



# EVALUATION OF DISPOSABLE SINGLE USE MIXING SYSTEMS FOR BUFFER PREPARATION

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It has been shown that biopharmaceutical manufacturing operations constitute approximately 20 percent to 25 percent of a biopharmaceutical company's operating costs, and that the two most significant costs involved in manufacturing are facility time – which can account for up to 55 percent of the total manufacturing costs – and validation – which can account for 10 percent to 20 percent of the company's operating costs. In order to reduce these costs, one method that is being adopted by biopharmaceutical manufacturers is the move to disposable manufacturing systems. Many biopharmaceutical manufacturing facilities have implemented this approach and, as a result, have been successful in improving facility throughputs by reducing downtime between campaigns.

The key disposable components for a biopharmaceutical production suite include filtration, tubing, clamps, connectors, storage vessels, small volume (<500L) bioreactors and now mixing systems. Traditional stainless steel tank-based mixing systems that have thus far been widely used in biopharmaceutical

processing require considerable time and cost investment for cleaning, sterilization and validation following a mixing run. The newly developed disposable systems offer the advantage of eliminating many, and sometimes all, of these costly steps from a manufacturing environment. Moreover, there are many different types of closed disposable mixing systems to select from and they are based on a wide range of mixing technologies. These include levitated magnetic impellers, magnetically driven stir bars, motor-driven impellers, small motor-driven wands and bellows systems that force a perforated plate through the solutions to be mixed. One thing that most of these systems have in common is that they are all relatively efficient for liquid/liquid mixing on a small to medium scale, but most are less efficient for powder mixing applications, which usually require high torque to overcome the flow resistance of the powder and to allow uniform dispersion.

The company that is testing the efficacy and cost efficiency of these new disposal systems is a European-based company comprised of two divisions. The first division is the Therapeutics Group, which develops and manufactures recombinant vaccines, treatments for pulmonary edema, sepsis and chronic venous leg ulcers. This division also operates a fully compliant contract development and manufacturing operation. The second division is Specialty Diagnostics, which develops test systems against infectious diseases and neurodegradation, as well as genetic testing systems for a range of disorders.

The company is looking to implement a disposable system for buffer preparation to both reduce costs and increase flexibility. The choice of the correct system is important for the implementation of new projects and especially important for the contract manufacturing activities at the company. The goal is to identify those systems that can undertake both liquid/liquid and liquid/powder mixing in a scalable format

| Mixing of 50L 5M NaCl |             |                      | Mixing of 200L 5M NaCl |                      |
|-----------------------|-------------|----------------------|------------------------|----------------------|
| Time (Mins)           | Speed (rpm) | Conductivity (mS/cm) | Speed (rpm)            | Conductivity (mS/cm) |
| 5                     | 50          | 168                  | 80                     | 159                  |
| 10                    | 50          | 170                  | 80                     | 168                  |
| 15                    | 50          | 171                  | 80                     | 176                  |
| 20                    | 50          | 171                  | 80                     | 176                  |
| 25                    | 50          | 171                  | 80                     | 176                  |

Table 1: Showing mix data results at 50L and 200L mix volumes

SCALABILITY

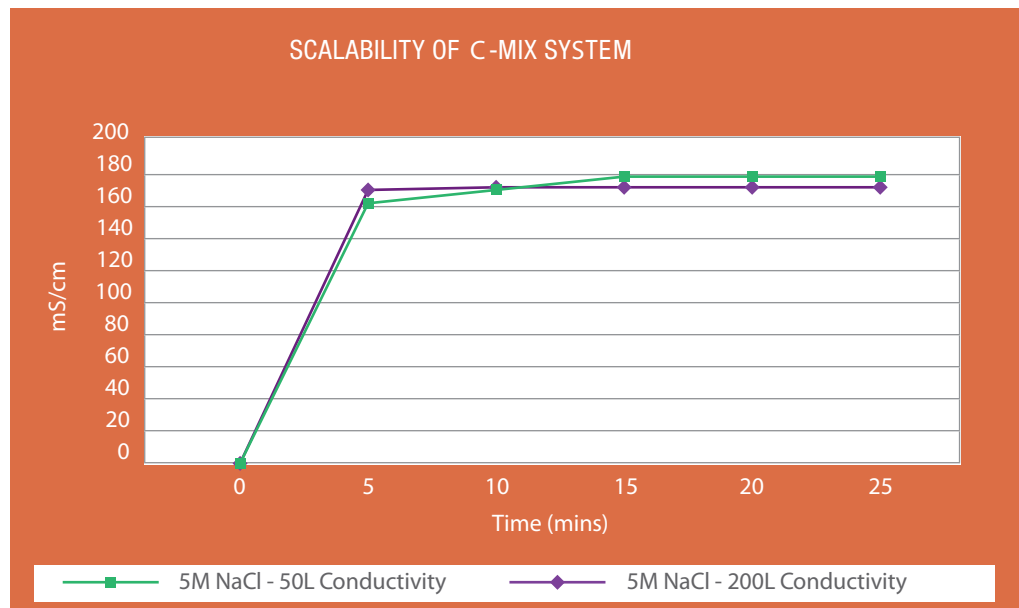


Figure 1: Showing scalability of mixing results from 50L to 200L



from R&D to full production volumes while offering cost-effective mixing. It was also important to find a system that was easy to integrate into the existing processes.

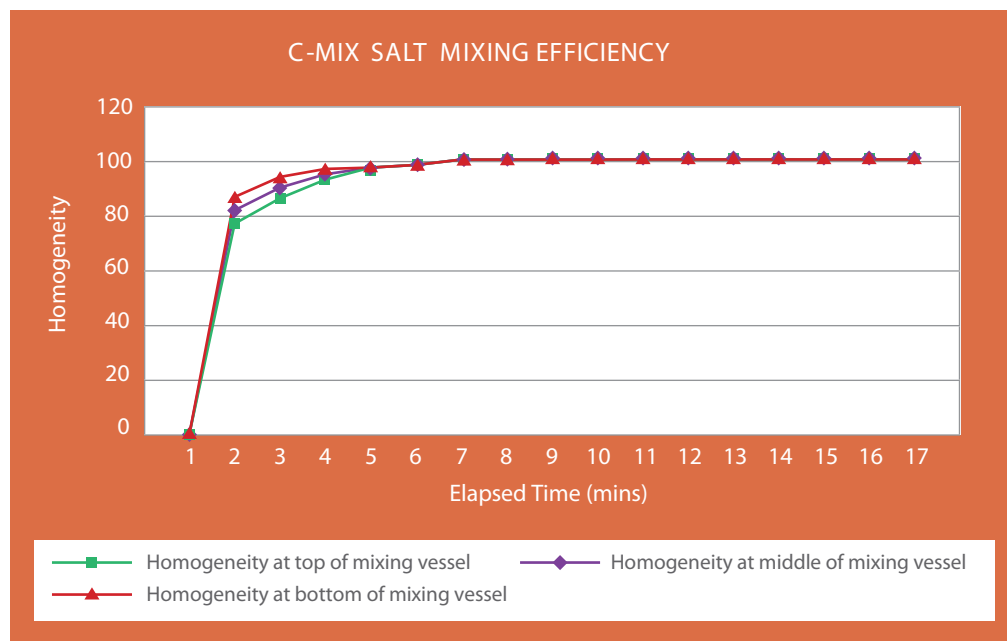
The Pad-Drive 1000, a non-invasive, single use, disposable, liquid/liquid and liquid/powder mixing system, has been evaluated as a system that potentially circumvents the time consuming and costly cleaning, sterilization and validation steps identified earlier. Initially, the mixing system was tested to determine its suitability and effectiveness for use for the scalable and homogeneous mixing of sodium chloride, which was used to simulate salt-based buffers. The description of the test protocols and results obtained in the evaluation are detailed in section one of this report.

## MATERIALS AND METHODS

In the evaluation, ATMI's Newmix Pad-Drive 1000 mixing system was used in combination with a 200L circular holding tank fitted with a standard 200L Newmix-C bag, a cylindrical, disposable mixing bag. The C-mix bag contained an integrated mixing paddle which was sheathed in the same film used in the construction of the bag to maintain a continuous and homogeneous fluid contact layer. The mix system consisted of an oscillating paddle within the liquid column. The effect of the paddle was similar in function

| Time (min) | Top  | Middle | Bottom |
|------------|------|--------|--------|
| 0          | 0    | 0      | 0      |
| 0.5        | 78   | 84     | 89     |
| 1          | 88   | 92     | 94     |
| 2          | 95   | 97     | 98     |
| 3          | 98   | 98     | 98.5   |
| 4          | 98.5 | 99     | 99.5   |
| 5          | 100  | 100    | 100    |
| 10         | 100  | 100    | 100    |
| 15         | 100  | 100    | 100    |

Table 2: Showing homogeneity results from top, middle and bottom of bag during mixing



HOMOGENEITY

Figure 2: Showing homogeneity of mixing through the liquid column of a C-Mix 200 bag

to a spoon when it is used to stir a cup of coffee. Thus, the paddle revolved within the liquid column, but the paddle and its associated shaft did not rotate around themselves. The Newmix Pad-Drive 1000 mixing system has a memory which is capable of saving up to 99 settings. Therefore, the direction of rotation, speed of rotation and frequency of change in direction of the paddle could be set and saved by the system operator within the system's memory. This design provided a highly efficient, low shear mixing system with no integrated bearings. This had the added benefit of placing no rotational torque on the film used in the construction of the bag, and therefore minimizing shear generated failure of the film paddle interface. Moreover, the coupling of the bag to the system was very easy to perform in that it required no tools and was capable of being operated by a single operator after minimal training.

#### SCALABILITY

Sodium chloride was weighted out to provide a final concentration, once dissolved and mixed, of 5M. Sodium chloride (292g/L NaCl) was dissolved and mixed at 50L (14.6Kg NaCl) and 200L (58.4 Kg NaCl) volumes by adding the appropriate amount of sodium chloride through the integral 6-inch diameter powder transfer port to the appropriate volume of water for injection (WFI) already filled into the disposable mixing bag. Then the mixing operation was started. Mixing efficiency was determined by measurement of conductivity at the top of the bag until the conductivity stabilized. Measurement was taken opposite the point of addition of the powder during the mixing operation.

#### HOMOGENEITY

The bag was pre-filled with 170L water for injection to which was added 60Kg of sodium chloride (353g/L) to give a final concentration, once fully dissolved, of 6.55M NaCl. Homogeneity of mixing was determined by measuring the conductivity of the solution at the top, middle and bottom of the bag until the conductivity measurements stabilized. A stable measurement was determined to represent 100 percent homogeneity.

The bags used in the scalability and homogeneity trials were standard 200L bags supplied by ATMI with integral filling lines, a 6-inch diameter powder transfer port, air supply line and bottom drain line.

#### SUMMARY

The Pad-Drive 1000 unit with the 200L Newmix-C tank and bag was found to be simple and easy to use, gave excellent mixing characteristics, and very quickly produced a homogeneous solution – determined by measurement of conductivity – of high molarity sodium chloride. The time taken to change the tank out of the system, install a new bag and proceed to the next mixing step was less than 10 minutes, required minimal training and supervision, and no tools – therefore making the system extremely user-friendly.

The tested system fulfilled all the parameters identified as important for the company prior to the start of the experimental trials. Several systems were subsequently purchased for use in both production and the contract manufacturing operation.



DEREK PENDLEBURY is Director of ATMI LifeSciences U.S., where he is responsible for the establishment of ATMI's North American Life Science division. He received his Ph.D. in marine microbiology before working at the University of London for five years, prior to moving into the industry. He has held sales and marketing management positions with Sartorius UK, North America, Biopath Inc., Pall Corporation and Charter Medical before joining ATMI. He has authored numerous papers on the use of disposable systems, and has presented at conferences in the U.S., Canada and Europe. He is a member of the marketing council of the Bioprocessing Systems Alliance.



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