User Manual

Micro-Flow Imaging™ (MFI)

MFI 5100/5200 Flow Microscope

October 2011

Part Number 4008-003-001

proteinsimple™

proteinsimple.com

Table of Contents

List of Figures

List of Tables

1.0 Introduction

This Manual describes the installation, operation and maintenance of the MFI 5000 series instruments. MFI 5000 series instruments use operating software called MFI View System Software (MVSS) which has a separate User Manual.

We make every attempt to ensure the accuracy and utility of the information in this User Manual but recognize that errors may occur. We also appreciate feedback from our customers on the design and use of the Manual. Please send any comments or questions to **mfi@proteinsimple.com**.

2.0 Safety Precautions

2.1 Cautionary Symbols and Conventions

The caution symbol is used throughout this manual to identify conditions or processes that may cause harm to individuals and/or equipment or result in inaccurate results.

2.2 Safety Warnings

The warning label shown in Figure 2.1 is located on the instrument.

Figure 2.1: Instrument Warning Label

RISK GROUP 2 LED
WARNING Possible hazardous optical radiation emitted from this product. Do not look at operating LED. Eye injury may result.

There are no user-serviceable parts inside the instrument. The removal of the main instrument shell can expose an individual to hazardous voltages. Please contact a qualified ProteinSimple technician for servicing.

The instrument uses **SINGLE POLE FUSING**. Always replace fuses with fuses of same type and rating. IEC60127-2-3 "Time Delay" fuse rated 3A, 250V is used on DC input of the instrument.

This product is pluggable equipment type A, Class 1. Protection against electric shock is achieved by using basic insulation and a connection to the protective earth conductor in the building wiring. It is intended for connection to the building wiring via a non-industrial plug and socket-outlet. As such, **the instrument must be connected to an earthed mains socket outlet for protection purposes**. Always ensure the ground on the outlet is of sufficiently low impedance. Refer to your local electrical code.

Only use the Power Supply and Line Cord supplied with the equipment or an alternative approved by ProteinSimple which satisfies local applicable electrical code(s).

Equipment may only be operated at the specified line voltage, frequency and environmental conditions. Refer to the Electrical Specifications and Environmental Specifications sections in this manual.

CAUTION: The output of the LED should not be viewed with certain optical instruments (for example, eye loupes, magnifiers and microscopes) within a distance of 100 mm may pose an eye hazard.

All peripheral components used in conjunction with this equipment must adhere to local safety regulations. Please consult local regulations and authorities for further details.

The instrument module weighs approximately 12 Kg. Handle with care. If required, lift the instrument using proper equipment and trained personnel.

Do not create chemical reactions in, or in the vicinity of, the instrument.

Equipment must be used in the manner specified in this User Manual. Operation of the equipment in a manner not specified in this User Manual could result in injury.

2.3 Electrical Supply

During setup and operation, fluids are in close proximity to electrical components. To avoid equipment damage, the Flow Cell and plumbing module tubing should be primed and thoroughly checked for leaks prior to installation into the instrument module. Although the instrument has been designed to withstand moderate exposure to moisture, users should take sufficient precautionary measures to prevent liquids contacting electrical components.

The MFI 5100/5200 requires three (3) 100-240V-AC 50-60Hz connections not exceeding 15A-AC total current. Do not exceed the rating of the supply used to power the instrument. See Electrical Specifications. It is recommended that the instrument be connected to an uninterruptible power supply (UPS) to prevent unintended shutdown in the case of a power failure.

3.0 Instrument Installation

The MFI 5100/5200 instruments can be supplied in different configurations depending on user requirements. A typical configuration is shown in Figure 3.1 (Control System and cables not shown). Unpack and inspect all components for possible shipping damage and notify ProteinSimple immediately of any damage. Retain packaging materials for future use.

Figure 3.1: Instrument Configuration

Additional Items Required (not provided)

- 100-240VAC 50-60Hz electrical service (3 outlets,15A-AC circuit total, with surge protection)
- Fluid volume measuring device for calibrating pump (scale or graduated cylinder)
- Waste vessels/beakers
- PCC-54™ cleaning solution (recommended glassware detergent)
- Pipetter (two-step operation with separate ejection and blow-out)

The instrument requires 100-240VAC, 50-60Hz electrical input for operation. Depending on the electrical distribution system in your area, it may be necessary to provide an intermediate power converter. Please consult your local electricity provider if you are unsure of power specifications. Only electrical components certified for use in the country of installation should be used.

3.1 Electrical and Communication Cables

STEP	DESCRIPTION
1	Verify all component power switches are **OFF**.
2	Connect Instrument power supply and Control System power cords to electrical source. Surge protection is recommended.
3	Connect USB cable between Instrument and Control System.
4	Connect IEEE 1394 (FireWire) cable between Instrument and Control System.

Table 3.1: Connecting Electrical and Communication Cables

3.2 Install Fluidic Components

All parts are provided. Refer to the cutaway view in the figure.

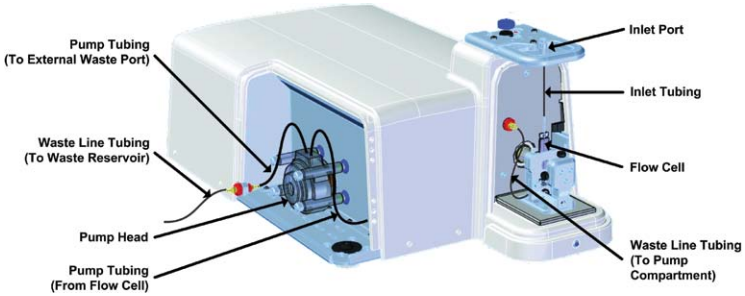

Figure 3.2: Fluidic Components

Flush fluids are particle-free, filtered (using a 0.22 µm filter) versions of the sample fluid. Where possible, the sample and flush fluid should be degassed.

Note: The surface of the instrument objective and the light source must remain clean at all times. If necessary consult the Maintenance chapter of this manual for the cleaning procedure.

3.3 Peristaltic Pump – Installation and Set Up

Once the system is fully installed, the Pump will automatically dispense samples based on the Run Configuration parameters.

3.3.1 Pump Head Tube Loading

STEP	DESCRIPTION
1	Hold pump-head with rotor (inside of head) facing you and tube pointing down. Pull apart to separate pump-head halves.
2	Feed middle of tube around rotor (ensure tubing is mounted properly on right-hand side at tube inlet port.
3	Insert loading key (provided) into right side of pump-head near tube inlet port. While securing tube in right-hand side of inlet port, rotate key towards left-hand inlet port.
4	Remove key and assemble, ensuring pump-head halves fit snugly together. The rotor should rotate freely when tube is properly installed.
5	Position pump-head into pump and align to key in rotor shaft.

Table 3.2: Pump-Head Tube Loading Procedure

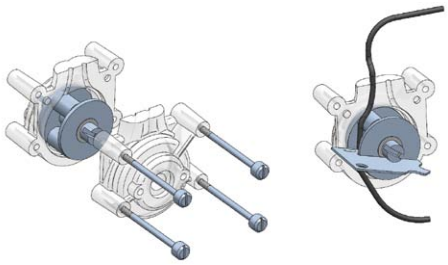

Figure 3.3: Pump Head Assembly (left) and Tubing Installation (right)

If the pump tubing is loaded incorrectly or the tubing requires replacement, the pump may emit a "knocking" noise when activated. Run pump for several minutes. If knocking continues, re-load or replace the pump head tubing.

3.3.2 Pump Head Calibration

The pump head calibration is performed using a wizard-based routine in the system software (MVSS). Please see the MVSS user manual for details on performing the pump head calibration.

3.4 Precision Stirrer Installation

The MFI 5000 instrument can be fitted with an optional precision stirrer for use with a syringe barrel. Please ensure that the software is shut down and the instrument is powered OFF before connecting or disconnecting the precision stirrer. Failure to do so may damage the instrument and void the warranty.

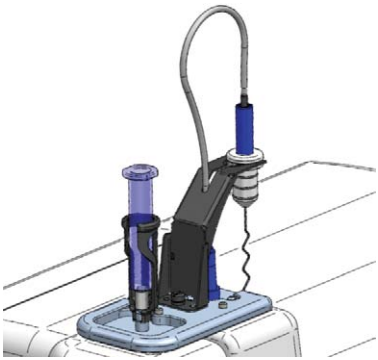

Figure 3.4: Optional Precision Stirrer and Holder

4.0 Verification

All MFI instruments are verified for size and concentration accuracy and repeatability during installation. The procedures can be re-performed as necessary. Refer to Section 6 for recommended Maintenance schedules. Prior to performing verification, flush the system and perform a clean baseline.

4.1 Verification Standards

NIST-traceable polystyrene bead standards are used during execution of this protocol. Prepare standards as follows:

SIZE	DROPS	DILUTION VOLUME
5 µm	1	15 mL
10 µm	2	10 mL

Table 4.1: Preparing Size Standards

Use the Method settings below to perform the size and concentration performance tests. During Method creation, if a parameter is not listed below, accept the default value.

PARAMETER/FIELD	VALUE
Method Name	"Size Verification, x um" (where x = particle size standard)
Method Type	Sample Analysis and Report
Morphological Filter - ECD	See Table 4.3 for range for each size standard
Chart Options – X Axis Step	0.25
Particle Statistics and Images	Uncheck
Include Raw Data File	Uncheck
Include Method Summary in Report	Check

Table 4.2: Method Parameters for Size Verification

4.2 Size Verification

After analyzing each size standard compare the results with Table 4.3. Repeat until the size standards pass for three (3) consecutive runs.

NOMINAL SIZE STANDARD	X-AXIS RANGE	X-AXIS RES.	VARIATION TO SIZE STD (+/-)
5 µm	3.5-6.5	0.25	5%
10 µm	8.0-12.0	0.25	5%

Table 4.3: Size Verification Criteria

4.3 Concentration

This procedure verifies the instrument's concentration accuracy. Use a 5 µm concentration standard.

STEP	DESCRIPTION
1	Flush Flow Cell and perform Baseline Run. Baseline particle count should typically be ≤2% of concentration standard's value.
2	Roll and tumble the standard 10 times. Fill a 1 ml pipette with the standard.
3	In **MFI View**, select or create an appropriate Method. Select 0.90ml for **Sample Dispensed** and 0.20ml for **Purge Volume**. Uncheck **Edge Particles Rejection**.
4	In Report Format, set X-Axis range and resolution per 5 µm entry in Table 4-3 .
5	Verify particle concentration is ±10% of standard's nominal concentration.

Table 4.4: Concentration Verification Procedure

Repeat until three (3) consecutive runs pass.

Always use flush fluids that are particle-free, filtered (0.22 µm) versions of the sample fluid. Where possible, the sample and flush fluid should be de-gassed.

5.0 Good Operating Practices

This chapter provides users with operating tips which can be used to obtain the best possible results using an MFI series instrument. Please see the MVSS user manual for details on the various operating procedures referenced in this section

5.1 Instrument Verification

5.1.1 Pump

Calibrate and verify the pump flow rates regularly, as per the preventative maintenance schedule in Table 6.1. The calibration procedure is given in the system software (MVSS) user manual.

5.1.2 Flow Cell Focus

Perform the **Flow Cell Focus** procedure for every Flow Cell. Record the result and retain with the Flow cell. Repeat as often as desired. Refer to the Flow Cell Focus Log file if required.

5.1.3 Performance Verification

Verify the size and counting accuracy for the instrument on a regular basis.

5.2 Flow Cell Integrity

5.2.1 Bubble Entrapment

The flow cell should be purged of air for proper operation. Residual micro-bubbles upstream of the flow cell may impact particle flow through the sensing zone and affect measurements. Trapped bubbles can be expelled by momentarily (1 to 2 seconds) pinching the input tubing while the pump is operating. If necessary, the flow cell can be removed and inverted during this process to aid bubble removal.

5.2.2 Flow Cell Fouling

Flow cells may become fouled over time. Proper care and storage will extend their lifetime. Perform the **Flow Cell Integrity Check** at regular intervals.

The interior surface of the flow cell has been treated with a hydrophobic silane coating to inhibit particle adherence. The coating will degrade with extended use, at which time the flow cell must be replaced. Regularly inspect stored images and the system display during sample analysis for evidence of repeating/stuck particles which are an indication of degradation of the interior coating. Contact ProteinSimple for additional information on verifying Flow Cell coatings.

5.3 Sample Preparation and Transfer

5.3.1 Homogenous Particle Dispersion

Consistent and sample-specific procedures for sample preparation and transfer (e.g. rolling, tumbling, inversion, pipette technique, etc.) should be specified. Sample homogeneity can be characterized using the **Time Resolved Mode** feature.

5.3.2 Sample Transfer/Introduction

Whenever possible, perform sample preparation and transfer procedures under a laminar flow hood.

Minimize the number of transfer vessels. Verify that transfer vessels are not contributing to baseline particulate level.

Perform multiple, successive tests to verify that the sample transfer procedures are achieving stable and consistent results.

5.3.3 Pipette Tip Insertion/Removal

The following procedures should be followed to minimize particle generation when using pipette tips:

- Gently insert pipette tip straight down into the luer, without twisting.
- For most sample types, it is good practice to run the pipette dry to prevent particles sticking on the luer fitting.
- After removing the tip, drip particulate free water into the luer fitting while the pump is running to clean the luer before inserting the next tip.

5.3.4 Diluents

Source particle-free diluents and use appropriate filtering (0.22 µm). Double-filter diluents if possible. Use the MFI 5100/5200 instrument to measure the particulate level of the diluents. Note that some samples may require time to stabilize after dilution.

5.3.5 Particle Formation/Degradation

The particle population in the sample itself may be unstable and susceptible to time-based changes (e.g. formation/degradation of protein particles). Use the **Time Resolved Mode** feature to determine the period in which the sample is stable. Select timelines for measurement to minimize the effects of any sample instability.

5.3.6 Gas Bubble Removal

If the presence of small gas bubbles is suspected, de-gas the sample by letting stand, uncovered, under a laminar flow hood. Sonication or vacuum can also be used (with suitable protocol verification). Optimize degassing procedure until bubbles are eliminated.

5.3.7 Particle Settling

Samples with larger, denser particles may require a syringe barrel and the Precision Stirrer to ensure homogenous sample introduction. Use the **Time Resolved Mode** feature to characterize the settling properties of the particles and to select the correct Stirrer RPM setting.

5.4 Run Configuration Parameters

5.4.1 Purge Volume

Use the **Time Resolved Mode** feature to determine the volume required to avoid mixing affects (e.g. Schlieren lines/false particles) at the beginning of each run. The default value (0.2 mL) is based on pipette tips and aqueous samples.

5.4.2 Preferences

Verify user and run preferences are consistently applied for all sample runs (e.g. **Edge Particle Rejection**, **Fill Hollow**, etc.). Use a consistent chart/histogram template specifying the bin width, size ranges, and size classes/statistics.

5.5 Sample Analysis

5.5.1 Baselines

Good laboratory practices will minimize contamination of the system. This includes wearing powder free gloves and working in a clean environment (including in a laminar flow hood whenever possible). Using clean, particle free flush/purge fluid is essential. Ensure that the Flow Cell does not run dry during flushing and measurement as this may promote adhesion if particles are present.

Replace inlet tubing and clean or replace the Inlet Port as required. Determine whether a fixed vessel should be connected to the Inlet Port by measuring particles before and after inserting pipette tips.

1. The sample container system should be filled with particle-free and bubble-free flush fluid. If stirring is required, insert the stir rod into the container.
2. The system should be well flushed (> 2 mL) using prime speed on the pump until no particles are visually observed on monitor.
3. Perform a baseline measurement including **Optimize Illumination**. If the stirrer will be used for samples, it should also be used for the baseline.

Using new components, baselines of < 100 particles per mL should be readily obtained.

5.5.2 Optimize Illumination

As indicated in the Sample Analysis menu, perform **Optimize Illumination** between each analysis run. Whenever possible, perform **Optimize Illumination** using particle-free versions of the sample (0.22 µm filter) to obtain optimal sensitivity, reduce Schlieren lines, and to minimize sample consumption.

5.5.3 Cleaning Between Samples

The degree and method of cleaning required to achieve a clean baseline between successive samples will depend on the sample type and the baseline count required. Normally, flushing with particle-free flushing fluid is sufficient.

When additional cleaning is required, purge with a detergent solution (for example 50% PCC-54® in filtered water) followed by purge fluid.

For some sample types, the stainless steel stir rod can be a source of contaminant. Keep a supply of clean rods available and use a new, clean rod for each new sample.

5.6 Special Sample Challenges

5.6.1 Incompatible Purge and Sample Fluids (Schlieren Lines)

Ideally the purge fluid is a filtered version of the sample fluid. If the purge and sample fluid have sufficiently different fluid properties, dark wavy Schlieren lines may be observed at the fluid boundaries at the start of the run. When Schlieren lines are observed, either select a different purge fluid which does not show Schlieren lines or determine the **Purge Volume** required to purge enough volume of fluid before Sample Analysis begins. Refer to the MVSS user manual for details.

In occasional cases, these may also appear at the end of the run. If Schlieren lines are observed at the end of a run, ensure that the volume dispensed/analyzed excludes these end-of-run effects.

5.6.2 Particle Transparency

Imaging requires sufficient contrast between the particle and the background fluid to fully resolve a particle. Particles with varying intensities nearing the threshold value may be only partially detected. This effect can be detected and assessed by viewing binary and grayscale particle images in their 'full frame' format.

5.6.3 High Particle Concentration

Particle concentrations in excess of the MFI concentration limits will lead to coincidence effects and may result in over-sizing and/or under-counting. This effect can be assessed by conducting a short dilution series, and verifying linear results are obtained.

5.6.4 High Viscosity

The default pump calibration is performed in water. If the sample has higher viscosity, it may be necessary to re-calibrate the pump with the sample to ensure proper flow rate accuracies.

5.6.5 Turbidity/Color

If the turbidity or color of the sample is too high, the system may not achieve sufficient illumination intensity or uniformity and will present a warning/failure message. When this occurs perform **Optimize Illumination** using the sample itself (or 0.22 µm filtered sample). Dilution may also be necessary to achieve suitable illumination conditions.

5.6.6 High mAb Concentration

When working with high concentration protein formulations (50 mg/mL+):

- Perform **Optimize Illumination** using sample or filtered sample (0.22 µm)
- Flush at the operational flow rate (vs. Prime Speed) to minimize Schlieren Lines
- Dilution may improve detection sensitivity, depending on the particle/carrier fluids

6.0 Maintenance

"When in doubt throw it out". After use, plastic luer pieces and tubing should be discarded and replaced.

Use only fresh, filtered (0.22µm) flush water and flush fluid. Fluids should be de-gassed whenever possible.

6.1 Preventative Maintenance

The following preventive maintenance schedule is recommended. This schedule may require modification (more or less frequently) depending on the sample types and operating environment.

TASK	DAILY	WEEKLY	MONTHLY	ANNUALLY
Flush System	x			
Backup Data	x			
Clean/Replace Inlet Port	x			
Replace Flow Cell Inlet Tubing/Stopcock		x		
System Verification Check (particle standards)		x		
Flow Cell Integrity Check		x		
Flow Cell Focus		x		
Replace Precision Stirrer Coupler Tubing		x		
Flow Cell Cleaning Procedure		x		
Replace Waste Line Tube Assembly and Calibrate Pump			x	
Objective Lens Cleaning			x	
LED Light Source Cleaning			x	
De-fragment Computer			x	
ProteinSimple Factory/ Site Service/Verification				x

Table 6.1: Preventative Maintenance Schedule

6.2 Instrument Housing Cleaning

The instrument may be wiped down with a clean, damp cloth. Use water only.

6.3 Flow Cell Cleaning

Refer to the following table for cleaning Flow Cells.

Do not clean Flow Cells in ultrasonic baths or cleaning solutions warmer than 40°C. If detergent is used, clean Flow Cell immediately with flush water to prevent detergent from drying out, and thus avoid Flow Cell damage.

This Cleaning Procedure is a guideline only. It may not be suitable for specific applications. ProteinSimple does not guarantee that the procedure will always be effective.

STEP	DESCRIPTION
1	Thoroughly flush Flow Cell using filtered (0.22 µm) water.
2	Replace water sample with 50% by volume "PCC-54® detergent to filtered water" solution (Substitute other detergent solutions according to specific requirements. See information below on other detergent options.)
3	Set pump to maximum flow rate or press Prime button
4	The exact time to clean will vary. Most samples require less than 2 minutes.
5	Flush detergent from system using 0.22 µm filtered water. The rule-of-thumb is to flush for as long as the detergent was used (detergent residue can form a precipitate that may damage Flow Cell).
6	Remove Flow Cell from holder and swab exterior surface using analytical grade Isopropyl Alcohol (IPA) and a lint-free swabbing component such as Texwipes® TX743B CleanTip® Swabs. After removing excess IPA from swab, pass swab in one direction over sensing zone area on exterior glass in one motion. Rotate swab to a fresh surface and repeat. Do not use same surface of swab twice. Ensure all IPA has evaporated from surface of Flow Cell prior to re-insertion to holder. If required, this step can be repeated using PCC-54® (20% to 50% solution to filtered water by volume) followed by rinsing the exterior with water to remove PCC-54®.

STEP	DESCRIPTION
7	Perform a Flow Cell Integrity Check. If the Flow Cell Integrity check fails or the cleanliness of the Flow Cell is visually unsatisfactory, replace Flow Cell.

Table 6.2: Flow Cell Cleaning

6.3.1 Cleaning solutions

The following detergents are effective, general cleaning solutions:

NAME	MANUFACTURER
PCC-54®	Thermo Scientific
Micro-90®	International Products Corporation
Triton®X-100	Sigma-Aldrich

Although the following detergents have not been used by ProteinSimple, they have also been reported as being effective cleaning solutions. This list is provided for information only.

NAME	MANUFACTURER
Contrad®70	Decon Labs
Hellmanex® II	Hellma
Liqui-Nox®	Alconox Inc.

Refer to MSDS papers for Safety, Handling, Storage, and Disposal information before working with these products. Always dispose of chemicals in accordance with all applicable government regulations and guidelines.

6.3.2 Triton®X-100 Cleaning Solution

Triton®X-100 is an effective degreaser and can be used to remove substances like silicone oil. Dilute acetic acid is added to aid the cleaning properties of the solution.

Triton®X-100 Cleaning solution (1 L) is prepared by combining the following materials:

a) Triton®X-100 (5%, v/v) = 50 mL
b) Acetic Acid (5%, v/v) = 50 mL
c) Water (0.22 µm filtered) = 900 mL

This solution should be kept refrigerated, and has a shelf life of ~ 1 month.

6.4 Flow Cell Coating Check

Flow cells may become fouled over time. Proper care and storage will extend their lifetime. Perform the **Flow Cell Integrity Check** at regular intervals.

The interior surface of the Flow Cell has been treated with a hydrophobic silane coating to inhibit particle adherence. The coating will degrade with extended use, at which time the Flow Cell must be replaced. A degraded coating may result in a higher percentage of repeating particles. Regularly inspect stored images and the system display during sample analysis for evidence of these repeating/stuck particles.

A simple experiment can be performed to determine the status of the interior coating. Flush the Flow Cell with particle-free water until clean. Run the pump at the normal Set Point speed until it is dry. Use **Preview Mode** to inspect the Flow Cell interior surface. An intact coating will repel water and appears relatively uniform (potentially some small droplets appearing as round beads). A degraded coating will cause irregularly-shaped water droplets, marks, or streaks to remain on the surface. Refer to the images below.

Note that the results for a dirty Flow Cell will resemble a degraded Flow Cell, so it is important that the Flow Cell is clean before starting this test.

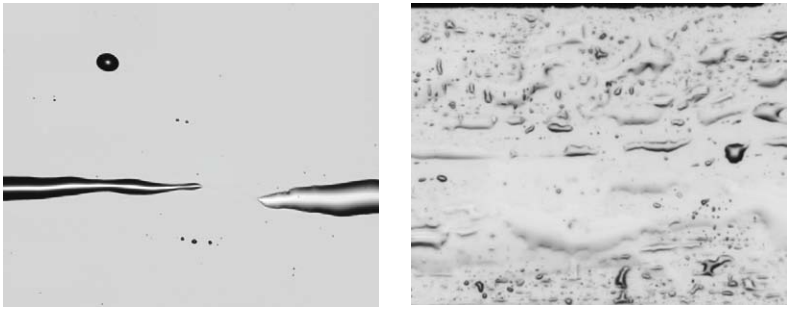

Figure 6.1: Flow Cell Images – Intact Coating (Left), Degraded Coating (Right)

6.5 Flow Cell Storage

Flow Cells can be stored in either a wet or dry mode. Regardless of the storage mode, ensure the Flow Cell is thoroughly flushed and clean before storing.

If the Flow Cell is stored dry, ensure that all residual fluid is purged from the Flow Cell before storing. See the figure 6.2 for drying the interior of the Flow Cell using inert, dry, optical-grade gas.

If the Flow Cell is stored wet, ensure the storage fluid is not permitted to evaporate.

Contact ProteinSimple for further information on storing Flow Cells.

6.5.1 Dry Flow Cell Storage

STEP	DESCRIPTION
1	Perform the Flow Cell Cleaning procedure. Ensure Flush water is in the sample vessel.
2	Turn pump ON, and press PRIME for 2 minutes to flush out remaining flushing water present in Flow Cell.
3	Remove Flow Cell from holder and disconnect attached tubing. Use Caution to avoid spilling any liquid.
4	Operating under a laminar flow hood, use a 10ml syringe barrel with tubing attached to a luer-to-barb adaptor or a blunt-end needle. Attach the free end of the tubing to the Flow Cell, and slowly flush Flow Cell with analytical grade Isopropyl Alcohol (IPA).
5	Operating under a laminar flow hood, evaporate any remaining IPA by gently flushing with inert, dry, optical grade gas (Nitrogen or, 1,1,1,2 Tetrafluoroethane such as MG Chemicals Super Duster134 Cat# 402A-450G). When using a compressed gas can to evaporate IPA, do not connect the flow cell directly to nozzle. Hold nozzle at a distance of ~5mm from Flow Cell input tube and gently apply gas into flow cell to evaporate IPA. Refer to figure 7.2. Note that high pressures can destroy the Flow Cell.

Table 6.3: Dry Flow Cell Storage Procedure

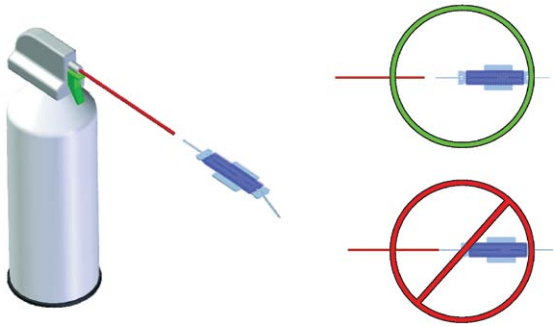

Figure 6.2: Drying Interior of Flow Cell

6.5.2 Wet Flow Cell Storage

STEP	DESCRIPTION
1	Perform the Flow Cell Cleaning procedure. Ensure Flush water is in the sample vessel.
2	Turn pump ON, and press PRIME for 2 minutes to flush remaining flushing water present in Flow Cell.
3	Insert Syringe with 5 mL of PCC-54®, or Triton®X-100 cleaning solution (see Section 6.3.2 for preparing Triton®X-100 cleaning solution).
4	Prime 2 mL of PCC-54®, or Triton®X-100 cleaning solution, through flow cell.
5	Cap syringe with Parafilm® M laboratory film, to prevent evaporation of detergent.

Table 6.4: Wet Flow Cell Storage Procedure

PCC-54® can remain within the Flow Cell for up to 1 week. Triton®X-100 can remain within Flow Cell for up to 2 weeks. After this period, the detergent should be removed from the Flow Cell which is then flushed with ~ 10 mL clean filtered water. The Flow Cell can then be re-filled with fresh detergent solution. To prevent damage to the Flow Cell, it is important to prevent the detergent solution from evaporating.

6.6 Sterilization

If required, the wetted components may be sterilized with a 1 ml flush of 10% bleach solution followed by a thorough flush with water to displace the bleach.

6.7 Inlet Port and Tubing

If the sample container is left in place during a series of analyses, contamination of the inlet port may occur. The inlet port can be cleaned "in situ" using detergent and a small swab followed by flushing. If the inlet port cannot be cleaned "in situ", replace or use the following procedure to clean the port.

STEP	DESCRIPTION
1	Swab the inside with a lint fee swab coated with detergent.
2	Rinse with clean filtered water.
3	Immerse in a beaker in a sonic bath with a stainless steel compatible detergent (ie 50% PCC-54® in filtered water).
4	Remove with clean forceps and rinse with clean filtered water followed by IPA.
5	Store in a clean dry location.

Table 6.5: Cleaning Inlet Port and Tubing

The input tubing can also be replaced as necessary.

6.8 Syringe Barrels

STEP	DESCRIPTION
1	Add detergent to the syringe barrel.(i.e. Micro-90®).
2	Scrub using a clean (dedicated) plastic bristle brush with detergent. The brush should have roughly the same diameter as the syringe opening. Move the brush in an up-and-down motion to agitate the detergent and produce a foamy consistency.
3	Clean the outside of the metal Luer section of the syringe with the detergent and brush.
4	Rinse the detergent from the syringe barrel and Luer section using clean filtered water.
5	Rinse the syringe barrel with Isopropyl Alcohol (IPA). The IPA will displace the water and help to dry the syringe barrel. The syringe barrel can then be left to air dry upside down on a stand.
6	Prior to storing, the top opening of the syringe should be covered with clean Parafilm®, or other particle-free material, to prevent airborne contamination

Table 6.6: Cleaning Syringe Barrels

6.9 Stir Rods

STEP	DESCRIPTION
1	Scrub using a clean plastic bristle brush with detergent.
2	Immerse in a beaker in a sonic bath with a stainless steel compatible detergent (i.e. 50% PCC-54® in filtered water).
3	Remove with clean forceps and rinse with clean filtered water followed by Isopropyl Alcohol (IPA).
4	Store in a clean dry location.

Table 6.7: Cleaning Stir Rods

6.10 Stirrer Assembly

In normal use, the Stirrer Assembly should only require a light rinse of the exterior of the white plastic syringe adaptor with clean filtered water. A more thorough cleaning procedure can be carried out periodically as follows:

STEP	DESCRIPTION
1	The stirrer assembly is made up of 3 parts: white syringe plastic adaptor, micro motor, and motor coupler tube.
2	Unscrew the motor from the plastic syringe adaptor.
3	Remove and discard the old motor coupler tube. The motor can now be carefully cleaned using a lint free swab by passing it over the motor shaft to pick up any debris. A low-pressure blast of compressed gas from a gas duster, or dry nitrogen supply, can also be used to blow off any residual dust from the shaft. Add a new coupler tube. (NOTE: do not immerse or clean the motor with fluids. This will damage the motor.)
4	The white plastic syringe adaptor (without motor) should be manually cleaned using a clean water/detergent solution, rinsed with clean filtered water followed by Isopropyl Alcohol (IPA) and then dried.
5	Re-assemble the stirrer components.

Table 6.8: Cleaning Stirrer Assembly

6.11 Optical Objective Cleaning Procedure

The objective lens may become contaminated by fingerprints, watermarks, dust etc. which will interfere with image quality. Use this procedure to clean the Objective:

STEP	DESCRIPTION
1	Power **OFF** instrument, electrical module and pump(s).
2	Use a lint-free swab such as Texwipes® TX743B CleanTip® Swabs.
3	Apply a small amount of isopropyl alcohol (IPA) to end of swab.
4	Starting from centre of objective lens, scrub exterior glass in a circular motion spiralling outwards.
5	Use a new, dry swab. Starting from centre of objective lens, wipe exterior glass in a circular motion, spiralling outwards.
6	Ensure all residual IPA has evaporated prior to powering **ON** instrument, electrical module and pump(s).

Table 6.9: Optical Objective Cleaning Procedure

6.12 LED Light Source Cleaning Procedure

Power **OFF** instrument, electrical module and pump(s). Wait approximately 2 minutes and then use a dry, inert compressed gas (e.g. a gas duster can or nitrogen) to gently blow any contamination from light source.

CAUTION: The output of the LED should not be viewed with certain optical instruments (for example, eye loupes, magnifiers and microscopes) within a distance of 100 mm may pose an eye hazard.

6.13 Customer Support

Please contact your local sales representative with any questions or concerns regarding your ProteinSimple product. You may also contact a ProteinSimple Customer Support Specialist directly via support@proteinsimple.com or telephone toll free in North America 888.607.9692, option 3 or +1.408.510.5500, option 3 internationally.

7.0 Safety And Other Compliances

7.1 Safety Certifications and Compliances

Safety Requirements for Electrical Equipment for Measurement, Control, and Laboratory Use

- Certified to IEC/EN/CSA/UL 61010-1 2nd Edition

EMC requirements – Electrical equipment for measurement, control and laboratory use

- Type tested and found to comply with the limits for a Class A digital device, pursuant to:
  - EN 61326-1:2005
  - FCC 47 CFR Part 15 (Class A – office use only)
  - ICES-003 Issue 4 February 2004 (Class A – office use only)

Photobiological Safety of Lamps and Lamp Systems

- Type tested to IEC62471-1:2006-07 and IEC62471-2:2006-07 (Risk Group 2 LED)

7.2 FCC Notice (U.S. Only)

This equipment has been tested and found to comply with the limits for a Class A digital device pursuant to Part 15 of the FCC Rules. These limits are designed to provide reasonable protection against harmful interference when the equipment is operated in a commercial environment. This equipment generates, uses, and can radiate radio frequency energy and, if not installed and used in accordance with the manufacturer's instruction manual, may cause harmful interference with radio communications. Operation of this equipment in a residential area is likely to cause harmful interference, in which case you will be required to correct the interference at your own expense.

Operation is subject to the following two conditions:

This device may not cause harmful interference.

This device must accept any interference received, including interference that may cause undesired operation.

Electromagnetic Interference (EMI) is any signal or emission, radiated in free space or conducted along power or signal leads, that endangers the functioning of radio navigation or other safety service or seriously degrades, obstructs, or repeatedly interrupts a licensed radio communications service. Radio communications services include but are not limited to AM/FM commercial broadcast, television, cellular services, radar, air-traffic control, pager, and Personal Communication Services (PCS). These licensed services, along with unintentional radiators such as digital devices, including laboratory equipment, contribute to the electromagnetic environment.

Electromagnetic Compatibility (EMC) is the ability of items of electronic equipment to function properly together in the electronic environment. While this laboratory equipment has been designed and determined to be compliant with regulatory agency limits for EMI, there is no guarantee that interference will not occur in a particular installation. If this equipment does cause interference with radio communications services, which can be determined by turning the equipment off and on, you are encouraged to try to correct the interference by one or more of the following measures:

Reorient the receiving antenna.

Relocate the equipment with respect to the receiver.

Move the instrument away from the receiver.

Plug the instrument into a different outlet so that the instrument and the receiver are on different branch circuits.

If necessary, consult a ProteinSimple Technical Support representative or an experienced radio/television technician for additional suggestions.

7.3 ICES-003 Notice (Canada only)

This Class A digital apparatus complies with Canadian ICES-003.

Cet appareil numérique de la classe A est conforme à la norme NMB-003 du Canada.

7.4 Recycling Information

This product must be recycled after its useful life as required by local laws or regulations. Please return the product for proper recovery/disposal to the authorized collection location nearest you. Always respect local recycling legislation.

7.5 Solvent Support

The MFI 5100/5200 instrument has been certified to IEC/EN/CSA/UL 61010-1 2nd Edition which meets the safety requirements for "Electrical Equipment for Measurement, Control, and Laboratory Use" when used with nonflammable or non-combustible materials. The MFI 5100/5200 instrument has not been certified to section 9.4 "Requirements for equipment containing or using flammable liquids" in IEC/EN/CSA/UL 61010-1. The MFI 5100/5200 instrument has not been certified for use in Hazardous (Classified) Locations.

7.6 EC Declaration of Conformity

DECLARATION OF CONFORMITY

Manufacturer's Name: ProteinSimple

Manufacturer's Address: 115 Terence Matthews Cres.
Unit 4
Ottawa Ontario K2M 2B2
CANADA
+1 (613) 591-7715
www.proteinsimple.com

Declares, that the product:

Product Name: MFI 5000
Model Number(s): MFI 5100 and MFI 5200
Product Description: The MFI 5100 and MFI 5200 measure the size, shape, transparency, count and concentration of liquid borne particles/cells through automated sample introduction, image acquisition and image analysis.

Conforms to the following Standards:

2006/95/EC (Low Voltage Directive)

This declaration is based on the full compliance with the standards:

EN 61010-1 ed2.0 (2001) (Safety Requirements for Electrical Equipment for Measurement, Control and Laboratory Use – Part 1: General Requirements)

EN 62471:2006 (Photobiological safety of lamps and lamp systems)

2004/108/EC (EMC Directive)

This declaration is based on the full compliance with the standards:

EN 61326-1:2005 (Electrical Equipment for Measurement, Control, and Laboratory Use. EMC Requirements)

This declaration is issued under the sole responsibility of ProteinSimple

Ottawa, Aug 8, 2011

Peter Oma, P.Eng.
Director of Research & Development

8.0 Specifications

8.1 Operating Conditions

PARAMETER	UNIT	MIN	MAX
Temperature	°C	5	40
Relative Humidity to 31°C decreasing linearly to 50 % relative humidity at 40°C. Non-condensing.	%	10	80
Altitude	m		2000
Pollution Degree (indoor use only)	Category		2
Installation Category	Category		II
Flush Fluids	1. Water filtered using a 0.22 µm filter 2. PCC-54® Detergent, 50% by volume		
Sample Fluids	Aqueous		
Enclosure Rating	IP22 as per IEC 60529		

Table 8.1: Operating Conditions

8.2 Storage Specifications

PARAMETER	UNIT	MIN	MAX
Temperature	°C	-10	60
Relative Humidity	%	10	80

Table 8.2: Storage Specifications

8.3 MFI 5100 Performance Specifications

PARAMETER	NOTE (TABLE 8.5)	UNIT	MIN	MAX
Particle Sizing Range	1	µm	2	300
Display Resolution		µm	0.25	
Concentration Limit – 2.5 µm	2	#/mL	0	175,000
Concentration Limit – 5 µm	2	#/mL	0	75,000
Concentration Limit – 10 µm	2	#/mL	0	20,000
Sizing Accuracy	3	-	±0.5 µm for particles <5 µm, ±5% for particles ≥5 µm	
Sizing Repeatability	4	%	± 5	
Concentration Accuracy	1,5	%	± 10	
Concentration Repeatability	1,4	%	± 5	
Analysis Rate		µl/min	185	193
Sampling Efficiency	6	%	85	
Linear Velocity (sample line)	7	mm/sec	4.49	
Linear Velocity (Flow Cell)	7	mm/sec	5.72	
Priming Volume		mL	0.25	
Pump Flow Rate		mL/min	0.22	

Table 8.3: MFI 5100 Performance Specifications

8.4 MFI 5200 Performance Specifications

PARAMETER	NOTE (TABLE 8.5)	UNIT	MIN	MAX
Particle Sizing Range	1	µm	1.0	70.0
Display Resolution		µm	0.25	
Concentration Limit – 2.5 µm	2	#/mL	0	900,000
Concentration Limit – 5 µm	2	#/mL	0	400,000
Concentration Limit – 10 µm	2	#/mL	0	250,000
Sizing Accuracy	3	-	±0.5 µm for particles <5 µm, ± 5% for particles ≥5 µm	
Sizing Repeatability	4	%	± 5	
Concentration Accuracy	1,5	%	± 10	
Concentration Repeatability	1,4	%	± 5	
Analysis Rate		µl/min	150	
Sampling Efficiency	6	%	85	
Linear Velocity (sample line)	7	mm/sec	7.36	
Linear Velocity (Flow Cell)	7	mm/sec	17.7	
Priming Volume		mL	0.20	
Pump Flow Rate		mL/min	0.17	

Table 8.4: MFI 5200 Performance Specifications

8.5 Notes for Performance Specifications

NOTE	DESCRIPTION
1	Assumes particles are in suspension.
2	Coincidence limit.
3	Percent deviation of measured mean from mean of PS size standard.
4	Relative standard deviation using PS size standard. Value shown is for 5 µm particles.
5	Relative to concentration of 5 µm PS concentration standard.
6	Nominal value (percentage of total sample through flow cell that is analyzed).
7	Nominal values based on laminar flow.

Table 8.5: Notes for Performance Specifications

8.6 Run Configuration Parameters

PARAMETER	NOTE	UNIT	MIN	MAX
Test Duration		hours	-	25
Quantity of Images per Run		images	-	20,000
CSV Output File Size (for 25 hour test)		Mb	-	90
Selective Storage Size Ranges		Ranges	0	8
Frame Rate	1	frames/sec	3 (MFI 5100)	10 (MFI 5200)
Particle Limit				1M

Table 8.6: System Parameters

NOTE:

1. Varies according to operating mode and hardware configuration.

8.7 Image File Specifications

PARAMETER	UNIT	MIN MAX
Sensor Array Size	Pixels	1280x1024
Grey Scale Resolution	bit	10
File Formats	-	JPG
File Types	-	binary, Grey Scale
Image File Size	Kb	140

Table 8.7: Image Storage Specifications

8.8 Mechanical Dimensions

SYSTEM COMPONENT	UNIT	SPECIFICATION
Instrument	mm	601x351
Flow Cell – Sample Channel Depth	µm	400 or 100 nominal
Silastic Tubing Inner Diameter	µm	1016 nominal

Table 8.8: Mechanical Dimensions

8.9 Minimum Computer Specifications

COMPONENT	SPECIFICATION
CPU	Intel® Core™2 Duo E7000
Motherboard Chipsets (tested)	Intel®: P35, P45, G41, H57, Express Chipsets
RAM	3GB DDR2
Hard Drive (OS)	500GB, 7200 RPM
Hard Drive (Repository)	1 TB, 7200 RPM
Video Card	PCIE, 128MB RAM, DirectX9c
Removable Storage Device	CD/DVD Burner
Firewire Port	One (1) IEEE 1394a 6-pin powered Firewire A
USB Port	One (1) available USB 2.0 port
Operating System	Windows® Xp Professional with Service Pack 3 (English, 32 bit), Windows® 7 Professional (English, 32 bit)
Monitor	17" or 19", 5:4 ratio only, 1280x1024 resolution

Table 8.9: Minimum Computer Specifications

8.10 Electrical Specifications

PARAMETER	UNIT	VALUE
Power Consumption	W	36
Voltage	V(DC)	12
Source Current	A(DC)	3

Table 8.10: Electrical Specifications

Note:

- Mains supply voltage fluctuations are not to exceed 10 percent of the nominal supply voltage.
- Voltage Supplied by External Power Supply:
  - Sinpro Model: SPU65-105: Input 100VAC to 240VAC, 1.9A, Output 12VDC, 6.66A

  or

  - Advanced Power Solutions Model: APS100EST-12/2.5: Input 100VAC to 240VAC, 2.5A, Output 12VDC, 8.5A

9.0 Intellectual Property

MFI 5100/5200 Flow Microscopes contain technology that is protected by the following patents.

1. Digital Optical Measurement of Particle Populations Using Reduced Magnification
    - Issued June 20, 2006, Patent # 7,064,826
2. Method and Apparatus for Particle Measurement Employing Optical Imaging
    - Issued May 27, 2008, Patent # 7,379,577
3. Automatic Identification of Suspended Particles
    - Issued May 15, 2007, Patent # 7,217,937
4. Particle Imaging System Having a Varying Flow Rate
    - Issued December 11, 2007, Patent # 7,307,721
5. Method and Apparatus for Analyzing Particle Populations in a Fluid
    - Issued October 20, 2009, Patent # 7,605,919
6. Plurality of Samples and Method for Selecting a Target Sample Therefrom
    - Issued December 28, 2010, US Patent # 7,859,664 B2
7. Particle Standard and Method of Calibrating or Validating an Optical Particle Analyzer
    - Filed September 2009 in US and Canada
8. Particle population measurement system having an extended range
    - Filed October 2010 in US and Canada

10.0 Warranty

ProteinSimple warrants only to the original Buyer (except to some extent in Quebec and where otherwise prohibited) that all products, other than software, made by ProteinSimple are free from defects in material and workmanship under normal use for a period of one (1) year from the date of install.

Under warranty, ProteinSimple will repair defective products or provide a replacement product at its sole option, for any product made by it, which upon inspection by ProteinSimple and in the sole opinion of ProteinSimple, is determined to be defective in workmanship or material and has in fact failed. Replacement products and parts may be new or remanufactured. Subject to applicable laws, all products repaired or replaced under warranty are only warranted for the remaining unexpired period of time in the original warranty for the particular defective product. ProteinSimple reserves the right, at its sole option, to issue a credit note for any defective product as an alternative to repair or replacement.

This warranty shall extend to any product which has proved defective and has failed through normal use, but excludes and does not cover:

- any products or parts thereof which have been accidentally damaged, disassembled, misused, used in applications which exceed their specifications or ratings, neglected, improperly installed or otherwise abused;
- damage or failure caused by unauthorized modification, accessories, or alteration, or if it is used for other than the intended purpose;
- improper repair or maintenance, including use of parts not approved or specified by ProteinSimple;
- installation, alteration, repair or service by anyone other than a licensed, authorized ProteinSimple contractor or other than pursuant to the manufacturer's instructions, including installation not in accordance with applicable laws, codes and standards applying in the jurisdiction in which the product is installed;
- failure of a product to perform during power failures and interruptions or inadequate electrical service;

- damage resulting from running the product in a corrosive atmosphere or contrary to the instructions outlined in the product operation manual; and
- products with original serial numbers that have been removed, altered or cannot be readily determined.

Buyer must claim under the warranty in writing not later than thirty (30) days after the claimed defect is discovered. Except as otherwise permitted by law (which may include to some extent Quebec), all claims under this warranty must be made by the Buyer and no claim will be accepted from any third party.

ProteinSimple will only accept returns for which an approved Return Material Authorization (RMA) has been issued by ProteinSimple. Defective products shall be returned prepaid and insured to ProteinSimple at the address shown herein. All products which have been returned to ProteinSimple but which are found to meet all previously applicable specifications for such products shall be subject to ProteinSimple's standard examination charge in effect at the time, which shall be charged to the Buyer. All products returned to ProteinSimple which are not accompanied by an itemized statement of defects, shall be returned to the Buyer at the Buyer's expense and no evaluation of such products shall be carried out by ProteinSimple.

Where products contain software, please refer to the licence agreement included with the software.

PROTEINSIMPLE'S SOLE LIABILIY IS LIMITED TO THE WARRANTY SET OUT ABOVE. THIS WARRANTY IS IN LIEU OF ANY OTHER WARRANTY OR CONDITION, EXPRESS OR IMPLIED, INCLUDING WITHOUT LIMITATION, ANY WARRANTY OR CONDITION OF MERCHANTABILITY OR (EXCEPT WHERE OTHERWISE PROHIBITED, WHICH MAY INCLUDE QUEBEC) FITNESS FOR A PARTICULAR PURPOSE. TO THE EXTENT ANY IMPLIED WARRANTY OR CONDITION IS REQUIRED BY LAW, IT IS LIMITED IN DURATION TO THE EXPRESS WARRANTY PERIOD ABOVE. PROTEINSIMPLE SHALL NOT BE LIABLE FOR ANY INCIDENTAL, CONSEQUENTIAL, DIRECT OR INDIRECT, SPECIAL, PUNITIVE OR EXEMPLARY DAMAGES OF ANY NATURE, INCLUDING WITHOUT LIMITATION, LOSS OF GOODWILL, LOST REVENUES OR PROFITS, OR ANY OTHER DAMAGE WHETHER ARISING DIRECTLY OR INDIRECTLY FROM ANY CONTRACTUAL BREACH, FUNDAMENTAL OR OTHERWISE, OR FROM ANY ACTS OR OMMISSIONS, TORT, OR OTHERWISE.

Some jurisdictions (which may include Quebec) do not allow the exclusion or limitation of incidental or consequential damages or limitations on how long an implied warranty lasts, so the above exclusions or limitations may not apply to the Buyer. This warranty gives the Buyer specific legal rights and the Buyer may also have other rights that vary from jurisdiction to jurisdiction. Any term of this warranty that negates or varies any implied condition or warranty under local laws is severable where it conflicts with local laws without affecting the remainder of this warranty.

t +1.408.510.5500 tf +1.888.607.9692 f +1.408.510.5599

proteinsimple.com