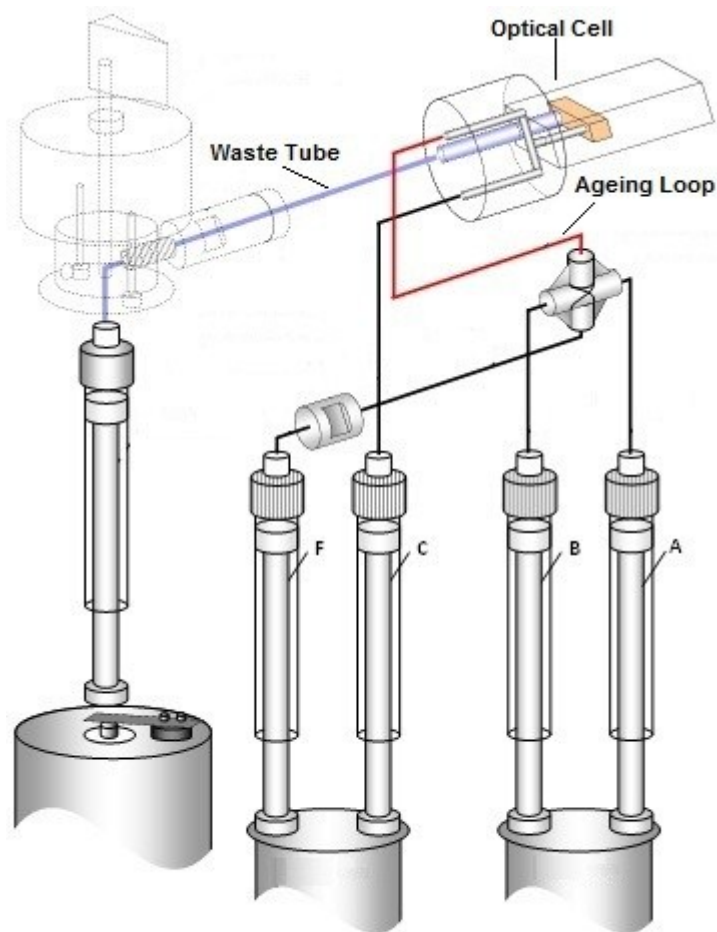


Cleaning the Observation Cell and Flow Circuit

Relevant instruments

The cleaning procedures described in this document can be applied to **all SX instruments**.

SF
SX18MV
SX18MV-R
SX20



Cleaning the Observation Cell and Flow Circuit

Introduction

This document is a guide to cleaning the observation cell and flow circuit on any SX instrument.

To maintain the high performance of the stopped flow instrument, cleaning of the observation cell and flow circuit should be part of the routine maintenance, particularly if protein or sub-cellular fragment work is being done. A build up of debris in the flow circuit and cell can cause serious errors in your results.

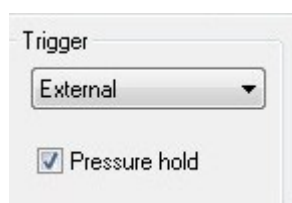
Required Cleaning procedures

- Daily Basis (see page 3)
- Monthly Basis (see page 3)

Signs of impurities

For the user a few points can be relevant for spotting a dirty cell.

- If at any point a fluorescence detection is experienced when pure water is run through the system this is an indication of impurities present in the cell.
- Look out for a small peak at 30ms which also can be an indication of impurities. This peak should disappear if the "Pressure Hold" is enabled (see screenshot below) and then appear again when "pressure hold" is disabled indicating the presence of impurities. If this is not the case the peak could also indicate the presence of an air bubble.



(NB: Screenshot from ProData Version 2.2.1)

Procedure

Cleaning of Cell and Flow Circuit on a Daily Basis

Cleaning of the cell and flow circuit should be carried out on a daily basis if the SX instrument is used regularly. This can easily be done with water, but if protein or sub-cellular fragment work is being done we strongly recommend to carry out the cleaning using "Hellmanex II liquid".

Hellmanex II is an alkaline liquid concentrate which must be diluted with water to yield an effective cleaning solution for the mixing circuit and cell. Details of dilution or concentration are given in the instructions. Just flush the mixing circuit and cell through with the Hellmanex II cleaning solution and leave it to soak for the prescribed length of time. Then flush through well with distilled water. Please refer to the instructions provided with Hellmanex II for full details.

This cleaning procedure should be performed at least once a week or more often.

Where to obtain Helmanex II liquid:

Hellmanex II liquid (catalogue number: 320.001) is supplied by a company called "HELLMA". They have outlets all over the world.

Cleaning of Cell and Flow Circuit on a Monthly Basis

Over time there is a tendency for debris to build up even though regularly cleaning using Hellmanex II is carried out. Hence the need for performing a more thorough cleaning. This is recommended to be done on a monthly basis.

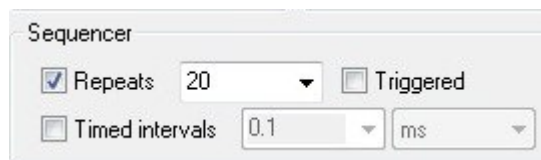
The cleaning procedure follows three steps that are repeated using different solutions. The solutions used should be in the following order;

- 1M NaOH
- 2M Nitric Acid (particular important for instruments used with protein solutions)
- Distilled Water (to clean out any old protein left in the flow circuit)

The three steps should be repeated with each of the chemicals listed above. The steps are as followed:

- 1) Using 10 ml reservoir syringes, flush the drive syringes three times with at least 30 ml of the required liquid.

- 2) Fill the drive syringes with fresh liquid. Turn the control valves to the drive (forwards) position. Set the time base to the fastest linear timebase available. Set the triggering to external trigger and choose to do 20 repeats as illustrated in the screenshot below. Make sure that there is no gap between the drive ram and the syringe plungers (see Figure 2). Click on Acquire. 20 shots will be fired through the optical cell and flow lines.



(NB: Screenshot from ProData Version 2.2.1)

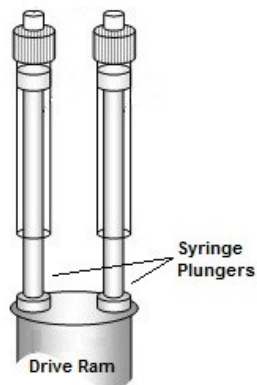


Figure 2

- 3) Refill the drive syringes with the liquid. Turn the control valves to the drive (forwards) position. Completely empty the stop syringe. Push the drive ram up, by hand, until the stop syringe is full (see Figure 3). Repeat this step at least three times.

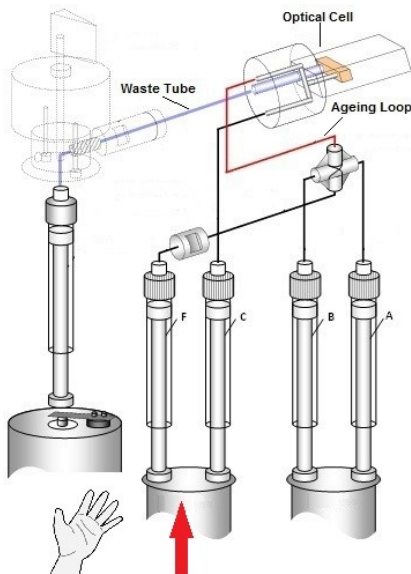


Figure 3