Single-Use Optical Sensors

Ushering in a New Era of PAT for Bio-Processing

Catching-Up – Despite the rapid adoption of the process analytical technology (PAT) initiative by the petrochemical and pharmaceutical industries, the implementation of PAT in biotechnology has been slow, primarily owing to the complexity of large molecular products and the living organisms that produce them. The benefits of embracing PAT in bio-processing are significant, because understanding the bioprocess and optimizing production efficiency directly result in improving the product and yield.

Traditionally, PAT involved the implementation of sensors or analytical instrumentation for process control. The stringent steam-in-place (SIP) and clean-in-place (CIP) sterilization requirements for traditional bioreactors precluded many sensor technologies from being applied, or if a sensor could be designed to satisfy the requirements, its high cost could not be economically justified. Moreover, in many cases, the bioreactors themselves, being of a 20-year-old design with a limited number of measurement ports, could not be easily modified to retrofit additional sensors.

More recently, the PAT initiative has evolved into a broader context, specifically the identification and control of sources of variability in bioprocesses. With this broader definition, PAT includes the bioreactor and control system design as well.

The ideal implementation of PAT is to introduce new technologies quickly into R&D, with the intent of implementing them early in the design of manufacturing process for new products, before they are validated, and provides three core capabilities to a bioprocess:

Analyze in real-time the uptake of nutrients, growth and viability of the cell population, as well as the concentration and composition of active and side products in bioprocess

Model and predict the process so as to actively control it in real-time

Manage and store all process data, from R&D through cGMP manufacturing, in order to generate a complete genealogy of the process.

The Single-use Revolution

Due to the requirements for handling hydrostatic pressure and the pressure associated with cleaning/sterilization procedures, bioreactors have generally been constructed from stainless steel such as 316L. The initial cost of such a bioreactor and associated plumbing is substantial. The energy cost

to run the impeller, the aerator and to cool/heat the bioreactor is also sizable. Finally, the costs of cleaning and sterilizing such a reactor after use and disposing of the waste water from the cleaning process, are non-negligible.

Given the high capital and operating costs associated with conventional bioreactors, single-use bioreactors have become increasingly prevalent in upstream processing. These are constructed using plastic films which have been proven to be biocompatible and animal-derived-component free. One popular design uses a rocking motion to mix and aerate the bag whereas the predominant design uses the more conventional stirred-tank approach with plastic impellers that essentially mimic traditional bioreactors. All disposable bag reactors include gas/liquid inlet and outlet ports, filters, pressure control valves and additional ports for sensors.

Single-use bioreactors represent a unique opportunity for implementing PAT ideology. They are at an early stage in the adoption cycle, being brought into R&D for evaluation, prior to being deployed into manufacturing. Therefore, new sensors and control methods can be tested without risking costly FDA approval. Any process transitioned from a traditional to a disposable bioreactor must be re-validated, so that measurement and control strategies can be changed. Moreover, most traditional sensor technologies are too large or cumbersome for single-use applications, creating an opportunity for new technologies.

Many off-line process analytical tools, such as basic HPLC or blood analyzers are in use with single-use bioreactors today. However, a fundamental need for in-process, real-time sensors remain.

Novel Optical Technologies

Single-use bioreactors have a flexible port design, utilize gamma radiation-based sterilization and are optically translucent, thereby lending themselves very well to the integration of optical sensors. Optical measurements for PAT can utilize any part of the electromagnetic spectrum including ultra-violet (UV), visible (VIS), near-infrared (NIR), and mid-infrared (MIR) radiation to probe the biochemical or chemical system inside the bioreactor bag.

The collected data can be directly or indirectly indicative of the state of the bioreactor medium. Virtually any method that can be directly correlated to an analyte of interest allows for a useful PAT sensor.

Two key optical sensor technologies for single-use bioreactors allow the measurement of dissolved oxygen (DO) and pH. Both techniques apply phase fluorimetry and utilize a dye which has a fluorescent lifetime that is quenched by the presence of the analyte in question. The spot must be in contact with the bioprocess inside the bioreactor bag. The excitation radiation must be delivered to the spot, and the emission radiation from the spot must be delivered to the detection system. The optical and electrical system resides outside of the bioreactor bag.

Most of the spot-based approaches involve either integrating a patch (that holds the spots) with the bag or using a fiber-based optical delivery system into the bag via a port. The former method has the disadvantage that the spots and the optical system must be mechanically aligned in close proximity, whereas the latter method has the disadvantage that fiber-based systems are optically inefficient and expensive. In both cases, significant optical radiation must be delivered to the spot, which results in accelerated photo-bleaching of the dye and long-term measurement drift. This issue is of particular concern for pH, and has delayed the adoption of optical sensors for PAT.

An alternative approach uses a micro-optic light engine for radiation delivery to and collection from the spot, and is already showing better drift results. The spot is housed on a sheath that is inserted into a 12 mm port in the bioreactor bag prior to sterilization. The seal between the port and sheath assures that sterility is preserved. The optical reader is rated for NEMA 4X, allowing it to be used throughout scale-up from R&D to production. RFID tags are used to enter calibration data for each sheath automatically and eliminate human error. The menu-driven transmitter is highly capable and has two analog outputs for easy integration into an automation system. Overall, TruFluor optical DO and pH sensor features are ideally suited for single-use PAT applications.

In the future, optical methods will also be applied for real-time, in-line measurements of nutrients, additives, waste products, and end products. A tremendous improvement in the new optical technologies for bio-processing is expected in the next two to five years. The advent of these technologies will fundamentally and permanently change PAT in single-use upstream manufacturing.

Finally, it should be noted that potential PAT measurement methods for single-use systems are not limited to optical methods, but can include a diversity of alternative physical mechanisms: electrical (RF and DC electrical fields), chemical, biochemical, acoustic, magnetic, and micro-fluidic techniques, or any combination thereof. Other measurements can involve miniaturized or on-chip mass spectrometry, liquid chromatography, flow cytometry, or nuclear magnetic resonance (NMR).

► Contact:
Barbara Paldus and Mark Selker
Finesse Solutions
Santa Clara, Calif., U.S.
Tel.: +1 408 327 6623
Fax: +1 408 327 6698
bpaldus@finesse.com
www.finesse.com