Bioprocess Advances Drive Vaccine Manufacturing in Developing Countries

by Ronald A. Rader and Eric S. Langer

Advances in bioprocessing technology hardware and genetic engineering are expanding the geographic options for biologics manufacturing to include developing and emerging economies. Such advances are beginning to permit biopharmaceutical production in regions that previously lacked the technical expertise or quality processes to permit complex operations, monitoring, record-keeping, and oversight. Global demand by countries for in-country production of biological vaccines is increasing, so those products tend to be leading the way in terms of adoption of modern bioprocessing in developing countries.

Ongoing bioprocessing advances are enabling a diverse spectrum of companies worldwide to develop biosimilars (**1**). Although biosimilar trends tend to be more newsworthy, the long-established worldwide vaccine market is actually much larger — currently ~US$30 billion/year (**2**). This includes access to vaccines that many health authorities consider to be absolutely essential, even a basic human right. Products include universal vaccines (e.g., polio), which are administered to nearly every infant worldwide. By comparison, products with biosimilar approvals are new, with a market only a small percent of that of vaccines and with not a single one yet in the major US market.

(WWW.PHOTOS.COM)

The worldwide spread of vaccine manufacturing is well under way. BioPlan Associates' *Top 1,000 Global Biopharmaceutical Facility Index*, which ranks facilities worldwide by estimated bioprocessing capacity, uses a source database (broader than just the top 1,000) that currently includes 430 vaccine facilities, including 183 (46%) outside the United States and Western Europe (**3**).

Many factors are driving the propagation of vaccine manufacture. The public health and economic needs of developing countries for vaccines are much greater than for biosimilars and therapeutic biopharmaceuticals. Most recombinant biosimilar therapeutics markets pale in comparison to virtually every country's vaccines markets. In some developing countries, healthcare is often solely provided through national governments (often with foreign aid or philanthropic assistance). In such regions, governments are often the sole source for pharmaceuticals and have incentive to seek the cheapest sources for those purchases that they cannot avoid. Some see the development of a manufacturing capability within their countries as health policy mandates. The governments of developing countries — with their ongoing need for vaccines and reduced spending — are driving the spread of vaccine biomanufacturing worldwide.

Vaccines are among the least expensive and most cost-effective class of pharmaceutical products. They save lives, improve public health, and reduce healthcare expenditures. Consider the effectiveness of smallpox, polio, diphtheria, and other vaccines. Many vaccines — particularly those most widely used — are inexpensive, costing just dollars or even pennies per dose to manufacture. But some can be expensive (e.g., a three-dose course of human papillomavirus vaccine can cost nearly $400 in the United States). Many vaccines, particularly those produced and used in large volumes (often hundreds of millions of doses per year) such as universal infant and influenza vaccines, are being targeted by application of new bioprocessing technologies for faster and less expensive manufacture. That includes US biodefense programs supporting development of diverse expression systems for rapid vaccine manufacture.

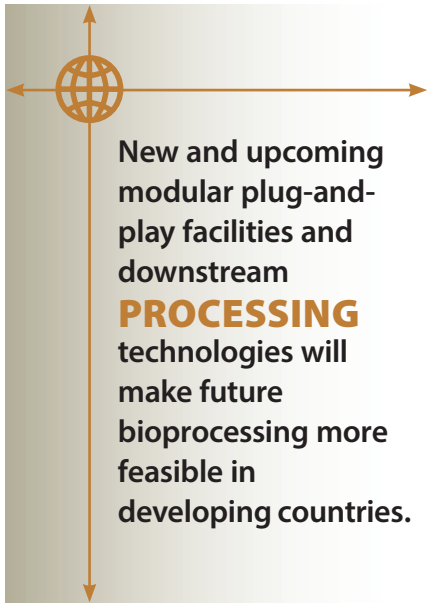
Nearly 300 vaccines are reported to be in development, including those in or for developed countries (4). Many are in development within developing countries and targeted for domestic use. The great majority of those vaccines in the pipeline involve recombinant technologies, with bioprocessing and equipment much the same as for conventional recombinant proteins and antibodies. Vaccines in development include next-generation, follow-on versions of current products as well as new vaccines for diseases without curret available vaccines. Such diseases primarily affect populations in developing countries (e.g., dengue and malaria). US and European foreign aid and philanthropies such as PATH (founded by the Bill and Melinda Gates Foundation) are providing funding for new vaccine development and vaccine manufacturing infrastructures in developing countries. For many developing countries, vaccines will be their first entry into biopharmaceutical manufacture.

Vaccines in Developing Countries

The great majority of current vaccines (both in terms of range of products and manufacturing scale) still originate from a few long-established companies. Current leading vaccine companies include Sanofi (the largest), Merck & Company, GlaxoSmithKline, and Pfizer (formerly Wyeth). Those companies are often the original developers of many current vaccines, and they dominate world vaccine markets. They also have been responsible for much effort to establish major vaccine facilities in emerging countries worldwide.

For example, most leading vaccine companies currently have major manufacturing facilities in China or have licensed or partnered with local manufacturers there. China effectively demanded domestic manufacture of vaccines to support its nearly a third of the world's population.

Many other countries worldwide are increasing domestic supply of vaccines for cost reasons and for the safety of their populations. This is becoming increasingly easier, inexpensive, and technologically feasible. More foreign governments and companies can be expected to bring vaccine manufacturing facilities online to support domestic needs and increasingly also for sales (cost recovery) internationally. And advancing bioprocessing technologies are making it simpler and more attainable to do so.



Technology Enabling Global Production

Technological advances in bioprocessing are coming together to make vaccine manufacture cheaper, faster, and simpler. Such advances include

- Single-use/disposable bioprocessing systems, providing easier operation
- Modular/transportable bioprocessing facilities
- Novel expression systems/ improved cell lines
- New purification technologies.

Those and other technologies are increasingly being adopted for commercial-scale manufacturing. Significant manufacturing improvements are now commonly being reported, particularly when the above advances are combined. In fact, a case can be made that vaccine (and other biopharmaceutical) manufacture may not be viable in many developing countries without combined adoption of multiple new(er) bioprocessing technologies, such as single-use systems with high-yield expression technologies and in prebuilt modular unit operations. New and upcoming modular plug-and-play facilities and downstream processing technologies will make future bioprocessing more feasible in developing countries.

Single-use bioprocessing systems will be a critical part of most new vaccine facilities in developing countries. Developing countries often cannot afford major investments in fixed, stainless-steel bioreactor-anchored facilities and lack the trained expert workforce required to operate and maintain them. By contrast, single-use systems with presterilized components allow flexibility and much lower capital investment and operator expertise. In addition, they need only basic utilities, with no steam and other complex piping requirements. They provide flexibility, enabling multiple product manufacturing in the same space, with expansion of capacity involving adding more systems in parallel.

Vaccines include products with a wide range of manufacturing methods, including live attenuated pathogens (viruses, bacteria, fungi); inactivated (killed) pathogens; and subunit vaccines composed of microorganism proteins/complexes, including recombinant and natural proteins. Newer vaccines in the development pipeline are mostly recombinant. There are as yet no synthetic vaccines, so conventional vaccine manufacture involves bioprocesses that are similar to those of many other biopharmaceutical products.

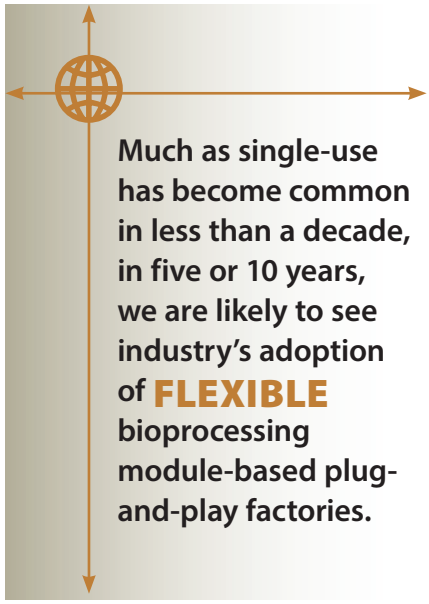
Impact of Single-Use Systems

In our *10th Annual Survey of Biopharmaceutical Manufacturing*, we document the increasing adoption of single-use (disposables) and other new technologies and their impact on biopharmaceutical manufacturers (5). This study shows that bioprocessing at precommercial scales (such as for clinical trials) is now thoroughly dominated by single-use systems use. That included 78% of those surveyed reporting current use of single-use bioreactors and 92% using single-use filter cartridges.

Single-use technology involves one-time use, then disposal, of bioprocessing equipment composed of plastics, thereby simplifying the establishment of new bioprocessing lines and facilities. Single-use technology allows flexible manufacturing, when and at the scale needed. It reduces costs and time to bring facilities online, especially because the equipment is presterilized. By contrast, stainless-steel bioreactor-anchored facilities require costly and complex infrastructure, including complex piping (such as for steam sterilization and cleaning). That can add weeks to batch turn-around time and cost $10s or even $100s of millions in construction costs and time for construction and validation.

For example, Novavax is developing a single-use-based facility for manufacturing 75 million doses/year of recombinant insect cell-cultured influenza vaccines reported to cost ~$40 million and $55/ft^2. By contrast, a comparable conventional egg-culture–based facility (which still has much lower costs than new cell-cultured influenza vaccines) could cost >$100 million and $140/ft^2.

Advancing single-use technologies and the increasing implementation of those systems at the precommercial R&D stage will broaden the adoption of single-use systems for commercial manufacturing (**6**). As a result, the market for single-use equipment at commercial scale is projected to grow 1,000% in five years to over $1.5 billion/year (**6**). Much of this involves capacity development in developing countries. With single-use systems already accepted by the bioprocess industry and regulatory authorities, and with proven cost and flexibility benefits, the only practical or feasible path for most new manufacturing facilities in developing countries will be installation of single-use systems. But some governments will still prefer to have domestic and fixed stainless-steel facilities — viewing them as tangible investments and accomplishments — compared with more "virtual," flexible, single-use facilities.



Much as single-use has become common in less than a decade, in five or 10 years, we are likely to see industry's adoption of FLEXIBLE bioprocessing module-based plug-and-play factories.

Major Impact of Modular Systems

Going modular is the next revolution in bioprocessing hardware, particularly in developing countries. This involves self-contained single-use equipment. Single-use equipment is housed within its own cleanrooms (portable prefabricated trailers or equipment sealed within dedicated isolator cabinets), and they increasingly are designed for plug-and-play simplicity. Bioprocess facilities that formerly required years for planning and construction can now be ordered and brought online in a matter of months. Much as single-use has become common in less than a decade, in five or 10 years, we are likely to see industry's adoption of flexible bioprocessing module-based plug-and-play factories. Vaccines will be a key product sector affected by this trend.

Modular technology will accelerate worldwide proliferation of vaccine manufacturing, including transfer to lesser-developed countries. Even easier than with single-use process manufacturing, modular systems allow plants to be identically cloned, potentially allowing replication of current good manufacturing practice (CGMP) manufacture in developing countries. Many countries outside the United States can be expected to increasingly demand local vaccine manufacture, particularly once modular facilities become commonplace; and a number of companies plan to actively pursue that market. Most established vaccine market leaders are currently adopting single-use technologies and planning to use modular systems for new vaccines in development. And their sales in an increasing number of countries are expected to require local manufacture. "Cloned" facilities are a way to achieve that while also maintaining product and CGMP standards worldwide.

Companies developing and already offering modular systems for vaccine manufacture include Pharmadule and G-Con (which is also working with Sartorius Stedim Biotech) and GE/Xcellerex. One example is Project GreenVax, a private–public consortium now constructing an influenza vaccine manufacturing facility (to be operated by G-Con, which is developing the modular units being used) in Texas. The project aims to manufacture recombinant tobacco-plant–expressed influenza vaccines, with "a projected final scale capacity of 100 million doses per month (1.2 billion doses per year)." Production costs are estimated at pennies per dose rather than dollars per dose for conventional egg culture manufacturing. That and other modular facilities are fully expected to be cloned by these and similar companies in developing countries.

Expression Systems Enable Cost-Effectiveness

Improved expression systems are also enabling developing countries to manufacture vaccines and other biopharmaceuticals. Higher yields allow manufacture of more product using the same equipment — or making the same amount of product at commensurately lower scale — with lower costs for equipment, facilities, and operation. The use of smaller-scale single-use equipment is largely enabled by and dependent on higher yields. And a wide range of new and improved expression systems, cell lines, and promoters are available (**7**).

New and improved versions of currently-predominating expression systems (Chinese hamster ovary cells,

yeast, and *Escherichia coli*) for recombinant protein expression are further making vaccine manufacture easier and more cost-effective and reducing the scale and investment required to manufacture products. The BioPlan annual survey shows a fairly consistent doubling of mammalian-cell protein expression and product yield about every five years, with yields at commercial scale now typically in the upper 2–3 g/L (bioreactor volume) range (**4**). New expression systems and continued incremental improvements coming online promise even higher yields and cost-effectiveness, with yields even as high as over 30 g/L being reported. With higher yields and using much the same manufacturing systems, the same amount of product can be manufactured at commensurately lower cost and often much faster. This has led US biodefense programs to support development of diverse expression systems for rapid vaccine manufacture.

Upcoming expression systems include plants (both laboratory- and field-grown) such as from iBio; transgenic animals, such as from rEVO Biologics (formerly Genzyme Transgenics); PER.C6 from DSM Biologics and other high-yield human cell lines; and bacteria other than the usual *E. coli*, such as *Pseudomonas fluorescens* from Pfenex and Corynex from Ajinomoto. An example of expression systems being implemented in developing countries was developed by Protalix BioTherapeutics, the developer and manufacturer of Elelyso (beta-glucocerebrosidase), the first plant-expressed (cultured carrot cells) biopharmaceutical. The company recently concluded an agreement with the Brazilian government to build a manufacturing plant in Brazil that will supply domestic needs and to fully transfer that facility and technology to Brazil after seven years.

Novel purification technologies are also in development, many of which are single-use. They are designed to require less space and infrastructure and be more plug-and-play. Such improvements are much needed for cases in which advanced upstream manufacturing cause capacity downstream constraints and problems. Purification has advanced nowhere near as rapidly as expressions systems and other upstream technologies. The BioPlan survey shows that many facilities are considering upgrading (adopting) new purification technologies. Those include 54% considering high-capacity chromatography resins; 44% single-use filters; 38% automated buffer dilution systems; and 35% single-use tangential flow filtration (TFF). Other advances being adapted for large-scale use include simulated moving bed (SMB) chromatography systems; single-use membrane adsorber filters; and cast-in-place "monolithic" chromatography media. Once pioneered in developed countries, these technologies can be expected to be rapidly adopted by developing countries.

New Technologies Are Key

Vaccines — many of which are required for all citizens and are an ongoing, recurring cost to health authorities — will continue to make advances as key products are adopted by manufacturers in developing countries. Those advances may come from developing countries' governments themselves or local proxies, joint ventures, or sponsors that subsidize or control local facilities manufacturing needed vaccines.

Use of the latest bioprocessing technologies is rapidly becoming a requirement in developing countries. Classic, fixed, commercial-scale, stainless steel-based facilities are simply too complex to construct or operate at rigorous CGMP quality levels in most developing countries.

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