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Bridging Polymer Science and Biotechnology Applications with Single-Use Technologies

Ekta Mahajan and Gary J. Lye

Implementation of single-use technology in the biotechnology industry is increasing every year. One major interest has been understanding the interaction of extractables with protein and cells for applications ranging from cell banking to biopharmaceutical manufacturing. In October 2015, the Engineering Conference International (ECI) organization hosted a conference in Leesburg, VA, to explore how the science of plastic applies to bioprocessing.

The "Single-Use Technologies: Bridging Polymer Science to Biotechnology Applications" meeting brought together experts from different fields to share issues, understanding, and solutions. About 100 attendees representing end users, academia, and suppliers (e.g, resin, film and single use components fabricators) participated in scientific and collaborative discussions regarding the use of disposables for different functional areas. Those discussions led to appreciation for each other's challenges and requirements. Representatives from other industries (e.g., food processing) also attended and participated in open communication. Attendees discovered clear similarities among industries as well as lessons learned in the food industry that can be leveraged for the biopharmaceutical industry.

Professor Govind Rao (director at the Center for Advanced Sensor Technology at the University of Maryland, Baltimore) gave the keynote presentation. He focused on how single-use technologies can support the future of bioprocessing and making "bench-to-bedside" processing possible. The keynote by Mitchell Cheeseman (managing director, regulatory and industry affairs at Steptoe & Johnson LLP) portrayed similarities between a risk-based extractables approach in the food industry and strategies used in the pharmaceutical industry. For example, the food industry uses the model solvent boundary approach and multiple time points similar to those in the pharmaceutical industry approach. The food industry extractables approach was mandated by the US FDA, further emphasizing the importance of collaboration between end users and suppliers to agree on standards. To maximize the potential of using disposables, different industries should work together. The conference included six sessions and a workshop (see "Workshop" box) that addressed topics requiring scientific focus to ensure successful implementation of single-use systems.

Properties of a Good Plastic

Session co-chairs Magali Barbaroux (vice president of R&D in fluid management technologies at Sartorius Stedim Biotech) and Sally Kline (director of material science at Amgen) led the first session. The final form, fit, and function of a plastic component is determined by the choice of polymer family and formulation, polymer processing techniques, and sterilization methods. All three factors must be considered during design and qualification of a plastic component. The targeted deliverable for this first session was to harmonize knowledge of polymer synthesis, processing, and sterilization considerations.

Presenters addressed four key points. First, plastic properties are the outcome of a combination of formulation, converting process, and post-treatment methods. Polymer properties and potential profiles of extractables and leachables are determined not just by the synthesis route and

selection of catalysts and additives, but also by the plastic converting (e.g., injection, extrusion, and lamination) and posttreatment processes (e.g., gamma irradiation and autoclaving). Additives can't be eliminated but must be controlled to ensure consistency. Reducing additive content affects polymer properties (e.g., reducing antioxidant contents increases risk of gelling). A good balance must be defined by product designers so that products are fit for their anticipated use. Understanding how interactions among sterilization energies and methodologies affect polymer properties is critical to delivering consistent polymeric components to the industry.

Second, regulatory status of plastics for biopharmaceutical applications is now focused on a risk-based approach rather than solely a quality focus. Over-engineering a risk-based approach may provide a false sense of advanced understanding. Regulatory guidance is regionally specific, thus adding complexity and confusion to end-user interpretation.

Third, difference in scales between plastics manufacturers and the single-use industry requires an open dialog among all supply chain stakeholders. Plastics manufacturers and their sub-suppliers may not be dedicated to the biopharmaceutical industry. As a result, those companies may have limited data relevant to or understanding of the applications in biomanufacturing. Openness, transparency, trust, and partnerships among all stakeholders in the chain (e.g. a polymer supplier, polymer converters, single-use systems manufacturer, sterilizer, and end users) are critical for controlling risk associated with the use of single-use systems.

Finally, plastics have been widely used in other industries for decades. The main issue now is determining how the bioprocess industry can benefit from that experience. Methodologies for applying plastics in other industries can be translated to the biopharmaceutical industry, thus saving time and resources required to invent new methods. Key examples of best practices that are translatable include additive migration modelling for packaging and effect of irradiation and sterilization on plastics (in the food industry).

Subsequent conference sessions on plastic properties focused on identifying current practices and procedures gaps and reviewed potential approaches to close those gaps by transferring best practices from other industries.

Workshop on Standards and Standardization

Christopher Smalley (director of engineering, biosterile validation at Merck) and James Vogel (founder and director of the Bioprocess Institute) led a workshop that used a real-time survey approach to focus on key issues faced by industry and factors impeding the progress of single-use technologies. Enthusiastic audience participation and surveys confirmed that extractables, particulates, and system integrity are key issues needing to be better addressed. As discussed in other sections of the ECI conference, applied science is the pragmatic approach to address these issues. A summary of the "Alphabet Soup" organizations related to single-use technology was presented to ensure alignment of efforts toward a common goal.

The Infamous Extractables and Leachables

Joseph St. Laurent (president and chief scientific officer at Chemic Laboratories) and Trishna Ray–Chaudhuri (senior QC associate in raw materials at Genentech) showed that extractables and leachables (E/L) studies continue to contribute to a deeper understanding of single-use technologies. Their presentations focused on how extractable studies can be used to facilitate

implementation, further develop new methods for detection, and standardize testing for single-use assemblies.

The first session presentation showed that clearance of single-use assembly extracts occurs during the ultrafiltration/diafiltration (UF/DF) step. This proof-of-concept study presented a simplified view of E/L assessments for qualification and implementation of single-use technologies. Study results showed that extracts derived from single-use assemblies are cleared in a UF/DF process step, thus further easing implementation of single-use before UF/DF.

The second presentation focused on development of a new extraction and analytical method. This method was used to facilitate detection of the degradant associated with cell death that originated from gamma irradiation of excess bis(2,4-di-tert-butylphenyl) phosphate (bDtBPP). It is a commonly used antioxidant in thermal processing of polyethylene films. If bDtBPP is not overloaded, it is consumed during that processing step. To facilitate development of a new film, the new method allowed detection of bDtBPP at very low concentrations, which are detrimental to cell growth in most cell lines.

The third presentation highlighted the fact that extractable studies under a range of conditions will reflect different results pertaining to extracts and particulates. For example, silicone tubing is commonly used in bioprocessing manufacturing. A study on particulates originating in silicone tubing showed that sample preparation and testing methodology must be closely controlled to obtain reproducible and relevant data. By contrast, use and simulation conditions are related to leachables originating from silicone tubing. Biological regulatory requirement tests measure extractables, and exhaustive conditions are considered true extractable studies. The study showed that testing parameters influenced the resulting extracts in solid or a liquid state in silicone tubing.

The final presentation in this session showed the transition to standardization of extractable studies. The Biophorum Operations Group (BPOG) Extractables Work Group has published a standardized extractable protocol that defines the sample conditions and analytical protocol for extractable testing. This presentation compared BPOG protocol results with previous studies performed under different conditions.

Interaction of Plastic with Cells and Proteins

Jincai Li (executive director at WuXi AppTec) and Weibing Ding (principal scientist of process development at Amgen) discussed the influence of E/L on cells and proteins. The effect of leachables on cell growth was observed several years ago. Significant efforts from suppliers and end users have led to much better understanding of the root causes of those effects. However, researchers are far from claiming complete victory over either understanding or preventing that impact.

Leachables caused by irradiation sterilization seems to be the “dark horse” of cell growth change, as evidenced by a case study presented by Zara Melkoumian (senior technology manager of bioprocesses at Corning Life Sciences). Shake flasks made of polycarbonate (PC) have been shown to affect cell growth.

Interactions of plastics with proteins are much less studied and understood than they are with cell growth. Interactions between plastics and proteins can be significant and far-reaching. Based on end-user experiences, such interactions can occur in any step during a manufacturing process, from cell culture to fill–finish.

Challenges of Single-Use Technologies
Martina Micheletti (senior lecturer at University College London) and Russell Wong (senior manager, manufacturing sciences and raw materials at Bayer HealthCare) led a session on the challenges of process implementation for single-use bag systems. Presenters discussed concerns related to the integrity of disposable bags, how it can be assessed (including types of tests), and when and where integrity tests should be performed to ensure robustness.

A strong multidisciplinary skills base is needed to link materials properties with mechanical testing, and bioprocessors need to address these challenges. Training on how disposable bags should be handled and on how to perform integrity tests is crucial. Presenters described results of a study from GE Global Research that addressed bag failures due to flex fatigue and how fundamental studies on plastic deformations can provide information for future bag designs or even enhance film properties. But such studies could be expensive and might not be scalable. Current available bag products do not scale geometrically, and studies would have to be repeated at scale.

Other major challenges include availability and robustness of sensor technologies, especially single-use sensors. Presenters proposed solutions based on an optical enzymatic system, solid state pH, and ultrasonic sensing for flow rates. They agreed that fundamental research in this area should be encouraged to provide the bioprocess industry with the needed reliability and accuracy.

Overlap with Other Industries
Yuh-Fun Maa (principal engineer at Genentech) and Peter Neubauer (professor of bioprocess technology Technische Universität Berlin, Germany) led a session covering single-use applications in different fields, including biologic drug substance development, biopharmaceutical processes, and medical devices.

Gary Lye (professor of biochemical engineering at University College London) presented two novel, single-use micro-photobioreactors as development tools for investigating microalgae cultivation. Both models were characterized to successfully translate into manufacturing-scale reactors. Implementing single-use bioreactors can help reduce bioprocess development timelines.

Benson Gikanga (technical development senior research associate at Genentech) presented an assessment of multiple single-use bottom-mounted mixing systems, which have different impeller configurations. Configurations with close contact or tight clearance between the impeller and the drive unit posed detrimental stress to multiple monoclonal antibodies, resulting in subvisible particle formation. The outcomes of this study may benefit scientists and engineers who develop biologic product manufacturing processes by providing a better understanding of mixing principles and challenges.

Jan Oberdoerster (WL Gore & Associates) provided an overview of biocompatibility of single-use medical devices in relation to patient safety. This presentation highlighted the importance of assessing the criticality of single-use materials of construction and product-contact surfaces used during production. Specific examples of material selection, E/L studies, and subsequent risk assessments also were provided and discussed.

How Flexible Is Flexible?
Robert Repetto (senior director at Pfizer) and Mohamad Awada (Thermo Fisher) led a session discussing the flexibility of single-use systems. The bioprocess industry's need for

interchangeability between supplier's designs is a common point of discussion, but is this feasible given a lack of standards and the evolving nature of single-use technology? Should suppliers be expected to support the use of their products in alternative applications chosen by an end user?

New innovative applications of single-use systems are possible, but the key enabler is that single-use components and materials of construction are truly designed for their intended functions. However, as the industry progresses, interchangeability will be required for success in good manufacturing practice (GMP) environments to address technical, supply chain, and quality issues. Interchangeability could be used to solve process problems if unexpected problems arise with a product or process. Industry and academia must work together to enable implementation of single-use technologies in commercial productions.

Session speakers provided examples of how single-use technology can offer flexibility and accommodate interchangeability in unique applications. Chris Smalley outlined how the interchangeability of single-use systems is enabled by well-established functional specifications and an understanding of materials of construction. Peter Neubauer discussed challenges associated with designing unique single-use bioreactors for microbial processes. Valentin Jossen (Zürich University of Applied Sciences) closed the session by presenting different applications of a number of expression systems including plant and stem-cell cultures, for which his laboratory has successfully designed single-use bioreactors.

Next Conference

Please visit the conference website for information about future conferences at www.engconf.org/conferences/biotechnology/single-use-technologies-ii-bridging-polymer-science-to-biotechnology-applications

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Corresponding author Ekta Mahajan is a senior process development engineer at Genentech, South San Francisco, CA; ektam@gene.com.

Gary J. Lye is a professor of biochemical engineering at University College London, London, UK , Inc.