

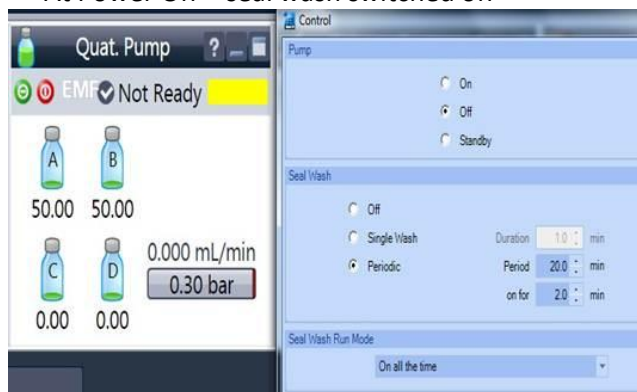
Seal Wash Operation:

- Use **10% Iso-Propanol in Water**
- Change solvent weekly
- Use 300-400ml Solvent bottle, put date on bottle
- Position wash bottle above pump
- Position Wash Solvent Waste below instrument
- Test Peristaltic Pump
 - Finger check for movement, look for drops to confirm delivery of solvents
- Operate **PERIODIC** with 0.3min each 3min
 - Peristaltic Pump moves 700ul/min
 - 0.3min/3min = 6min/h = 4.2ml/hour
 - 12h = 50ml/day
- 5days = 250ml/week solvent needed
- Setting in Software is **NOT a method parameter**, it stays with the pump.

***Do not** recycle seal wash solvent

Seal Wash Dialog in OpenLab Software:

- Seal Wash is a software setting NOT a method parameter
- Find dialog under CONTROL
- On ERROR – seal wash is switched off
- At Power Off – seal wash switched off



System Shutdown:

- If another sequence will be run after oen previously is complete (within 12 – 16 hours) schedule a transitional method that reduces the flow rate to ~0.1ml/min.
- End of a sequence when the system will not be used for a day use a water flush shutdown method
- When system will be ideal longer than 2 days place all bottle heads in fresh solvents
 - Water must have no buffers but must have some organic to avoid algae growth
 - Purge each channel at 2.5-3ml/min for 5min
 - Shut down the system

*** Remember** to remove column if needed.

Housekeeping: Daily / System Start-up

- Replace Aqueous mobile phase (buffer, water) with fresh solvents
 - Always use a clean bottle, never top-off a bottle
- Replace Organic mobile phase at least every 2nd day
- Check seal wash pump function, finger/drops, check software settings
- Purge each pump head at 2.5-3ml/min for 5min each
 - Oxygen will re-defuse into the solvent channel when the system is idle, for example if turned off over night
- Condition pump for ~15 min
 - Conditioning will use the current pump settings of solvent composition, flow rate and maximum pressure
 - Ideally system pressure during conditioning should be >200 bar
 - Start/Stop manually with right mouse in **Instrument GUI**
 - Pump status will be Not Ready while running
 - Pump returns to Ready when conditioning time is complete
 - Conditioning removes remaining small air bubbles in pump head.

Recommended Housekeeping: Weekly

- New seal wash solvent and bottle (10% Iso-Propanol in water)
- Flush out buffer with 1L of HPLC grade warm water
- Visual inspection of Solvent Filters
 - * Look for yellowing, broken and dirty **glass** filters, **replace** 'do not sonicate'
 - * SS Solvent Inlet Filters may be sonicated to clean



- Purge with composition of your application
- Run conditioning with composition of your application
- * Stainless Steel Solvent Inlet Filter Part #: 01018-60025

Purging the Pump:

The purging procedure below is described for the 1290 Infinity Binary Pump. It can be executed in an equivalent way for the 1290 Infinity Quaternary Pump. Recommended time to perform pump purging:

- After the pump has been primed for the first time.
- If the pump has been idle for a few hours or more (air may have diffused into the solvent lines).
- If the solvent reservoirs are refilled and the pump requires purging to fill the system with fresh solvent. If different solvents are to be used, ensure that the new solvent is miscible with the previous solvent and if necessary use an intermediate step with a co-miscible solvent. Isopropanol is often a good choice, check with a solvent miscibility table.

The inlet tubes to the pump should already be full of solvent. If the inlet tubes are partially or totally dry, follow the full priming procedure (see "Priming the Pump"). The purge valve allows both pump heads to be connected to waste at the same time. Purge each pump-head with 2.5 – 3ml/min for 5 minutes, until the fresh solvent reaches the pump-head and through the pump.

Conditioning the Pump:

The conditioning function is useful for removing small air bubbles inside the pump flow path. Condition the pump with a restriction in place that provides at least 200 bar of back pressure (> 500 bar is better). You may run conditioning with the normal load such as the column under conditions of the analysis. The conditioning process should decrease the pressure ripple over time. If the ripple remains unchanged and higher than approx 10% of the pressure; try using LabAdvisor to run the pump\system diagnostics to look for another cause.

Condition the pump if you see:

- Excessive pressure ripple
- Excessive composition ripple (baseline noise\mixing noise – noise level changes with the composition), when you are sure that the solvent type (aqueous/organic or specific solvent/solvent mix) is correctly set, and there is no evidence of out of specification leakage in the pump.

Conditioning may be necessary if the pump contains air:

- After running out of solvent
- After a long period of standby
- After service or repair

With the introduction of the G4220A pump, Agilent also introduced a mode of operation called "Fast defill", which makes the secondary piston the main piston responsible for delivery of solvent to the system. This leads to a problem getting air out of the primary chamber due to the short period of time it is under pressure. In order to compensate for this a different mode of operation called "Conditioning" was introduced. This makes the primary piston run at full stroke length thereby delivering the majority of time.

Additional Action to Help with ACN:

- Filter ACN using a 0.45um nylon filter depending on initial solvent quality. Not good for High Sensitivity LCMS.
- Put ACN in an amber bottle and keep amount of ACN in the bottle to a minimum
 - a. Only put on as much fresh ACN as will be needed for that sequence plus a 20-30% allowance for time issues. Remember to include in between run time into the solvent calculation.
 - b. Put on fresh ACN for every sequence and always use a clean bottle (Never add additional ACN into a bottle)
- Don't leave ACN on the system for more than a few days (2-3)
- If able, it helps to have 5-10% water in the ACN, especially with LCMS when 0.1% formic acid is present
- If system was shut down for more than a few days (2-3), flush the system with 10% Iso-Propanol in water
- Setting up and getting ready to start a new sequence
 - a. Put on fresh solvents in fresh clean bottles
 - b. Purge **each** channel individually for 5 minutes @ 2.5-3ml/min. each
 - c. Run conditioning - 15 minutes automated, requires a load, column or restriction capillary, backpressure must be greater than 200bar, even better over 500bar and can be done with customer mobile phases
- Perform a periodic warm water (60-70 deg C) wash - 1L pumped at 2ml/min. on a monthly basis depending on usage.

When turned on, the conditioning runs for 15 minutes or until "Conditioning off" is selected. The conditioning feature is a "not ready" condition and does not allow a run to be started when turned on.