Technical Brief

Characterization of Mobius® FlexReady Solution for Clarification

INTRODUCTION

The Mobius FlexReady Solution for Clarification is an easy-to-use system featuring an optimized single-use flowpath for primary and secondary clarification. The Mobius FlexReady Solution for Clarification features single-use Flexware® assemblies, innovative depth filtration devices and process-ready hardware systems to deliver optimal operational flexibility. The use of Millipore's innovative, high-performance Millistak+® Pod depth filters permits sizing for typical process volumes of up to 250 L of high cell density, low viability cell culture. The system is available in two sizes ideally suited for lab scale processing (CL-1 for up to 20 L) and pilot scale (CL-2 for up to 250 L) Pod devices.

In this study, three criteria for successful installation and operation of the system were tested: 1) ease of installation and product recovery, 2) scaling and filtration performance compared to historical values for Millistak+ HC Media in Pod format, and 3) system sterility following clarification. Data indicates ease of installation and operation, combined with scaling and filtration performance consistent with the published performance of Millipore's Millistak+ and Durapore® filters.

EXPERIMENTAL DESIGN

Hardware Configurations

The Mobius FlexReady CL-1 and CL-2 hardware systems were configured as shown in Figures 1 and 2, respectively.

Consumables

For the CL-1 trials, Millistak+ HC A1HC 0.11 m^2 depth filters (2 x 0.054 m^2 in parallel) followed by a Durapore 0.22 µm sterile filter (Catalogue No. KHGEG003FH3) were used in a Flexware CL-1 flowpath. For the CL-2 trials, a single Millistak+ HC A1HC 1.1 m^2 depth filter was followed by a Durapore 0.22 µm sterile filter (Catalogue No. KHGEG05TH1). The A1HC media was chosen due to its optimal media design for post-TFF clarification of low-solid fluids.

Feed Stream and Liquids

Each experiment used 30 g/L Kraft® Dairy whey solution (Laktodou™ Lot No. 861758 from Lacto Serum, France). WFI was used for rinsing and as buffer. Other standard cleaning liquids were used for proper cleaning and decontamination of equipment (where applicable).

Experimental Design

For the application test, a whey protein solution was used as a model feed stream. This solution was filtered through Millistak+ A1HC filters followed by a sterile Durapore 0.22 µm filter. The filtered product was analyzed for bioburden removal to test system sterility, and protein breakthrough and maximum loading to demonstrate scalability from bench trials across both the CL-1 and CL-2 platforms. Each configuration (CL-1 and CL-2) was run in duplicate. A representative data set is presented for each system.

Scale-up was measured using terminal pressure and turbidity. Turbidity was significantly reduced from ~1100 NTU to 5.5 NTU and 1.36 NTU for the 0.11 m^2 and 1.1 m^2 devices, respectively. Sizing to filter 500 L in 2 hours reveals that the Millistak+ A1HC filters are scalable. For the 1.1 m^2 device, only 7.7 m^2 is required to filter the 500 L in 2 hours with a safety factor of 1.86, while only 9.9 m^2 is required to filter 500 L in 2 hours for the 0.11 m^2 device (Table 1). Data also indicates no pressure drop for the 0.22 µm Durapore filter, suggesting that any sizing can be used for sterile filtration.

RESULTS

Ease of Installation and Product Recovery

The first set of criteria for successful operation was rapid installation and ease of product recovery. In these trials, installation of all Flexware and filters required only 25 minutes. The installation allows an easy recovery of the product by air blow down. After the clarification, an air blow down at 10 psi allowed recovery of 5.7 kg on the 1.1 m^2 Millistak+ Pod and 0.78 kg on the 0.11 m^2 Millistak+ Pod.

Scaling and Filtration Performance

Filter capacity of each filter was determined using the P_{max} method. The P_{max} method uses the pressure endpoint in a constant flow test to size filters, with the endpoint determined as the point of pressure loss. For this experiment, 500 L of 30 g/L whey with a starting turbidity of ~1100 NTU was filtered for 2 hours with 20 psi pressure endpoint (Table 1). For both the 0.11 m^2 and 1.1 m^2 devices, there was no filter plugging, indicating appropriate capacity for each filter.

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Millistak+ Pod disposable depth filters are ideal for primary and secondary clarification in lab, pilot and process-scale applications

Table 1. **P_{max} results for filter with 500 L solution in 2 hours with 20 psi pressure endpoint (starting turbidity around 1100 NTU).**

	Filter 1	
	A1HC	A1HC
Area of tested devices (m²)	0.11	1.1
Trial flowrate LMH	172	145
Trial volume (l)	7.130	80
Initial pressure (psi)	3.2	1.2
Final pressure (psi)	26	12
Trial loading (L/m²)	66	73
Final turbidity NTU	5.5	1.36
Proposed configuration (m²)	9 x 1.1	7 x 1.1
Area of configuration (m²)	9.9	7.7
Loading in process (L/m²)	51	65
Estimated safety factor	1.57	1.87

Figure 1. **P&ID for Mobius FlexReady CL-1 Small Scale (2 x 0.054 m²)**

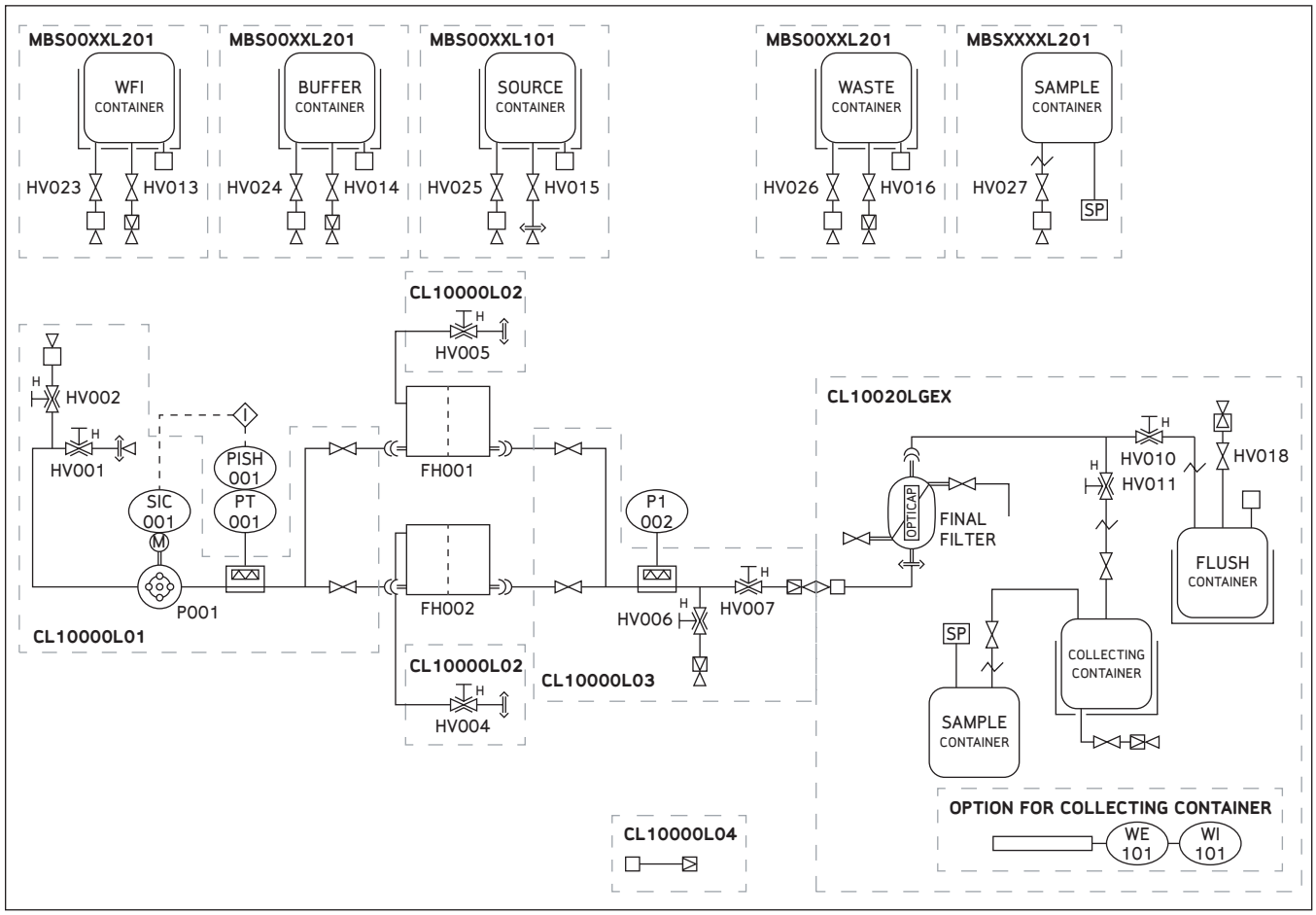


Figure 2. **P&ID for Mobius FlexReady CL-2 Large Scale Pod Holder (up to 2.2 m²)**

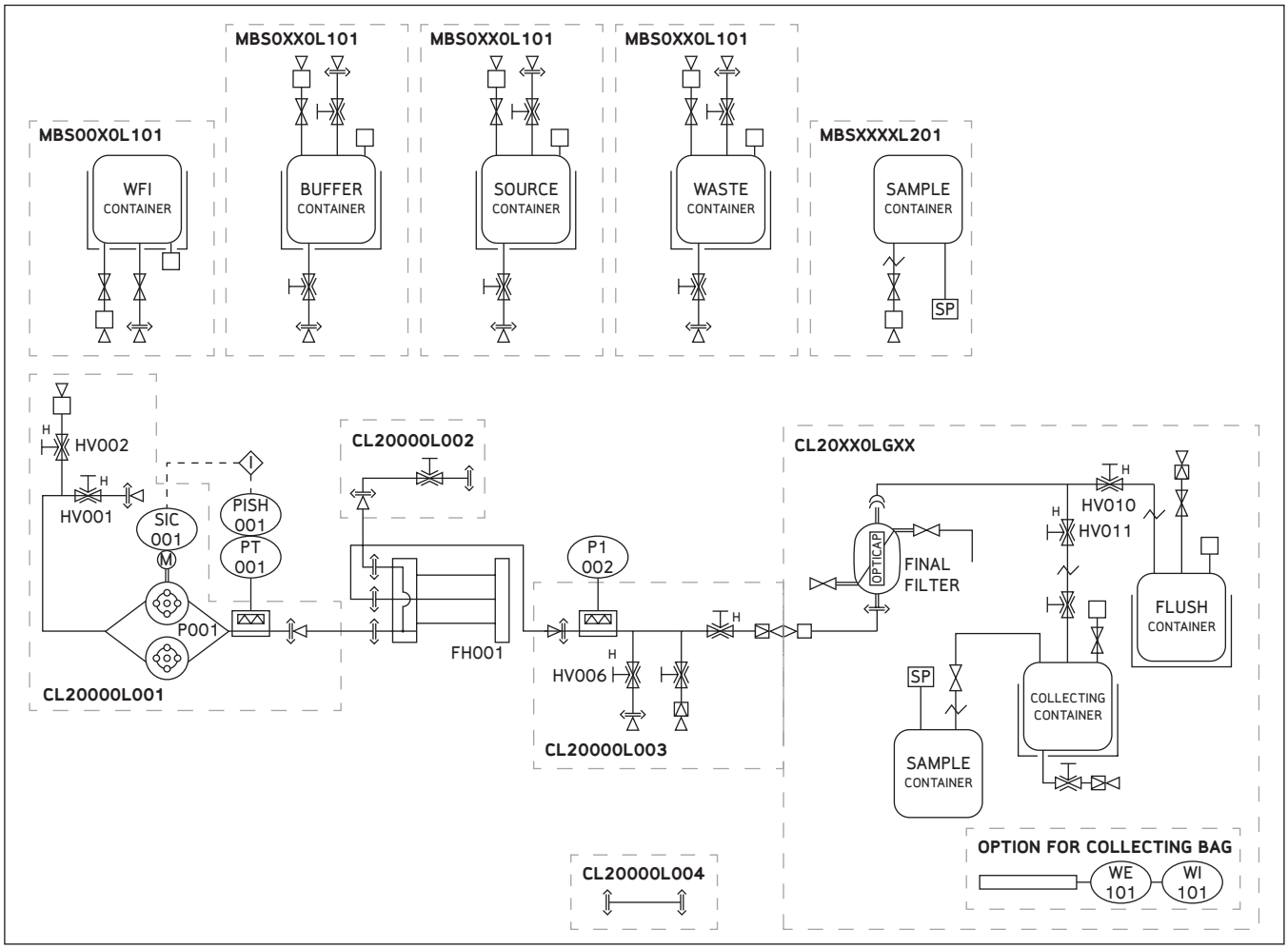
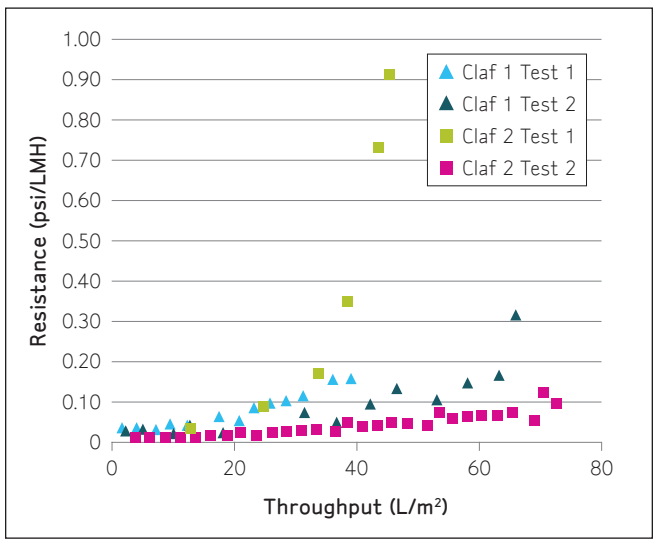


Figure 3. **Sterile filter capacity of a 0.22 µm Durapore filter following filtration by Millistak+ A1HC prefilter**



Sterility of the System

The sterility of the product contained in the final container was tested over 14 days. Sterility testing indicated no detectable contamination at the end of the clarification run (data not shown).

DISCUSSION

The objective of this study was to characterize the performance of the Mobius FlexReady Solution for Clarification. The use of Flexware flowpath assemblies for the CL-1 and CL-2 systems enable users to quickly and easily perform all steps necessary for a clarification unit operation. There are minimum field connections required to be made by the operator, reducing the risk of contamination to the sterile part of the system.

Data presented here indicate that the solution is ideal for performing primary and secondary clarification with comparable results to the published performance of Millipore's Millistak+ Pod single-use depth filters. As the P_{max} data confirms, the systems are scalable across the CL-1 and CL-2 platforms, in agreement with prior data reported in "Using Millistak+ HC Filter for Mammalian Cell Culture Clarification" (Millipore Application Guide, Lit. No. AN1100EN00). Although only the Millistak+ HC filters were tested in these experiments, similar results would be expected for the Millistak+ DE and CE media, enabling clarification in a variety of applications, including cell culture, yeast and *E. coli* lysates post-centrifuge, *E. coli* refolds, media, vaccines, plasma proteins and sera.

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