

**Frederick National Laboratory
for Cancer Research**

sponsored by the National Cancer Institute

Vaccine, Immunity and Cancer Directorate
Standard Operating Procedure

SOP Title: Use and Maintenance of the Agilent Bioanalyzer

Document ID: 26023

Version

2.0

Page 1 of 16

Supersedes

1.0

Effective Date: 27Aug21

Written by:

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1. PURPOSE

- 1.1. The purpose of this procedure is to describe the general use and maintenance of the Agilent 2100 Bioanalyzer.

2. SCOPE

- 2.1. This procedure applies to all Agilent 2100 Bioanalyzers.

3. REFERENCES

- 3.1. Agilent 2100 Bioanalyzer User Manual
- 3.2. Agilent 2100 Expert Software Install User Manual
- 3.3. 10007: Non-Routine Equipment Maintenance
- 3.4. 10009: General Record Review
- 3.5. 15000: Waste Disposal at the Advanced Technology Research Facility
- 3.6. 26016: Operation, Use and Maintenance of the Water Purification Systems
- 3.7. 30011: Protein Analysis of Virus-Like Particles (VLPs) using the Agilent 2100 Bioanalyzer

4. RESPONSIBILITIES

- 4.1. The Research Associate, hereafter referred to as Analyst, is responsible for reviewing and following this procedure, and documenting performance of equipment maintenance.
- 4.2. The Scientific Manager or designee is responsible for training personnel in this procedure and reviewing associated documentation.
- 4.3. The Quality Assurance Specialist is responsible for quality oversight and approval of this procedure.
- 4.4. Trained personnel perform assay record review per "10009: General Record Review".

5. REAGENTS, MATERIALS, AND EQUIPMENT

- 5.1. Agilent 2100 Bioanalyzer
- 5.2. Chips, Protein 230 Kit (Agilent, Cat # 5067-1517)
- 5.3. Chips, RNA 6000 Nano Kit (Agilent, Cat # 5067-1511)

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- 5.4. Fuse, 1A 250V TD FE UL-L (Agilent, Cat # 2110-0007)
- 5.5. Isopropanol, Reagent-Grade
- 5.6. Kimwipes, Lint-Free Lens Wipe (VWR, Cat # 21905-026 or equivalent)
- 5.7. RNaseZap (Ambion, Inc Cat # 9780 or equivalent)
- 5.8. Type I Water
- 5.9. Soft brush (toothbrush)
- 5.10. Oil-free compressed air
- 5.11. Needle
- 5.12. Screwdriver or equivalent
- 5.13. Forceps
- 5.14. Gasket kit (Agilent, Cat # G2938-68716 or equivalent)

6. HEALTH AND SAFETY CONSIDERATIONS

- 6.1. Proper safety precautions should be taken while working in a laboratory setting. This includes, but is not limited to, proper protective equipment such as lab coats, safety glasses, closed-toe shoes, and non-latex gloves.
- 6.2. Refer to the respective Safety Data Sheet (SDS) when working with any chemicals.
- 6.3. Refer to "15000: Waste Disposal at the Advanced Technology Research Facility" regarding waste disposal processes at the Advanced Technology Research Facility (ATRF).
- 6.4. Never fill more than 350 µl of water in the electrode cleaner. Liquid spill may cause leak currents between the electrodes.
- 6.5. The Bioanalyzer should be kept clean using a lint-free cloth. Foreign materials in reagents, samples, or in chip wells will interfere with assay.
- 6.6. Do not touch the Bioanalyzer instrument during run and never place unit on a vibrating surface.
- 6.7. Do not leave the assay chip in the Bioanalyzer for longer than an hour (such as overnight) as the electrodes may get contaminated.

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- 6.8. Do not force Bioanalyzer lid closed after inserting chip as this can damage electrodes or result in liquid spills. If the lid does not close smoothly the chip is not properly seated.

7. DEFINITIONS

- 7.1. As Needed Maintenance – maintenance that is performed outside of routine maintenance but is not performed in response to equipment malfunction.
- 7.2. Non-Routine Maintenance – maintenance that is performed in response to equipment malfunction or failure.
- 7.3. Routine Maintenance – maintenance that is performed at planned intervals to identify and prevent problems before they result in equipment failure.
- 7.4. TOC – Total Oxidizable Carbon
- 7.5. Type I Water – Ultrapure/Reagent Grade/critical applications (Resistivity >18 MΩ-cm and TOC ≤ 50 ppb)

8. EQUIPMENT USE

- 8.1.1. Log into 2100 Expert Software Select appropriate Bioanalyzer for run under Instrument > All Instruments.
- 8.1.2. Open Bioanalyzer lid and seat chip securely so sample wells are located to the top right. The chip only fits within the receptacle in this position.
- 8.1.3. Close Bioanalyzer lid and software will show a graphical representation of the chip at the top left of the Instrument Context.
- 8.2. Starting Chip Run
- 8.2.1. Within Instrument Context menu select Assay Selection to choose assay.
- 8.2.2. Accept the default File Save location or update as needed.
- 8.2.3. Click Start in the upper right of the window.
- 8.2.4. Input your log in credentials to authorize the run.
- 8.3. Completing Chip Run
- 8.3.1. Run time will be 25 minutes for Protein 230 chips and 30 minutes for RNA 6000 chips.

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8.3.2. Once run has finished remove chip from Bioanalyzer.

8.3.3. Refer to 15000 for disposal of chip.

8.3.4. Clean Electrodes see 9.1.

9. MAINTENANCE

9.1. Cleaning Electrodes Daily During Use

9.1.1. Fill Electrode Cleaner (clear chip included in kit) with 350 µL of Type I Water for Protein 230 chips or RNaseZap for RNA 6000 chips.

Note: Never fill more than 350 µl of water in the electrode cleaner. Liquid spill may cause leak currents between the electrodes.

9.1.2. Open Bioanalyzer lid and seat Electrode Cleaner.

9.1.3. Close lid and leave it closed for approximately 10 seconds for Protein 230 chips or 1 minute for RNA 6000 chips.

9.1.4. Open lid and remove Electrode Cleaner.

9.1.5. Wait for approximately 10 seconds for electrodes to dry before closing the lid.

9.1.6. Empty the electrode cleaner after every cleaning procedure and refill the electrode cleaner.

9.1.7. If using RNA 6000 chips, repeat steps 9.1.1 – 9.1.5 as with Protein 230 chips using RNaseZap in a RNaseZap dedicated Electrode Cleaner.

Note: After 25 chip runs, the contents of a chip kit, replace the Electrode Cleaner with an Electrode Cleaner from a new chip kit.

9.2. Monthly Maintenance

9.2.1. Checking the Chip Priming Station for Proper Performance – Seal Test

9.2.1.1. Make sure the syringe is tightly connected to the chip priming station.

9.2.1.2. Pull the plunger of the syringe to the 1.0 mL position (plunger pulled back).

9.2.1.3. Place an unused chip in the chip priming station.

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9.2.1.4. Close the chip priming station. The lock of the latch will audibly click when it closes.

9.2.1.5. Press the plunger down until it is locked by the clip. This is shown in Image 8.

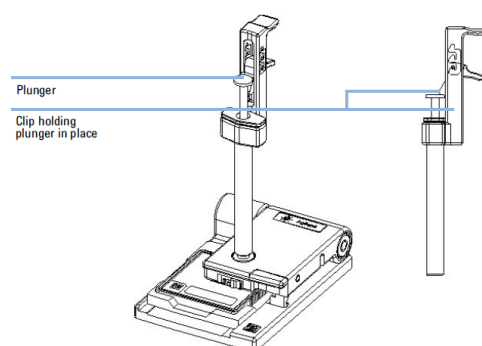


Figure 8 Locking the plunger of the syringe with the clip

9.2.1.6. Wait for 5 seconds and lower latch of the clip to release the plunger as shown in Image 9.

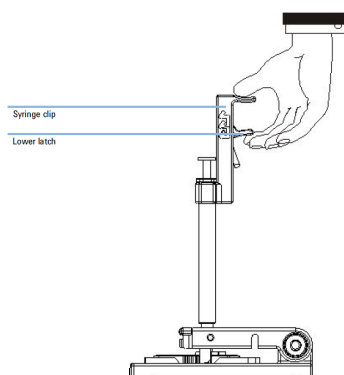


Figure 9 Releasing the plunger from the syringe

9.2.1.7. To indicate an appropriate sealing, the plunger should move back up at least to the 0.3 mL mark within less than 1 second.

9.2.1.8. If the plunger does not move up to the 0.3 mL mark within a second, the syringe-chip connection is probably not tight enough. Retighten the syringe or replace the syringe adapter, syringe or gasket to fix the problem.

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9.2.1.9. Document maintenance performed on “26023-01: Bioanalyzer Maintenance Form”.

9.2.2. Cleaning the Pin Set

9.2.2.1. Turn off power to the 2100 Bioanalyzer instrument.

9.2.2.2. Open the lid and pull the metal lever on the inside left of the lid to the vertical position as shown in Image 1. When the lever is in the vertical position, the cartridge is released from the lid by about 10 mm.

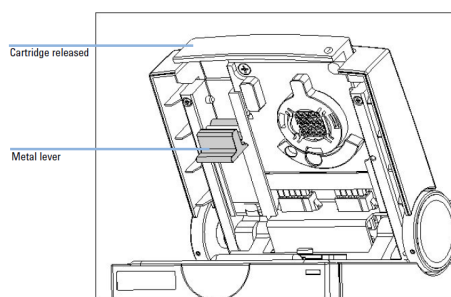


Figure 1 Remove/replace the electrode cartridge

9.2.2.3. Gently pull the cartridge out of the lid as shown in Image 1.

9.2.2.4. Open the bayonet socket of the pin set by turning the plastic lever to the left as shown in Image 2.

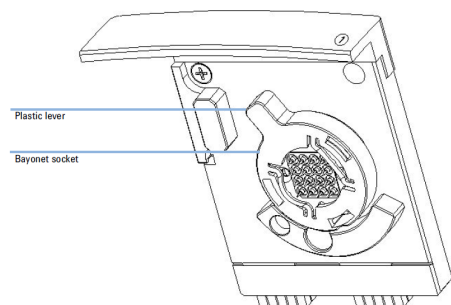


Figure 2 Bayonet socket of the electrode pin set

9.2.2.5. Remove the cover of the bayonet socket by gently pulling the plastic lever. The pin set may stick to the electrode base. Remove it by carefully pulling it off. See Image 3.

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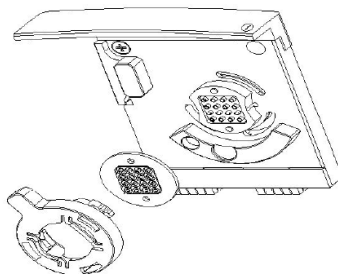


Figure 3 Bayonet cover and pin set

- 9.2.2.6. With soft brush gently clean pin set with deionized water or isopropanol when using Bioanalyzer for DNA and Protein Assays. In case of RNase contamination, use RNase Zap.

Note: Bending or misaligning the pins will lead to poor quality results or prematurely terminated assay runs.

Note: Make sure that the pin set is fully dry before placing back into the electrode base. Even small amounts of liquid on the pin set can damage the high voltage power supply.

- 9.2.2.7. Let the pin set completely dry in a desiccator overnight or use oil-free compressed air.
- 9.2.2.8. Place the pin set on the cartridge base and the bayonet cover over the pin set.
- 9.2.2.9. Lock the pin set to the electrode base by turning the plastic lever of the bayonet cover to the right.
- 9.2.2.10. Slide the electrode cartridge with the pin set into the 2100 Bioanalyzer instrument lid and move the metal lever to the flat (closed) position.
- 9.2.2.11. Push the metal front of the electrode cartridge to ensure a tight connection to the 2100 Bioanalyzer instrument.
- 9.2.2.12. To verify that the electrodes are completely dry, perform the Short circuit diagnostic test from the Diagnostics tab in the Instrument context. This test takes approximately three minutes.

Note: Heat can permanently damage the electrode cartridge. Do not dry the electrode cartridge in an oven.

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- 9.2.2.13. If the short circuit test fails, the electrode assembly may still be wet. Take the pin set out of the instrument, dry it with oil-free compressed air, then repeat the test.

Note: The limits of the short circuit test specify an ambient temperature of 25°C and relative humidity less than or equal to 50%. Higher temperatures and relative humidity could result in a leak current. Unused chip required.

- 9.2.2.14. Document maintenance performed on 26023-01.

9.3. Quarterly Maintenance

9.3.1. Lens cleaning

Note: Cleaning the lens is required every three months.

- 9.3.1.1. To clean the lens, switch the instrument off.
- 9.3.1.2. Open the lid of the instrument and wipe with a lint-free lens wipe dampened with reagent-grade isopropanol.
- 9.3.1.3. Gently swab the surface of the lens.
- 9.3.1.4. Allow the alcohol to evaporate prior to closing lid or use.

Note: Liquid dripping into the instrument could cause a shock or damage the instrument.

- 9.3.1.5. Use 26023-01 to record the maintenance performed.

9.3.2. Cleaning and Replacing the syringe adapter and gasket.

- 9.3.2.1. Open the priming station.
- 9.3.2.2. Move the mounting ring holding the adapter in place to the left as shown in Image 4. The ring will come off.

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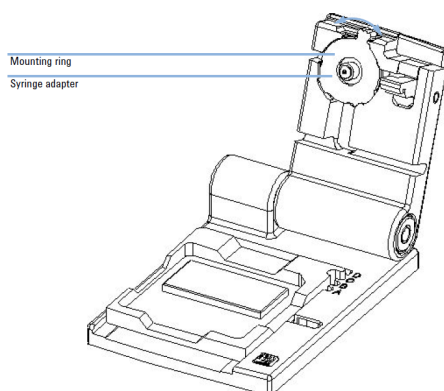


Figure 4 Mounting ring of the syringe adapter

9.3.2.3. Press the syringe adapter out of its mount as shown in Image 5.

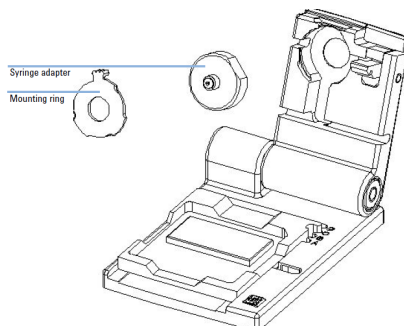


Figure 5 Removing/replacing the syringe adapter

- 9.3.2.4. Remove dried gel at the opening of the adapter with a needle.
- 9.3.2.5. If cleaning adapter, screw on syringe and flush water through the adapter several times.
- 9.3.2.6. If cleaning syringe adapter, flush syringe with isopropanol.
- 9.3.2.7. Allow adapter to dry fully.
- 9.3.2.8. Insert the cleaned syringe adapter or replace syringe adapter and gasket see Image 5 and 6.

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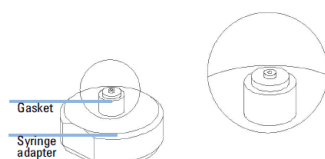


Figure 6 Syringe adapter with gasket

- 9.3.2.9. Pull out the old silicone gasket with fingers or forceps. See Image 7.

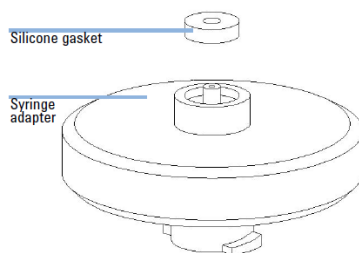


Figure 7 Syringe adapter with disassembled gasket

- 9.3.2.10. Assemble the priming station.
- 9.3.2.11. Close the chip priming station.
- 9.3.2.12. Screw a dry syringe tight into the luer lock adapter.
- 9.3.2.13. Perform a seal test described below in 9.2.1.

9.3.3. Replacing the syringe

- 9.3.3.1. Unscrew the old syringe from the top of the chip priming station.
- 9.3.3.2. Remove clip from the old syringe. Dispose syringe according to good laboratory practices.
- 9.3.3.3. Slide new syringe into the clip. Ensure syringe and clip are flushed together.
- 9.3.3.4. Screw the syringe tight into the luer lock adapter
- 9.3.3.5. Perform a seal test described in 9.2.1

9.4. As Needed Maintenance

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Note: Changing two fuses is required when the status indicator light is off and the cooling fan is not running.

9.4.1. To change the fuse, switch the instrument off then disconnect the power cord from the electrical socket.

9.4.2. Using a screw driver or other flat-edged tool, gently lift the outer plastic housing of the power inlet socket see Image 10.

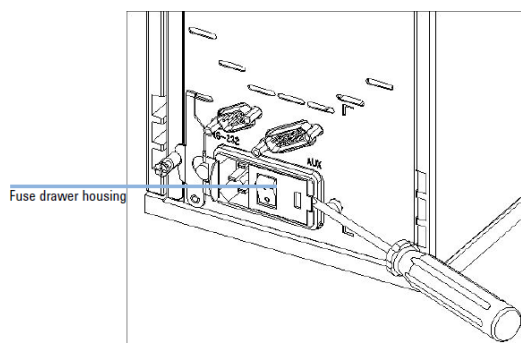


Figure 10 Remove power inlet housing

9.4.3. Pull out the fuse drawer, then replace the two fuses (see Image 12.)

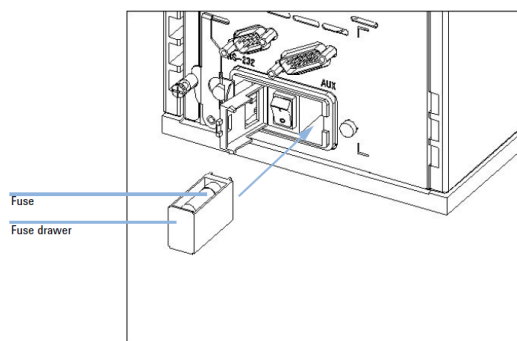


Figure 12 Insert fuse drawer

9.4.4. Return the fuse drawer and push until it fits tightly in the instrument.

9.4.5. Close the fuse drawer housing and reconnect the power cord.

9.4.6. Document maintenance performed on 26023-01.

9.5. Annual Calibration

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9.5.1. Facilities, Maintenance, and Engineering (FME) or a contracted vendor shall certify and perform annual calibration maintenance of the Agilent 2100 Bioanalyzer.

9.5.2. Print and file calibration report.

9.6. Non-Routine Maintenance

9.6.1. In the case that the equipment is not operating correctly, transition processes being performed to another unit (when applicable), post a sign stating the equipment is out of service and initiate non-routine maintenance documentation per "10007: Non-Routine Equipment Maintenance."

9.6.2. Document the nature of any failures or malfunctions, how and when it was discovered, and the personnel involved on "10007-01: Non-Routine Equipment Maintenance Form."

9.6.3. Initiate a service request and complete the non-routine maintenance process following 10007.

10. ATTACHMENTS

10.1. Attachment 1. 26023-01: Bioanalyzer Maintenance Form

11. REVISION HISTORY

Version	Change	Reason
1.0	Create new SOP for the Agilent 2100 Bioanalyzer.	New SOP.
2.0	<ol style="list-style-type: none">1. Section 2.1 removed ATRF2. Section 3 removed/updated references3. Section 4 record review added4. Section 5 added additional reagents5. Section 6 Added Safety Conditions6. Section 7 removed/updated definitions7. Section 8 Equipment use updated8. Section 9 Update Maintenance	<ol style="list-style-type: none">1. Clarification2. Reflect current practices3. Reflect current practices4. Reflect Manual Instructions5. Reflect Manual Instructions6. Reflect current practices7. Reflect current practices8. Reflect Manual Instructions

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Attachment 1. 26023-01: Bioanalyzer Maintenance Form

Frederick National Laboratory for Cancer Research <small>sponsored by the National Cancer Institute</small>		HPV Serology Laboratory Standard Operating Procedure Form	
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Maintenance Year:		Equipment ID:	HSL_
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Monthly Maintenance						
Month:	January	February	March	April	May	June
Seal Test (✓)						
Clean Pin Set (✓)						
Isopropanol Lot Number:						
Isopropanol Expiration Date:						
Performed By/Date:						
Reviewed By/Date:						

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Month:	July	August	September	October	November	December
Seal Test (✓)						
Clean Pin Set (✓)						
Isopropanol Lot Number:						
Isopropanol Expiration Date:						
Performed By/Date:						
Reviewed By/Date:						

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Quarterly Maintenance

Quarter:	Q1	Q2	Q3	Q4
Isopropanol Lot Number:				
Isopropanol Expiration Date:				
Performed By/Date:				
Reviewed By/Date:				

As Needed Maintenance: ☐ N/A

Date	Activity Performed	Performed by/date	Reviewed by/date
<input type="checkbox"/> N/A			
<input type="checkbox"/> N/A			

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