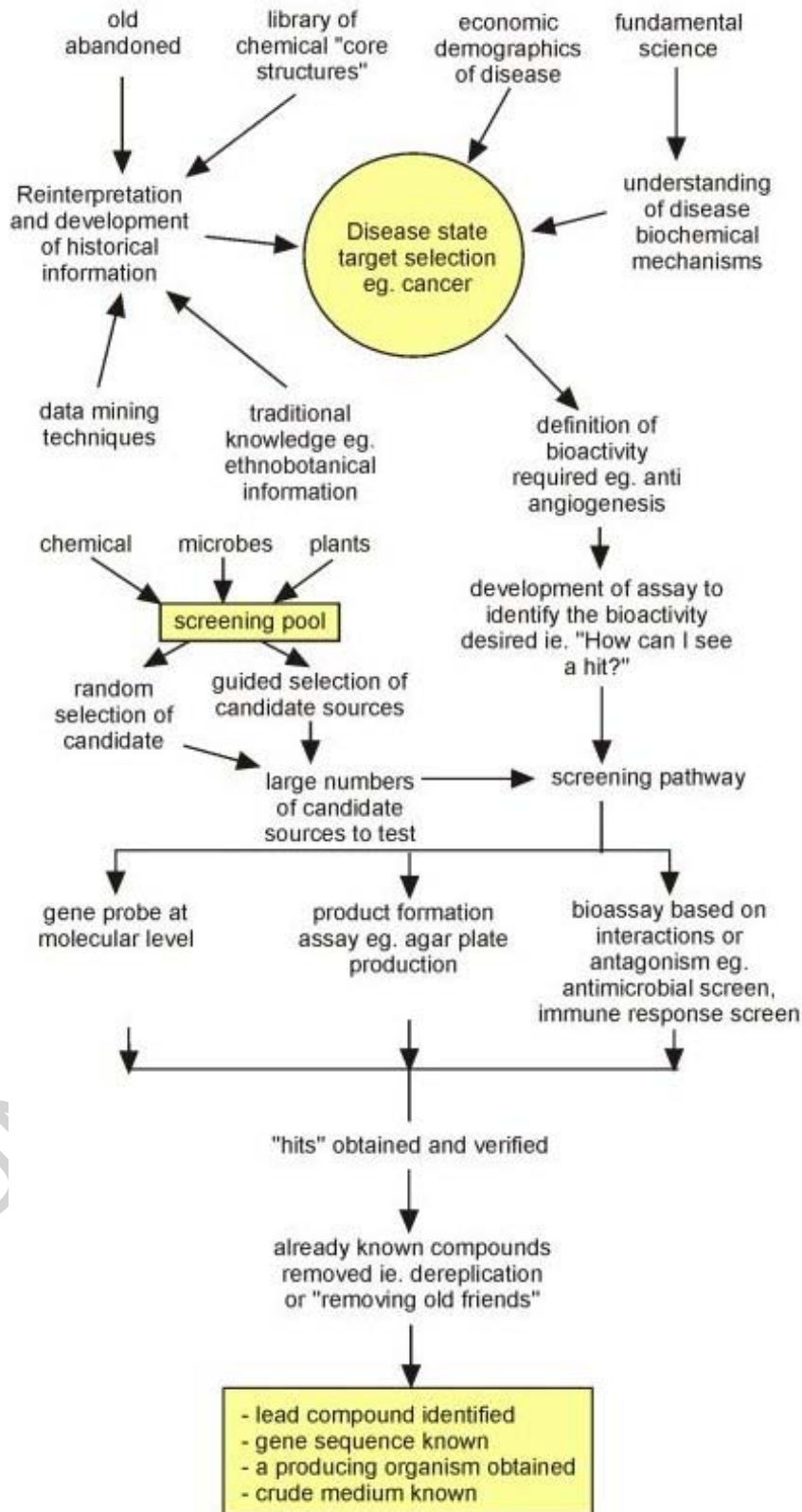


Figure 2: The discovery phase of a generic drug development pathway. Many of the steps are iterative and interdependent on each other.

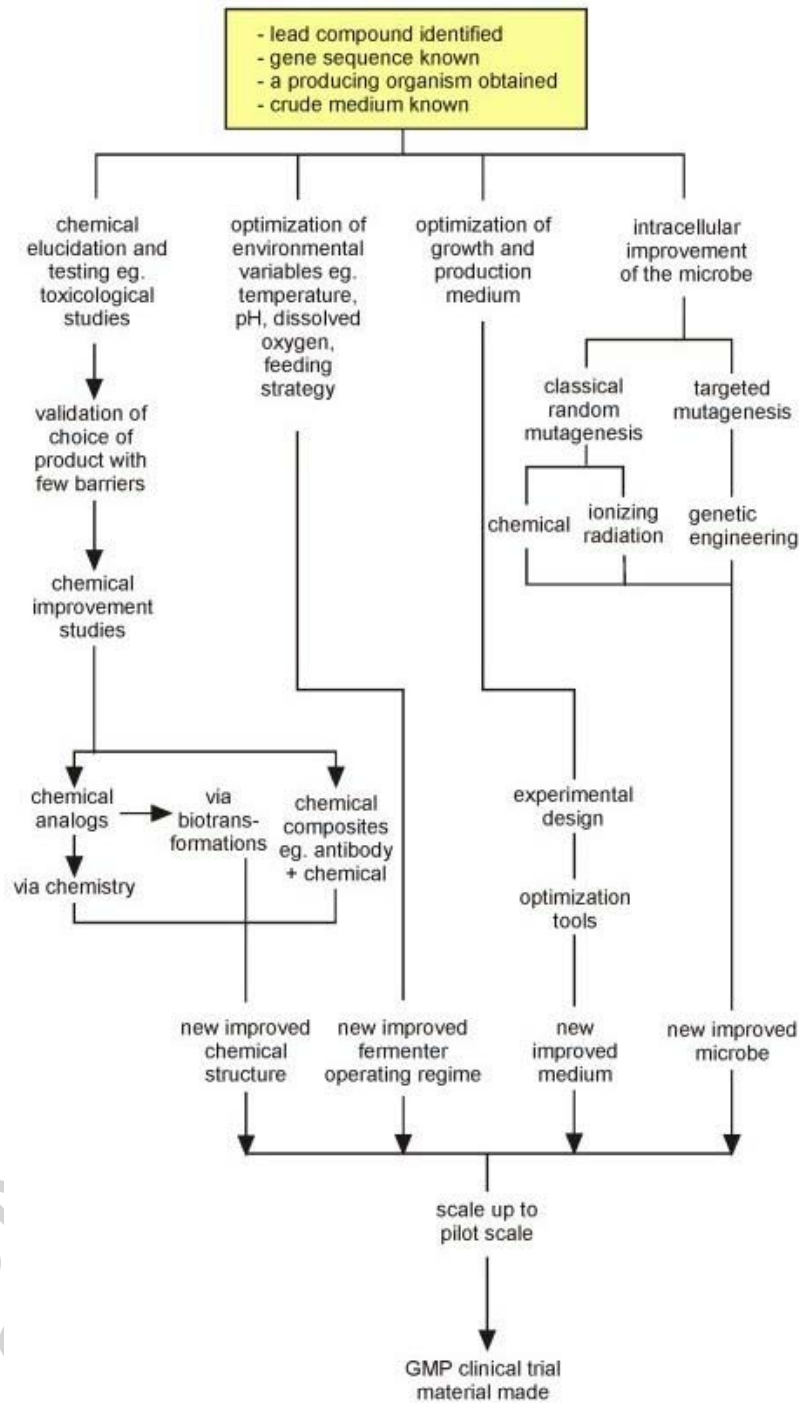


Figure 3: The Optimization phase of a generic drug development pathway. Many of the steps are iterative and interdependent on each other.

Sources of candidate materials to test can be from the animal, plant or microbial kingdoms. Recently there has been much interest in combinatorial chemistry to supply large numbers of source chemicals to screen. The sources can be chosen at random or using a guided strategy, for example, choosing one plant species from each plant Phylla to test, or choosing microbes from extreme environments to test.

The screening pathway used is either one of two main types:

- A gene level or pre-expression screen
- A product/activity level or post expression screen

A pre-expression screen is one that searches for genes rather than products or activities, the most popular being a gene probe (see *Molecular Methods in Diagnostics*). The product of this screen is an organism, which has the potential to produce a given product. The gene for this product may be put into a new host that has a reliable expression system to ensure that the gene makes a product (see *Methods in Gene Engineering*). Gene level screening is fast, inexpensive and independent of the medium used to grow the organism. However, it is very targeted and requires knowledge of the gene sequence one is seeking.

The post-expression screens rely on detecting product formation such as an agar plate screen or a bioassay involving antagonism or response e.g. antimicrobial sensitivity or immune response. The post-expression screens have almost opposite advantages and disadvantages to the pre-expression screens. Post-expression screens require no prior knowledge, depend heavily on the medium and conditions used to grow the organisms (see *Microbial Cell Culture*), and are very broad in their scope. This has led to screening having a reputation of being very risky, expensive and taking a long time. Estimates of screening success rates vary wildly.

After the initial screening process where “hits” have been obtained, the hits have to be dereplicated and verified. Dereplication is an important, but often-arduous task. Companies do not want to invest time progressing a chemical structure only to find that someone else has already discovered and patented it before them, or that it is already in the public domain. Removing “old friends” is often accomplished through the use of databases and is a key skill for the discovery company to master. At the end of this process a lead compound should have been identified, a gene sequence obtained, a producing organism found and a crude sub-optimal growth medium identified.

From this point the optimization process begins in earnest. The optimization process consists of 4 separate pathway developments along which progress occurs simultaneously (in parallel) to cut the development time down as much as possible. The four pathways are:

- Testing and improving the chemical structure of the product under consideration.
- Improving the intracellular genetics of the producing microbe.
- Improving the nutrient medium that grows the microbe.
- Designing operating environments for growing the microbe, for example, process control strategies.

Each of these optimization pathways will be considered in more detail below. When the four pathways converge into a manufacturing process, the next step is to scale up the process to the pilot scale usually 10–1000L in size (see Figure 4) and lastly to produce clinical trial material under GMP conditions.





Figure 4: Pilot-scale process development such as that shown at Industrial Research Ltd is a crucial step in the process optimization strategy

One key question that needs to be answered at the onset of process optimization studies is, what is the target variable to be optimized? There are many target variables to choose from, for example, product yield, volumetric productivity, medium cost, number of processing steps, capital cost and fermenter size to name a few. The answer to this important question was considered at the 1999 US Bioprocessing Symposium, which stated “To achieve economic viability increasing production yield is the single most important factor in the production of biotherapeutic drugs... improving yield has four to five times the impact on reducing the cost per gram than does reducing operating costs” (Langer, 1999). This clearly signals the dominant importance of yield increases. The next question to arise is, when is the yield high enough to stop the development effort. As a rule of thumb for secondary metabolites,  $100\text{mgL}^{-1}$  is usually sufficient to enter into pilot scale production, with  $1\text{-}100\text{gL}^{-1}$  being an economically significant yield. This obviously depends on the particular product involved, but the above can be considered “ball park” estimates.



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## Biographical Sketches

**Dr Max Kennedy** completed his doctorate in biochemical engineering at the Massachusetts Institute of Technology and is currently the Biopharmaceuticals Operations Manager at Industrial Research Ltd, New Zealand where he manages the contract manufacturing of pharmaceuticals up to Good Manufacturing Practice (GMP) standard for human clinical trials. He has long standing interest and experience in process development and optimization, particularly in the fermentation medium design area.

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