



# Intermountain Forensics

SOP # AMP-201

Revision # 01

Forensic DNA Technical Leader Approval

Issue Date

11/30/2020

## MiSeq FGx Operation

### 1. Purpose

This document describes setup and load of the Verogen MiSeq FGx


### 2. Summary

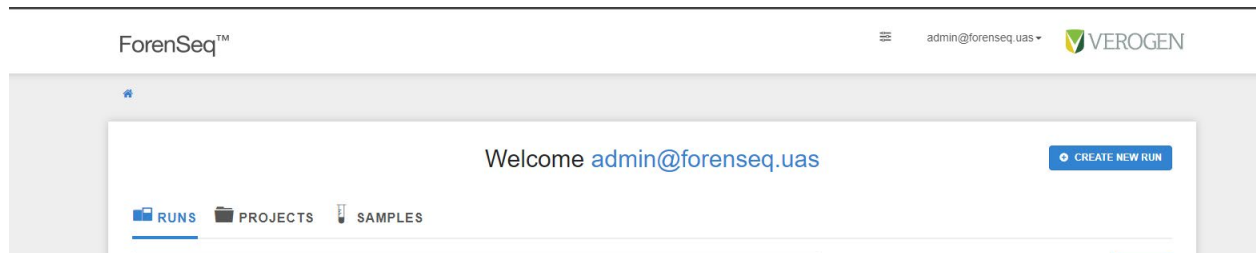
Next Generation Sequencing is the primary amplification assay of the IMF laboratory. The Verogen FGx comparison of evidentiary items with known profiles and potential upload to CODIS. This describes the setup and load of the Verogen FGx instrument.

NOTE: When utilizing the Verogen MiSeq FGx with the Verogen ForenSeq DNA Signature Prep kit, the laboratory will attempt to load 12 (or less) DNA containing samples if utilizing the MiSeq FGx Reagent Micro kit (Verogen Item #20021681) and 32 (or less) DNA containing samples if utilizing if the MiSeq FGx Reagent Kit (Verogen Item #15066817)

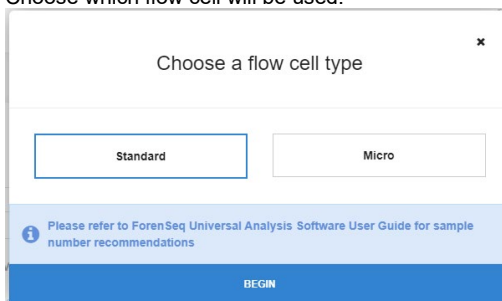
### 3. Procedure

#### UAS

1. Log into the Universal Analysis Software (UAS)
2. Select the Home icon  if the home screen is not displayed.



3. Select **CREATE NEW RUN**.
4. Choose which flow cell will be used.



5. Enter the run name.
6. Import sample information or enter sample information manually.
7. Click **SAVE RUN**

#### Instrument



# Intermountain Forensics

SOP # AMP-201

Revision # 01

Forensic DNA Technical Leader Approval

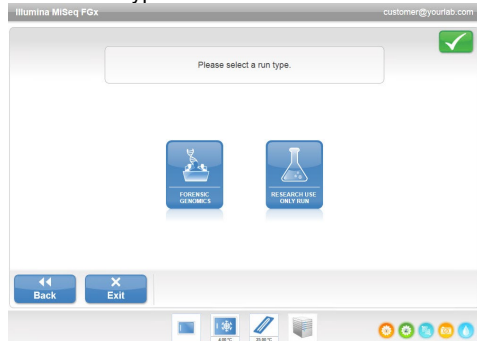
Issue Date

11/30/2020

## Log In and Select Run Type

- From the log in screen, enter your ForenSeq Universal Analysis Software username and password, and then select **Next**.
- From the Welcome screen, select **Sequence**.
- When the Select Run Type screen appears, select **Forensic Genomics**.

### Select Run Type



- Follow the prompts to load the flow cell and reagents and set up the run (described in the following sections).

## Cleaning the Flow Cell

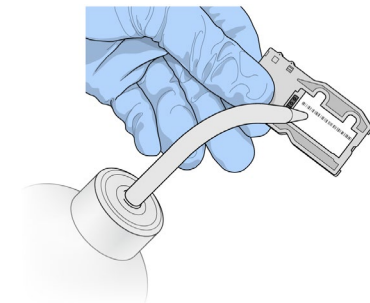
- The flow cell is immersed in storage buffer in a flow cell container. Remove it from the flow cell container.

### Remove Flow Cell



- Lightly rinse the flow cell with nuclease-free water, making sure that both the glass and plastic cartridge are thoroughly rinsed of excess salts. Excess salts can affect flow cell seating on the instrument. If salts dry in the imaging area, imaging can also be affected.

### Rinse Flow Cell



- Using care around the black flow cell port gasket (outlined in orange in the following illustration), thoroughly dry the flow cell and cartridge using a lint-free lens cleaning tissue. Gently pat dry in the area of the gasket and adjacent glass.



# Intermountain Forensics

SOP # AMP-201

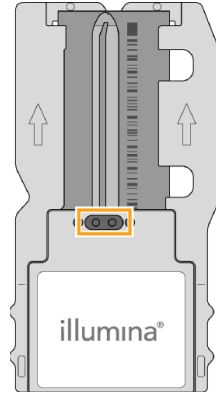
Revision # 01

Forensic DNA Technical Leader Approval

Issue Date

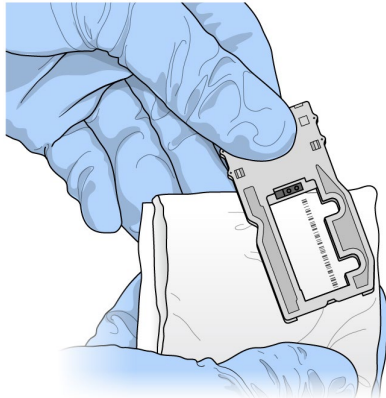
11/30/2020

Flow Cell Ports and Gasket



15. Make sure that the glass is free of streaks, fingerprints, and lint or tissue fibers.

Dry Flow Cell

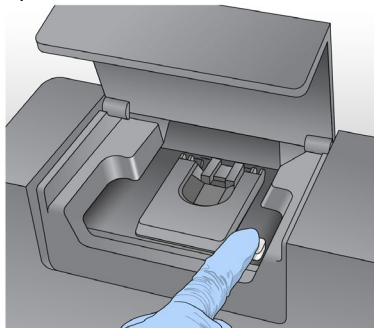


16. Visually inspect to make sure that the flow cell ports are free of obstructions and that the gasket is well-seated around the flow cell ports. If the gasket appears to be dislodged, gently press it back into place until it sits securely around the flow cell ports.

## Performing a Run

17. Raise the flow cell compartment door and press the release button to the right of the flow cell latch.

Open Flow Cell Latch





# Intermountain Forensics

SOP # AMP-201

Revision # 01

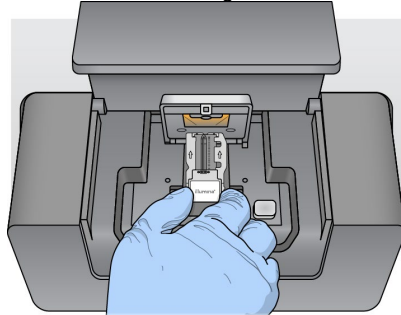
Forensic DNA Technical Leader Approval

Issue Date

11/30/2020

18. Visually inspect the flow cell stage to make sure that it is free of lint. If lint or other debris is present, clean the flow cell stage using an alcohol wipe or a lint-free tissue moistened with ethanol. Carefully wipe the surface of the flow cell stage until it is clean and dry.
19. Holding the flow cell by the edges of the flow cell cartridge, place the flow cell on the flow cell stage.

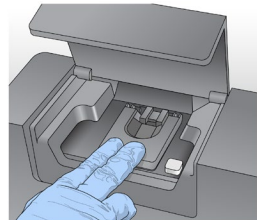
Place Flow Cell on Stage



20. Gently press down on the flow cell latch to close it over the flow cell.

**NOTE:** As the flow cell latch is closed, two alignment pins near the hinge of the flow cell latch properly align and position the flow cell. An audible click indicates that the flow cell latch is secure.

Close Flow Cell Latch



21. Check the lower-left corner of the screen to confirm that the flow cell RFID was successfully read.

**NOTE:** If the RFID cannot be read, identifying information can be entered manually. However, the software allows only one of the three RFID-labeled components (flow cell, reagent cartridge, or SBS Solution) to fail.

22. Close the flow cell compartment door.
23. Select **Next** on the Load Flow Cell screen. The Load Reagents screen opens.

## Loading Reagents

24. Load the SBS Solution (PR2) bottle and make sure that the waste bottle is empty, and then load the reagent cartridge.

### Load SBS Solution (PR2) and Check the Waste Bottle

25. Remove the bottle of SBS Solution (PR2) from 2° to 8°C storage. Gently invert the bottle to mix, and then remove the lid.
26. Open the reagent compartment door.
27. Raise the sipper handle until it locks into place.
28. Place the SBS Solution (PR2) bottle in the indentation to the right of the reagent chiller.

Load the SBS Solution (PR2) Bottle



# Intermountain Forensics

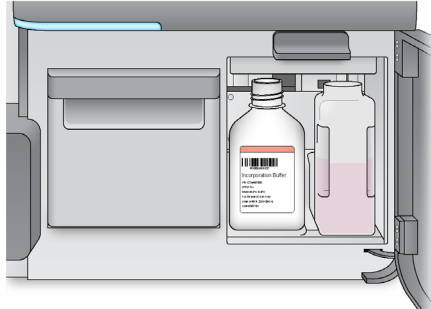
SOP # AMP-201

Revision # 01

Forensic DNA Technical Leader Approval

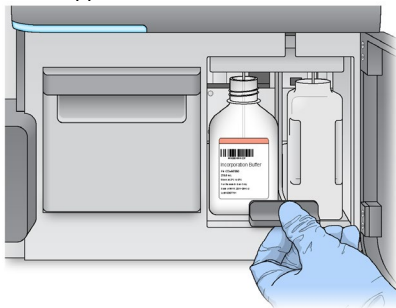
Issue Date

11/30/2020



29. Make sure that the waste bottle is empty. If it is not empty, empty the contents into the appropriate waste container.
30. Slowly lower the sipper handle. Make sure that the sippers lower into the SBS Solution (PR2) and waste bottles.

Lower Sipper Handle



31. Check the lower-left corner of the screen to confirm that the RFID of the SBS Solution (PR2) bottle was read successfully.

**NOTE:** If the RFID cannot be read, identifying information can be entered manually. However, the software allows only one of the three RFID-labeled components (flow cell, reagent cartridge, or SBS Solution) to fail.

32. Select **Next** on the Load Reagents screen.

## Load the Reagent Cartridge

**NOTE:** Do not leave the reagent chiller door open for an extended period of time.

33. Open the reagent chiller door.
34. Hold the reagent cartridge on the end with the Illumina label, and slide the reagent cartridge into the reagent chiller until the cartridge stops.

Load Reagent Cartridge





# Intermountain Forensics

SOP # AMP-201

Revision # 01

Forensic DNA Technical Leader Approval

Issue Date

11/30/2020

35. Close the reagent chiller door.
36. Check the lower-left corner of the screen to confirm that the RFID of the reagent cartridge was read successfully.

**NOTE:** If that the RFID cannot be read, identifying information can be entered manually. However, the software allows only one of the three RFID-labeled components (flow cell, reagent cartridge, or SBS Solution) to fail.

37. Close the reagent compartment door.
38. Select **Next** on the Load Reagents screen. The Review screen opens.

## Starting the Run

39. After loading the flow cell and reagents, review the run parameters and perform a pre-run check before starting the run.

### Review Run Parameters

40. Review Worklist Name, Analysis Workflow, and Read Length.
41. Select **Next**. The Pre-Run Check screen opens.

### Review Pre-Run Check

42. The system performs a check of all run components, disk space, and network connections before starting the run.

If any items do not pass the pre-run check, a message appears on the screen with instructions on how to correct the error.

When all items successfully pass the pre-run check, select **Start Run**.

**WARNING: The MiSeq FGx is sensitive to vibration. Touching the instrument after starting a run could adversely affect sequencing results.** After selecting **Start Run**, do not open the flow cell compartment or the reagent compartment doors, or touch the instrument monitor except to pause the run.

## 4. References

---

MiSeq-FGx-Reference Guide

## 5. Definitions

---

N/A