F2 BioSAXS Late Night Software Restart Procedure

On data collection computer (Underline):

1) kill ADX – from ADX window push “EXIT” then select “stop all”.
2) Click on “Collect data (SAXS Pilatus) robot” icon.
3) Is there a TigerVNC remote desktop window open? If not, open a terminal window and type >vncviewer 192.168.0.72
4) close robocon window on remote netbook desktop (red X) and, if necessary, close terminal window underneath.
5) click on “robocon-Shortcut” icon to relaunch Robocon
6) reconfigure Robocon
   set target directory, filename, and exposure time. Re-enter your well volumes and labels, if needed.
7) open a Terminal window and set the Adxv viewers again:
   >adxvA&
   >adxvB&
Recovering from problems with BioSAXS at F2

A. Lost contact with robot (“Connection to Robot Failed”)

cause: unknown

solution: Restart the Robocon software. In the vnc viewer on Underline, close the Robocon window by clicking the x-button in the upper left of the window. Also close the black window that says "robot control." To re-launch Robocon, simply double click on the "Robot Control Software" (RoboCon shortcut) icon on the desktop. (See Restart Procedure)

It may also be necessary to disconnect and reconnect USB plugs into laptop. In rare cases, power-cycle the robot. **IMPORTANT: if RoboCon asks you to RESET motors, take the pipette tip off first.**

B. ADX/SPEC Busy - blinking red light won't go out, can't take snapshots, and can't find any ADX windows

cause: user probably typed a bogus file name (space in name, nonexistent folder, or lack of world write/execute permissions on folder)

solution: write down any critical settings in the Robocon GUI and do the emergency software restart procedure at the beginning of this document.
C. Connection to remote desktop with Robocon lost

*Cause:* internet cable to netbook in hutch may have come loose or user may have accidentally closed TigerVNC window.

*Solution:* check the green cable connection to the netbook in the hutch. From a terminal window on the data collection computer desktop, restart the vncviewer:

Example:

```
>vncviewer 192.168.0.72
```

D. Current images are being written to disk using *previous* file names.

*Cause:* probably collecting images while the synchrotron storage ring was being refilled.

*Solution:* kill ADX – from the proper ADX window push “exit” then select “stop all”. Click on “Collect data (SAXS Pilatus) robot” on desktop.

E. capillary video image seems very dim and fuzzy

*cause:* condensation on cell window

*solution:* check to make sure the nitrogen jet is blowing on the sample-cell vacuum window. Wipe window with a Q-tip if necessary.
F. liquid plug will not oscillate very much or stops oscillating in the middle of a run.

*cause:* liquid drops not fully cleared from tube entering waste vial.

*solution:* make sure the tube entering the waste vial is cleared of any hanging droplets and is touching the side of the glass vile.

G. bubbles appear in capillary when sample moves into position.

*cause:* blowoff value in Robocon may be set too high. This can be a problem when loading volumes so large that the pipette tip is submerged when the funnel is filled.

*solution:* Try setting blowoff to 5 or even 0.

H. One side of meniscus drags behind, causing sample to split as sample is positioned. Bubble is introduced as a result.

*cause:* damaged protein deposits forming on capillary surface or, in new capillaries, uneven surface phobicity.

*solution:* try the CLEAN procedure. If that doesn’t work, load a plug of 25 ul of Hellmanex (2%) and let it soak for 20 min at a warm temperature. You can also try a DDMAB (detergent) rinse.

I. Liquid suddenly refluxes within the capillary during what should be steady flow or cannot get liquid to suck into funnel at all (rare).

*cause:* Check the waste vial. If waste vial is full to overflowing then liquid has been sucked into pump causing failure. Cause may not be obvious.
solution: Install spare Aurora pump if available.

Note: pump solenoid valve can be cleaned or replaced if necessary.

J. **Buffer curves taken at beginning of run do not agree with those taken later in the run.**

cause: incomplete rinse or, more likely, deposition of damaged protein on capillary wall.

solution: try an aggressive CLEAN of capillary with detergent or Hellmanex. If this fails, tweak the sample capillary stage (samx) left or right by 500 microns (call Richard first). Otherwise, change capillary.

K. **Adxv image display window(s) gone**

cause: maybe software was restarted?

Solution: open a Terminal window on underline and type
>adxvA&
>adxvB&
Important F2 BioSAXS Reminders

- Avoid collecting data while the synchrotron storage ring is being filled. You are welcome to ask the CHESS operator for an extension on the run if you need a little extra time.

- *Use only allowed characters in file names.* Letters, numbers, underscores “_”, and dashes “-“ are ok. Spaces “ “, periods “.”, and other characters will cause trouble. Very long filenames will be truncated.

  **ok:** lysobuffer_3mg_01  
  **trouble:** lysobuffer 3.0 mg/ml

- Check waste vial and empty when full

- Make sure sample clears completely from tubing into waste vile before loading a new sample

- Use “Clean” regularly to prevent flow problems

- Keep a close eye on sample cell when oscillating sample plug to make sure sample does not drift out of beam

- Use the proper pipette tips to refill the robot! Use VRW brand 200 μl size NOT Rainin!

- Make sure to SAVE your profiles and .csv (Guinier spreadsheet) files.