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Culture vulture: Building a better bioreactor

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A single use bioreactor with an alternative design offers improved performance over previous systems. David Wilson reports

For many years, bioreactors have been used to culture mammalian cells for the production of pharmaceuticals and vaccines. To do so, such systems sustain and grow populations of genetically modified cells in a culture medium contained in a vessel inside the bioreactor. These devices precisely control environmental conditions such as temperature, dissolved oxygen levels, pH, and the speed at which the medium is agitated.

Traditionally, bioreactor vessels were cylindrical stainless steel designs with thick walls built to ensure that they can be adequately sterilised by high pressure steam prior to use. Needless to say, such reactors also sported a lot of stainless steel piping for adding media, cells, and reagents as well as harvesting cells – process piping that must also be sterilised to guarantee that no viable microorganisms remained that could contaminate the culture medium and thus spoil the process.

Just over ten years ago, a solution to the sterilisation issue was introduced onto the market – the "single-use" bioreactor. Eschewing the use of pressure-rated stainless steel designs, these systems incorporate disposable plastic vessels that can be sterilised with gamma irradiation and used just once. Such single-use bioreactor systems had the advantage that they not only reduced capital equipment costs but also operational cost too, since heavy-walled stainless-steel vessels and associated high-temperature cleaning were no longer required.

Building upon the single-use concept, the next-generation platform of bioreactors developed by Camarillo, California-based PBS Biotech is claimed to not only perform better than earlier single-use designs but is also simpler to use as well. To achieve these goals, PBS' bioreactors are designed to be compact integrated units, comprising a culture chamber, an integrated controller and liquid handling all in one pre-configured modular unit.

One of the distinctive features of the PBS bioreactor is the way that the designers have chosen to agitate the medium inside the single-use vessel. Rather than use a conventional impeller connected to an external motor via a shaft, the PBS system makes use of an Air-Wheel impeller instead. The Air-Wheel, which rotates freely about a shaft anchored to the bag, is powered by the buoyant force of gas bubbles introduced from the bottom of the vessel.

A gas mixture, comprised of varying amounts of air, carbon dioxide and nitrogen, is sparged into an orifice at the bottom of the bag via mass flow controllers. The exact composition is dictated by the pH and dissolved oxygen requirements for the culture at any given time. A supplementary supply of pure oxygen is also introduced into the culture as needed through a microsparger element to maintain high cell densities during the later stage of a production cell culture process. use bioreactors.

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According to Daniel Giroux, the Director of Product Development for PBS Biotech, the use of the Air-Wheel impeller, rather than a motor driven impeller, provides users with a number of distinct advantages over earlier single use bioreactors.

‘Since all the rotational parts of the mixing system are self-contained within the sealed plastic vessel, it is unnecessary to provide a mechanical seal between the shaft and the plastic vessel. This is an issue on earlier designs, since the seals must be made inexpensively enough for single use as well as capable of being sterilised by gamma irradiation,’ he said.

What is more, the PBS system is inherently scaleable - because the impeller is being driven from its circumference, the torque imparted by the gases on the impeller wheel increases as a function of the impeller’s diameter, unlike conventional systems where larger motors are required to provide the necessary torque to rotate larger impellers.

‘While it was previously impossible to keep mixing times constant across bioreactors of different sizes simply by increasing the size of the impeller, the PBS design has no such disadvantages. As our impellers grow larger to keep up with the size of our vessels, mixing times do not increase as rapidly when the system is scaled up as they would in a more traditional single-use system,’ said Giroux. Indeed, Giroux claims that the mixing time for a 5000 litre PBS reactor is lower than a traditional 2000 litre scale reactor.

While it provides relatively constant mixing times at whatever the size of the system, the use of the Air-Wheel in the bioreactor means that fast mixing times can also be achieved without compromising the amount of shear stress that is imparted to the culture by the impeller. Traditional designs, on the other hand, improved their mixing times at the expense of increasing the shear stress that the impeller imparted to the culture as a process was scaled up.

‘If you want to provide adequate mixing at scale, you typically have to endure higher shear forces, but we achieved exceptional mixing while keeping the shear very low. The shear profile across all scales of our reactors is practically constant across the spectrum of bioreactors that we offer,’ claimed Giroux.

To enhance the mixing process further, the development team at PBS also chose to design two vanes in the middle of the impeller – one of them drives the culture content in one axial direction, while the other drives it in the opposite direction. The effect is that the fluid is folded in on itself as the impeller rotates, mixing the medium even more effectively.

According to Giroux, another advantage to the design is that since the gas mixture moves circumferentially with the direction of the impeller, the downward flow of gases on one side of the reactor tends to draw the oxygen bubbles from the microsparger downward with it, entraining them in the solution for a longer period of time, enhancing the efficiency of oxygenation.

Regulating the temperature of such media is a critical part of the process. In the PBS system, the temperature of the culture is measured non-invasively by a resistive temperature detector on the non-disposable housing unit. Once the bag has been installed, the detector on the housing comes in direct contact with the back portion of the bag and is sufficiently insulated from the conductive metal housing to ensure that it accurately measures the temperature of the culture.

In order to validate their pneumatic mixing technology at large scale, the engineers at the company built a 3,500 litre prototype system with 5,000 litre maximum working volume. Biological tests using a mammalian cell line were performed at 2 litre, 10 litre, 50 litre, and 250 litre scales, and the results showed that the PBS bioreactors achieved comparable or superior performance to that of conventional stainless-steel systems or existing single-use systems.

‘Because the system is scaleable, we can offer bioreactors from bench-top research scales all the way up to a full-scale production. . As such, the technology has the potential to accelerate the time to market of a pharmaceutical or vaccine by eliminating the potential technology transfer challenges that are often encountered as a process moves from the research and development phase through to clinical and commercial phases,’ said Giroux.

PBS Biotech is currently developing 3 litre, 15 litre, 80 litre and a 500 litre bioreactors based on its technology. A 2500 litre reactor is currently in design and will be custom configured to suit each client. The company is also assessing the market demand for their larger 5000 litre version of the system.