

# MFI Silane-Coated Flow Cell Cleaning and Care

## Introduction

The use of Micro-Flow Imaging (MFI) for subvisible particle detection and characterization relies on precise and accurate imaging of a solution as it passes through the flow cell. However, proteins and other particles could potentially adhere to the interfaces and surfaces that make up the flow cell; a sort of sticking that inevitably weakens imaging precision and, subsequently, data quality. As such, ProteinSimple offers flow cells that have been treated with a hydrophobic silane coating to prevent particle adhesion. Proper care and cleaning are essential to minimize degradation of this coating. Generally speaking, flow cells can be sufficiently cleaned by a simple flush with filtered (0.22  $\mu\text{m}$ ) water. However, cleaning with a commercial cleaning solution may at times be required to remove residual sample (see the MFI 5100/5200 [Quick Reference Guide](#) for cleaning instructions). In this technical note, we've examined the effect of several popular cleaning solutions for silane coating used in the MFI flow cell, with the goal of making safe-to-use recommendations for users, both in terms of cleaning solution type and exposure time.



## Image-Based Particle Differentiation with MFI

Obviating contamination and immunogenicity concerns during biopharmaceutical formulation and testing is an essential step to on-track regulatory approval. To ensure the product's safety and efficacy, you'll need to accurately assess the product for the presence of subvisible particles and protein aggregates. And as an image-based approach, MFI gives you the advantage over compendial techniques for this type of analysis. MFI can discriminate protein aggregates from micro-droplets of silicon, air bubbles and other contaminants commonly found in complex biopharmaceutical samples. In the end, you'll have more confidence in the important decisions you make during development and manufacturing.

## Putting Cleaning Solutions to the Test

Glass slides used in the assembly of MFI flow cells were treated and coated with silane according to the ProteinSimple manufacturing process. Prior to testing both the harshness and effectiveness of a cleaning solution, a 5  $\mu\text{L}$  droplet of water was deposited onto each slide ( $t = 0$ ), visually inspected for droplet cohesion – indicative of surface hydrophobicity – and recorded via a series of images (**Figure 1**).



**FIGURE 1.** A 5  $\mu\text{L}$  water bead on the glass surface of a flow cell at  $t = 0$ , showing the intact silane coating.

Next, glass slides were immersed in various cleaning solutions diluted in 0.22  $\mu\text{m}$ -filtered water at the concentrations listed in **Table 1**. Slides were exposed for increasing periods of time, ranging from 30 minutes to 1 week (168 hours). Following the incubation period, slides were removed, rinsed with ultrapure water, dried and re-tested for water beading using 5  $\mu\text{L}$  of water.

CLEANING SOLUTIONS TESTED		VENDOR
Hellmanex™ III	1%	Hellma GmbH & Co. KG
	2%	
Liquinox®	1%	Alconox, Inc.
	2%	
	5%	
PCC-54™	2%	Thermo Fisher Scientific
	5%	
Micro-90®	2%	International Products Corp.
	4%	
	6%	
Contrad® 70	5%	Decon Labs, Inc.
	10%	

**TABLE 1.** Five popular cleaning solutions were tested in this technical note.

If a silane-coated glass slide exposed to a cleaning solution failed the water-beading test at any of the time points tried, longer time points were not considered. Water-beading test failure was defined as a loss of droplet cohesion, suggesting degradation of the silane surface coating and thus its hydrophobicity attribute (**Figure 2**).

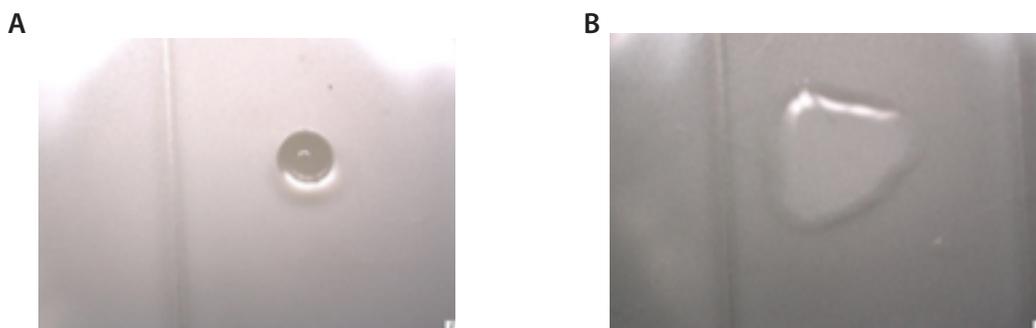


**FIGURE 2.** Water bead test failure, indicating a degraded silane coating.

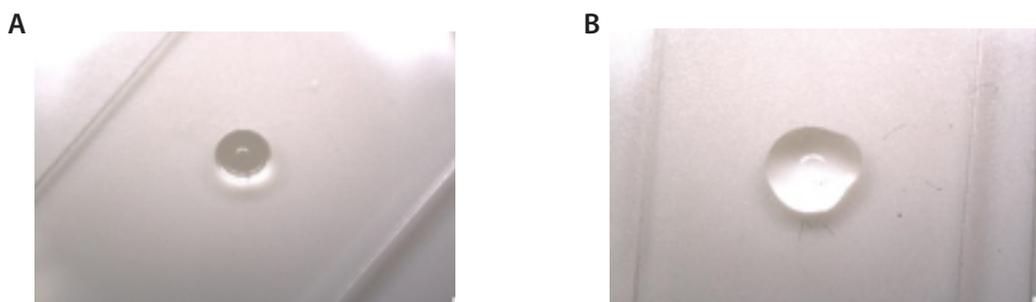
## Working Toward a Recommendation

We've included representative images of the visual inspection of surface stability and silane coating hydrophobicity for three of the five cleaning solutions tested, namely Contrad 70 (**Figure 3**), Hellmanex III (**Figure 4**) and Liquinox (**Figure 5**). **Figure 6** is a comprehensive summary of the results obtained from all concentrations of cleaning solutions included.

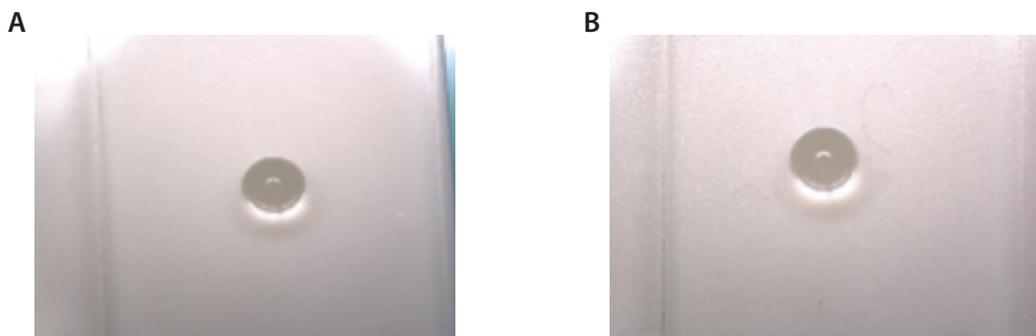
- **Contrad 70:** 5% and 10% solutions were tested, of which the 5% solution resulted in degradation of the silane coating within one day of exposure to the cleaning solution (**Figure 3, B** versus **A**). This was deemed too harsh of a cleaning solution, as our goal was to identify cleaning solutions that maintain the hydrophobicity of the silane coating for at least a week's time. Therefore, a longer time point for Contrad 70 was not assessed.
- **Hellmanex III:** 1% and 2% solutions were evaluated. Although not as severe as Contrad 70, this cleaning solution did compromise the flow-cell silane coating after a one-week immersion period, as observed by a failure of the water-beading test (**Figure 4, B** versus **A**).
- **Liquinox:** 1%, 2% and 5% solutions all maintained the integrity of the silane coating after even one week of exposure (**Figure 6**), making this cleaning solution a good recommendation for cleaning and flow cell storage. **Figure 5** is a typical visual water-bead test result observed using the 2% solution.



**FIGURE 3.** Water bead test on flow-cell glass after a 30-minute exposure to 5% Contrad 70, (A) and after a 24-hour exposure (B).



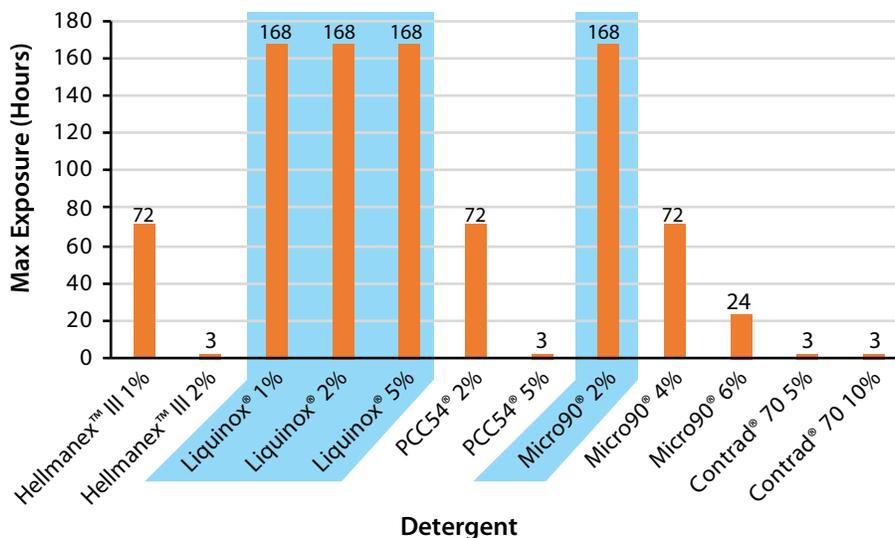
**FIGURE 4.** Water bead test on MFI flow-cell glass after a 30-minute exposure to 1% Hellmanex III (A) and after one week (B), showing some degradation of the water bead and silane coating.



**FIGURE 5.** Water bead test on MFI flow-cell glass after a 30-minute exposure to 2% Liquinox (A) and after one week (B), showing intact water bead and silane coating.

All cleaning solutions tested, their respective concentrations, and time to surface degradation are summarized in **Figure 6**. From these data, we can conclude that Contrad 70 (5% and 10% solution) is the most stringent of the cleaning solutions included and should not be used for cleaning and care of MFI flow cells. Both 1% Hellmanex III and 2% PCC-54 had the same time to test failure (72 hours), as did 2% Hellmanex III and 5% PCC-54 (3 hours) and thus cannot be recommended for long-term storage of flow cells. Liquinox (1%, 2% and

5%) performed as the best in this study, maintaining the surface integrity for as long as 168 hours (one week) at all concentrations tested, and is therefore supported for use as a cleaning and storage solution when used as  $\leq 5\%$  solutions. Additionally, a 2% solution of Micro-90 performed just as well as Liquinox (1%, 2%, and 5%), and is safe to use. However, Micro-90 solutions  $> 2\%$  are not recommended, as 4% and 6% failed the water-bead test, with a compromised surface observed at 72 hours and 24 hours, respectively.



**FIGURE 6.** Silane coating stability. Cleaning solutions tested and the respective concentrations (x-axis) are graphed as a measure of time in hours to surface degradation (y-axis). Recommended solutions are outlined in a blue box.

## Conclusions and Recommendations

Sometimes you need more than filtered water to remove residuals from the MFI flow cell completely. But if you just reach for any cleaning solution off the shelf, you'll also risk damaging the silane coating that's there to prevent particle adhesion, preserve imaging precision and maintain the level of data quality you rely on MFI for. So, after testing some of the more commonly used cleaning solutions we present our recommendations in **Table 2**. Do note that this is not an exhaustive list. When considering cleaning solutions for use, those with higher pH values will be more likely to damage or degrade the flow cell silane coating, even at low concentrations, and should be avoided. For a quick rundown of cleaning guidelines using the recommended cleaning solutions, refer to the [MFI Quick Reference Guide for Flow Cell Cleaning](#).

FLOW CELL PRODUCT NUMBER	CLEANING SOLUTION
4002-002-001 4002-003-001	<b>Recommended</b> <ul style="list-style-type: none"> <li>Liquinox® (Alconox, Inc.) ≤5%</li> <li>Micro-90® (International Products) ≤2%</li> </ul>
	<b>Not Recommended</b> <ul style="list-style-type: none"> <li>PCC-54™</li> <li>Hellmanex™ III</li> <li>Contrad® 70</li> </ul>

**TABLE 2.** Cleaning solutions recommended for cleaning and care of MFI silane-coated flow cells.