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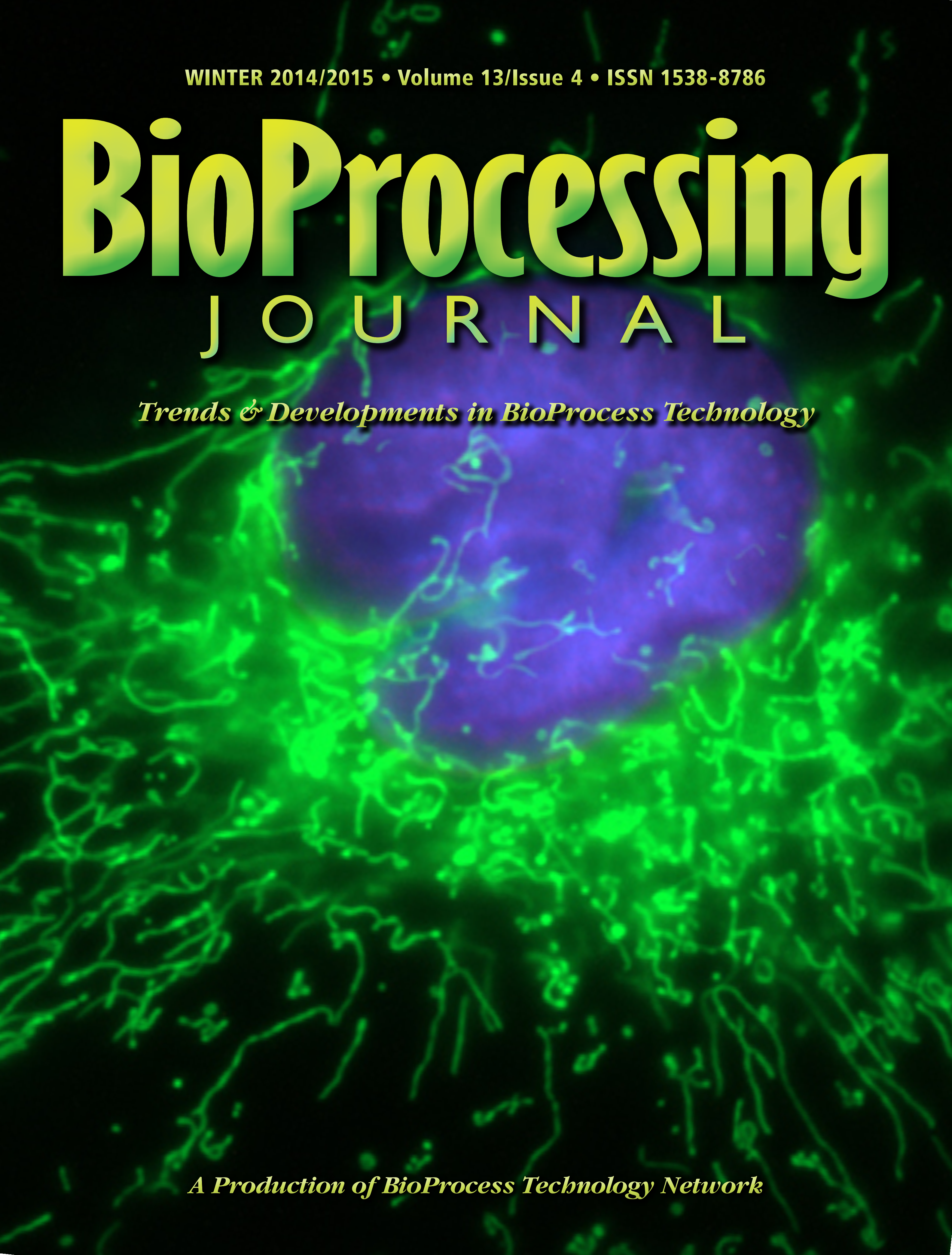
Trends & Developments in BioProcess Technology



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SUPPLIER SIDE

Streamline Your Cell Line Screening: *With the Automated, Microscale, Stirred Tank, Single-Use Bioreactor*

By TIM WARD

Abstract

The price per patient for protein-based and monoclonal antibody (mAb) therapies runs into thousands of dollars per patient each year. These therapies cost considerably more to manufacture than small molecules. Hence, if mammalian or insect cell lines expressing high protein titres can be selected and optimized for protein expression using microscale bioreactor models early in development, then manufacturing costs can be reduced significantly.

ambr™15 System

During cell line screening, the chances of isolating a high-producing clone for proteins such as mAbs is improved by maximizing the number of cell lines that are evaluated. Traditionally, shake flasks and benchtop bioreactors have been used to select the most productive cell lines, as well as define optimal media, feed, and bioprocessing conditions. However, shake flasks provide no control over pH and dissolved oxygen (DO), and the mixing environment is unlike that in a bioreactor. Additionally, shake flasks are often manipulated by hand so it is difficult to perform nutrient feeding or sampling without introducing variability. The use of shake flasks can often result in different cell growth and protein expression profiles to those seen after process scale-up.

Currently, cell line ranking requires several rounds of batch and fed-batch, and shake-flask studies to reduce the initially large number of clones to a more manageable number for evaluation in benchtop bioreactors. These vessels are resource-intensive and, due to their scale, expense, and limited throughput, researchers are restricted in the number of benchtop bioreactors that can be run in parallel. Typically, only three-to-four cell lines can be taken through to evaluation in small (2 L–10 L) vessels. Hence, the final choice of cell line can sometimes perform suboptimally upon scale-up, adversely affecting outcomes such as yield and product quality.

This need to conduct many experiments under benchtop bioreactor conditions has resulted in the development of miniaturized, high-throughput culture technologies. The drawback with many of these approaches is that they do not mimic the sparged and stirred action of a bioreactor because they do not have internal impellers but rely instead on other methods for mixing, such as shaking.

Automated, Stirred and Sparged Microbioreactor

With the need to imitate bioreactor mixing and gassing parameters in mind, the advanced microbioreactor system (ambr15) was introduced by TAP Biosystems (now part of the Sartorius Stedim Biotech Group). The ambr15 microbioreactor system has three components: the disposable bioreactor, the workstation, and the software. The system offers automated parallel processing and control of 24 or 48 bioreactor experiments using just one operator, and can be used with mammalian or insect cells.

This stirred, sparged microbioreactor system is a technology that mimics benchtop bioreactors, with the microbioreactor providing better environmental control and thus, more representative culture performance than shake flasks.

ambr15 System

Single-Use Bioreactor Mimic

The ambr15 workstation controls 24 or 48 single-use microbioreactors, each with a working volume of 10–15 mL. Whereas conventional glass vessels require post-run cleaning and sterilization, the single-use reactor is free of such concerns. Key to its success as a mimic is that its contents are stirred by an internal impeller, and gases are supplied by sparging. To maintain sterility, the workstation is housed in a biological safety cabinet and liquid handling automation facilitates aseptic culture set-up, inoculation and feeding, as well as sampling into vessels such as 24/96-well plates and ViCell cups. These operating protocols can be configured for individual control of up to 24 or 48 reactors operated in parallel. Each reactor incorporates individual DO and pH sensors to enable closed loop control of these parameters. Automated pH regulation is achieved by CO_2 sparging or alkali addition, while O_2 is sparged whenever needed to maintain a DO set point. There is also the option to attach an analysis module to the workstation for at-line pH calibration, thus enabling accurate culture control.

Single-Use Microbioreactor With Impeller

Software for Full Control

The ambr15 system is supported by easy-to-use software which enables the simple set-up of protocols for the workstation to carry out. The protocol defines operating parameters such as DO/pH set points, stirrer speed and temperature, as well as tasks such as inoculation, feeding, and sampling. Protocol timings and details can be edited at any time if steps need to be added, modified, or deleted, including while an experiment is running. Real-time data such as gas flow rates, volumes, and pH/DO values are logged continuously during a run, and external values such as cell counts, metabolites, and product titres can also be imported into the software. Data can be generated as graphs within the software or exported for production of graphical or tabular results in spreadsheet software. None of these automation features is available with shake flask models. Automation delivers consistency while also enabling scientists to spend less time on repetitive laboratory tasks and instead, focus on value-adding activities such as assessing data or planning future experiments.

The Future of Cell Line Screening

The ambr15 stirred, microscale bioreactor system can be used instead of shake flask models or conventional benchtop reactors, saving considerable time and money in set-up, operation, shut down, cleaning, and sterilizing tasks. Since this microscale system is fully automated and utilizes smaller cell culture volumes, scientists can perform many more experiments with the same amount of resources such as media or feeds, making this a cost-effective and high-throughput way to select higher expressing cell lines and to optimize the culture process. Consequently, bioprocess optimisation is no longer limited by availability of benchtop bioreactors, operator time, or facility infrastructure, so the use of the ambr15 microscale technology can make a significant contribution in reducing development timelines and manufacturing costs of protein-based therapies.

An Interview With the Author, Tim Ward

Q: *Tim, as one of the team responsible for developing the ambr technology, can you give us an industry-wide perspective on the need for scale-down bioprocess models?*

A: There has been a search for tools to improve cell line screening and selection as using shake flasks and benchtop bioreactors have proven to be slow, labor intensive, and unlikely to generate optimal clones. As a result, scientists frequently only take their most promising clones forward for runs in benchtop bioreactors and then into process development. These clones often perform as expected in scale-up, but scientists are never certain if they would have identified a more stable or higher-expressing clone if they had been able to screen more clones under bioreactor conditions.

Q: *How have scale-down bioprocess models developed since?*

A: Recently, there has been a focus on developing high throughput microscale bioreactor technologies. The first system used a custom-designed, multiple-chamber cassette. This was manipulated in a large, automated system that could run hundreds of plates in parallel.

Although initially tested by a small number of companies, the large size and complexity led to limited market adoption. Smaller, benchtop systems were also developed based on 24-well shaken plate designs. Some initially focused on microbial strain selection and were then tested for cell cultures, but they lacked the ability to automatically control pH through liquid base addition.

Q: *Why is the ambr15 different?*

A: ambr15 is the first true, microscale mimic of a benchtop bioreactor, which includes an impeller, full pH control using CO_2 and automated liquid base addition, and gas sparging. This ability to mimic bioreactors is validated by many published reports, which show ambr15 models all key aspects of a stirred bioreactor from 1 L–200 L scale.

Q: *What has happened with this technology since its launch in 2010?*

A: The ambr15 system has rapidly become the standard tool for cell line and media development within the biopharm industry, with over 100 systems installed. Additionally, a number of further system developments have been introduced, including cooling, integrated cell counting, and software enhancements.

About the Author

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