PAT
IN-DEPTH FOCUS

PROCESS ANALYTICS EXPERIENCES IN BIOPHARMACEUTICAL MANUFACTURING

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Process analytics play a key role in achieving the objectives towards control strategy per Quality by Design in biopharmaceutical process development and manufacturing. It also has the potential to enhance continued process verification and make it closer to real-time. While the common perception of process analytics could be more focused on the analysers or on-line assays, our approach in this paper is more towards a systems thinking which is aligned with regulatory guidelines. Experiences in multivariate tools for data analysis, process analysers, process control and continuous improvement and knowledge management tools are summarised. The role of multivariate statistical process monitoring in understanding the sources of variation and detecting weak signals is articulated with industrial examples.

A lot of data are generated during biopharmaceutical process development and during the commercial process runs within the product lifecycle. Effective management of these data and development of insightful control strategies are critical for ensuring process consistency and verification. Recent guidelines[1,2] from the agencies also promote the lifecycle concept linking product and process development with the commercial manufacturing process. Availability of the right data is critical to achieve this endeavour while at the same time having systems in place to effectively monitor and control the manufacturing process to obtain repeatable and reproducible runs. Process analytical technology (PAT) with its tools helps address the monitoring and control aspirations summarised above. These tools are summarised in the FDA's PAT guidance[3] as follows:

1. Multivariate tools for design, data acquisition and analysis
2. Process analysers and process chemistry tools
3. Process and end-point monitoring and control tools
4. Continuous improvement and knowledge management tools

We will summarise our approach in data management, storing and organising the vast amounts of data generated during commercial manufacturing, multivariate tools and monitoring technologies, some of the experiences with process analysers and other process chemistry tools combined with process control opportunities and a short discussion on the knowledge management tool in capturing the learning during commercial campaigns via advanced multivariate monitoring systems.

Systems thinking

A combined use of aforementioned technologies in the PAT toolbox provides an effective means towards understanding and controlling process variability as well as supporting continued process verification and process improvements. Managing the data is crucial to enable the PAT toolbox. Therefore, establishing robust data capture and management systems has been our preliminary focus on the journey to PAT-enabled manufacturing.

Data Management Approach

It is imperative to capture process and product data coming from various source systems. Different levels of data/information hierarchy are described in ISA S95 model in five levels. Level 0 being the I/O level where analysers and sensors are, level 1 above it is where the distributed control systems (DCS) typically are found, level 2 has manufacturing execution

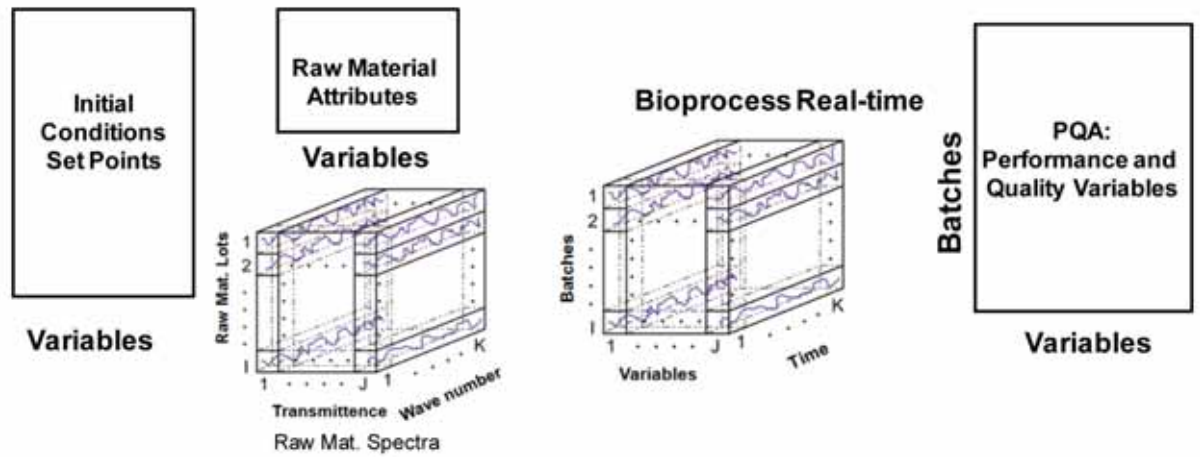

FIGURE 1 Multiple data blocks generated in a typical biopharmaceutical manufacturing process

systems (MES) and other batch data collection and Plant Data Historians, level 3 contains manufacturing operations and supervisory control systems and finally level 4 at the top has the business planning and logistics systems (such as enterprise planning). While we take data management as a lifecycle approach, even within the commercial manufacturing setting, there are various sources of data that require organisation and access in the cGMP environment. In a typical biopharmaceutical manufacturing process, these include operational parameters measured in real-time and made available in Plant Data Historians, and process parameters such as daily cell counts, metabolic indicators, other process set points and performance parameters captured in Manufacturing Execution Systems (MES) (e.g., electronic Batch Records) and lastly, product quality attributes that are usually tested by Quality Control and captured in Laboratory Information Management Systems (LIMS) (**Figure 1**). In addition, there might be raw materials-related data captured in other source databases. If we multiply these data sources with multiple plants, multiple molecules and multiple geographical areas, it quickly becomes a complicated task to manage and access the data.

We have successfully deployed a virtual Plant Data Warehouse (PDW) application that provides a single point of access to all of the data sources mentioned above for multiple plants and products[4]. PDW design is based on (but not limited to) the S88 physical and procedural models as outlined in **Figure 2**. For data management purposes, we have made some additions to the standard S88 models. For instance, an additional data element 'Product' has been incorporated into the Physical Model to accommodate multi-product facilities. We have also added an additional data element 'Step' to the Procedural Model for processes that require data management at a fine level of granularity.

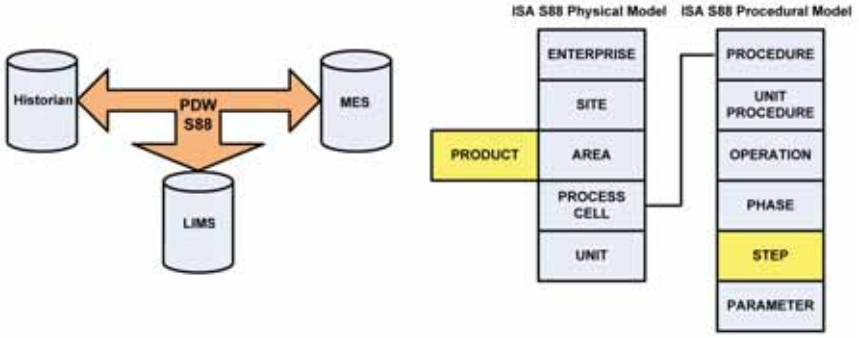

FIGURE 2 Manufacturing Data Management and use of S88 Model to tie the data together

This infrastructure is an important enabler for accessing historical data to support development of process models, generation of automated cGMP reports, and understanding of process variability via process monitoring. Furthermore, it supports the Lifecycle Data Management paradigm.

Process analysers and process chemistry tools for end-point monitoring and control

There have been a lot of process analysers and chemistry tools proposed for use in biopharmaceutical process development and manufacturing[5-7]. While we will not be covering

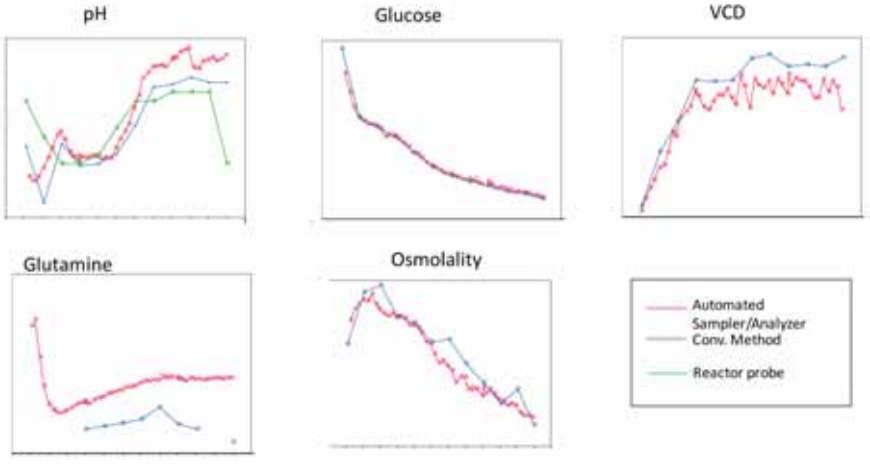

FIGURE 3 Comparison of sterile automated sampler and analyser and conventional offline methods for cell culture

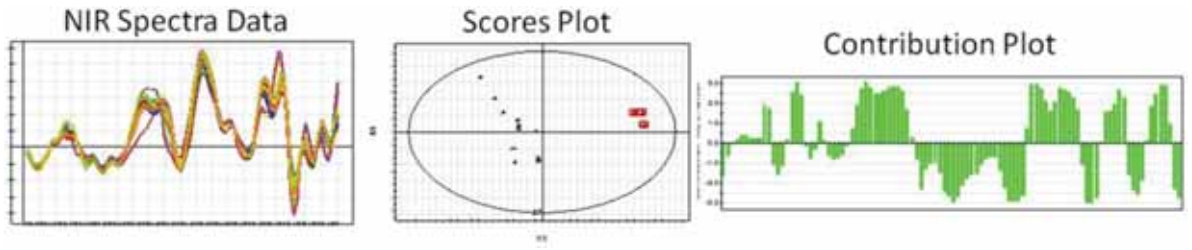

FIGURE 4 Left – Processed NIR spectra data of different lots of the same raw material; Middle – Scores plot of a number of different lots of the same raw material; Right – Contribution plot of NIR spectra showing the differences of the raw materials lots highlighted in Red in the Scores plot vs. the rest of the lots

the entire domain here, it has been our experience that when implemented right, these tools can help achieve variability reduction and end-point monitoring / control goals. They also include automated sterile sampling capabilities to reduce the time required to take offline samples. For instance, in a typical cell culture bioreactor setting, offline samples are taken once or twice a day during many days of cultivation to monitor process performance. This involves sterile manual sampling and quickly analysing the samples using bench-top equipment and finally entering the results into batch records (and into MES). This could be fully automated by using compact on-line analysers that are available today (direct method). Another approach has been to use spectral probes (NIR and Raman are the most common) and developing calibration models to relate the spectra to the measurement of interest (indirect method). In our bench-top testing, automated sterile sampling and analysis has worked despite the refinement opportunities. Note that it has given us more frequent sampling and automation capabilities that otherwise required additional effort to collect the same amount of data manually.

While direct methods have their advantages and wide area of applicability, the indirect method has also shown a lot of promise and industrial applications especially in cell culture monitoring such as NIR and Raman-based optical probes which give real-time information about the culture[8-10]. Successful application of an NIR-based cell density probe has shown variability reduction in cell culture performance[8]. Another process chemistry tool that has found effective utility is on-line HPLC for detecting aggregate levels in chromatography operation to determine the stop of the elution phase to achieve consistent purity levels[11]. This application is also considered as end-point monitoring and control.

Furthermore, advances in Raman and NIR-based spectroscopy have made these technologies available in handheld units. One application of this development involves non-intrusive rapid identification and variance monitoring of raw materials. As processes advance and become more controlled, the impact of raw material variation becomes more pronounced. Due to the excellent functional group identification capability of the Raman and NIR spectroscopy methods, rapid identification can be used throughout the process for fast and reliable identification of critical materials. The unique spectra information for the raw material can also be fed into multivariate analysis (MVA) to monitor lot-to-lot variability and gain greater understanding of the incoming

raw material. **Figure 4** on page 25 shows how a few outlier lots of a raw material can be easily identified by using NIR spectroscopy.

Real-Time Multivariate Statistical Process Monitoring (RT-MSPM) used as a continuous improvement and knowledge management tool

RT-MSPM provides an effective means of monitoring many variables measured at many different unit operations across many batches in real-time. This data-driven technology has allowed us to monitor process consistency and identify weak signals about equipment issues and process trends early. This helps with quick process troubleshooting and prevents process failures and losses in product yield. Some of the benefits of this technology include:

- Prevention of process and equipment issues (e.g., during the process and after the process is complete)
- Process performance improvement (e.g., yield increase and impurity reduction)
- Operational excellence (e.g., faster troubleshooting, purposeful presence on the manufacturing floor)
- Predictive monitoring

The methodology of RT-MSPM involves data mining of historical batches that are representative of desired process performance and inherent process variability. Once the rational subgroup of historical batches is determined and data mined for the parameters monitored, various multivariate models are developed after appropriate data pretreatment (i.e., outlier detection / removal, scaling, etc.) is applied. Multivariate charts are constructed to monitor new batches. Further details of multivariate modelling and monitoring algorithms can be found in our other technical publications[5-7,12].

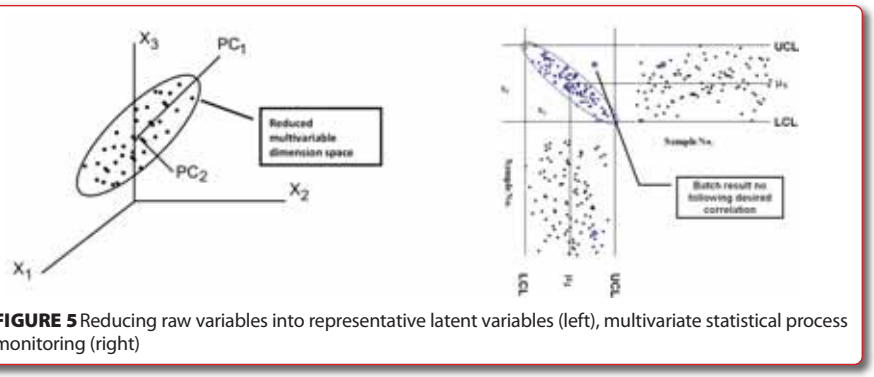

FIGURE 5 Reducing raw variables into representative latent variables (left), multivariate statistical process monitoring (right)

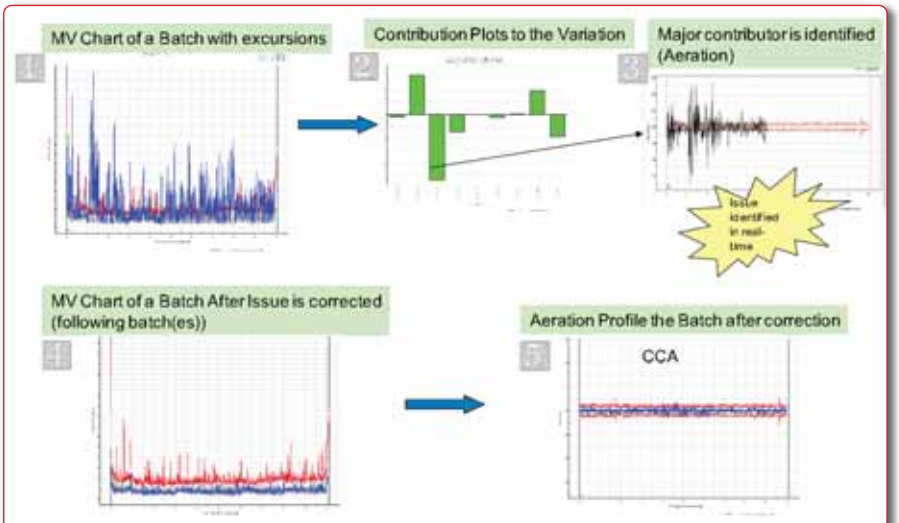

FIGURE 6 (1) A batch showed high degree of variation violating its multivariate chart limits, (2) Contribution plot indicates the major variability offender being aeration rate in the bioreactor, (3) inspecting on the raw data confirms the noisy signal, (4) after recalibrating the mass flow controller before the next batch, process noise is significantly reduced and (5) confirmed by inspecting the univariate chart

We also make use of high-level dashboards that are based on multivariate chart run rules to make issue identification effective for the manufacturing and other process support staff. Multivariate modelling technology helps by reducing large number of variables monitored to a few latent variables and establishes monitoring on those few latent variables. It also provides us with the ability to monitor the interactions between the variables.

An example of equipment issue identification and batch-to-batch correction is provided in **Figure 6**. This is a representative case of how this technology is used on the manufacturing floor by the operators. It helps by identifying variability in real-time and quickly brings the staff's attention to the issue. The staff can then triage it and take the appropriate action (by following cGMP procedures).

Another means of using the tool is to prevent issues by anticipating the potential process or equipment issues. In **Figure 7**, a case where a salient increase across batches in chromatography differential pressure is presented. The RT-MSPM tool allowed staff to quickly identify across-batch trend that was

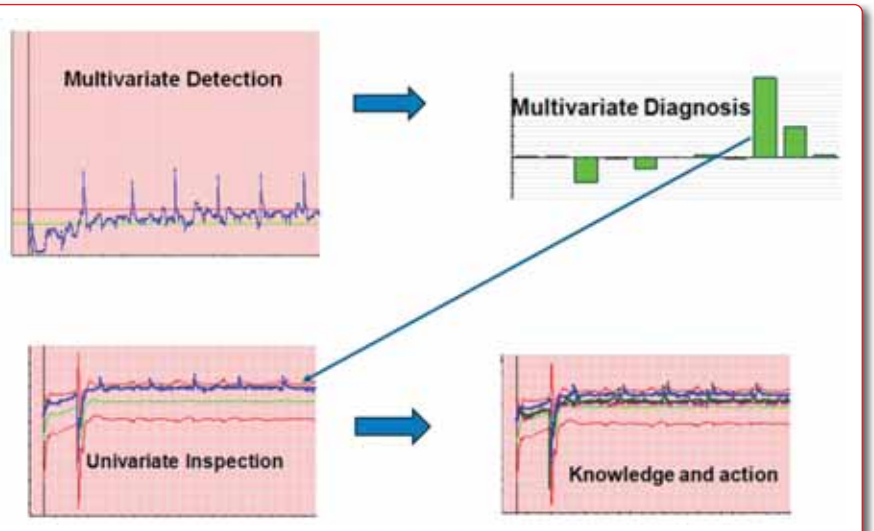

FIGURE 7 Real-time detection of a developing trend followed by across-batch analysis to anticipate a long-term trend to prevent further issues in chromatography column operations

developing (bottom right chart in **Figure 7**) and in a timely manner to allow re-packing of the column before any further issues. Trends identified via RT-MSPM can be weak signals which, if not checked, may develop into real process problems.

Figure 8 demonstrates this on a signal that was picked up using the multivariate charts and helped identify the potential root cause of a transmembrane control issue in an ultra-filtration skid which was addressed in real-time. Notice that the process TMP was well within the defined quality alarm ranges for subsequent duration of the batch after the issue was corrected in real time.

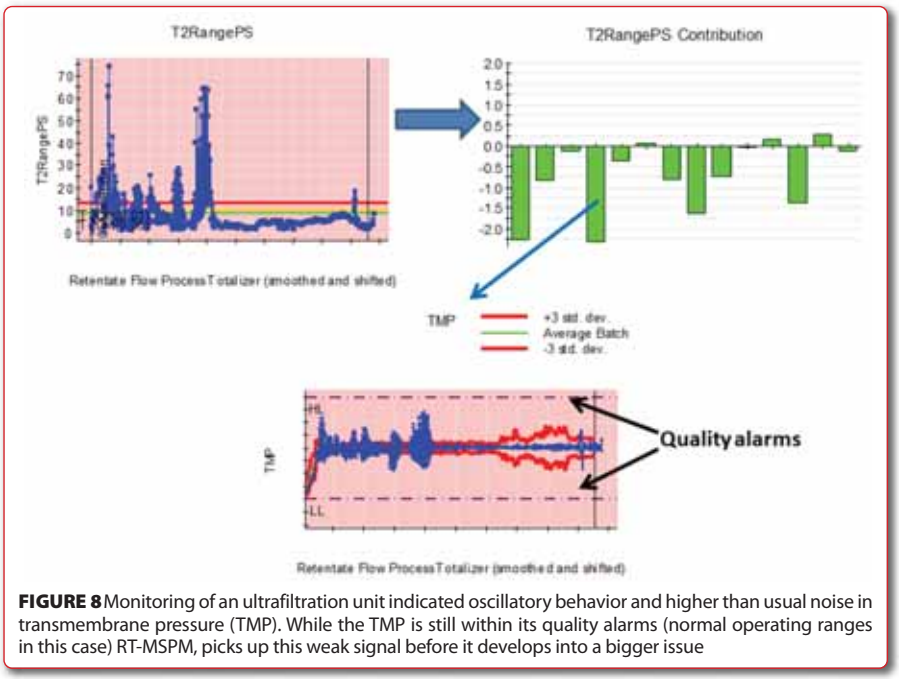

FIGURE 8 Monitoring of an ultrafiltration unit indicated oscillatory behavior and higher than usual noise in transmembrane pressure (TMP). While the TMP is still within its quality alarms (normal operating ranges in this case) RT-MSPM, picks up this weak signal before it develops into a bigger issue

Visual factory and knowledge management

RT-MSPM has also made use of Visual Factory principles to improve the end user interaction with the multivariate tool. To that end, it was not only made available in client desktops/laptops, it was also made available in large touchscreen displays and iPads that are accessible on the manufacturing floor (**Figure 9**, page 28).

In order to capture the key trends identified during production campaigns, we have also developed a knowledge management tool. Once the trends are reviewed and triaged (**Figure 10**, page 28), the key findings are entered and catalogued using this tool (**Figure 11**, page 28). It is designed in such a way to allow advanced filtering to quickly find related information. For instance, one can quickly get a summary of all the observed trends in a given unit operation, (including a given phase) across campaigns and products to get any systemic trends or cyclic events. This then allows systems analysis, review of the equipment and event process holistically. **Figures 10 and 11** (page 28) are representative views from the system, though it has more information and links to the relevant reports to enrich the knowledge management experience.

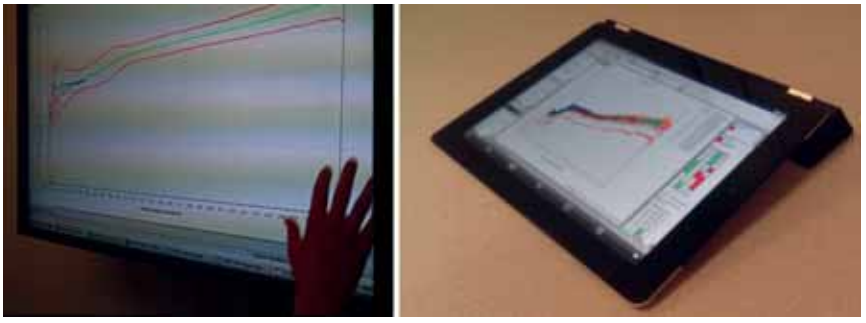

FIGURE 9 Large touchscreen display used on the manufacturing floor (on the left), iPad for remote and mobile access (on the right)

Predictive monitoring

Multivariate predictions using RT-MSPM adds value across many aspects of processing from efficiency to yield savings and improvements. Having early knowledge of process performance lends itself to opportunities such as shifting the process operating space within validated limits and scheduling future activities which are dependent on time, performance and / or batch size. In one example, a bioreactor is required to meet certain viable cell density (VCD) levels in order to inoculate the next step (Lower Acceptance Limit, LAL in this example). For improved process consistency and higher capability, statistical control limits are also followed (Lower Control Limit, LCL in the example). The top left chart in **Figure 12** (opposite) illustrates a low growth situation in a bioreactor where two final samples were required as the first was below the LCL. The culture was extended to meet the LCL resulting in additional time spent sampling, analysing and re-scheduling manufacturing activities. The top right chart in **Figure 11** demonstrates this new batch was in the lower growth quadrant of the batch level model on the second to last day. Evaluating the associated prediction (bottom bar chart in **Figure 11**) shows the Final VCD prediction was slightly lower than the LCL for the target culture duration. Efficiency and improved seeding can be achieved through accurate predictions of the final process performance by adjusting the schedule and process duration,

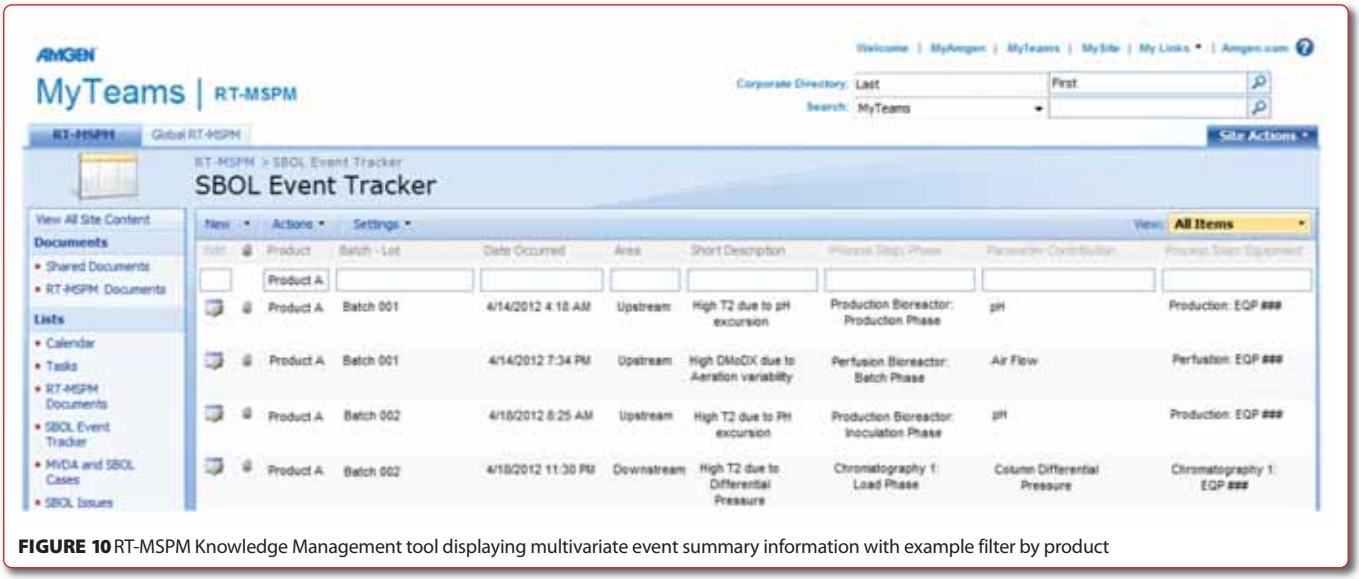

FIGURE 10 RT-MSPM Knowledge Management tool displaying multivariate event summary information with example filter by product

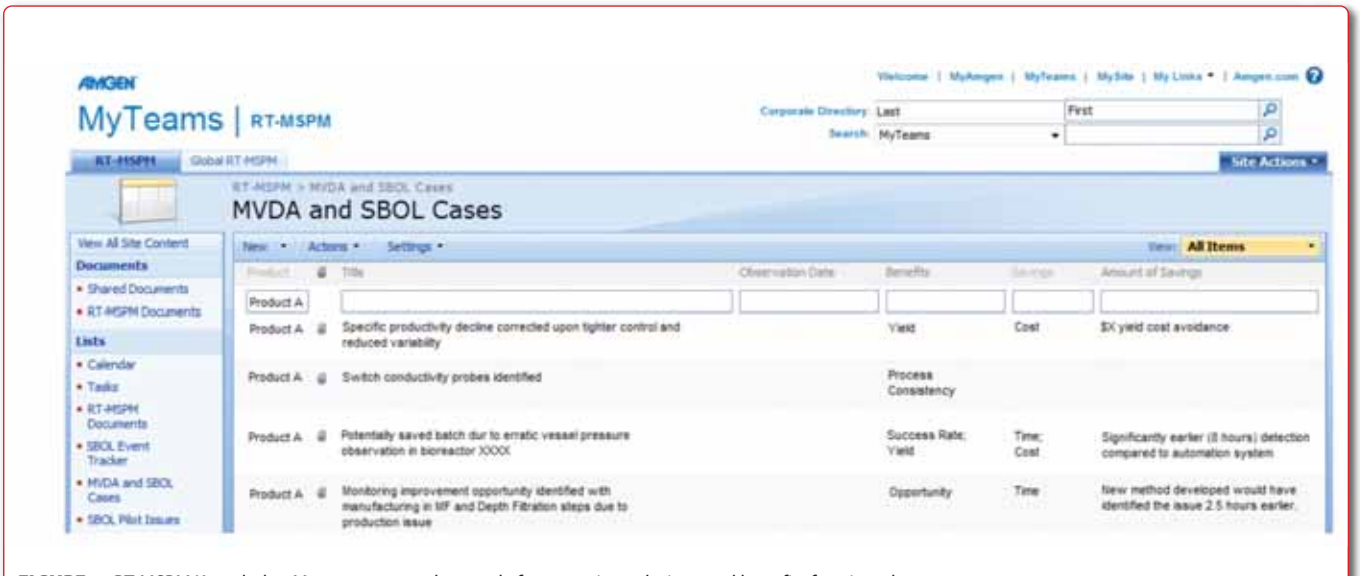

FIGURE 11 RT-MSPM Knowledge Management tool example for capturing solutions and benefits for triaged events

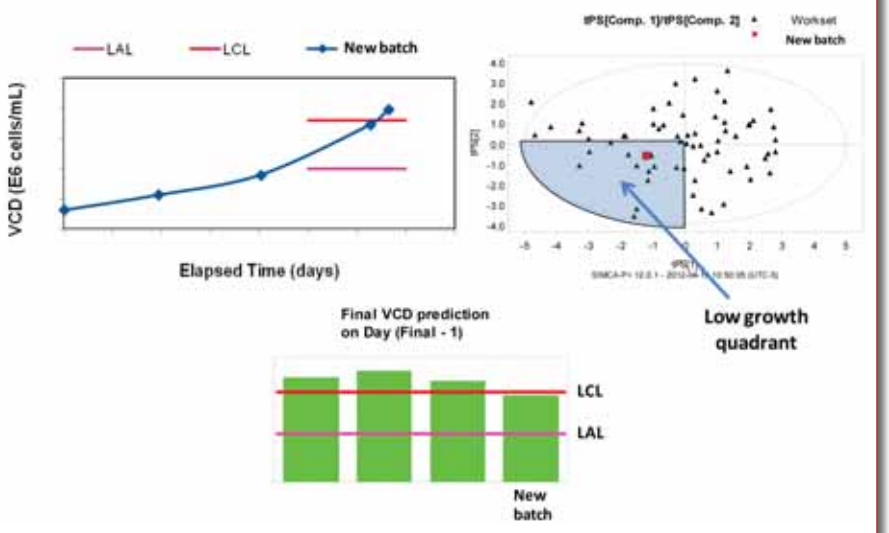

FIGURE 12 Sampling points for VCD across the culture duration (top left), batch level model for VCD prediction indicating a new batch associated with lower growth historical batches (top right), Final VCD predictions indicating a new batch will be lower than the LCL at the target duration (bottom)

within validated limits, before the process reaches the sampling time.

In summary, applying process analytics, RT-MSPM technology in particular, has provided notable business benefits. It also supports the paradigm shift towards enhanced continued process verification and establishes the necessary platform to bring process and product data together for process end-point monitoring and control. The use of RT-MSPM technology in preventing process or equipment issues has proven successful. While the initial use of RT-MSPM was limited to monitoring the consistency of the process and the equipment, with the advent of process analysers and process analytical chemistry tools the monitoring capability can evolve into a predictive end-point monitoring and control tool and used as a continued process verification tool.

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BIOGRAPHY

Cenk Undey, PhD, is a Director of Process Development in Process and Product Engineering at Amgen. He leads the Process and Systems Analysis group within the Network Process Engineering, advancing the Process Analytical Technologies, implementation of real-time multivariate statistical process monitoring and control for biopharmaceutical manufacturing processes as well as applications in formulation and finish, and in monitoring and control of variation in raw materials including primary containers and media. He has a BS, MS and PhD all in Chemical Engineering from Istanbul University in Turkey.

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Bryan Looze is a Senior Engineer in Process and Product Engineering at Amgen. His responsibilities in Process Analytical Technologies include implementing real-time multivariate statistical process monitoring, multivariate data analysis, and raw material information management and analysis. He obtained his BSc in Chemical Engineering from the University of Massachusetts and is currently pursuing his MS in Chemical Engineering from the Illinois Institute of Technology.

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BIOGRAPHY

Tony Wang is a Senior Engineer in the Process and Product Engineering at Amgen. He is responsible for multivariate data analysis and advancing Process Analytical Technologies, including Raman, NIR applications in raw materials and cell culture monitoring, as well as model predictive control. Previously he has worked in cell culture within Process Development and also as a process engineer within Facility Engineering at Amgen. Tony earned his BS degree in Chemical Engineering and MS degree in Biomedical Engineering both from University of Calgary in Canada. He is a licensed Professional Engineer within APEGGA.

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Rob Woolfenden is a Principal Engineer in Process and Product Engineering at Amgen. He currently leads data management efforts which reach across the global Amgen network. Prior to joining Amgen, he worked in plant management, quality assurance, engineering, and technical roles for Universal Foods (now Sensient Technologies), Fleischmann's Yeast and Seragen. Rob's background is in microbiology, and he holds an MS from the University of Hawaii.

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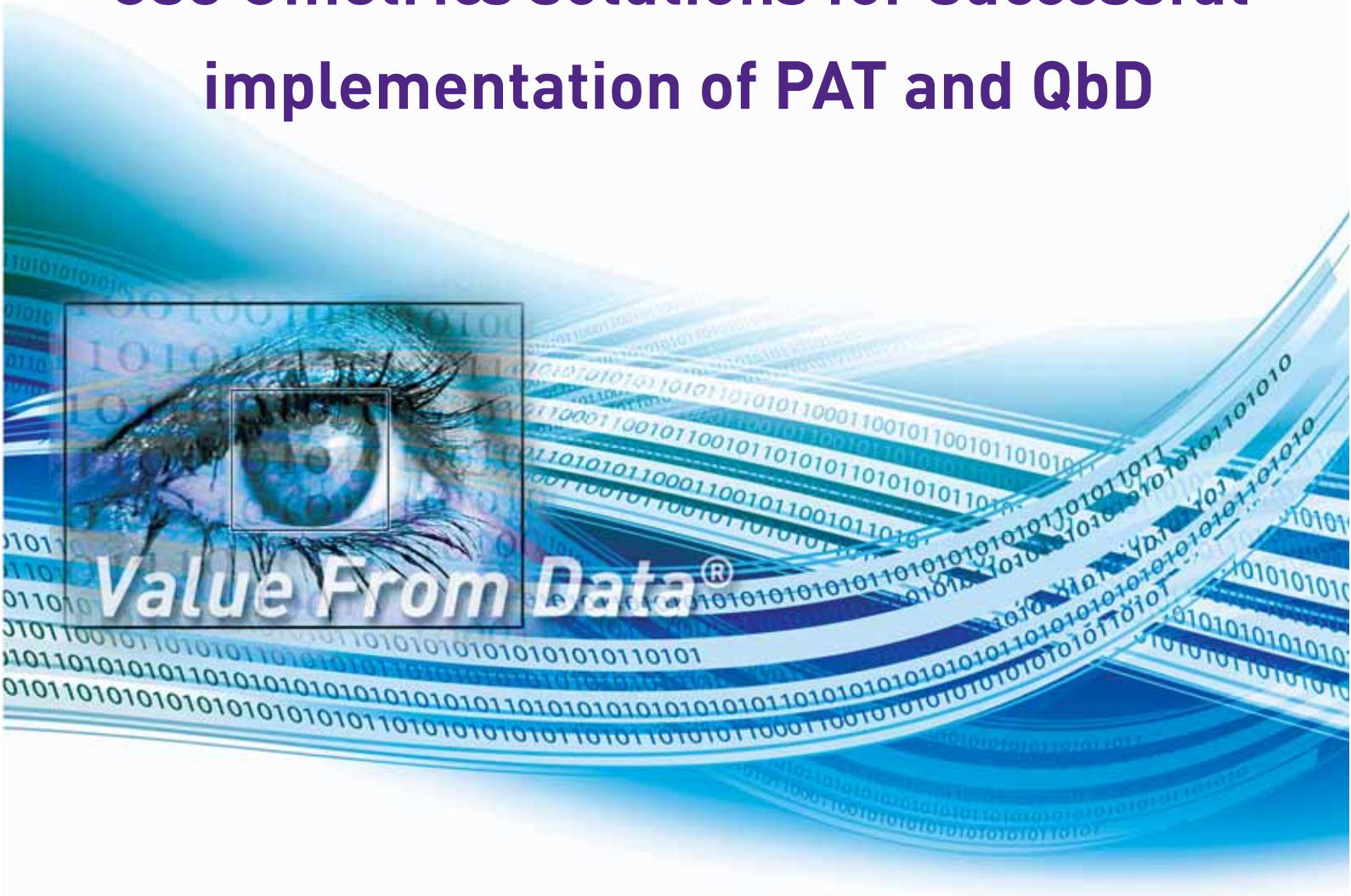

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