## SeqStudio<sup>™</sup> Genetic Analyzer Instrument and Software USER GUIDE

for use with: SeqStudio<sup>™</sup> Data Collection Software v1.1.4 SeqStudio<sup>™</sup> Genetic Analyzer Cartridge (Cat. No. A33671) SeqStudio<sup>™</sup> Genetic Analyzer Cartridge v2 (Cat. No. A41331) SeqStudio<sup>™</sup> Plate Manager SeqStudio<sup>™</sup> Remote Monitoring App

Publication Number MAN0016138 Revision B.0





For Research Use Only. Not for use in diagnostic procedures.

Manufacturer: Life Technologies Holdings Pte Ltd   Block 33   Marsiling Industrial Estate Road 3   #07-06, Singapore 739256	Products: SeqStudio <sup>™</sup> Genetic Analyzer SeqStudio <sup>™</sup> Data Collection Software SeqStudio <sup>™</sup> Plate Manager (desktop) SeqStudio <sup>™</sup> Genetic Analyzer Cartridge SeqStudio <sup>™</sup> Genetic Analyzer Cartridge v2
Manufacturer: Life Technologies Corporation   200 Oyster Point Blvd   South San Francisco, CA 94080   USA	<b>Products:</b> SeqStudio <sup>™</sup> Remote Monitoring App
Manufacturer: Life Technologies Corporation   35 Wiggins Avenue   Bedford, MA 01730	<b>Products:</b> SeqStudio <sup>™</sup> Genetic Analyzer Cathode Buffer Container

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#### Revision history: Pub. No. MAN0016138

Revision	Date	Description
B.0	18 March 2019	<ul> <li>Update for v1.1.4 of the SeqStudio<sup>™</sup> Data Collection Software.</li> <li>Add the SeqStudio<sup>™</sup> Genetic Analyzer Cartridge v2.</li> <li>Add information on Contiguous read length and Quality Values (QVs).</li> </ul>
A.0	19 October 2017	New document for v1.1 software. Quick references only ( MAN0016144 and MAN0017087) were provided for v1.0 software.

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## **Product information**

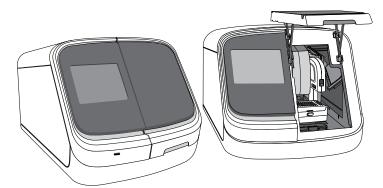
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#### Instrument overview

The Applied Biosystems<sup>™</sup> SeqStudio<sup>™</sup> Genetic Analyzer with SeqStudio<sup>™</sup> Data Collection Software is a fluorescent dye-labeled genetic analysis system using capillary electrophoresis technology. It enables both sequencing and fragment analysis applications without the need to switch polymer type or capillary array length.

The instrument uses a self-contained, replaceable cartridge with:

- A 4-capillary array
- A universal polymer capable of performing sequencing and fragment analysis
- A polymer delivery system (PDS)
- Anode buffer



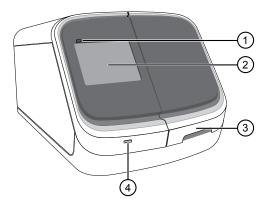
The SeqStudio<sup>™</sup> Genetic Analyzer automatically:

- Performs an optical alignment each time a cartridge is inserted.
- Performs an automatic spectral calibration adjustment (auto calibration) for each sample to correct for spectral overlap.

The instrument is compatible with 96-well Standard plates and 8-strip Standard tubes.

The SeqStudio<sup>™</sup> Genetic Analyzer is a stand-alone instrument. It is run directly from the touchscreen with SeqStudio<sup>™</sup> Data Collection Software and does not require a computer. Plate setup can be done directly on the touchscreen, on a computer with SeqStudio<sup>™</sup> Plate Manager, or on the Thermo Fisher Cloud. A run can be monitored directly on the instrument touchscreen or remotely on the Thermo Fisher Cloud.

#### Parts of the instrument



- **Figure 1** Front of the instrument
  - (1) Front panel indicator—Shows the status of the instrument
- Touchscreen—User interface
- (3) Door—Provides access to the cartridge, the cathode buffer, and sample plate or tubes
- ④ USB port

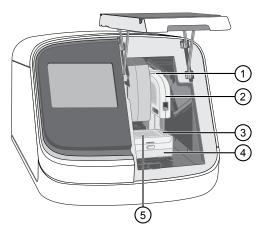


Figure 2 Interior of the instrument

- 1 Cartridge rails
- 2 Cartridge
- ③ Cathode buffer (located inside the autosampler)
- ④ Autosampler (contains the plate or tube holder and the cathode buffer)
- (5) Plate or tube holder

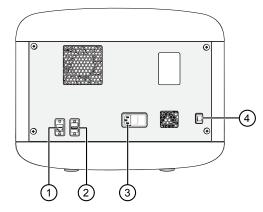


Figure 3 Rear of the instrument

- (1) RJ45 ethernet port
- 2 USB port
- ③ Power receptacle
- ④ On/Off switch

#### Instrument status indicator

Indicator	Instrument status
All lights off	Powered off or in <b>Cartridge storage mode</b> .
Blue light (blinking)	Starting up.
Blue light	Ready to start a run or run is in progress.
Amber light (blinking)	Run is paused, door is open, or error state.

## SeqStudio<sup>™</sup> Genetic Analyzer consumables

Description	Cat. No.	Amount	Storage
SeqStudio <sup>™</sup> Genetic Analyzer Cartridge	A33671	1 cartridge with an optical cover and a SeqStudio <sup>™</sup> Integrated Capillary Protector attached for shipment and storage.	See Table 1 on page 17
SeqStudio <sup>™</sup> Genetic Analyzer Cartridge v2	A41331	1 cartridge with an optical cover and a SeqStudio <sup>™</sup> Integrated Capillary Protector attached for shipment and storage.	See Table 2 on page 18
SeqStudio <sup>™</sup> Genetic Analyzer Cathode Buffer Container	A33401	1 package of 4	See page 149
SeqStudio <sup>™</sup> Integrated Capillary Protector	A31923	1 (single-use) for future storage	See page 148



## **Cartridge** The SeqStudio<sup>™</sup> Genetic Analyzer uses a 4-capillary, self-contained, replaceable cartridge. The following cartridges are available:

Cartridge	No. of injections	No. of samples	Storage
SeqStudio <sup>™</sup> Genetic Analyzer Cartridge (Cat. No. A33671)	125 injections	500 samples	See Table 1 on page 17
SeqStudio <sup>™</sup> Genetic Analyzer Cartridge v2 (Cat. No. A41331)	250 injections	1,000 samples	See Table 2 on page 18

Each cartridge:

- Contains a capillary array, a polymer delivery system, polymer, and anode buffer. See Figure 4 and Figure 5.
- Is shipped with an optical cover and an Integrated Capillary Protector. See Figure 6.
- Has a radio frequency identification (RFID) tag, which is used by the instrument to track remaining usage and expiration.

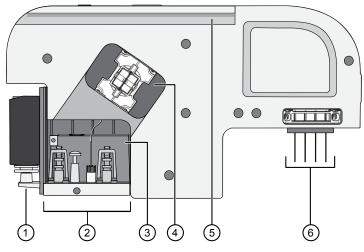


Figure 4 Parts of the cartridge

- ① Universal polymer—Supplies polymer to the polymer delivery pump.
- 2 Polymer delivery system—Pumps polymer into the capillary array.
- ③ Anode buffer reservoir—Genetic Analysis running buffer to support electrophoresis.
- (4) Optical detection window
- (5) Cartridge track—To insert the cartridge into the instrument.
- (6) Capillary array (cathode end)—Tips of the four capillaries that enable electrophoretic separation of fluorescent-labeled DNA fragments.

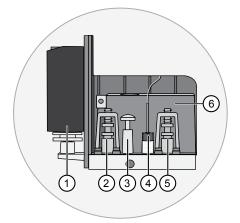


Figure 5 Parts of the polymer delivery system

- (1) Universal polymer
- 2 Polymer valve
- ③ Syringe
- (4) Capillary fitting (anode end)
- 5 Buffer valve
- 6 Anode buffer reservoir

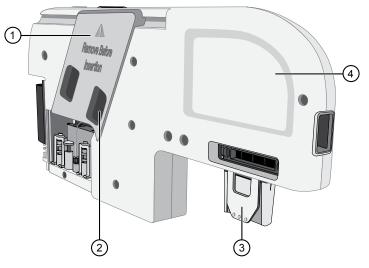


Figure 6 Cartridge with optical cover and Integrated Capillary Protector

- 1 Optical cover
- (2) Optical cover hand hold (for removing the optical cover)
- ③ Integrated Capillary Protector
- (4) Cartridge hand hold

**IMPORTANT!** Remove the Integrated Capillary Protector before installing the cartridge into the instrument. Installing the cartridge with the ICP in place can damage the capillary array.



#### Cartridge storage

 Table 1
 Storage information for the SeqStudio<sup>™</sup> Genetic Analyzer Cartridge (Cat. No. A33671)

Condition	Description
Shipping	Shipped at 2–8°C.
	Store upright at 2–8°C upon receipt.
	Save the white storage box and optical cover for off-instrument cartridge storage.
On-instrument storage	For routine use, can be used and stored on the instrument for up to 4 months. If you store the cartridge on-instrument:
	The instrument must be powered on.
	A Cathode Buffer Container must also be installed.
	The instrument keeps the components under the following conditions when it is powered on and in <b>Cartridge storage mode</b> :
	Optical detection window—Covered
	• <b>Capillary array electrodes</b> —Submerged in cathode buffer (buffer must be above the fill line in the Cathode Buffer Container)
	Polymer—Chilled
	Anode buffer—Ambient temperature
	<b>IMPORTANT!</b> The instrument does not maintain the correct temperature conditions for the cartridge when it is powered off. Avoid cartridge exposure to ambient temperature.
Off-instrument storage	For intermittent use, can be stored off-instrument until the expiry date on the label or up to 4 months after first use. Store upright at 2–8°C, with an integrated capillary protector (ICP) and optical cover installed (see "Store the cartridge" on page 146).
	<b>Note:</b> After you remove the cartridge from the instrument, install an ICP within a few minutes. Avoid cartridge exposure to ambient temperature.
Reuse	Can be removed from an instrument then inserted again on the same instrument or a different instrument, if it was stored properly at 2–8°C and has not expired or exceeded 125 injections.
	Information about the cartridge installation and usage is retained in the cartridge history ( <b>Settings &gt; Cartridge &gt; Instrument-cartridge history</b> ).



		Th		
Table 2	Ctorogo information f	on the Ceartudie	Constic Analyzan	Contridge v2 ICot No. A/12211
	Storaue iniornation in		Genetic Analyzer	Cartridge v2 (Cat. No. A41331)
	J			

Condition	Description
Shipping	Shipped at 2–8°C.
	Store upright at 2–8°C upon receipt.
	Save the white storage box and optical cover for off-instrument cartridge storage.
On-instrument storage	For routine use, can be used and stored on the instrument for up to 6 months. If you store the cartridge on-instrument:
	• The instrument must be powered on.
	A Cathode Buffer Container must also be installed.
	The instrument keeps the components under the following conditions when it is powered on and in <b>Cartridge storage mode</b> :
	Optical detection window—Covered
	• <b>Capillary array electrodes</b> —Submerged in cathode buffer (buffer must be above the fill line in the Cathode Buffer Container)
	Polymer—Chilled
	Anode buffer—Ambient temperature
	<b>IMPORTANT!</b> The instrument does not maintain the correct temperature conditions for the cartridge when it is powered off. Avoid cartridge exposure to ambient temperature.
Off-instrument storage	For intermittent use, can be stored off-instrument until the expiry date on the label or up to 6 months after first use. Store upright at 2–8°C, with an integrated capillary protector (ICP) and optical cover installed (see "Store the cartridge" on page 146).
	<b>Note:</b> After you remove the cartridge from the instrument, install an ICP within a few minutes. Avoid cartridge exposure to ambient temperature.
Reuse	Can be removed from an instrument then inserted again on the same instrument or a different instrument, if it was stored properly at 2–8°C and has not expired or exceeded 250 injections.
	Information about the cartridge installation and usage is retained in the cartridge history ( <b>Settings &gt; Cartridge &gt; Instrument-cartridge history</b> ).



#### SeqStudio<sup>™</sup> Genetic Analyzer Cathode Buffer Container

The SeqStudio<sup>™</sup> Genetic Analyzer Cathode Buffer Container (CBC) contains running buffer for capillary electrophoresis. The container has two compartments. The rear compartment provides the cathode buffer for electrophoresis. The front compartment is for capillary wash and waste.

The CBC requires a Reservoir Septa.

See "Assemble the SeqStudio<sup>™</sup> Genetic Analyzer Cathode Buffer Container (CBC)" on page 149.

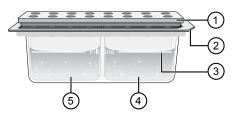


Figure 7 SeqStudio<sup>™</sup> Genetic Analyzer Cathode Buffer Container with Reservoir Septa (1) Reservoir Septa

- Notch (inserted towards the rear right in the autosampler)
- (3) Fill line (replace the CBC when the buffer is at the fill line)
- (4) Cathode buffer compartment
- (5) Waste and wash compartment

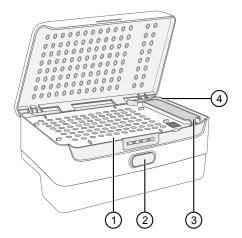


Figure 8 Autosampler

- (1) Location of plate or tubes
- 2 Release button
- (3) Location of Cathode Buffer Container
- (4) Location to position the Cathode Buffer Container notch

## Radio frequency identification

The cartridge and cathode buffer have radio–frequency identification (RFID) tags. The instrument reads and tracks:

- Expiry date (shelf life)
- Remaining injections (usage)
- Serial number (cartridge only)
- Lot number (cathode buffer only)



Component	RFID read	RFID write	Consumables status update
SeqStudio <sup>™</sup> Genetic Analyzer Cartridge SeqStudio <sup>™</sup> Genetic Analyzer Cartridge v2	When a cartridge inserted. The first time a new cartridge is loaded into an instrument, the manufacturer expiry date is listed for <b>Expiration</b> <b>time</b> in the <b>Consumable</b> <b>status</b> screen.	<ul> <li>The Expiration time is reset from the manufacturer expiry date to 4 months from the current date of installation.</li> <li>After the cartridge has been loaded and before it is ejected, its cartridge history record is updated to include its usage on this instrument.</li> <li>During each injection, injection count and remaining polymer volume are updated.</li> <li>The Expiration time is reset from the manufacturer expiry date to 6 months from the current date of installation.</li> <li>After the cartridge has been loaded and before it is ejected, its cartridge history record is updated to include its usage on this instrument.</li> <li>During each injection, injection count and remaining polymer</li> </ul>	<ul> <li>Every 8 hours and/or before each run<sup>[1]</sup>:</li> <li>When a cartridge is within 2 weeks of expiry date.</li> <li>When the number of injections is approaching the limit of 250 injections.</li> </ul>
Cathode buffer	<ul> <li>When the autosampler initializes.</li> <li>When a plate is retracted.</li> </ul>	<ul> <li>volume are updated.</li> <li>The first time a new CBC is loaded on an instrument, the installation date is recorded on the CBC.</li> <li>After each injection, the injection count is updated.</li> </ul>	<ul> <li>Every 8 hours and/or before each run<sup>[1]</sup>:</li> <li>When the CBC is within 2 days of expiry date.</li> <li>When the number of injections is approaching the limit of 125 injections.</li> </ul>

#### Table 3 RFID tag read/write events and consumables status updates

<sup>[1]</sup> If either limit is met, an email notification is also sent to any Cloud users who are linked to this instrument

#### Important notice regarding use of consumables that exceed supported limits

#### BEFORE DISMISSING THE WARNING THAT THE CONSUMABLES HAVE REACHED SUPPORTED LIMITS AND CONTINUING WITH OPERATION OF THE INSTRUMENT, PLEASE READ AND UNDERSTAND THE FOLLOWING IMPORTANT NOTICE AND INFORMATION:

Life Technologies does not recommend the use of consumables that exceed supported limits. The recommended limits are designed to promote the production of high quality data and minimize instrument downtime. Reagent and consumable lifetime minimum performance are based on testing and studies that use reagents and consumables that have not exceeded supported limits.

The use of consumables beyond the supported limits may impact data quality or cause damage to the instrument or capillary array. The cost of repairing such damage is NOT covered by any Life Technologies product warranty or service plan. Customer



use of expired consumables is at customer's own risk and without recourse to Life Technologies. For example, product warranties do not apply to defects resulting from or repairs required due to misuse, neglect, or accident including, without limitation, operation outside of the environmental or use specifications or not in conformance with Life Technologies instructions for the instrument system, software, or accessories.

Please see your specific service contract or limited product warranty for exact language regarding coverage and ask your Life Technologies representative if you have further questions.

#### Software features

The instrument can be operated directly from the touchscreen using the SeqStudio<sup>™</sup> Data Collection Software. The touch screen allows scrolling and zooming by pinch.

Plates can be set up and saved on a computer using the SeqStudio<sup>™</sup> Plate Manager running on a desktop or on the Thermo Fisher Cloud. These plate setups can be saved to and accessed from the instrument from:

- A network drive
- A USB drive
- The Thermo Fisher Cloud

You can monitor runs directly from the instrument or from the SeqStudio<sup>™</sup> Remote Monitoring App.

Feature	SeqStudio <sup>™</sup> Data Collection Software	SeqStudio <sup>™</sup> Plate Manager (desktop)	SeqStudio <sup>™</sup> Plate Manager (Cloud)	Remote Monitoring App
Plate setup				
Create a new plate setup	$\checkmark$	$\checkmark$	$\checkmark$	_
Enter plate properties	$\checkmark$	$\checkmark$	$\checkmark$	_
Set up plate wells	$\checkmark$	$\checkmark$	$\checkmark$	_
Advanced options	for plate setup			
Edit sample properties	$\checkmark$	$\checkmark$	$\checkmark$	_
Edit plate setup	$\checkmark$	$\checkmark$	$\checkmark$	_
Manage size standards	$\checkmark$	$\checkmark$	$\checkmark$	_

#### Table 4 Features of the software



Feature	SeqStudio <sup>™</sup> Data Collection Software	SeqStudio <sup>™</sup> Plate Manager (desktop)	SeqStudio <sup>™</sup> Plate Manager (Cloud)	Remote Monitoring App
Manage run modules (including edit a run module)	~	$\checkmark$	$\checkmark$	_
Advanced options	for plate propertie	S		
Adjust fragment analysis parameters	$\checkmark$	$\checkmark$	$\checkmark$	_
Adjust sequence parameters	$\checkmark$	$\checkmark$	$\checkmark$	_
Adjust the file naming format	$\checkmark$	$\checkmark$	$\checkmark$	_
Select injection options	$\checkmark$	$\checkmark$	$\checkmark$	_
Create custom dye set	~	— (to import, open a plate setup containing a custom dye set)	— (to import, open a plate setup containing a custom dye set)	_
Edit during a run	or after a run is cor	nplete		
Monitor run	~	_	_	$\checkmark$
Pause or cancel a run	$\checkmark$	_	_	$\checkmark$
Edit injection parameters and re-inject samples	$\checkmark$	_	_	$\checkmark$
View results				
View run result details	$\checkmark$	_	_	$\checkmark$
View PUP score	$\checkmark$	_	_	$\checkmark$
View trace score (sequence analysis only)	~	_	_	$\checkmark$
View size quality (fragment analysis only)	$\checkmark$	_	_	$\checkmark$

	ж		
	112	18	
	84	ε.,	

Feature	SeqStudio <sup>™</sup> Data Collection Software	SeqStudio <sup>™</sup> Plate Manager (desktop)	SeqStudio <sup>™</sup> Plate Manager (Cloud)	Remote Monitoring App
View Contiguous Read Length (sequence analysis only)	~	_	_	$\checkmark$
View and export r	esults			
View real-time results	~	_	_	$\checkmark$
Adjust the graphical view	~	_	—	$\checkmark$
Auto export or manually export data files to the Thermo Fisher Cloud, a network drive, or a USB	✓ 	_	_	_
Export a results report	~	_	_	$\checkmark$

## **SeqStudio**<sup>™</sup> **Plate Manager overview**

The SeqStudio<sup>™</sup> Plate Manager is a stand-alone software. It allows you to set up and save plates that you can open and run on the instrument.

The SeqStudio<sup>™</sup> Plate Manager is available:

- On the Thermo Fisher Cloud as an app, with access to the SeqStudio<sup>™</sup> Remote Monitoring App
- At thermofisher.com, for download and installation on a computer
- On a USB, for installation on a computer

Install the Plate Manager by following the instructions in the install wizard.



rify	and	Assig	in Sa	ampl	es									 ≣	Actions
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1	2	3 4	5	6	7	8	9	10	11	12	Injec	tion Gro	pup		
$\square$		76	) É		$\square$	$\bigcirc$		$\square$	$\bigcirc$	$\bigcirc$	Well #	ŧ	Sample name	Samp	le type
X		56			X	X	X	X	X	g	A1	A1			
	IQ(	JL	JĻ,	)Q	IQ	Q	IQ	IQ	Q	Q	B1	B1			٧
-	M	76	hr		M	A		m	$\overline{\Box}$	$\overline{\Box}$	C1	C1			
Ý		25	12	19	P	9			Y	9	D1	D1			
		DC													
X		56			R	X	Ĩ	K	X	X					
Ļ	IQ (	JL	JL.	JQ	IQ	Q	IQ	IQ	Q	Q					
5	$\square$	76	76		$\square$	$\square$			$\square$	$\bigcirc$	Siz	ze			~
4		25	12	P	Y	Y	Y	Y	Y	¥.	sta	andard			~
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ž		57			F	F	F	F	F	Ä	Dy	e set			~
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											Ri	ın mod	ule		

Figure 9 Plate Manager on Cloud or desktop

Computer and
operating system
options

- PC with Windows<sup>™</sup> 7
- PC with Windows<sup>™</sup> 10
- $Macintosh^{TM}$  with OS X •

Recommended browsers

- The following browsers are recommended to use the Plate Manager app on Thermo Fisher Cloud or a desktop computer:

  - ⊘ Apple<sup>™</sup> Safari<sup>™</sup> Version 7+ ٠
  - $\bigcirc$  Microsoft<sup>TM</sup> Edge 10+ (Windows<sup>TM</sup> 10)
  - Microsoft<sup>™</sup> Internet Explorer<sup>™</sup> 11 (Windows<sup>™</sup> 7) •

The use of other browsers or other versions can result in reduced functionality and improper display of information.

## $\mathbf{SeqStudio}^{^{\mathrm{M}}} \mathbf{Remote} \ \mathbf{Monitoring} \ \mathbf{App} \ \mathbf{overview}$

The Remote Monitoring App allows you to monitor the status of instrument runs from a remote location.

The Remote Monitoring App is available:

- On the Thermo Fisher Cloud as an app, with direct access from the Plate Manager or the InstrumentConnect.
- At thermofisher.com, for download and installation on a mobile device.

RM	Run	
sec	studio-CK is running	Actions 🗸
	and the second se	Plate_20170804_223088
	end ~U7:38 AM	1 2 3 4 5 6 7 8 9 10 11 12 A O O O O O O O O O O O O O O O O O O O
2 of 2	4 injection completed	
1	Injection Group : S : A1-D1 ● ● ● ● ■ Run module: ShortSeq, Dye Set: E_BigDye <sup>™</sup> Terminator v1.1	
2	Injection Group : S : E1-H1 ● ● ● ● Run module: ShortSeq, Dye Set: E_BigDye™ Terminator v1.1	G
3	Injection Group : S : A2-D2 Running Run module: ShortSeq, Dye Set: E_BigDye™ Terminator v1.1	
4	Injection Group : S : E2-H2 ★ Queued Run module: ShortSeq, Dye Set: E_BigDye™ Terminator v1.1	Quality Legend     Good     Caution     Alarm       Dye set     D     E
5	Injection Group : S : A3-D3 ★ Queued Run module: ShortSeq, Dye Set: E_BigDye™ Terminator v1.1	Run module FragAnalysis ShortSeq

Figure 10 Remote Monitoring App on Cloud



#### **Network connection options**

The SeqStudio<sup>™</sup> Genetic Analyzer can be connected to a network or computer in the following configurations:

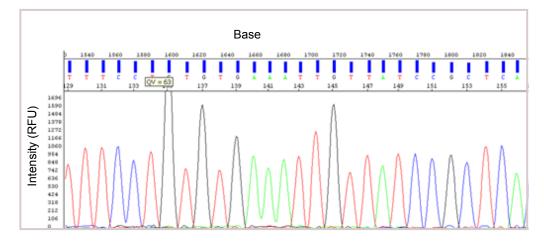
Direct connection	Local area network (LAN) connection	Thermo Fisher Cloud connection
Wired	Wired or wireless	Wired or wireless

#### **Experiment types**

#### Sequencing

Sequencing is the determination of the base-pair sequence of a DNA fragment by the formation of extension products of various lengths amplified through PCR.

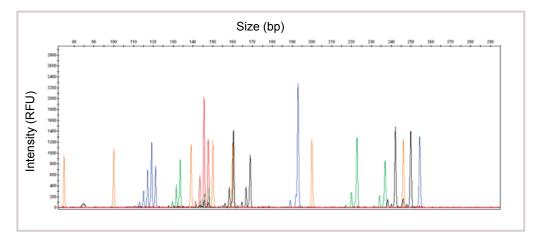
For more information, see *DNA Sequencing by Capillary Electrophoresis Chemistry Guide Second Edition* (Pub. No. 4305080).



#### **Fragment analysis**

Fragment analysis is the determination of the size of fragments. It uses the size standard in each sample to create a standard curve for each sample. It then determines the relative size of each dye-labeled fragment in the sample by comparing fragments with the standard curve for that specific sample.

For more information, see *DNA Fragment Analysis by Capillary Electrophoresis User Guide* (Pub. No. 4474504).



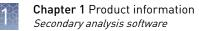
#### Data output

SeqStudio<sup>™</sup> Data Collection Software generates an electropherogram (intensity plot) for each dye that is based on the migration of DNA fragments through the capillaries during a run.

The format of the sample data files is determined by the type of experiment that is specified at the time of plate setup.

- Sequencing experiments use basecalling (the algorithms and settings required to determine the fragment base sequences) and generate an AB1 file.
- Fragment analysis experiments use sizecalling (the algorithms and settings required to determine the fragment sizes) and generate an FSA file.

The AB1 and FSA file formats can be analyzed by secondary analysis software.



#### Secondary analysis software

Secondary analysis software is available for desktop computers and on the Thermo Fisher Cloud.

Visit **thermofisher.com/cloud** for the latest available secondary analysis applications.

**Note:** Data from the SeqStudio<sup>™</sup> Genetic Analyzer may be labeled as "3200" in secondary analysis software.

Analysis	Арр	Description
Sequencing	Quality Check (QC) module	<ul> <li>Automatically checks sequence trace quality.</li> <li>Provides a results summary that is based on quality parameter settings.</li> <li>Auto-flags lower-quality traces for further inspection.</li> </ul>
	Variant Analysis (VA) module	<ul> <li>Finds variants in samples that are sequenced on Applied Biosystems<sup>™</sup> genetic analyzers.</li> <li>Reports variants at genomic coordinates.</li> <li>Allows export of variant calls in standard Variant Call Format.</li> </ul>
	Next-generation Confirmation (NGC) module	<ul> <li>Confirms next-generation sequencing (NGS) variants using CE technology.</li> <li>Allows visualization of the variants that are detected by both NGS and CE platforms.</li> <li>Allows export of confirmed variants in standard Variant Call Format.</li> </ul>
Fragment analysis	Sizing Analysis Module Peak Scanner <sup>™</sup> Software	Performs peak sizing.

#### Thermo Fisher Cloud secondary analysis apps

#### Desktop secondary analysis software

**IMPORTANT!** Older versions of the desktop secondary analysis software cannot analyze data files generated by the SeqStudio<sup>™</sup> Genetic Analyzer. Contact Support for information on obtaining the latest versions of software.

Analysis	Software	Minimum version required
Sequencing	Sequencing Analysis Software	6.2
	SeqScape <sup>™</sup> Software	3.2
	Variant Reporter <sup>™</sup> Software	2.2
	Minor Variant Finder Software	1.2
Fragment analysis	GeneMapper <sup>™</sup> Software	5.1



#### Workflow: Cloud or desktop

#### Get started

Prepare the instrument (page 38)

Prepare the samples (page 36)

#### Create a plate setup on the Cloud or desktop

Access the Plate Manager on the Cloud (page 55)

Access the Plate Manager on the desktop (page 55)

Create or open a plate setup PSM file (page 55)

Enter plate properties (page 55)

Assign wells: Sample and run information (page 57)

#### Start and monitor a run

On the instrument: Load the plate or the tube assembly [page 76]

Select a plate setup and start a run (page 77)

Monitor a run from the Thermo Fisher Cloud (page 79)

Monitor a run from a mobile device (page 87)

Monitor a run from the instrument (page 89)

#### View and analyze results

View results in the Remote Monitoring App on the Cloud (page 83)

View results on the instrument (page 95)

Export results from the instrument (sample data files and QC reports) (page 104)

Analyze data in a secondary analysis software application



#### Workflow: instrument

#### Get started

Prepare the instrument (page 38)

Prepare the samples (page 36)

#### Create a plate setup on the instrument

Create or import a plate setup (page 66)

Enter plate properties (page 67)

Assign wells: run module, size standard, and dye set (page 68)

Assign wells: sample name, sample type, and custom fields (page 70)

#### Start and monitor a run

On the instrument: Load the plate or the tube assembly (page 76)

Select a plate setup and start a run (page 77)

Monitor a run from the instrument (page 89)

#### View and analyze results

View results on the instrument (page 95)

Export results from the instrument (sample data files and QC reports) (page 104)

Analyze data in a secondary analysis software application



# Prepare the samples and the instrument

Precautions for use	32
Power on the instrument	33
Sign in	33
Sign in with the Guest instrument profile	34
Sign out	34
Parts of the home screen	35
Prepare the samples	36
Prepare the instrument	38

#### **Precautions for use**

**CAUTION!** PHYSICAL INJURY HAZARD. Do not remove the instrument cover. There are no components inside the instrument that you can safely service yourself. If you suspect a problem, contact technical support.



**CAUTION!** Moving parts.



**CAUTION! FIRE HAZARD**. For continued protection against the risk of fire, replace fuses only with listed and certified fuses of the same type and rating as those currently in the instrument.

<u>\$\$</u>

**CAUTION!** Hot surface.



**CAUTION!** Piercing hazard.



**CAUTION!** Potential biohazard.

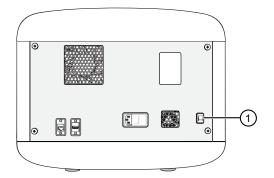


**CAUTION!** Risk of electrical shock.

#### Power on the instrument

**IMPORTANT!** Do not power on the instrument until it has been installed and set up by a Thermo Scientific<sup>M</sup> representative.

Press the On/Off switch on the rear panel.



**IMPORTANT!** The instrument does not maintain the correct temperature conditions for the cartridge when it is powered off. Avoid cartridge exposure to ambient temperature.

(1)  $_{\rm On/Off\ switch}$ 

#### Sign in

- 1. If the another user is signed in, touch (2) in the home screen, then touch **Sign out**.
- **2.** In the **Sign in** screen, touch **Sign In**, then select your instrument profile and enter your PIN.

**Note:** If the instrument is left unattended for 120 minutes, the instrument profile is signed out.

Sign	In
Sign in	New to this instrument?
admin 🗸	Connect to the Thermo Fisher Cloud or create a local instrument profile.
PIN	
Sign in	Get started

SeqStudio<sup>™</sup> Genetic Analyzer Instrument and Software User Guide



### Sign in with the Guest instrument profile

In the home screen:

- 1. In the Sign In screen, touch Sign In.
- 2. Touch Guest.

#### Sign out

In the home screen:

- **1.** Touch **()**.
- 2. Touch Sign out.

**Note:** If a run is in progress, **Lock the instrument** is displayed instead of **Sign out**.

3. Touch Yes to confirm.





2 Help

- (3) Instrument name
- ④ Status dial—Touch to create a plate setup.
- (5) Settings—Touch to view previous results (Run History) or configure the instrument.
- (6) Status of consumables—See "Check the consumables status" on page 38.
- Current user
- (8) Connectivity icons

Connectivity icon	Status
ය or 🤶	The instrument is connected to a wired or wireless network.
	The instrument is connected to network drive.
	The current instrument profile is linked to Cloud.





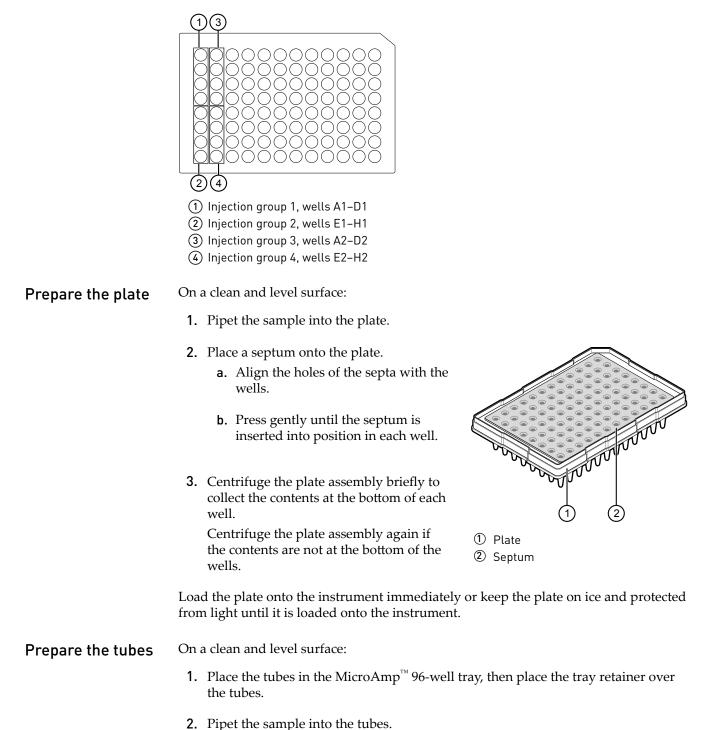
## Prepare the samples

#### Sample preparation guidelines

Item	Guidelines
Plates and tubes	<ul> <li>Use MicroAmp<sup>™</sup> Optical 96-Well Reaction Plate or MicroAmp<sup>™</sup> Reaction Tubes with a tray and retainer set.</li> <li>IMPORTANT! Fast plates are not compatible with the SeqStudio<sup>™</sup> Genetic Analyzer. Fast plates will damage the cartridge.</li> <li>Use the appropriate septa for plates and tubes.</li> <li>See "Required materials not supplied" on page 198 for more information.</li> </ul>
Fragment analysis sample preparation	<ul> <li>Prepare the samples as recommended by the kit for fragment analysis.</li> <li>Use a 10-20 μL sample volume.</li> <li>Ensure that Hi-Di<sup>™</sup> Formamide is fresh. <ul> <li>Hi-Di<sup>™</sup> Formamide should not undergo more than two freeze-thaw cycles (one to aliquot and one for use).</li> <li>Use the same day after thawing.</li> </ul> </li> <li>For more information, see DNA Fragment Analysis by Capillary Electrophoresis User Guide (Pub. No. 4474504).</li> </ul>
Sequence analysis sample preparation	<ul> <li>Prepare sequencing reactions according to kit instructions, and purify the extension products with ethanol precipitation, spin columns, or the BigDye XTerminator<sup>™</sup> Purification Kit.</li> <li>If ethanol precipitation or spin columns are used, dry the samples in a vacuum centrifuge without heat or at low heat for 10–15 minutes or until dry. Note: Do not over dry the samples.</li> <li>Resuspend in 10–20 µL of Hi-Di<sup>™</sup> Formamide.</li> <li>Use a 65 µL or 130 µL sample volume for samples that are prepared with the BigDye XTerminator<sup>™</sup> Purification Kit. See <i>BigDye XTerminator<sup>™</sup> Purification Kit Protocol</i> (Pub. No. 4374408).</li> <li>IMPORTANT! Use the appropriate run modules for samples prepared with BigDye XTerminator<sup>™</sup> Purification Kit. See "Run modules, read lengths, size ranges, and run times" on page 123.</li> <li>Use a 10–20 µL sample volume for samples that are prepared with Hi-Di<sup>™</sup> Formamide.</li> <li>Ensure that Hi-Di<sup>™</sup> Formamide is fresh.</li> <li>Hi-Di<sup>™</sup> Formamide should not undergo more than two freeze-thaw cycles (one to aliquot and one for use).</li> <li>Use the same day after thawing.</li> <li>Do not resuspend samples in water, which can decrease sample stability.</li> <li>For more information, see <i>DNA Sequencing by Capillary Electrophoresis Chemistry Guide Second Edition</i> (Pub. No. 4305080).</li> </ul>

# Plate layout and loading guidelines

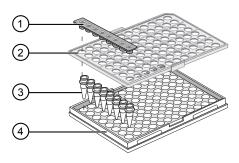
- Samples are stable for 16–24 hours on the instrument.
- Load a maximum of 48 samples per plate if you use a long run module (Long Seq, Long Seq BDX, and Long Frag Analysis). The long fragment analysis run modules can take >24 hours to run an entire 96-well plate.
- Add samples to plates in columns. The default injection order is: A1–D1, E1–H1, A2–D2, E2–H2....A12–D12, E12–H12.





- **3.** Place a septum on the tubes.
  - **a.** Align the holes of the septa with the tubes.
  - **b.** Press gently until the septum are inserted into position in each tube.
- **4.** Centrifuge the tube assembly briefly to collect the contents at the bottom of each tube.

Centrifuge the tube assembly again if the contents are not at the bottom of the tubes.



- Septum
- ② 96-well retainer
- ③ 8-tube strip
- ④ MicroAmp<sup>™</sup> 96-well tray

Load the tube assembly onto the instrument immediately or keep the tubes on ice and protected from light until they are loaded onto the instrument.

### Prepare the instrument

Check the consumables status

- 1. Touch 🌣 Settings > Consumable status.
- **2.** Ensure that:
  - Sufficient consumables are installed for the run.
  - Installed consumables have not exceeded their expiry date.

	Consuma	ble Status	?	
	Cartridge information			
Serial I	number:	3075208-013		
Expirat	ion time:	2019/08/03 12:30:32		
Injectio	ons remaining:	250		
Installe	ed date:	2019/02/04 11:30:32		
(	Cathode buff	er information		
Lot nu	nber:	20170731		
Expirat	ion time:	2019/02/18 11:31:44		
Injectio	ons remaining:	125		
Installe	ed date:	2019/02/04 11:31:44		
Cartridge history			Close	

Display	Cartridge	Cathode buffer
NA	Not installed on the instrument.	Not installed on the instrument.
White OK for use.		OK for use.

Display	Cartridge	Cathode buffer
Yellow	<ul> <li>OK for use, but:</li> <li>It will exceed the manufacturer's expiry date within 2 weeks <i>or</i></li> <li>It will exceed the maximum number of days allowed on the instrument within 2 weeks<sup>[1]</sup></li> </ul>	<ul> <li>OK for use, but:</li> <li>It will exceed the manufacturer's expiry date within 2 days <i>or</i></li> <li>It will exceed the maximum number of days allowed on the instrument (14 days) within 2 days</li> </ul>
Red	<ul> <li>Expired because either of the following conditions have occurred:</li> <li>It has exceeded the manufacturer's expiry date <i>or</i></li> <li>It has exceeded the maximum number of days allowed on the instrument<sup>[1]</sup></li> </ul>	<ul> <li>The cartridge is expired.</li> <li>It has exceeded the manufacturer's expiry date <i>or</i></li> <li>It has been installed on the instrument for &gt;14 days</li> </ul>

[1] The maximum number of days allowed on the instrument is 120 days for the SeqStudio<sup>™</sup> Genetic Analyzer Cartridge (Cat. No. A33671) and 180 days for the SeqStudio<sup>™</sup> Genetic Analyzer Cartridge v2 (Cat. No. A41331). For more information, see "Cartridge storage" on page 17.

See "Insert the Cathode Buffer Container" on page 150 if a new Cathode Buffer Container is required.

See "Insert the cartridge" on page 144 if a new cartridge is required.

**3.** Touch **Close**, then touch  $\textcircled{\bullet}$ .

#### Load the CBC, the sample plate, and the cartridge

The **Eject plate** command is disabled for a few minutes after you insert a cartridge. If you are loading the CBC, sample plate, and cartridge at the same time, you can save time by loading the CBC and sample plate before you insert the cartridge.

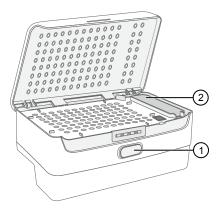
For information on loading the individual components, see:

- "Install cathode buffer" on page 149
- "Load the plate or the tube assembly" on page 76
- "Insert the cartridge" on page 144

In the home screen:



- 1. Touch (a), touch IIII Eject plate, then open the instrument door when prompted.
- **2.** Press the release button on the autosampler to open the lid, then remove the CBC.



Release button
 Location of CBC

- -
- **3.** Check the buffer fill level:
  - **a.** Remove the CBC.
  - **b.** Ensure that the level of buffer is above the fill line.

If the buffer is at or below the fill line, see "Assemble the SeqStudio<sup>™</sup> Genetic Analyzer Cathode Buffer Container (CBC)" on page 149 and "Insert the Cathode Buffer Container" on page 150. If the buffer is above the fill line, reinsert the CBC.

**4.** Place the plate or tube assembly firmly in the autosampler.



① Replace if buffer is at or below the fill line



New CBC buffer level

**5.** Close the autosampler lid: Press down on the center of the lid or press down on both sides of the lid with equal pressure until the lid clicks shut.



6. Touch Retract plate, then close the instrument door.

- 7. Touch Eject cartridge, then open the instrument door when prompted.
- **8.** Hold the cartridge at the hand hold above the capillaries, then pull to remove it from the instrument.
- **9.** Insert a new cartridge (see "Insert the cartridge" on page 144).
- **10.** Close the instrument door.



# Use the instrument with the Thermo Fisher Cloud

Understanding instrument and Cloud interaction	42
Thermo Fisher Connect administrators for an instrument	47
Register and obtain a Thermo Fisher Connect account	49
Link the instrument to your Thermo Fisher Cloud account	50
Change your own Cloud instrument profile PIN	51
Set up email notifications from the instrument	51

# **Understanding instrument and Cloud interaction**

"Connect" versus "link"	The words "connect" and "link" are used interchangeably in the software.
	In one location you touch a <b>Connect</b> button, in another location you touch a <b>Link</b> button.
	Both actions do the same thing:
	• Connect the instrument to the InstrumentConnect on the Thermo Fisher Cloud <i>and</i>
	• Link your local instrument profile to your Thermo Fisher Cloud account.
First user who links is assigned administrator role	The first user who links the instrument to the Thermo Fisher Cloud is automatically assigned Cloud administrator role for the instrument (even if the user has a standard local profile).
	Additional instrument administrators can be assigned, and user roles can be changed after linking.
	For more information, see "Cloud instrument profile roles and functions" on page 46 and "Thermo Fisher Connect administrators for an instrument" on page 47.

Local versus Cloud instrument profiles	• Local instrument profile—All plates and results are stored on the instrument under a local instrument profile.
	<ul> <li>Cloud instrument profile – When you link your local instrument profile to your Cloud account, a Cloud instrument profile is created. With a Cloud instrument profile:</li> </ul>
	<ul> <li>You can save results and data files directly to your Cloud account, access plate setups that you create in the SeqStudio<sup>™</sup> Plate Manager on the Cloud or desktop, and monitor instrument runs from the Cloud.</li> </ul>
	<ul> <li>All plate setups, data files, and results are automatically copied to your Cloud account if the plate setup Save location is set to Cloud.</li> </ul>
lf vou link when	In this scenario, your local instrument profile name is created manually on the

#### If you link when you *are* signed in to the instrument

In this scenario, your local instrument profile name is created manually on the instrument before you link. Your local instrument profile name differs from your Cloud instrument profile name.

If you link to the Cloud when you *are* signed in to the instrument:

Phase	Steps that occur		
Before you link:	• You enter your local instrument profile name in the <b>Sign In</b> screen.		
	Sign In		
	Sign in admin Admi		
	<ul> <li>Sign in Get started</li> <li>Your local instrument profile (<i>UserABC</i>) is displayed in the home screen of the</li> </ul>		
	<ul> <li>All plates and results that you create are accessible only when you are signed in with your local instrument profile.</li> </ul>		
When you link:	• You use an option described in "Link the instrument to your Thermo Fisher Cloud account" on page 50 to link.		
	• If this is the first time you link, a Cloud instrument profile is created using the <i>FirstNameLastInitial</i> of the user name from your thermofisher.com account.		
	Example: <i>User1@thermofisher.com</i> First name is <i>User</i> , Last name is <i>Gray</i> . Cloud account username is <i>User G</i> .		
	<ul> <li>Your local instrument profile (<i>UserABC</i>) is linked to your Cloud account (<i>User1@thermofisher.com</i>).</li> </ul>		
	• Your Cloud instrument profile ( <i>User G.</i> ) replaces your local instrument profile.		



Phase	Steps that occur	
After you link:	• Your Cloud instrument profile ( <i>User G.</i> ) and $ riangle$ is displayed in the home screen of the instrument.	
	<ul> <li>Plates and results from your local instrument profile can be copied to the Cloud (see "Export results from the instrument (sample data files and QC reports)" on page 104).</li> </ul>	
	• New plates and results are saved under your Cloud instrument profile.	
	<ul> <li>Your Cloud instrument profile name (User G. △) is available for selection in the Sign In screen.</li> </ul>	
If your Cloud account is unlinked:	• Your local instrument profile ( <i>UserABC</i> ) is displayed in the home screen of the instrument.	
	<ul> <li>Plates and results that were saved under your Cloud instrument profile are accessible under your local instrument profile.</li> </ul>	
	<ul> <li>New plates and results are saved under local instrument profile and can be copied to the Cloud (see "Export results from the instrument (sample data files and QC reports)" on page 104).</li> </ul>	
	<ul> <li>Your local instrument profile name (<i>UserABC</i>) is available for selection in the Sign In screen.</li> </ul>	

If you link to the Cloud when you are not signed in to the instrument:

Phase	Steps that occur		
Before you link:	• In the Sign In screen, you touch Get Started ➤ Connect.		
	Sign In		
	Sign in     New to this instrument?       admin     Connect to the Thermo Fisher Cloud or create a local instrument profile.		
	PIN		
	Sign in Get started		

3

Phase	Steps that occur		
When you link:	• You use an option described in "Link the instrument to your Thermo Fisher Cloud account" on page 50 to link.		
	• If this is the first time you link, a Cloud instrument and a local instrument profile (with standard role) are created with the same name using the <i>FirstNameLastInitial</i> of the user name from your thermofisher.com account.		
	Example: <i>User1@thermofisher.com</i> First name is <i>User</i> , Last name is <i>Gray</i> . Cloud account username is <i>User G</i> .		
	• Your local instrument profile ( <i>User G.</i> ) is linked to your Cloud account ( <i>User1@thermofisher.com</i> ).		
	• Your Cloud instrument profile ( <i>User G.</i> ) replaces your local instrument profile.		
After you link:	• Your Cloud instrument profile ( <i>User G.</i> ) and		
	<ul> <li>Plates and results from your local instrument profile can be copied to the Cloud (see "Export results from the instrument (sample data files and QC reports)" on page 104).</li> </ul>		
	New plates and results are saved under your Cloud instrument profile.		
	<ul> <li>Your Cloud instrument profile name (User G. △) is available for selection in the Sign In screen.</li> </ul>		
If your Cloud account is unlinked:	• Your local instrument profile ( <i>User G.</i> ) is displayed in the home screen of the instrument.		
	<ul> <li>Plates and results that were saved under your Cloud instrument profile are accessible under your local instrument profile.</li> </ul>		
	• New plates and results are saved under local instrument profile and can be copied to the Cloud (see "Export results from the instrument (sample data files and QC reports)" on page 104).		
	• Your local instrument profile name ( <i>User G.</i> ) is available for selection in the <b>Sign In</b> screen.		

**IMPORTANT!** If you sign in with a local profile, without linking to the cloud, sign out, then link using **Get Started** > **Connect**, you can potentially have two local instrument profiles with different names. Plates are results created when you are signed in with one local instrument profile are not accessible when you are signed in with the other local instrument profile.

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#### Cloud instrument profile roles and functions

Instrument profile	Location	Functions allowed
Standard	Cloud <sup>[1]</sup>	Create, save, open, import, and run plate setups
		Create and modify run settings
		View and export results
Administrator	Cloud <sup>[1]</sup>	All the permissions of a local administrator profile, plus the following functions performed in InstrumentConnect:
		• Access the <b>Manage users</b> function on the Cloud to see a list of all instrument profiles that are linked to the instrument.
		Assign Cloud administrator role to one or more users.
		Remove a user from an instrument.
		Disconnect the instrument from InstrumentConnect.
		Change the instrument name.

[1] The first user who links their local instrument profile to their Cloud account is assigned a Cloud profile with administrator role.

#### For more information on using the Thermo Fisher Cloud

In the top left of the screen you are viewing in the Thermo Fisher Cloud, click  $\equiv$ , then select **Help guide**.

	InstrumentConnect	Thermo		Thermo Fisher Cloud Help
	Home Connect your instrum	Contents Search		Home > View instruments, PC's, and smarl
1	Files	Select a topic to display:		
	Apps Instruments	View instruments, PC's, and smartphones, linked to your Thermo Fisher Cloud account	•	View instruments, PC's Fisher Cloud account
*	Groups BEDL1	<ul><li>2 Add an instrument</li><li>2 Connect your smartphone</li></ul>		Thermo Fisher Cloud enables use smartphones, linked to their acco
••••		10 Update your PIN number		From the dashboard, click 📟 Inst
	Help guide	<ul> <li>View instrument details</li> <li>Make an instrument a favorite</li> </ul>		smartphones to view connected ( tile in the respective Instrument
		Remove yourself from an instrument		What do you want to do?

Manage the users and

administrators of your

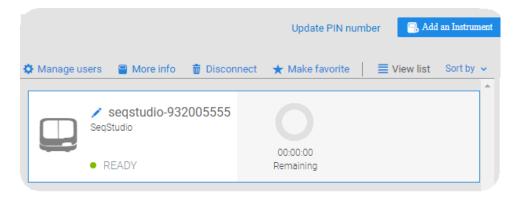
· Add an instrument

3

### Thermo Fisher Connect administrators for an instrument

Cloud administrator functions	The first user who links the instrument to the Thermo Fisher Cloud is automatically assigned Cloud administrator role for the instrument (even if the user has a standard local profile).		
	At least one Cloud administrator is required for each instrument.		
	A Cloud administrator can perform the following tasks from the InstrumentConnect:		
	• Access the <b>Manage users</b> function on the Cloud to see a list of all instrument profiles that are linked to the instrument.		
	Assign Cloud administrator role to one or more users.		
	Remove a user from an instrument.		
	Disconnect the instrument from InstrumentConnect.		
	Change the instrument name.		
Assign instrument administrator role to other users	<ul><li>A Cloud administrator for an instrument can assign instrument administrator role to other users.</li><li>1. Sign in to thermofisher.com/cloud.</li></ul>		
	2. Click 🛢 to access InstrumentConnect.		

**3.** Select the instrument.



**Note:** The **Manage users** and other administrator functions are not displayed until you select an instrument.

**4.** To assign the Admin role to a user, click **Administrator** then select the Administrator check box for the user.



#### Manage the users and administrators of your instrument

Any user with a Cloud administrator profile can manage users for an instrument or disconnect an instrument from InstrumentConnect.

If a Cloud administrator	The software
Assigns Admin role to a user	Allows the user to perform all Cloud administrator functions (see "Cloud administrator functions" on page 47).
Removes a user	Unlinks the instrument from their Thermo Fisher Cloud account.
Disconnects the instrument	<ul> <li>Unlinks the instrument from all user Thermo Fisher Cloud accounts.</li> <li>Removes the instrument from InstrumentConnect.</li> </ul>

- 1. Sign in to **thermofisher.com/cloud**.
- **2.** Click **access InstrumentConnect.**
- **3.** Select the instrument.

			Update PIN nu	mber	🔒 Add	l an Instrun	nent
🌣 Manage u	isers 📄 More info	🗑 Disconnect	★ Make favorite	≣\	/iew list	Sort by	<b>v</b>
	🖍 seqstudio-93 SeqStudio	2005555	0				
	• READY		00:00:00 Remaining				

**Note:** The **Manage users** and other administrator functions are not displayed until you select an instrument.

**4.** To assign the Admin role to a user or to remove a user, click **Admin and Second Second Manage users**, then:

То	Do this
Assign the Admin role to an additional user	Select the <b>Admin</b> checkbox, then click <b>Close</b> .
Remove a user	Click 🗑 , then click <b>Confirm</b> .

Disconnect individual users from an instrument	You cannot disconnect individual users from an instrument. To <i>disconnect</i> a user, or to unlink the instrument from a Cloud account, you must disconnect the instrument from InstrumentConnect. Doing so unlinks <i>all</i> instrument profiles and removes the instrument from InstrumentConnect.
	You can <i>remove</i> a user from the instrument, however, doing so deletes the user data from the instrument.
	For more information, see "Manage the users and administrators of your instrument" on page 48

# **Register and obtain a Thermo Fisher Connect account**

- 1. Go to **www.thermofisher.com**.
- 2. On the home page, select **Sign In Register**.

SCIENTIFIC	Search All	•	Contact Us	Sign In In Quick Order	Ä
Popular Applications 8	& Techniques Shop A		🗅 🙆 Cloud	Sign In	
				Register	

**3.** Fill in all information, then click **Create account**.



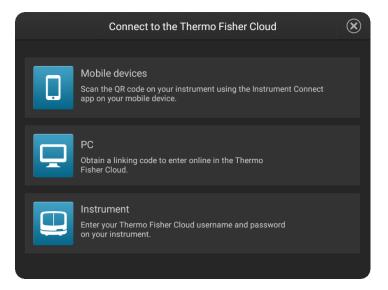
### Link the instrument to your Thermo Fisher Cloud account

**Note:** For detailed information on linking the instrument to your Cloud account, see Appendix B, "Link the instrument to your Cloud account—detailed instructions".

- **1.** If a user is signed in, touch **(a)**, then touch **Sign out**.
- 2. In the **Sign In** screen, touch **Get started > Connect**.

$\bigotimes$

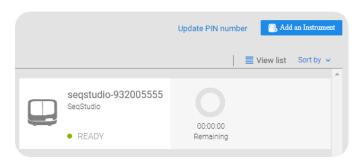
3. In the Connect to the Thermo Fisher Cloud screen, touch a connection option.



Option	Action	
Devices	<b>Note:</b> Before selecting this option, install and sign in to the InstrumentConnect app on your mobile device.	
	In the Connect to the Thermo Fisher Cloud screen:	
	1. Touch <b>Mobile devices</b> .	
	<ol><li>Hold the camera on your mobile device over the QR code that is displayed on the touchscreen.</li></ol>	
	3. Click <b>Close</b> .	
PC	In the <b>Connect to the Thermo Fisher Cloud</b> screen, a link code is displayed. On a computer:	
	1. Access the Thermo Fisher Cloud.	
	2. Access InstrumentConnect.	
	3. Click Add instrument.	
	4. Select <b>SeqStudio</b> .	
	5. Enter the link code.	
🖵 Instrument	In the <b>Connect to the Thermo Fisher Cloud screen</b> , enter your account information, then click <b>Link account</b> .	

### Change your own Cloud instrument profile PIN

- **1.** Sign in to **thermofisher.com/cloud**.
- **2.** Click **a** to access InstrumentConnect.
- 3. Click Update PIN number.

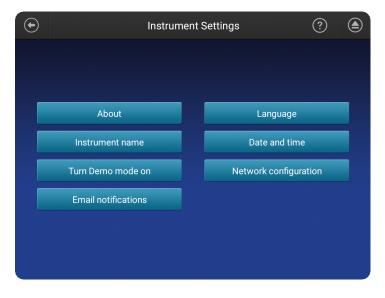


## Set up email notifications from the instrument

When an instrument is linked to your Cloud account, email notifications are automatically sent to your Cloud account email address.

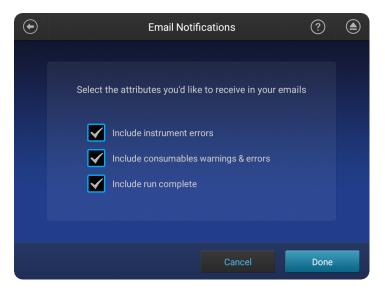
Perform this procedure to disable any of the default notifications.

- 1. Sign in to the instrument with your Cloud instrument profile and PIN.
- In the home screen of the instrument, touch 
   Settings > Instrument
   settings > Email notifications.



**Note:** If you are signed in with a local instrument profile instead of a Cloud instrument profile, the **Email notifications** button is not displayed on the **Instrument Settings** screen.

**3.** In the **Email notifications** screen, select or deselect the options for which you want to receive email notifications, then touch **Done**.





# Create or modify a plate setup from the Plate Manager

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Set up a plate using default settings (Plate Manager)	55
Additional plate settings (Plate Manager)	59

## Overview of plate setup settings

Category	Setting	Notes
Plate           000000000000000000000000000000000000	<ul> <li>Plate properties</li> <li>Plate set up security (Shared or Hidden)</li> <li>File name convention</li> <li>Analysis settings</li> </ul>	<ul> <li>Analysis settings that are saved with the plate setup:</li> <li>Hidden plates—No analysis settings are saved with the plate setup. The last-used analysis settings for a user are applied to a plate setup when it is opened.</li> <li>Shared plates—The analysis settings specified for a plate setup when it is created are saved with the plate setup.</li> <li>User-created analysis settings: <ul> <li>Users can name and save analysis settings.</li> <li>In the instrument software and in the Plate Manager (Cloud), user-created analysis settings are accessible only to the user who saves them.</li> <li>In the Plate Manager (desktop), user-created analysis settings are accessible to all users.</li> </ul> </li> </ul>

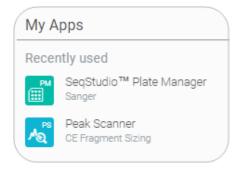


Category	Setting	Notes
Injection group (a set of 4 wells) O	<ul> <li><i>(Fragment analysis only)</i> Size standard</li> <li>Run module</li> <li>Application type (mixed plate only)</li> <li>Dye set</li> </ul>	<ul> <li>User-created size standards and dye sets are accessible to all users.</li> <li>User-created run modules: <ul> <li>Users can name and save run modules.</li> <li>In the instrument software and in the Plate Manager (Cloud), user-created run modules are accessible only to the user who saves them.</li> <li>In the Plate Manager (desktop), user-created run modules are accessible to all users.</li> </ul> </li> </ul>
Well O	<ul> <li>Sample name</li> <li>(Fragment analysis only) Sample type</li> <li>(Sequencing only) Specimen and amplicon</li> <li>Custom fields 1–5</li> </ul>	<ul> <li>Specimen and Amplicon fields are useful in secondary analysis software applications that organize sample data files based on amplicon and specimen information.</li> <li>Custom fields are text fields in which you can include additional sample attributes or identifiers. Custom fields can be used by some secondary analysis applications.</li> </ul>

## Set up a plate using default settings (Plate Manager)

Access the Plate Manager on the Cloud

- 1. Sign in to **thermofisher.com/cloud**.
- In the My Apps list, select SeqStudio<sup>™</sup> Plate Manager.
   If SeqStudio<sup>™</sup> Plate Manager is not listed under My Apps, scroll down in the All Apps list.



Access the Plate Manager on the desktop

Create or open a plate setup PSM file Select Start > All Programs > Applied Biosystems > Plate Manager > Plate Manager.

- 1. Click **PM** to display the home screen.
- **2.** In the **Plate setup** screen, create or open a plate setup:

If you are running the Plate Manager on the Thermo Fisher Cloud:

Click	То
New	Create a new plate setup or to create a plate setup from a template.
Open from cloud	Open a plate setup that you created in Plate Manager on the Thermo Fisher Cloud.
Open from local drive	Open a plate setup that you created in Plate Manager on your computer (PSM file) or in another application (CSV file).

If you are running the Plate Manager on the desktop:

Click	То
New	Create a new plate setup or to create a plate setup from a template.
Open	Open a plate setup that you created in Plate Manager (PSM file) or in another application (CSV file).

Enter plate properties

In the **Properties** tab:

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1. (*Optional*) Edit the **Plate name**, **Barcode**, or **Owner**.

late Properties		Actions N
Plate name	Plate_20170723_080538	I am analyzing my data with Sanger variant analysis software.
Barcode	(optional)	You will be prompted to assign an amplicon and specimen to each well. This will automatically
Owner	Marty V.	organize your files in a way that is compatible with the analysis software.
Plate setup security	Hidden	
	Hidden: Only you can view or access the plate setup.	
Application type	plate setup. Shared: Other users may view or edit the plate setup.	
Application type	plate setup. Shared: Other users may view or edit the	
Application type Sequencing analysis settings	plate setup. Shared: Other users may view or edit the plate setup. Mixed plate (sequencing & fragment) ~ Mixed plate combines both Sequencing and Fragment Analysis applications on the same plate. Plate properties are	Is

- 2. Select an option in the Plate setup security field.
  - Hidden—Prevents other users from using or accessing the plate on the instrument. The last settings specified by the signed-in user are applied when a Hidden plate setup is opened or imported on the instrument.
  - **Shared**—Allows other users to access and edit the plate on the instrument. Analysis settings saved in the plate setup.
- **3.** Select the **Application type**: **Sequencing**, **Fragment analysis**, or **Mixed plate** (sequencing & fragment).

A mixed plate allows you to specify fragment analysis and sequence analysis settings on the same plate.

 (Optional for Sequencing or Mixed plate) Select the I am analyzing my data with Sanger variant analysis software checkbox.

The amplicon and specimen fields are added to the **Plate** view, and the attributes

I am analyzing my data with Thermo Fisher Scientific Sanger analysis software.

You will be prompted to assign an amplicon and specimen to each well. This will automatically organize your files in a way that is compatible with the analysis software.

are automatically added to the default file name conventions (see "Modify the default file name convention" on page 72).

This feature is useful in secondary analysis software applications that organize files based on amplicon and specimen information (Cloud applications: Variant Analysis (VA) module, Next-generation Confirmation (NGC) module; desktop

applications: SeqScape<sup>™</sup> Software, Variant Reporter<sup>™</sup> Software, Minor Variant Finder Software).

5. Click Next.

Assign wells: Sample and run information

- In the **Plate** screen:
  - 1. Click a well to select an injection group, or Shift-click to select multiple injection groups.

Each set of 4 wells on the plate is referred to as an injection group. The default injection order is: A1-D1, E1-H1, A2-D2, E2-H2....A12-D12, E12-H12.

erify and A	Assign Samples					Actions
te_20170723_		Injec	tion Group			
s s s		Well	# Sample n	ame Amplicon	Specimen	Custom Field 1
		A1	A1			Custom 1
S S S		B1	B1			Custom 2
S S S		C1	C1			Custom 3
		D1	D1			Custom 4
	E_BigDye™ Terminator v1.1	Ru M	odule 1	E_BigDye™ T ShortSeq LongSeq		·
Dye set	Z_BigDye™ Terminator v3.1 E5 (DS-02)™ ShortSeq LongSeq	m	odule 2	Longood		-
Run module	FragAnalysis MediumSeq					

- **2.** If you are creating a mixed plate, select the **Application type** for the injection group.
- **3.** (*Fragment analysis only*) Select the sample type for each well: **Allelic ladder**, **Negative control**, **Positive control**, or **Sample**.
- 4. (Fragment analysis only) Select a size standard for the injection group.
- 5. Select a dye set for the injection group.



- Select a run module for the injection group.
   For more information, see "Run modules, read lengths, size ranges, and run times" on page 123
- 7. Click Next and proceed to "Save a plate setup in the Plate Manager" on page 58.

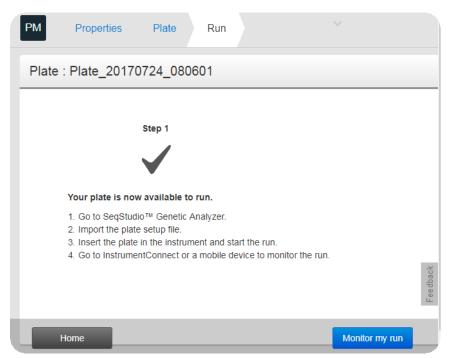
After you assign wells to a plate setup:

#### Save a plate setup in the Plate Manager

1. In the **Save the plate setup** dialog box, modify any settings as needed.

_	
*	
~	
	0723_061724.fsa

2. Click Save.



<b>Note:</b> The <b>Monitor my run button</b> is available only in the Cloud app.			
If you are running the Plate Manager on the	The plate setup is saved as a		
Cloud	Plate setup that you can open from the Cloud and run on the instrument.		
Desktop	PSM file that you can open from a network drive or a USB and run on the instrument.		

Proceed to Chapter 6, "Start and monitor a run".

# Additional plate settings (Plate Manager)

Specify replicate	In the <b>Plate</b> screen:	
injections	1. Select Actions > Add injection.	
	<b>2.</b> Select an injection group, then select a run module for the replicate injections.	
Edit analysis settings	Factory-installed items cannot be edited or deleted. To create a new item from a factory-installed item, copy, edit, then save the new item.	
-	In the <b>Plate properties</b> screen:	
	1. Click Setting details.	
	<b>2.</b> To create new analysis settings:	
	<b>a</b> . Select the default analysis settings or user-created analysis settings, then click <b>Copy</b> .	
	b. Enter a name and edit settings as needed (see "Fragment analysis settings (size calling)" on page 109 or "Sequencing settings (base calling)" on page 112).	
	<b>c.</b> As needed, click <b>Edit</b> or <b>Delete</b> (user-created settings only).	
	For more information, see "Manage analysis settings" on page 125.	
	3. Click Close.	



Modify the default file name	The file name convention determines how the data files (AB1 or FSA) associated with a plate are named.
convention	The default file name convention is:

Application	Default settings
Fragment analysis	<well>_<sample name="">_<sample type="">_<date and="" timestamp="">.fsa</date></sample></sample></well>
Sequence analysis	<well>_<sample name="">_<date and="" timestamp="">.ab1</date></sample></well>
Sequence analysis with the Sanger variant analysis option selected	<well>_<sample name="">_<amplicon>_<specimen>_<date and="" timestamp="">.ab1</date></specimen></amplicon></sample></well>

To change the default settings, in the **Properties** tab:

- 1. Select Actions > Edit the file name convention.
- **2.** Click **File attributes**, then select the attributes to include in or exclude from the data file name.

Well position _Sample name _S	Sample type _ 20170929_121858.fsa
	File attributes (3) -
Well position	✓ Well position
Sample name	Sample name
Sample type	Capillary number
	Injection number
	Sample type
	Custom field 1
	Custom field 2
	Custom field 3
	Custom field 4
	Custom field 5
	Instrument name
	Cartridge serial Cancel Save

For information on creating custom fields to include in file name conventions, see "Define custom fields" on page 73.

- **3.** (*Optional*) Click-drag is to move an attribute to another position.
- 4. Click Save.

Hide or share a	In the <b>Properties</b> screen:		
plate ( <b>Plate setup</b>	Select:		
security)	• <b>Hidden</b> —Prevents other users from using or accessing the plate on the instrument. The last settings specified by the signed-in user are applied when a <b>Hidden</b> plate setup is opened or imported on the instrument.		
	• <b>Shared</b> —Allows other users to access and edit the plate on the instrument. Analysis settings saved in the plate setup.		
Define custom fields	Custom fields are text fields in which you can include additional sample attributes or identifiers. Custom fields can be used by some secondary analysis applications. In the <b>Plate</b> tab:		

- 1. Select Action > Add custom field.
- **2.** Enter information in the custom field in the table at the right of the plate.

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 $\Sigma_{1}$ 



# Create or modify a plate setup from the instrument

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Set optional plate settings (instrument)	71

5

## PSM and CSV plate setup files for import into the instrument

Format	Contains	When to use
PSM		
Create by: Saving a plate setup with the SeqStudio <sup>™</sup> Plate Manager desktop or Cloud	<ul> <li>Size standard names and definitions</li> <li>Run module names and settings</li> <li>Dye set names and settings</li> <li>Analysis settings names and settings</li> <li>All remaining plate setup field settings: Well ID, Sample name, Application type and so on.</li> </ul>	<ul> <li>To create a plate setup that contains all the properties needed to save and run the plate setup.</li> <li>Note the following: <ul> <li>If the size standard, analysis settings, or run module in the PSM file do not exist on the instrument, they are automatically created when the PSM file is imported.</li> <li>If the <b>Plate setup security</b> is set to <b>Hidden</b>: <ul> <li>The plate setup is available for selection on the instrument only to the user who created the PSM file.</li> </ul> </li> </ul></li></ul>
CSV		
<ul> <li>Create by:</li> <li>Saving as CSV in the SeqStudio<sup>™</sup> Plate Manager desktop or Cloud</li> <li>Downloading a template from the SeqStudio<sup>™</sup> Plate Manager desktop or Cloud</li> </ul>	<ul> <li>Size standard names only</li> <li>Dye set name only</li> <li>Run module names only</li> <li>All remaining plate setup field settings: Well ID, Sample name, Application type and so on.</li> </ul>	<ul> <li>To update properties of the plate setup (such as sample name) after the size standard, dye set, and run module for the plate setup have been created or imported on the instrument.</li> <li>When you import a CSV file: <ul> <li>If the size standard, dye set, or run module in the CSV file do not exist on the instrument:</li> <li>An error message is displayed.</li> <li>The CSV file is not imported on the instrument.</li> </ul> </li> <li>If the size standard or run module in the CSV file do exist on the instrument: <ul> <li>The CSV file is imported on the instrument.</li> </ul> </li> <li>If the size standard or run module in the CSV file do exist on the instrument: <ul> <li>The CSV file is imported on the instrument.</li> </ul> </li> <li>The Settings from the size standard, dye set, and run module that exist on the instrument are used (because the CSV file contains the names of these elements, but not the settings).</li> <li>The Plate setup security is set to Hidden.</li> </ul>



# Shared (public), hidden (my plates), and guest plate setup files

Create by setting Plate setup security to	Accessible to users	Stored in folder on instrument	Analysis settings used	File name convention used
Shared	All users, including guest	Public	Analysis settings saved in the plate setup	Last settings specified by the signed-in user (not saved with plate setup)
	st user edits a shared p to <b>Hidden</b> and cannot b		st be saved under a new name,	and the <b>Plate setup</b>
Hidden	Only the user who created the plate setup	My plates	Last settings specified by the signed-in user (not saved with plate setup)	Last settings specified by the signed-in user (not saved with plate setup)
Guest plate setup security is automatically set to <b>Hidden</b> and cannot be changed	Guest user only	My plates	Last settings specified by the signed-in user (not saved with plate setup)	Last settings specified by the signed-in user (not saved with plate setup)

Category	Setting	Notes
Plate           000000000000000000000000000000000000	<ul> <li>Plate properties</li> <li>Plate set up security (Shared or Hidden)</li> <li>File name convention</li> <li>Analysis settings</li> </ul>	<ul> <li>Analysis settings that are saved with the plate setup:</li> <li>Hidden plates—No analysis settings are saved with the plate setup. The last-used analysis settings for a user are applied to a plate setup when it is opened.</li> <li>Shared plates—The analysis settings specified for a plate setup when it is created are saved with the plate setup.</li> <li>User-created analysis settings: <ul> <li>Users can name and save analysis settings.</li> <li>In the instrument software and in the Plate Manager (Cloud), user-created analysis settings are accessible only to the user who saves them.</li> <li>In the Plate Manager (desktop), user-created analysis settings are accessible to all users.</li> </ul> </li> </ul>
Injection group (a set of 4 wells) O	<ul> <li><i>(Fragment analysis only)</i> Size standard</li> <li>Run module</li> <li>Application type (mixed plate only)</li> <li>Dye set</li> </ul>	<ul> <li>User-created size standards and dye sets are accessible to all users.</li> <li>User-created run modules: <ul> <li>Users can name and save run modules.</li> <li>In the instrument software and in the Plate Manager (Cloud), user-created run modules are accessible only to the user who saves them.</li> <li>In the Plate Manager (desktop), user-created run modules are accessible to all users.</li> </ul> </li> </ul>
Well O	<ul> <li>Sample name</li> <li>(Fragment analysis only) Sample type</li> <li>(Sequencing only) Specimen and amplicon</li> <li>Custom fields 1–5</li> </ul>	<ul> <li>Specimen and Amplicon fields are useful in secondary analysis software applications that organize sample data files based on amplicon and specimen information.</li> <li>Custom fields are text fields in which you can include additional sample attributes or identifiers. Custom fields can be used by some secondary analysis applications.</li> </ul>

# Overview of plate setup settings



# (Optional) Set up for auto export of sample data files (AB1 and FSA)

By default, sample data files (AB1 and FSA) are saved to the instrument.

When you create a plate setup, you can also set the **Save location** to **Cloud**, **Network Drive**, and/or **USB**.

•	Plate Properties	?	
Properties			
Plate name Plate_20170906_200855	Save location Network Drive; Cloud Instrument	1; USB;	-1

<sup>1</sup> Save location

When the plate is run, the instrument automatically exports the sample data files to the save locations.

Before you can select these save locations, set up the instrument:

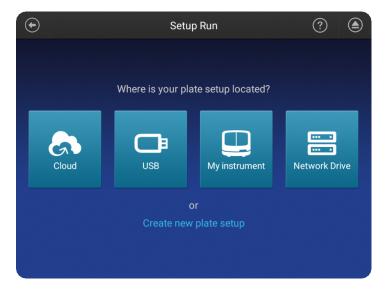
- "Link the instrument to your Thermo Fisher Cloud account" on page 50
- "Connect to a network drive" on page 115
- Insert a USB into the USB port on the front of the instrument (Figure 1 on page 13)

### Set up a plate using default settings (instrument)

Create or import a plate setup

In the home screen:

- 1. Touch Set up run.
- 2. Create, open, or import a plate setup:



То	Procedure
Create a new plate setup	<ol> <li>Touch Create new plate setup.</li> <li>See "Enter plate properties" on page 67.</li> </ol>
Open an existing plate setup on the instrument	<ol> <li>Touch  My Instrument.</li> <li>Touch:         <ul> <li>My plates folder to select hidden plates that you have created.</li> <li>Public folder to select (1) shared plates that were created by any user or (2) any plates that were created by a Guest user.</li> </ul> </li> <li>For more information, see "Shared (public), hidden (my plates), and guest plate setup files" on page 64.</li> </ol>
Import a plate setup	<ol> <li>Touch  Scloud, □ USB, or SNetwork Drive.</li> <li>Select:         <ul> <li>PSM file</li> <li>CSV file</li> </ul> </li> <li>For more information, see "PSM and CSV plate setup files for import into the instrument" on page 63</li> </ol>

# Enter plate properties

In the **Plate properties** screen:

#### 1. Touch the **Plate name** field, then enter the plate name.

	Plate P	roperties	?	۲
Pro	operties			
PI	te name ate_20170721_233015 lication agment analysis	Save location Instrument I am analyzing		~
Bar	code			
Мо	re options Injection options		Start ru	n

- **2.** Touch **Applications**, then select Sequencing, Fragment analysis, or Mixed (allows you to specify fragment analysis and sequence analysis settings on the same plate).
- 3. (*Optional*) Touch the **Barcode** field, then use a scanner to scan the barcode.
- **4.** (*Optional*) Touch the **Owner** field, then enter the plate owner name.

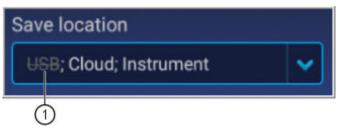
SeqStudio<sup>™</sup> Genetic Analyzer Instrument and Software User Guide

- **5.** (*Optional*) Touch the **More options** to check the **Plate setup security**, **Analysis settings**, and **File name convention**.
- **6.** If you want to save the plate to a location in addition to the instrument, touch **Save location**, then select a location for the run results.

The plate setup is always saved to the instrument. In addition, you can save the plate to the Cloud, a network, or a USB, which will auto export the sample data files.

**IMPORTANT!** To view analyzed data in the Remote Monitoring App on the Cloud, you must save the plate setup to the Cloud.

**Note:** If you save a plate setup to the Cloud, a network, or a USB, then access the plate setup at a later time when the instrument is not linked to the Cloud, a network, or a USB, the save location is displayed with strikethrough text.



(1) Original location to which the plate was saved, but is no longer accessible by the instrument.

7. (Optional for Sequencing or Mixed plate) Touch the I am analyzing my data with Sanger variant analysis software checkbox.

The amplicon and specimen fields are added to the **Plate** view, and the attributes are automatically added to the

In the **Plate properties** screen:

I am analyzing my data with Sanger variant analysis software.

Files will be configured compatible with Sanger variant analysis software. You will be prompted to assign an amplicon and specimen to each

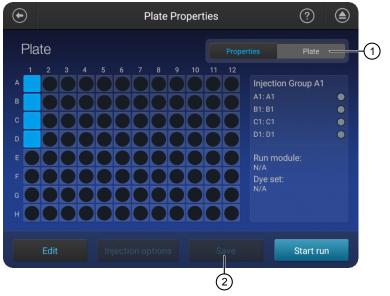
default file name conventions (see "Modify the default file name convention" on page 72).

This feature is useful in secondary analysis software applications that organize files based on amplicon and specimen information (Cloud applications: Variant Analysis (VA) module, Next-generation Confirmation (NGC) module; desktop applications: SeqScape<sup>™</sup> Software, Variant Reporter<sup>™</sup> Software, Minor Variant Finder Software).

Assign wells: run module, size standard, and dye set 1. At the top-right of the screen, touch **Plate**.



② Save button is not enabled until you create an injection group



Each set of 4 wells on the plate is referred to as an injection group. An injection group is identified by the first well in the set of 4 (for example, Injection Group A1 contains wells A1–D1).

The default injection order is: A1-D1, E1-H1, A2-D2, E2-H2....A12-D12, E12-H12.

- 2. Select injection groups, then touch Edit.
  - Touch a well to select a single injection group.
  - Touch and drag to select multiple injection groups or the entire plate.

$   \mathbf{\bullet} $		Edit Plate	?	
	Selected Injections	A1-D1		
	Sample name	A1, B1, C1, D1		
	Run module	Run module		
	Dye set			
	Dye set	Dye set		
	Clear wells	Cancel		

3. If you are creating a mixed plate, select the **Application type** for the wells.

- Touch Run modules, then select a run module. For more information, see "Run modules, read lengths, size ranges, and run times" on page 123.
- 5. Touch **Dye set**, then select a dye set for the injection group.
- **6.** (*Fragment analysis only*) Touch **Size standard**, then select a size standard for the injection group.

In the Edit plate screen:

1. Touch **Sample name** to display the well attributes fields.

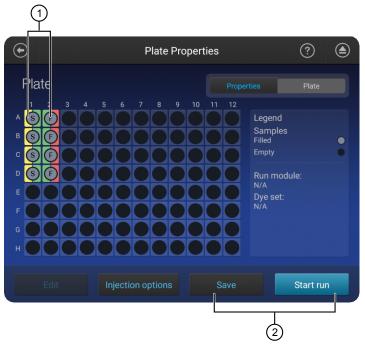
Plate\_20170722\_225336

Image: Sample

Im

- 2. Touch a setting, then enter the definition for the selected wells:
  - (Fragment analysis only) Sample type—Sample, Positive Control, Negative Control, or Allelic Ladder.
  - **Custom fields**—Text fields to include additional sample attributes or identifiers that can be used by secondary analysis applications.
  - (*Sequencing only*) **Amplicon and Specimen**—Amplicon and Specimen names for Sanger Sequence analysis, if you selected the option in the **Plate Properties** screen.

Assign wells: sample name, sample type, and custom fields 3. Touch Done to close the screen then Done to close the Edit Plate screen.



Application type—Sequencing or Fragment, designated by S or F
 Save the plate or start the run

4. Touch Save to save the plate to run at a later time, or touch Start Run.

### Set optional plate settings (instrument)

Specify replicate injections

#### In the Plate properties screen:

- 1. Touch Injection options.
- **2.** Touch an injection group, then touch **Edit and re-inject** to add replicates to the injection list.
- **3.** (*Optional*) Modify **Run module**, **Injection time**, **Injection voltage**, **Run time**, or **Run voltage** for the injections.

**IMPORTANT!** Changes to run conditions for replicate injections are not saved to the plate.

4. Touch Done.

Modify analysisIn the Properties tab of the Plate properties screen:settings1. Touch More options > Analysis settings.

**2.** Select a setting.

See "Manage analysis settings" on page 125 for detailed information.

Note: The last setting selected is used as the default for new plates.

3. Touch Done.

Modify the default The default file name convention determines how the data files (AB1 or FSA) associated with a plate are named. file name convention

The default file name convention is:

Application	Default settings	
Fragment analysis	<well>_<sample name="">_<sample type="">_<date and="" timestamp="">.fsa</date></sample></sample></well>	
Sequence analysis	<well>_<sample name="">_<date and="" timestamp="">.ab1</date></sample></well>	
Sequence analysis with the Sanger variant analysis option selected	<well>_<sample name="">_<amplicon>_<specimen>_<date and="" timestamp="">.ab1</date></specimen></amplicon></sample></well>	

1. Access the File name convention screen:

From	Action
Plate properties screen	Select the <b>Properties</b> tab, then touch <b>More options &gt; File name convention</b> .
Home screen	Touch

- 2. Touch Attributes.
- **3.** Select the attributes to include in the data file name.

Select Attributes		
Sequencing Fragment		
Sequencing		
Well number Amplicon Custom Field 1		
Sample name Specimen Custom Field 2		
Capillary # Instrument name Custom Field 3		
Injection number 🗹 Cartridge serial # 📃 Custom Field 4		
Custom Field 5		
Reset default Cancel Done		

For information on creating custom fields to include in file name conventions, see "Define custom fields" on page 73.

4. Touch Done.

5

- 5. Touch and drag attributes up or down in the list.
- 6. Touch Done.

Hide or share a plate ( <b>Plate setup</b> <b>security</b> )	<ul><li>For more information, see "Shared (public), hidden (my plates), and guest plate setup files" on page 64.</li><li>1. Touch More Options &gt; Plate Setup security.</li></ul>			
	<b>2.</b> Touch an option:			
	• <b>Hidden</b> —Prevents other users from using or accessing the plate on the instrument. The last settings specified by the signed-in user are applied when a <b>Hidden</b> plate setup is opened or imported on the instrument.			
	• <b>Shared</b> —Allows other users to access and edit the plate on the instrument. Analysis settings saved in the plate setup.			
	<b>3.</b> Touch <b>Done</b> , then touch <b>Save</b> .			
Define custom fields	Custom fields are text fields in which you can include additional sample attributes or identifiers. Custom fields can be used by some secondary analysis applications.			
	In the <b>Plate</b> tab of the <b>Plate properties</b> screen:			
	1. Select injection groups, then touch <b>Edit</b> .			
	<ul><li>Touch a well to select a single injection group.</li><li>Touch and drag to select multiple injection groups or the entire plate.</li></ul>			
	2. Click Edit.			

**3.** Touch **Sample name** to display the injection group and well attributes fields.



#### 1 Touch the sample name field

$   \mathbf{\bullet} $	P	lat	e_201707:	22_225336
	Sample		Well ID	Sample
	Run module			A1
	Dye set			B1
	Custom field 1			C1
	Custom field 2			D1
	Custom field 3			
	Custom field 4			
	Custom field 5			
				Cancel Done

- 4. Touch a custom field, then enter the definition for the selected wells.
- 5. Click Done.

In the **Plate properties** screen:

*(Optional)* View the injection list, change injection settings or order, or specify replicates and reinjections

#### 1. Touch Injection options.

- 2. Touch an injection group, then configure the injection list:
  - Touch and drag an injection group to a new location in the injection list.
  - Touch **Inject first**—Moves the selected injection group to the top of the injection list.
  - Touch **Edit and re-inject**—Adds replicates or re-injections to the injection list. You can also modify **Run module**, **Injection time**, **Injection voltage**, **Run time**, or **Run voltage** for these injections.

**Note:** Changes to **Injection time**, **Injection voltage**, **Run time**, or **Run voltage** are not saved to the plate setup and will be used during the current plate run only.

3. Touch Done.

5

# Start and monitor a run



Load the plate or the tube assembly	76
Select a plate setup and start a run	77
Lock the touchscreen	79
Monitor a run from the Thermo Fisher Cloud	79
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Unload the plate or the tube assembly	92

# Load the plate or the tube assembly

In the home screen:

- 1. Touch (a), touch IIII Eject plate, then open the instrument door when prompted.
- 2. Press the release button on the autosampler to open the lid.
- **3.** Place the plate or tube assembly firmly in the autosampler.
- **4.** Check the buffer fill level:
  - **a.** Remove the CBC.
  - b. Ensure that the level of buffer is above the fill line.
    If the buffer is at or below the fill line, see "Assemble the SeqStudio<sup>™</sup> Genetic Analyzer Cathode Buffer Container

(CBC)" on page 149 and "Insert

the Cathode Buffer Container" on page 150. If the buffer is above the fill line, reinsert the CBC.

**5.** Place the plate or tube assembly firmly in the autosampler.



① Replace if buffer is at or below the fill line



New CBC buffer level

- 6
- **6.** Close the autosampler lid: Press down on the center of the lid or press down on both sides of the lid with equal pressure until the lid clicks shut.



7. Touch Retract plate, then close the instrument door.

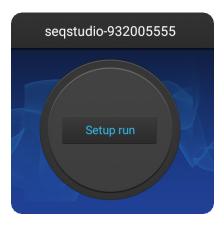
# Select a plate setup and start a run

After you load the plate in the instrument (see "Load the plate or the tube assembly" on page 76):

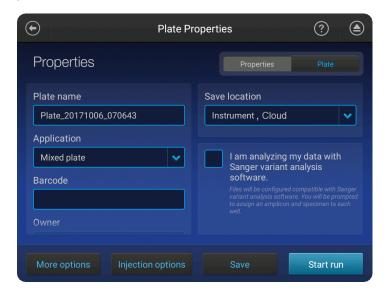
- 1. In the instrument home screen, touch **Setup run**.
- **2.** Select the location of your plate setup, then select the plate setup.

For information on plate setup files, see "PSM and CSV plate setup files for import into the instrument" on page 63 and "Shared (public), hidden (my plates), and guest plate setup files" on page 64.

**3.** Verify that settings are as needed.



The **Save location** must specify **Cloud** if you want to view analyzed data when you monitor the run from the Cloud.



#### 4. Touch Start run.

When the run starts, the instrument automatically:

- Performs an optical alignment each time a cartridge is inserted.
- Performs an automatic spectral calibration adjustment (auto calibration) for each sample to correct for spectral overlap.

During a run, an administrator can lock the touchscreen to prevent other users from using the instrument. Only the user who started the run or an administrator can sign in to the instrument if the touchscreen is locked.

Proceed to:

- "Monitor a run from the Thermo Fisher Cloud" on page 79
- "Monitor a run from a mobile device" on page 87
- "Monitor a run from the instrument" on page 89

Automatic file Before starting a run, the instrument calculates the total amount of storage space required to save the run. If the required storage space is not available, the instrument deletes files associated with the oldest exported plates until sufficient space is available.

Note: Only complete plates that have been auto exported (saved to Cloud, network, or USB) or manually exported (using (❀) Settings > Run History > plate name > Export) are deleted.

If the required storage space is not available and no plates have been exported, the instrument displays a notification indicating that there is not enough storage space.

You can export plates and delete plates, then start the run again.

# Lock the touchscreen

During a run, you can lock the touchscreen to prevent other users from using the instrument. This feature is not available to Guest users.

Only the user who locked the touchscreen or an administrator can sign in to the instrument if the touchscreen is locked.

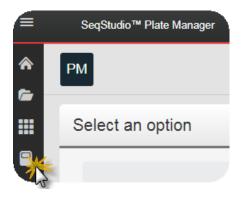
- **1.** Touch **()**.
- 2. Touch Profile.
- 3. Touch Lock instrument.

**Note:** If a run is not in progress, **Sign out** is displayed instead of **Lock instrument**.

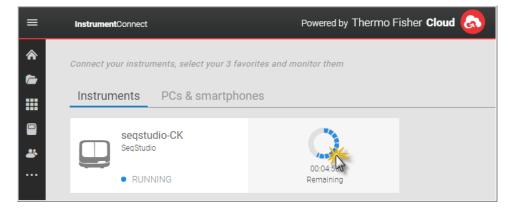
# Monitor a run from the Thermo Fisher Cloud

Open the Remote Monitoring App from Instrument Connect App A run is accessible from InstrumentConnect for 24 hours after the run is complete, or until another run is started.

- 1. Sign in to **thermofisher.com/cloud**.
- **2.** Click **access InstrumentConnect**.



**3.** Click the run status dial to display the Remote Monitoring App.





RM

Run

seqstudio-CK is running

Actions 🗸

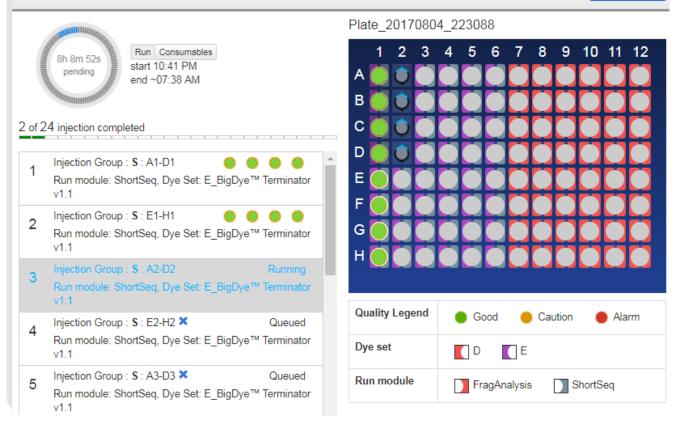


Figure 11 Remote Monitoring App



#### Open the Remote Monitoring App from the Plate Manager

A run is accessible from the Plate Manager for 24 hours after the run is complete, or until another run is started.

You can open the Remote Monitoring App immediately after you save a plate setup or at a later time.

In the Plate Manager on the Thermo Fisher Cloud:

• To open the Remote Monitoring App immediately after you save a plate setup and start the run, click **Monitor my run**.

	PM Prop	erties	Plate	Run			
	Plate : Plate	_201707:	24_080	601			
		st	ep 1				
	Your pl	ate is now a	vailable to	o run.			
	2. Impo 3. Inser	o SeqStudio™ ort the plate se t the plate in o InstrumentC	etup file. the instrur	nent and sta	tor the run.		
							Feedback
Į	Home					Monitor my run	



• To open the Remote Monitoring App at a later time, click **PM**, then select an instrument or click to access InstrumentConnect.

ATE SETUP	MONITOR RUN	
lew	Detected instruments	
eate a plate from scratch or create from a template	VTS-101	
	SVT-201	
pen from cloud	CTC-301	
a previous plate layout	PRC-401	
pen from local		
e a previous plate layout		

(1) Select an instrument or click to access InstrumentConnect.

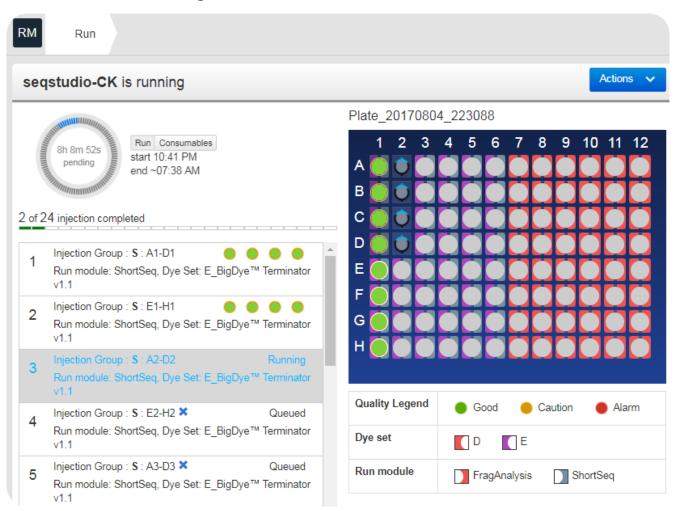


Figure 12 Remote Monitoring App



View notifications from the instrument on the Cloud

View results in the Remote Monitoring App on the Cloud 1. In any screen in the Thermo Fisher Cloud, click 🔲.



- 2. Click a notification, then click **Dismiss** or **Dismiss all** to dismiss the notification.
- 1. Open the Remote Monitoring App (see "Open the Remote Monitoring App from Instrument Connect App" on page 79).
- 2. Click an injection group in the injection list or the plate view.

The status dials are color-coded for quality alerts:

- - All QC tests passed.
- — At least 1 warning quality alert was triggered.
- At least 1 failing quality alert was triggered.

For information on quality alerts, see:

- "Data quality alerts" on page 102
- "Sizecalling and basecalling quality alerts" on page 102

pplication Dye Set PragAnalysis GS800LIZ				
Well Details	Raw EPT	Analyzed		
Status	Well #	Sample Name	Sample Type	Quality Alerts
Good	E2	E2	Sample	
Good	F2	F2	Sample	
Good	G2	G2	Sample	
Good	H2	H2	Sample	

**Note:** The **Analyzed** tab is disabled if the **Save location** for the plate setup is not set to **Cloud** or if the injection group has not finished running.

- 3. In the Quality alerts screen, click the Raw, EPT, or Analyzed tab to view data.
- **4.** As needed, select **Actions** ▶ **Re-inject group**, select the **Run module** and settings, then click **Inject**.

Pause or cancel an injection in the Remote Monitoring App

Select:

- Actions Pause plate to pause the run after the currect injection is complete.
- Actions Stop current injection to immediately stop the injection.

Edit injection group run settings

In the **Results** screen:



- 1. Select Actions > Edit injection group.
- 2. Edit settings as needed, then click OK.

#### Re-inject or delete an injection group

In the **Results** screen:

Select Actions > Re-inject group or Actions > Delete injection group.

#### Export a QC report

The **QC Report** command is not available for an injection until the injection is complete. The **QC Report** command is not available for any injections if the plate setup **Save location** does not specify the Cloud.

In the **Remote Monitor** screen:

#### Select Actions > QC Report.

A PDF of the plate QC report is generated.

#### Remote Monitoring App raw trace

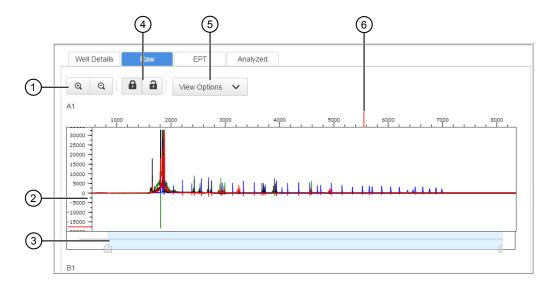


Figure 13 Fragment analysis raw trace

- 1 Zoom in/out.
- Raw trace.
- ③ Thumbnail trace—Click-drag to view another region of the trace.
- (4) Lock/unlock trace zooming for all traces in the injection group.
- 5 View Options—Select the dye color to display; set vertical scaling.
- 6 Cursor position indicator (red line).

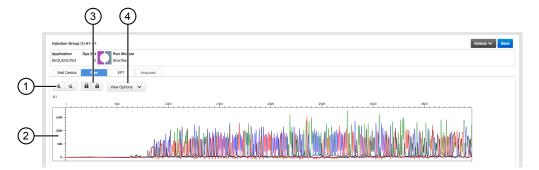
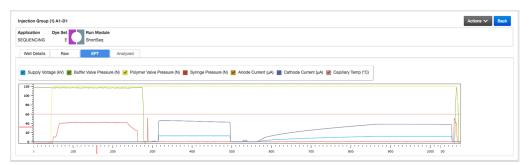


Figure 14 Sequence analysis raw trace

- 1 Zoom in/out.
- Raw trace.
- ③ Lock/unlock trace zooming for all traces in the injection group.
- 4 View Options—Select the basecalls to display; set vertical scaling.

#### Remote Monitoring App EPT trace

The EPT view (ElectroPhoresis Telemetry) shows instrument data conditions (currents, temperatures, electrophoresis voltage) as a function of time.



#### Remote Monitoring App analyzed trace

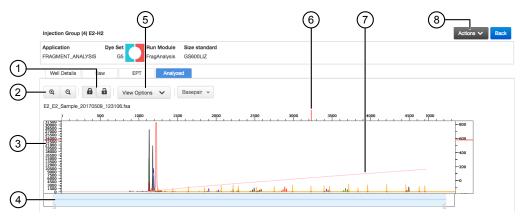
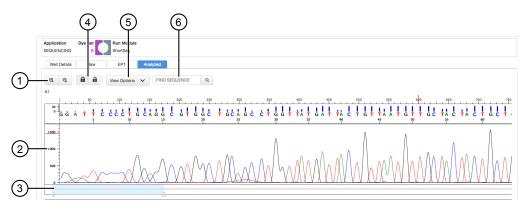


Figure 15 Fragment analysis analyzed trace

- ① Lock/unlock trace zooming for all traces in the injection group.
- 2 Zoom in/out.
- ③ Analyzed trace.
- ④ Thumbnail trace—Click-drag to view another region of the trace.
- (5) View Options—Select the dye colors to display; set vertical scaling.
- 6 Cursor position indicator (red vertical and horizontal tick marks outside trace).
- Size standard curve (red line).
- (8) Actions—Select commands to pause and cancel injections.



#### Figure 16 Sequence analysis analyzed trace

- 1 Zoom in/out.
- Analyzed trace.
- ③ Thumbnail trace—Click-drag to view another region of the trace.
- (4) Lock/unlock trace zooming for all traces in the injection group.
- (5) View Options—Select the basecalls to display; show/hide quality bars and values; set vertical scaling.
- 6 Search for a sequence.

# Monitor a run from a mobile device

Before you begin, see "Link the instrument from a mobile device" on page 168.

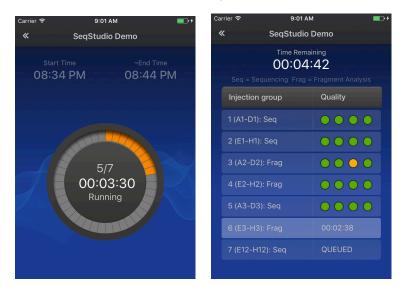
- 1. On your mobile device, launch InstrumentConnect.
- **2.** Touch the instrument to monitor.



**3.** Swipe left to view consumable status.



**4.** Touch the status dial to view the injection list.



**5.** Touch an injection group to display quality alerts, then touch **View raw plot** to view the data.



- Swipe left to view the entire trace.
- Pinch-zoom to expand the trace.

# Monitor a run from the instrument

*(Optional)* View the injection list, change injection settings or order, or specify replicates and reinjections In the **Plate properties** screen:

1. Touch Injection options.

**2.** Touch an injection group, then configure the injection list:

- Touch and drag an injection group to a new location in the injection list.
- Touch **Inject first**—Moves the selected injection group to the top of the injection list.
- Touch **Edit and re-inject**—Adds replicates or re-injections to the injection list. You can also modify **Run module**, **Injection time**, **Injection voltage**, **Run time**, or **Run voltage** for these injections.

**Note:** Changes to **Injection time**, **Injection voltage**, **Run time**, or **Run voltage** are not saved to the plate setup and will be used during the current plate run only.

3. Touch Done.

#### In the home screen:

status

View the run

View the run time information and the status dial for each capillary.



The status dials are color-coded for quality alerts:

- 🔹 😑 All QC tests passed.
- — At least 1 warning quality alert was triggered.
- 🛑 At least 1 failing quality alert was triggered.

If an injection group is set to re-inject, the number of the current injection is displayed on the status dials.

For information on quality alerts, see:

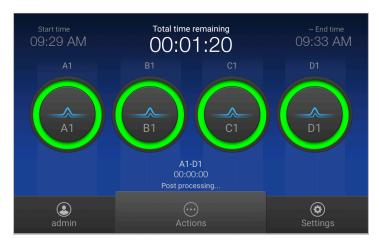
- "Data quality alerts" on page 102
- "Sizecalling and basecalling quality alerts" on page 102



## View real-time results

During a run, in the home screen:

Touch one of the injection dials to display the trace for the selected capillary. See "Fragment analysis trace" on page 96 or "Sequence analysis trace" on page 98 for information.



The status dials are color-coded for quality alerts:

- 😑 All QC tests passed.
- – At least 1 warning quality alert was triggered.
- — At least 1 failing quality alert was triggered.

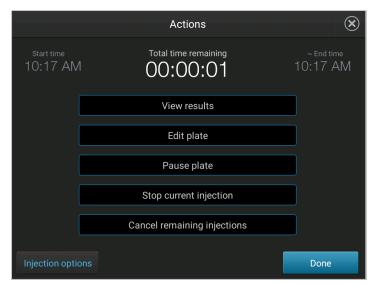
For information on quality alerts, see:

- "Data quality alerts" on page 102
- "Sizecalling and basecalling quality alerts" on page 102

Pause a plate or cancel or stop injections

In the **home** screen:

**1.** Touch  $\odot$  **Actions**.



2. Manage the plate or injections:

Touch	То
View results	View the list of injections and status.
Edit plate	For injections that have not yet been run, edit <b>Sample Name</b> , <b>Run Module</b> , <b>Dye Set</b> , <b>Size Standard</b> , <b>Sample Type</b> , and custom fields.
Pause plate	Stop the run after the current injection is complete. Touch <b>Resume</b> to continue the run.
Stop current injection	Stop the current injection immediately. Touch <b>Resume</b> to continue the run.
Cancel remaining injections	Specify whether to cancel the run immediately or after the current injection is complete.
Injection options	Move an injection to the top of the injection list, edit run module information, and/or reinject samples.

View results for the current plate When a run is complete, in the home screen:

## 1. Touch View Results.

2. Touch List view.

Each injection group displays a QC color for each capillary:

- All QC tests passed.
- At least 1 warning quality alert was triggered.
- — At least 1 failing quality alert was triggered.

For information on quality alerts, see:

- "Data quality alerts" on page 102
- "Sizecalling and basecalling quality alerts" on page 102 ٠
- **3.** Touch an injection group.
- 4. View the results in the **Run Result Details** screen, or touch 脂 for well details. See "Fragment analysis results" on page 96 or "Sequence analysis results" on page 98 for detailed information.
- **5.** Touch a sample file name.

If the data triggered any quality alerts, a QC alerts screen is displayed. See "Data quality alerts" on page 102 and "Sizecalling and basecalling quality alerts" on page 102 for detailed information.

Touch View data to display the analyzed trace for the sample. See "Fragment analysis trace" on page 96 or "Sequence analysis trace" on page 98 for detailed information.

**6.** Touch and drag the thumbnail view of the analyzed trace (below the trace) to scroll left or right.

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- 7. (Optional) Adjust the graphical view (see "Adjust the trace display" on page 92).
- Touch > or < to scroll to the raw data or EPT Plot (ElectroPhoresis Telemetry). See "EPT plot" on page 100.

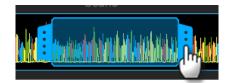
#### Adjust the trace display

See "View results for the current plate" on page 91 to access results.

- Drag one finger to pan to the left or right.
- Zoom in and out by pinching and expanding with two fingers.
- Touch on the left border of the trace, then touch a dye to deselect.
- Touch I on the right border of the trace, then touch ④ Zoom In, Zoom Out, or
   ▶ Fit to screen to adjust the display.
- Drag the center of the pane in thumbnail view to scroll left or right.



• Drag the right or left handle of the pane to zoom horizontally.



# Unload the plate or the tube assembly

When the run is complete:

- 1. Touch (a), touch IIII Eject plate, then open the instrument door when prompted.
- 2. Press the release button on the autosampler to open the lid.
- 3. Remove the plate or tube assembly.

- 6
- 4. Close the autosampler lid: Press down on the center of the lid or press down on both sides of the lid with equal pressure until the lid clicks shut.



5. Touch Retract plate, then close the instrument door.





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# View results in the Remote Monitoring App on the Cloud

- 1. Open the Remote Monitoring App (see "Open the Remote Monitoring App from Instrument Connect App" on page 79).
- 2. Click an injection group in the injection list or the plate view.

The status dials are color-coded for quality alerts:

- — All QC tests passed.
- At least 1 warning quality alert was triggered.
- — At least 1 failing quality alert was triggered.

For information on quality alerts, see:

- "Data quality alerts" on page 102
- "Sizecalling and basecalling quality alerts" on page 102

pplication Dye Set PagAnalysis G5 FragAnalysis G5600LZ				
Well Details	Raw EP1	Analyzed		
Status	Well #	Sample Name	Sample Type	Quality Alerts
Good	E2	E2	Sample	
Good	F2	F2	Sample	
Good	G2	G2	Sample	
Good	H2	H2	Sample	

**Note:** The **Analyzed** tab is disabled if the **Save location** for the plate setup is not set to **Cloud** or if the injection group has not finished running.

- 3. In the Quality alerts screen, click the Raw, EPT, or Analyzed tab to view data.
- **4.** As needed, select **Actions** → **Re-inject group**, select the **Run module** and settings, then click **Inject**.

# 7

# View results on the instrument

View results for the current plate

When a run is complete, in the home screen:

- 1. Touch View Results.
- 2. Touch List view.

Each injection group displays a QC color for each capillary:

- — All QC tests passed.
- — At least 1 warning quality alert was triggered.
- — At least 1 failing quality alert was triggered.

For information on quality alerts, see:

- "Data quality alerts" on page 102
- "Sizecalling and basecalling quality alerts" on page 102
- **3.** Touch an injection group.
- 4. View the results in the **Run Result Details** screen, or touch **h** for well details. See "Fragment analysis results" on page 96 or "Sequence analysis results" on page 98 for detailed information.
- **5.** Touch a sample file name.

If the data triggered any quality alerts, a QC alerts screen is displayed. See "Data quality alerts" on page 102 and "Sizecalling and basecalling quality alerts" on page 102 for detailed information.

Touch **View data** to display the analyzed trace for the sample. See "Fragment analysis trace" on page 96 or "Sequence analysis trace" on page 98 for detailed information.

- **6.** Touch and drag the thumbnail view of the analyzed trace (below the trace) to scroll left or right.
- 7. (Optional) Adjust the graphical view (see "Adjust the trace display" on page 92).
- **8.** Touch **>** or **<** to scroll to the raw data or **EPT Plot** (ElectroPhoresis Telemetry). See "EPT plot" on page 100.

#### Adjust the trace display

See "View results for the current plate" on page 91 to access results.

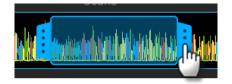
- Drag one finger to pan to the left or right.
- Zoom in and out by pinching and expanding with two fingers.
- Touch on the left border of the trace, then touch a dye to deselect.
- Touch on the right border of the trace, then touch ⊕ Zoom In, ♀ Zoom Out, or
   Fit to screen to adjust the display.



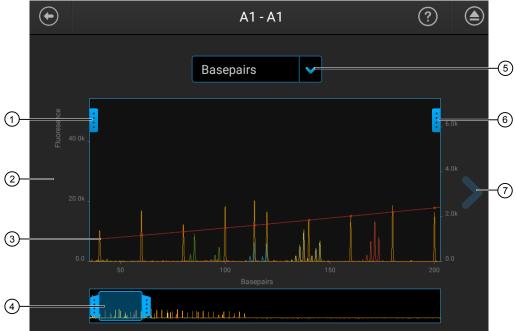
• Drag the center of the pane in thumbnail view to scroll left or right.



• Drag the right or left handle of the pane to zoom horizontally.



# Fragment analysis Fragment analysis trace results



- ① Trace color hide/show—Touch to open, then touch a color to hide or show.
- Analyzed trace
- ③ Size standard curve (red line)
- (4) Thumbnail trace—Drag the center of the pane in the thumbnail trace to display another trace area in the top pane. Drag the right or left handle of the pane to zoom horizontally.
- (5) Basepair or scan display selection.
- 6 Zoom tools—Touch to open.
- ⑦ Next trace tool—Touch to view the raw trace or EPT for the well.



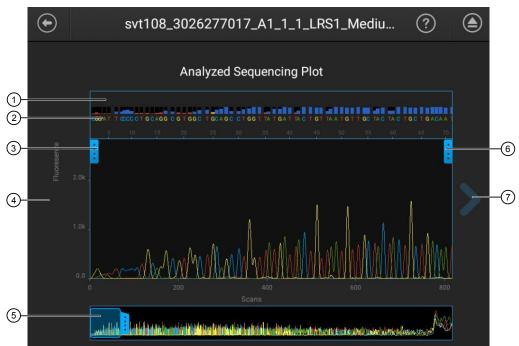
#### Fragment analysis results

Setting	Description
Quality	QC flags <ul> <li>—All QC tests passed.</li> <li>—At least 1 warning quality alert was triggered.</li> <li>—At least 1 failing quality alert was triggered.</li> </ul>
SQ	<ul> <li>Size Quality</li> <li>If SQ is ≤0.75, it passes the QC test and does not trigger a quality alert ().</li> <li>If SQ is 0.25-0.74, it triggers a warning quality alert ().</li> <li>If SQ is &lt;.25, it triggers a failing quality alert ().</li> <li>How Size Quality is determined</li> <li>The Size Quality algorithm evaluates the similarity between the fragment pattern for the size standard dye specified in the size standard definition and the actual distribution of size standard peaks in the sample, calculates an interim SQ (a value between 0 and 1).</li> </ul>
Exported	The sample has been auto exported (saved to Cloud, network, or USB) or manually exported (using () Settings > Run History > <i>plate name</i> > Export).
ĥ	Well details (see below).
Well details	
Sample file name, QC f	lag, sample name, sample type, settings used to acquire the data (size standard, run module

and dye set), Sizing quality value, and export status.



# Sequence analysis Sequence analysis trace results



(1) Quality Value bars and values:

- Pure base with  $QV \ge 20$
- Pure base with QV 15–19
- Pure base with QV <15
- Mixed base
- (2) Bases—Mixed basecalls are highlighted in red (if they exceed the **Mixed base threshold** specified in analysis settings; see "Sequencing settings (base calling)" on page 112).
- 3 Trace color hide/show—Touch to open, then touch a color to hide or show.
- 4 Analyzed trace
- (5) Thumbnail trace—Drag the center of the pane in the thumbnail trace to display another trace area in the top pane. Drag the right or left handle of the pane to zoom horizontally.
- 6 Zoom tools—Touch to open.
- ⑦ Next trace tool—Touch to view the raw trace or EPT for the well.

#### Sequencing results

Result	Description	
Quality	<ul> <li>QC flags</li> <li>—All QC tests passed.</li> <li>—At least 1 warning quality alert was triggered.</li> <li>—At least 1 failing quality alert was triggered.</li> </ul>	



Result	Description
CRL (Contiguous Read	The longest uninterrupted segment of bases with an average Quality Value (QV) $\geq$ 20.
Length)	In addition to evaluating the QV of a base call, the software considers the QV of adjacent bases within a $\pm 21$ -bp moving average to determine a contiguous read length based on quality values.
	<b>Note:</b> The contiguous read length passing criteria for install checks is an uninterrupted segment of bases with an average Quality Value (QV) of 30.
Exported	The sample has been auto exported (saved to Cloud, network, or USB) or manually exported (using <b>③ Settings → Run History → <i>plate name</i> → Export</b> ).
<u>b</u>	Well details (see below).

#### Well details

Sample file name, QC flag, sample name, CRL, signal strength, Median PUP, Trace score, run module and dye set used to acquire the data, and export status.

Signal strength	The average relative fluorescence unit (RFU) for all dyes across the electropherogram in the raw data.
Trace score	The average basecall Quality Value (QV) of bases in the clear range sequence of a trace.
	The <i>clear range</i> is the region of the sequence that remains after excluding the low- quality or error-prone sequence at the 5' and 3' ends. The clear range is calculated by the KB basecaller using QVs.
Median PUP (pull-up peak)	A measure of noise or pull-up that is determined by taking the mean of the ratios of signal strength calculated for each base-called peak: primary peak/secondary peak under the primary peak.
	A higher value indicates less baseline or secondary noise. A lower value indicates an elevated baseline or secondary noise.
	Example 1: Main called base signal strength is 1,000 RFU and the largest secondary peak beneath it is 10 RFU; PuP=100
	Example 2: Main called base signal strength is 1,000 RFU and the largest secondary peak beneath it is 100 RFU; PuP=10

#### Understanding Quality Values (QVs)

#### Quality value ranges

The color of a QV bar indicates the QV of a base.

- Pure base with  $QV \ge 20$
- Pure base with QV 15–19
- Pure base with QV <15
- Mixed base



#### Pure base versus mixed base QVs

Pure bases and mixed bases have the same probability of error for the associated basecall  $(10^{-q/10})$ . Note the following:

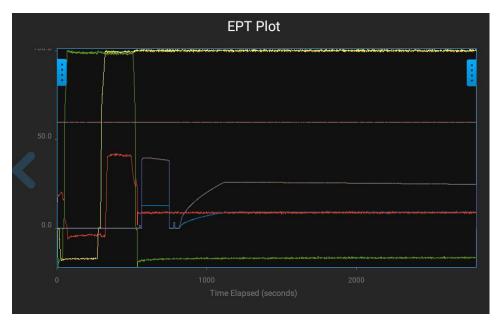
- High-quality pure bases typically have QVs of 20 or higher.
- The distribution of quality values for mixed bases differs dramatically from that of pure bases.
- Mixed bases have a maximum QV of 20.
- Review all mixed base calls.

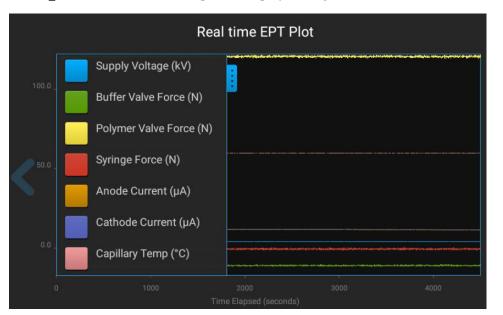
## Quality values (QV) and probability of error (Pe)

QV	Ре	QV	Pe
1	79.0%	30	0.10%
5	32.0%	35	0.032%
10	10.0%	40	0.010%
15	3.2%	45	0.0032%
20	1.0%	50	0.0010%
25	0.32%	60	0.00010%

#### EPT plot

The EPT view (ElectroPhoresis Telemetry) shows instrument data conditions (currents, temperatures, electrophoresis voltage) as a function of time.





Touch on the left border of the plot to display the legend.

View results for a previously run plate (run history)

In the home screen:

- **1.** Touch <sup>(⊕)</sup> **Settings** ▶ **Run history**.
- **2.** Touch a plate name, then touch **View**.
  - If you select more than one plate name, the **View** button is dimmed. The **Run History** screen is displayed.

$   \bullet $	Run	History		?	
	Plate	Туре	Run date		
	Plate_20170830_SVT126_Seq_BDT1	SEQUENCING	2017/08/30	18:29:53	
	Plate_20170830_SVT126_Seq_BDT1	SEQUENCING	2017/08/30	12:46:56	
	Plate_20170825_SVT126_Seq_BDT3	SEQUENCING	2017/08/25	18:34:06	
	Plate_20170825_SVT126_Frag_BVM	FRAGMENT	2017/08/25	15:34:20	
	Plate_20170825_SVT126_Seq_BDT3	SEQUENCING	2017/08/25	12:33:23	
	Plate_20170825_SVT126_Seq_BDT3	SEQUENCING	2017/08/25	09:54:50	
	Plate_20170825_SVT126_Seq_BDT1	SEQUENCING	2017/08/25	07:54:04	
	Select all Delete				

- **3.** Touch a sample file name.
- **4.** View the results in the **Run history** screen, or touch **b** to view well details. See "Fragment analysis results" on page 96 or "Sequence analysis results" on page 98 for information.



**5.** Touch a sample file name, then touch **View**.

If you select more than one sample file name, the **View** button is dimmed. If the data triggered any quality alerts, a QC alerts screen is displayed. For information on quality alerts, see:

Click **View data** to display the trace for the sample. See "Fragment analysis trace" on page 96 or "Sequence analysis trace" on page 98.

- **6.** Touch and drag the thumbnail view of the analyzed trace (below the trace) to scroll left or right.
- **7.** Touch **>** or **<** to scroll to the raw data or **EPT Plot**.

#### Data quality alerts

Quality alert	Description	Action
Offscale peaks. Adjust the injection parameters and/or the sample concentration.	At least 10 scans have saturated the CCD camera.	<ul><li>Reduce the injection voltage.</li><li>Dilute the sample.</li></ul>
No sample was detected.	Poor signal-to-noise ratio with low signal detected.	<ul> <li>Verify that the sample volume follows recommendations in the user manual.</li> <li>Troubleshoot upstream PCR and sequencing steps.</li> </ul>

#### Sizecalling and basecalling quality alerts

#### Table 5 Sizecalling quality alerts

Quality alert	Description	Action
Sizing quality value is low due to poor size standard peak quality. Peak height uniformity is low or the fitting quality in sizing is poor.	Low resolution or poor quality data is present.	<ul> <li>Re-inject the sample.</li> <li>If the problem persists, check the sample quality.</li> </ul>
Sizecaller found broad peak(s) in the size standard peak(s).		
Sizing quality value is in the intermediate range; check size standard data quality.		
The number of size standard peaks detected is less than what is defined in the size standard.	Size standard definition includes peaks that are not present in the sample. Example: Sample peaks are detected up to 500 bp, but the size standard definition includes peak sizes that are >500 bp.	Use or create a size standard definition with the appropriate number of peaks and peak sizes.



Quality alert	Description	Action
The analysis range is too small. Correct the analysis range in analysis settings and re- analyze in secondary analysis software or re- inject sample.	Various causes.	<ul> <li>Analyze the data in a secondary analysis software with a corrected analysis range.</li> <li>Re-inject the sample.</li> </ul>

## Table 6 Basecalling quality alerts

Quality alert	Description	Action
Basecalling failed due to poor quality data.	Poor quality data is present.	<ul> <li>Re-inject the sample.</li> <li>If the problem persists, prepare fresh sample.</li> <li>Troubleshoot upstream PCR and sequencing steps.</li> </ul>

Edit injection parameters and	You can edit injection parameters and re-inject samples during a run or after a run is complete.
re-inject samples	1. Access Injection options.
	<ul> <li>During a run — Touch ⊕ Actions ▶ Edit plate.</li> </ul>
	• After a run—Touch <b>Results</b> .
	<b>2.</b> Touch an injection group, then configure the injection list:
	<ul> <li>Touch and drag an injection group to a new location in the injection list.</li> </ul>
	<ul> <li>Touch Inject first—Moves the selected injection group to the top of the injection list.</li> </ul>
	<ul> <li>Touch Edit and re-inject—Adds replicates or re-injections to the injection list. You can also modify Run module, Injection time, Injection voltage, Run time, or Run voltage for these injections.</li> </ul>
	<b>3.</b> Touch <b>Done</b> .
	<b>Note:</b> The changes are not applied until you touch <b>Done</b> .
Export a report (QC report)	This function allows you to export a QC report for the current plate. To export a QC report for a previously run plate, export a run history (see "Export results from the instrument (sample data files and QC reports)" on page 104).
	When a run is complete, in the home screen:
	1. Touch <b>Results</b> .
	2. Touch Export report.
	<b>3.</b> Select a storage location.
	/ Navigate to then select a location then touch Export

**4.** Navigate to, then select a location, then touch **Export**.



# Export results from the instrument (sample data files and QC reports)

In the home screen:

- 1. Touch ( Settings > Run history.
- 2. Select one or more plates from the Run History table.
- 3. Touch Export.
- 4. Select a storage location.

The following data is exported for the plate:

- Fragment analysis—FSA file for each sample.
- Sequencing—AB1 file for each sample.
- Plate QC report in CSV and PDF format.

**Note:** If you select a plate, select **View**, then select **Export**, only an FSA or AB1 file for each analyzed sample is exported.

# Analyze data

- **1.** Export results (see "Export results from the instrument (sample data files and QC reports)" on page 104) or use auto exported data.
- **2.** Use an appropriate fragment analysis or sequencing application to analyze the data.

**Note:** Data from the SeqStudio<sup>™</sup> Genetic Analyzer may be labeled as "3200" in secondary analysis software.

For more information, see "Secondary analysis software" on page 28



# Manage the software (Plate Manager)

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# Add a custom dye calibration to the Plate Manager

On the instrument:

- 1. Open a plate setup that specifies the custom dye set of interest.
- **2.** Export a plate setup that specifies the custom dye.
- **3.** Open the exported plate setup in the Plate Manager. The custom dye is imported and is available for selection when you create new plates.

# Download a plate setup template as a CSV file

- 1. Click **PM** to display the home screen.
- 2. Click New, select Create from template, then click Download for the template of interest.

**Note:** You cannot add plate setups to the list of templates. However, you can create a plate setup with "Template" suffix, and use it as a starting point for creating new plate setups.



# Save a plate setup as a PDF

In the Plate tab:

Select Actions > Print plate view.

The plate view and table view are exported.

# Save a plate setup as a CSV file

In the **Plate** tab:

- 1. Select Actions > Save as CSV.
- 2. Navigate to and select a storage destination.

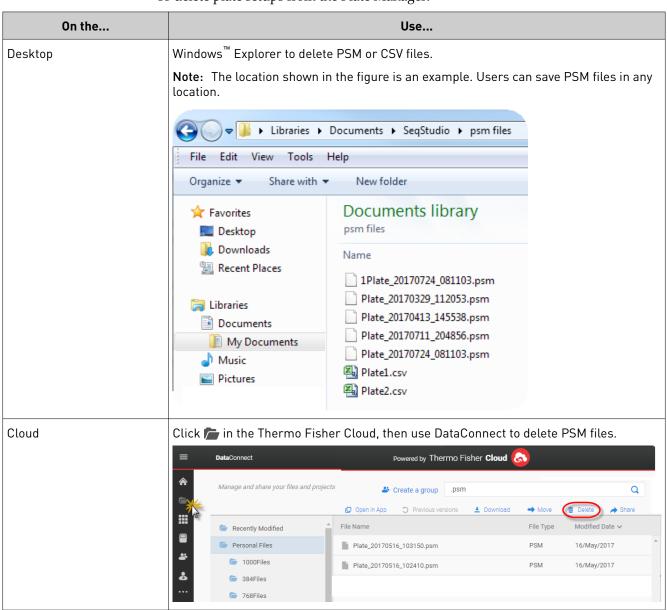
The plate setup is saved as a CSV file. The names of the dye set, run module, and size standard are included in the CSV file.

# Create a plate setup template

**Note:** You cannot add templates to the list of templates you select when you create a plate setup. However, you can create a plate setup with "Template" suffix, then use it as a starting point for creating new plate setups.

- 1. Click **New**, select **Create from template**, then click **Download** for the template of interest.
- **2.** Modify the CSV file as needed.
- **3.** Save the plate setup with a "Template" suffix (example: Plate\_FragAnalysis\_Template), or other identifier.

# **Delete plate setups**



To delete plate setups from the Plate Manager:

# Manage email notifications in the Thermo Fisher Cloud

1. In any screen in the Thermo Fisher Cloud, click 🔲.



- 2. Click Settings.
- 3. Select or deselect your email address.

# Manage run modules

This function manages the list of run modules that you can select from when you create a plate setup.

To assign a run module to a plate, see "Assign wells: run module, size standard, and dye set" on page 68.

Factory-installed items cannot be edited or deleted. To create a new item from a factory-installed item, copy, edit, then save the new item.

For more information, see "Run modules, read lengths, size ranges, and run times" on page 123.

- 1. In the **Plate** tab, select **Actions** > **Manage run modules**.
- 2. To create a new run module:
  - a. Select a default run module or a user-created run module, then click **Copy**.
  - b. Enter values, click Advanced to enter additional settings, then click Save.

See "Run module settings" on page 123 for detailed information.

**3.** As needed, select a run module of interest, then click **Edit** or **Delete** (user-created run modules only).

## Manage size standards

This function manages the list of size standard definitions that you can select from during plate set up.

To assign a size standard definition to a plate, see "Assign wells: run module, size standard, and dye set" on page 68.

A size standard defines the sizes in basepairs of known fragments. It is used to generate a standard curve. The standard curve is used to determine the sizing of fragments in unknown samples.

Factory-installed items cannot be edited or deleted. To create a new item from a factory-installed item, copy, edit, then save the new item.

Size standard definitions are accessible to all users.

- 1. In the Plate tab, select **Actions** > **Manage size standards**.
- 2. To create a new size standard:
  - a. Select a default size standard or a user-created size standard, then click **Copy**.
  - **b.** Enter a name and select a dye color, then edit the fragment sizes (basepairs).

- c. Click Save:
- **3.** As needed, select a size standard of interest, then click **Edit** or **Delete** (user-created size standards only).

## Manage analysis settings

#### In the **Properties** tab:

- **1.** Select Actions > Analysis settings.
- **2.** To create new analysis settings:
  - **a.** Select the default analysis settings or user-created analysis settings, then click **Copy**.
  - Enter a name and edit settings as needed (see "Fragment analysis settings (size calling)" on page 109 or "Sequencing settings (base calling)" on page 112).
  - c. As needed, click **Delete** (user-created settings only).
- 3. Click Save.

#### Fragment analysis settings (size calling)

Setting	Description		
Size calling method	• <b>Local Southern</b> — <i>(default)</i> Determines the fragment sizes using the reciprocal relationship between fragment length and electrophoretic mobility.		
	• <b>Global Southern</b> —Compensates for standard fragments with anomalous electrophoretic mobility (similar to least squares methods).		
	• <b>2nd LSQ</b> (2nd Order Least Squares)—Uses regression analysis to build a bestfit size calling curve.		
	• <b>3rd LSQ</b> (3rd Order Least Squares)—Uses regression analysis to build a bestfit size calling curve.		
	• <b>Cubic Spline Interpolation</b> —Forces the sizing curve through all the known points of the selected size standard.		
Analysis range	• <b>Full Range</b> — <i>(default)</i> To analyze the entire scan region as collected by the genetic analysis instrument, including the primer peak.		
	• <b>Partial Range</b> —To analyze only data points within a specified range. Enter Start Point in data points after the primer peak and before the first required size standard peak. Enter a Stop Point after the last required size standard fragment. Start and Stop points may vary from instrument to instrument and platform to platform. View raw data to determine the appropriate analysis range. Data points outside the specified analysis range are ignored.		
	<b>Note:</b> Ensure the <b>Analysis Range</b> contains all size standard fragments included in the <b>Sizing Range</b> .		



Setting	Description		
Sizing range	<ul> <li>The size range (in base pairs) appropriate for the kit you are using:</li> <li>Full Range for the software to analyze fragments of all sizes in the Analysis Range.</li> <li>Partial Range for the software to analyze only fragments within a specified range. Enter a Start Size and a Stop Size appropriate for the size standard used.</li> </ul>		
Peak amplitudes	The peak height threshold (RFU) for peak detection for each dye color. Peaks below the threshold are not detected. For example, if you use the default values of 175 RFU, peaks with heights equal to or greater than 175 RFU are detected. Peaks with heights below 175 RFU are still displayed in the electropherogram plots but are not detected or labeled. <b>Note:</b> Use the same peak amplitude thresholds in secondary analysis software.		
Primer peak	If the primer peaks in your application obscure peaks of interest, select <b>Present</b> . This instructs the algorithm to ignore primer peaks. Primer peaks are still displayed in the trace. If this setting does not allow detection of the 20- and 40-mer peaks for samples that use the GS600 LIZ <sup>™</sup> size standard, running samples with the GS600_LIZ_(60-600) or other size standards that include lower bp starting points may allow detection of the peaks.		
Common settings			
Smoothing	<ul> <li>Select an option to smooth the outline of peaks and reduce the number of false peaks detected:</li> <li>None (default) to apply no smoothing. Best if the data display sharp, narrow peaks of interest.</li> <li>Light to provide the best results for typical data. Light smoothing slightly reduces peak height.</li> <li>Heavy for data with very sharp, narrow peaks of interest. Heavy smoothing can significantly reduce peak height.</li> </ul>		
Baseline Window	<ul> <li>Specify a window to adjust the baseline signals of all detected dye colors to the same level for an improved comparison of relative signal intensity. Note the following: <ul> <li>A small baseline window relative to the width of a cluster, or grouping of peaks spatially close to each other, can result in shorter peak heights.</li> <li>Larger baseline windows relative to the peaks being detected can create an elevated baseline, resulting in peaks that are elevated or not resolved to the baseline.</li> </ul> </li> </ul>		
Minimum Peak Half Width	<ul> <li><b>h</b> Specify the minimum full peak width at half maximum <b>Peak Height</b> required for peak detection. The range is 2 to 99 data points.</li> </ul>		

Setting	Description
Peak Window Size	Enter a window width in data points for peak detection sensitivity. If more than one peak apex is within the window, all are labeled as a single peak. Note the following:
	• The maximum value is the number of data points between peaks.
	The <b>Peak Window Size</b> setting is limited to odd numbers.
	To increase peak detection sensitivity: Increase polynomial degree, decrease peak window size. To decrease peak detection sensitivity: Decrease polynomial degree, increase peak window size.
Polynomial Degree	Polynomial Degree cannot be greater than Peak Window Size.
	Adjust to affect the sensitivity of peak detection. You can adjust this parameter to detect a single base pair difference while minimizing the detection of shoulder effects and/or noise.
	The peak detector calculates the first derivative of a polynomial curve fitted to the data within a window that is centered on each data point in the analysis range.
	Using curves with larger polynomial degree values allows the curve to more closely approximate the signal and, therefore, captures more of the peak structure in the electropherogram.
Slope Thresholds Peak Start and End	• <b>Peak Start</b> —The peak starts when the first derivative (slope of the tangent) in the beginning of the peak signal before the inflection point becomes equal to or exceeds the <b>Peak Start</b> value. This threshold is set to 0 by default, which means that the peak will normally start at the leftmost point where the slope of the tangent is closest to 0° (horizontal line). A value other than 0 moves the peak start point toward its center. The value entered must be non-negative.
	• <b>Peak End</b> —The peak ends when the first derivative (slope of the tangent) in the end of the peak signal after the inflection point becomes equal to or exceeds the <b>Peak End</b> value. This value is set to 0 by default, which means that the peak will normally end at the rightmost point where the slope of the tangent is closest to 0° (horizontal line). A value other than 0 moves the peak end point toward its center. The value entered in this field must be non-positive.
	Using curves with larger polynomial degree values allows the curve to more closely approximate the signal and, therefore, captures more of the peak structure in the electropherogram.

SeqStudio<sup>™</sup> Genetic Analyzer Instrument and Software User Guide



#### Sequencing settings (base calling)

The default settings are optimized for sequencing of PCR amplicons from diploid genes, which are expected to have Mixed bases and which should end At PCR Stop.

For sequencing from plasmid templates, which are of pure sequence (no Mixed bases) and have longer sequence read times than PCR products, create new analysis settings and *disable* (uncheck) **At PCR Stop** and **Mixed base threshold** checkboxes.

Setting	Description	
Quality Threshold	<ul> <li>Basecall assignment (ambiguous bases):         <ul> <li>Do not assign Ns to basecalls</li> <li>Assign Ns to basecalls with QV&lt;5— Bases with a QV less than the threshold display N instead of the base letter</li> </ul> </li> <li>End base—Last base on which to perform basecalling:         <ul> <li>At PCR Stop</li> <li>After X number of bases</li> <li>After X number of Ns in X number of bases</li> <li>After X number of Ns</li> </ul> </li> <li>Note: If you have PCR products with sequences that end while data is still being collected, select the At PCR Stop checkbox.</li> </ul>	
Mixed bases threshold	When enabled, determines the secondary peak height ratio where the secondary peak is considered a potential mixed base. Reaching the threshold is a necessary but not sufficient condition for the basecalling algorithm to call a mixed base.	
Clear range methods	<ul> <li>Use quality values—Sets a window with a specified number of allowed low-quality bases by removing bases until there are <x <y.<="" bases="" li="" number="" of="" per="" qv="" with="" z=""> <li>Use base positions—Specifies the first and last base in the range to consider, or trims the specified number of bases from the 3' end.</li> <li>Mask base positions before—Specifies the base position before which to disregard bases.</li> </x></li></ul>	



# Manage the software (instrument)

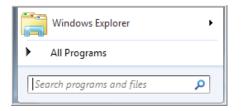
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## Connect the instrument to a network drive

Determine IP address for a computer on a network

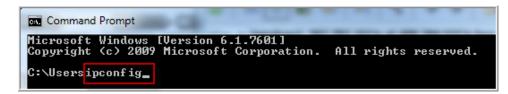
On the destination computer:

- 1. In the Windows<sup>™</sup> desktop, click 🛜.
- **2.** In the search field at the bottom of the pane, type **command prompt**, then press **Enter**.





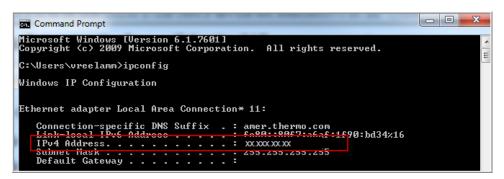
**3.** At the command prompt, type **ipconfig**, then press **Enter**.





**4.** Note the IP address listed.

**Note:** The location and number of digits in your IP address may differ from the **IPv4 Address** example shown below.



**Create folders and** In the Windows<sup>TM</sup> desktop:

enable network folder sharing

- 1. On a Windows<sup>™</sup> computer, server, or network drive, create a folder to store your plates and results. Example: C:/Users/Your Name/SharedData.
- 2. Create subfolders in the SharedData folder. Example: PlateSetups and Results.



- 3. Right click on a folder, then select **Sharing** ► **Advanced** or **Share** with ► **Advanced sharing**.
- 4. Select the **Sharing** tab.
- 5. Click Advanced sharing.
- 6. Select Share this folder.
- 7. Click Permissions, then select Full Control or Read, Write, and Delete options.

**IMPORTANT!** Without these permissions, the instrument cannot autoexport results when a run is complete.

8. Click OK, click OK, then click Close.

See your laboratory administrator for the information you need to connect to a Connect to a network drive. network drive

From any screen that displays 
Retwork drive or Save location as an option, you can connect to the drive for the first time.

IMPORTANT! Before saving to the network drive, ensure that the folder is shared (see "Create folders and enable network folder sharing" on page 114).

- 1. Touch **E** Network drive or Save location field.
- 2. Touch the Destination field, then touch the appropriate field to enter the IP address and shared folder name, then touch Done. For more information, see "Determine IP address for a computer on a network" on page 113.
- 3. If necessary, touch the appropriate fields to enter a domain name, username, and password.
- 4. Touch Connect.

Set up a default **Cloud location for** opening plate setups

Before you begin, create a plate setup (PSM file or CSV file) in the Cloud location that you want to set as the default location.

To set the default location, you must select a plate setup.

1. In the home screen of the instrument, touch **Setup run**, then touch **& Cloud**. The Setup Run screen is displayed.



2. Select a folder (if needed), then select a plate file. The plate file is imported.





Set up a default Cloud location for	<ol> <li>In the home screen of the instrument, touch Setup run, then touch Create new plate setup.</li> </ol>
saving results (auto export)	2. Touch Save location.
	3. Touch the Cloud <b>Destination</b> field.
	The <b>Select Directory</b> screen is displayed.
	<b>4.</b> Select or create a folder.
	5. Touch Done.
Set up a default network location	Before you begin, create a plate setup (PSM file or CSV file) in the shared directory on the computer that you want to set as the default location.
for opening plate	To set the default location, you must select a plate setup.
setups	<ol> <li>Determine the IP address of the computer on which you created shared folders (see "Determine IP address for a computer on a network" on page 113 and "Create folders and enable network folder sharing" on page 114 ).</li> </ol>
	<b>2.</b> In the home screen of the instrument, touch <b>Setup run</b> , then touch <b>B Network</b>

If your instrument profile	This screen is displayed	How to proceed
Is not connected to a network drive	Connect	<ol> <li>In the Network Destination field, enter the IP address of the computer followed by the folder names you created. Example: 10.43.32.82/SharedData/Results.</li> <li>If required by your patyork, enter</li> </ol>
		<ol> <li>If required by your network, enter Domain, User Name, and Password to access the shared location.</li> </ol>
		<b>3.</b> Touch <b>Connect</b> . The <b>Setup Run</b> screen is displayed.
Is connected to a network drive	Select Directory	Proceed to step 3.

3. In the **Setup Run** screen:

Drive.

Touch	То	
Set up import	Change the <b>Network Destination</b> you specified in the previous step.	
A folder name	Navigate to the location you want to set as the default.	
A plate name	Import the plate.	

#### The **Setup Run** screen is displayed.

•		Setup Run 10.43.32.82/SharedData/Pl Select a plate name from the		
	Туре	Plate file name	Last modified	
		Plate_20170720_112109_J6	2017/07/20 11:22:05	
		Plate_20170713_100908	2017/07/13 10:09:37	
		Plate_20170713_100202	2017/07/13 10:02:43	-2
		Plate_20170711_144447	2017/07/11 14:45:08	
			2017/07/11 14:43:11	
Set	t up imp	ort	Cancel	



1 Example IP address and folder locations

(2) List of plates (will be blank if you have not saved plates to this location)

- Determine the IP address of the computer on which you created shared folders (see "Determine IP address for a computer on a network" on page 113 and "Create folders and enable network folder sharing" on page 114 ).
- 2. In the home screen of the instrument, touch **Setup run**, then touch **Create new plate setup**.
- 3. Touch Save location.
- 4. Touch the network **Destination** field.

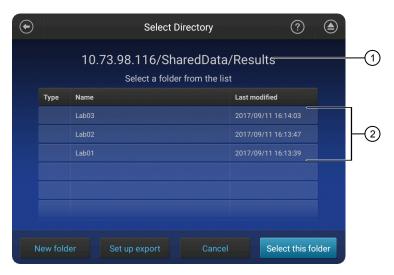
lf your instrument profile	This screen is displayed	How to proceed
Is not connected to a network drive	Connect	<ol> <li>In the Network Destination field, enter the IP address of the computer followed by the folder names you created. Example: 10.43.32.82/SharedData/Resu lts.</li> <li>If required by your network, enter Domain, User Name, and Password to access the shared location.</li> <li>Touch Connect. The Select Directory screen is displayed.</li> </ol>
Is connected to a network drive	Select Directory	Proceed to step 5.

Set up a default network location for saving results (auto export)



#### 5. In the Select Directory screen:

Touch	То	
New folder	Create a subfolder.	
Set up export	Change the <b>Network Destination</b> you specified in the previous step.	
Select this folder	Select the directory shown at the top of the screen.	



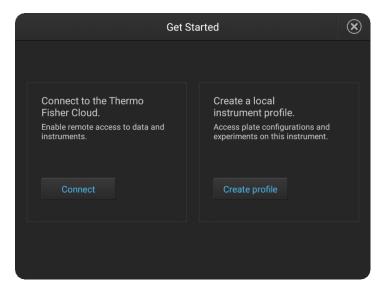
#### Figure 18 Select Directory screen example

- (1) Example IP address and folder locations
- List of folders
- 6. In the Save Destination screen, touch Done.

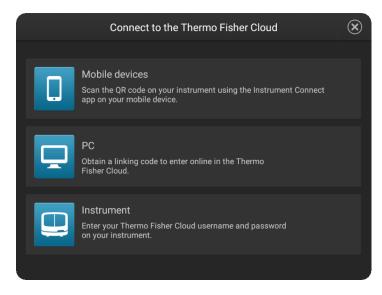
## Link the instrument to your Thermo Fisher Cloud account

**Note:** For detailed information on linking the instrument to your Cloud account, see Appendix B, "Link the instrument to your Cloud account—detailed instructions".

- 1. If a user is signed in, touch (2), then touch **Sign out**.
- 2. In the Sign In screen, touch Get started > Connect.



3. In the Connect to the Thermo Fisher Cloud screen, touch a connection option.





Option	Action	
Mobile devices	<b>Note:</b> Before selecting this option, install and sign in to the InstrumentConnect app on your mobile device.	
	In the Connect to the Thermo Fisher Cloud screen:	
	1. Touch <b>Mobile devices</b> .	
	<ol><li>Hold the camera on your mobile device over the QR code that is displayed on the touchscreen.</li></ol>	
	3. Click <b>Close</b> .	
PC	In the <b>Connect to the Thermo Fisher Cloud</b> screen, a link code is displayed.	
	On a computer:	
	1. Access the Thermo Fisher Cloud.	
	2. Access InstrumentConnect.	
	3. Click Add instrument.	
	4. Select SeqStudio.	
	5. Enter the link code.	
🖵 Instrument	In the <b>Connect to the Thermo Fisher Cloud screen</b> , enter your account information, then click <b>Link account</b> .	

## Lock the touchscreen

During a run, you can lock the touchscreen to prevent other users from using the instrument. This feature is not available to Guest users.

Only the user who locked the touchscreen or an administrator can sign in to the instrument if the touchscreen is locked.

- **1.** Touch **(.)**.
- 2. Touch Profile.
- 3. Touch Lock instrument.

**Note:** If a run is not in progress, **Sign out** is displayed instead of **Lock instrument**.



## Manage plate setups

Export or delete a plate setup (PSM file)

- 1. Touch **Set up run**, then touch **Wy instrument**.
- 2. Touch Manage at the bottom left of the screen.

$\bigcirc$	Setup Run			
		My Plates Public Plate		
		Select a plate name from th	e list	
P	Plate name		Last modified	
F	Plate_20170720_112	2109_J6	2017/07/20 12:22:41	
F	Plate_20170713_100908		2017/07/13 10:11:20	
	Plate_20170713_100	0202	2017/07/13 10:08:50	
	Plate_20170616_141		2017/06/16 14:34:24	
	Plate_20170616_143	3344	2017/06/16 14:34:10	
	Plate_20170616_141			
Mai	nage		Cancel	

- **3.** Touch a plate, then touch **Export** or **Delete**.
- 4. If you touched:
  - **Export**, select a storage location, then touch **Export**.
  - **Delete**, then touch **Yes** to delete the plate setup.



#### Import a plate setup from a CSV or PSM file

**Note:** You can create a plate setup in CSV or PSM file in the Plate Manager (desktop or Cloud).

Before importing a CSV file, see "PSM and CSV plate setup files for import into the instrument" on page 63.

In the home screen:

1. Touch **Set up run**, then touch **S Cloud** , **E Network**, or **D USB**.

		Setu	?	۲	
	1	Where is your pl	ate setup located?		
Clou	d	USB	U My instrument	Network Dr	ive
			or v plate setup		

2. Navigate to, then select a CSV or PSM file.

## Manage run settings (instrument)

Manage run modules This function manages the list of run modules that you can select from when you create a plate setup.

To assign a run module to a plate, see "Assign wells: run module, size standard, and dye set" on page 68.

Factory-installed items cannot be edited or deleted. To create a new item from a factory-installed item, copy, edit, then save the new item.

Run modules are accessible only to the user who creates them.

For more information, see "Run modules, read lengths, size ranges, and run times" on page 123.

1. Access the Manage run modules screen:

From	Action
Plate properties tab	Touch More options > Manage run modules.
Home screen	Touch

- **2.** To create a new run module:
  - **a.** Touch a default run module or a user-created run module to use as a starting point, then touch **Copy**.
  - **b.** Enter values, then touch **Next**.
  - c. Enter a name, touch Advanced to enter additional settings, then Done.

See "Run module settings" on page 123 for detailed information.

**3.** As needed, touch a run module of interest, then touch **Edit** or **View**. **Note:** The **Edit** button is dimmed if a run is in progress.

#### Run module settings

Setting	Description
Run module name	Name of the module
Capillary temperature (°C)	Temperature setting for the capillary array throughout run
Prerun voltage (Volts)	Voltage setting for pre-run before sample injection
Prerun time (seconds)	Prerun time
Injection voltage (Volts)	Voltage for sample injection
Injection time (seconds)	Sample injection time
Run voltage (Volts)	Final sample electrophoresis separation run voltage
Run ramp duration (seconds)	Time required to reach the <b>Run voltage</b>
	<b>Note:</b> Data collection does not start until this time elapses.
Run time (seconds)	Length of time that data is collected after the <b>Run ramp duration</b> elapses

#### Run modules, read lengths, size ranges, and run times

 Table 7
 Sequencing run modules for standard sequencing

Run module	Contiguous read length (CRL) <sup>[1]</sup>	QV threshold	Approximate run time
ShortSeq	≥350	QV30	30 minutes
ShortSeq_BDX			
MediumSeq	≥500	QV30	45 minutes
MediumSeq_BDX			

9



Run module	Contiguous read length (CRL) <sup>[1]</sup>	QV threshold	Approximate run time
LongSeq	≥800	QV20	~ 2 hours
LongSeq_BDX			

<sup>[1]</sup> CRL was determined using the Long Read Sequencing standard. A minimum of 90% of analyzed sequences with an average  $QV \ge QV$  threshold were observed.

IMPORTANT! Use BDX run modules only if you prepare samples with BigDye XTerminator<sup>™</sup> Purification Kit. Use non-BDX run modules for samples purified with other methods.

#### Table 8 Fragment analysis run modules

Run module	Resolution range	Approximate run time	Sizing precision	Compatible size standards
SNaPshot	40–120 bp	25 minutes	40–120: <0.5	GeneScan <sup>™</sup> 120 LIZ <sup>™</sup> Size Standard
FragAnalysis	60–460 bp <sup>[1]</sup>	45 minutes	60-460: <0.15	All except GeneScan <sup>™</sup> 1200 LIZ <sup>™</sup> Size Standard
LongFragAnalysis <sup>[2]</sup>	60-600 bp <sup>[1]</sup>	< 2 hours	60-460: <0.15 461-600: <0.3 601-800: >0.45	<ul> <li>GeneScan<sup>™</sup> 600 LIZ<sup>™</sup> Size Standard v2.0</li> <li>GeneScan<sup>™</sup> 1200 LIZ<sup>™</sup> Size Standard</li> </ul>

[1] Resolution Range: The range of bases over which the resolution (peak spacing interval divided by the peak width at half-max in a GS600 or GS1200 LIZ size standard sample sized with a third order fit) is ≥1. The table shows the resolution range in ≥90% of samples.

<sup>[2]</sup> Load a maximum of 48 samples per plate if you use a long run module.

Note: The following size standards have not been validated for use with the instrument. A default size standard definition is not provided in the software.

- GeneScan<sup>™</sup> 500 LIZ<sup>™</sup> Size Standard
   GeneScan<sup>™</sup> 350 ROX<sup>™</sup> Size Standard
- GeneScan<sup>™</sup> 400HD ROX<sup>™</sup> dye Size Standard

Manage size standard definitions	This function manages the list of size standard definitions that you can select from during plate set up. To assign a size standard definition to a plate, see "Assign wells: run module, size standard, and dye set" on page 68.
	A size standard defines the sizes in basepairs of known fragments. It is used to generate a standard curve. The standard curve is used to determine the sizing of fragments in unknown samples.
	Factory-installed items cannot be edited or deleted. To create a new item from a factory-installed item, copy, edit, then save the new item.
	Size standard definitions are accessible to all users.

1. Access the Manage size standards screen:

From	Actions	
Plate properties tab	Touch More options > Manage size standards.	
Home screen	Touch	

- **2.** To create a new size standard:
  - **a.** Touch a default size standard or a user-created size standard, then touch **Copy**.
  - **b.** Enter a name and select a dye color.
  - c. As needed, touch any of the following, then touch Done:

**Note:** To change an existing value, add a new value, then delete the original value.

- One or more fragment size values, then touch Delete.
- Add to add a value.
- **3.** As needed, touch a size standard of interest, then touch **Edit** or **View**.

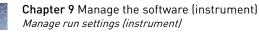
# Manage analysisEdit fragment analysis settingssettingsFactory-installed items cannot be edited or deleted. To create a new item from a<br/>factory-installed item, copy, edit, then save the new item.

Analysis settings are accessible only to the user who creates them.

1. Access the Analysis settings screen:

From	Action
Plate properties tab	Touch <b>More options ▶ Analysis settings</b> , then select the settings to assign to the plate setup.
Home screen	Touch

- 2. To create new analysis settings:
  - **a.** Touch the default analysis settings or user-created analysis settings, then touch **Copy**.
  - b. Enter a name and edit settings as needed (see "Fragment analysis settings (size calling)" on page 109 or "Sequencing settings (base calling)" on page 112).
  - c. As needed, touch View or Delete (user-created settings only).
- 3. Touch Done.



#### Edit sequencing settings

Factory-installed items cannot be edited or deleted. To create a new item from a factory-installed item, copy, edit, then save the new item.

Analysis settings are accessible only to the user who creates them.

1. Access the **Analysis settings** screen:

From	Action
Plate properties tab	Touch <b>More options ▶ Analysis settings</b> , then select the settings to assign to the plate setup.
Home screen	Touch

- **2.** To create new analysis settings:
  - **a**. Touch the default analysis settings or user-created analysis settings, then touch **Copy**.
  - **b.** Enter a name and edit settings as needed (see "Sequencing settings (base calling)" on page 112).
  - c. As needed, touch View or Delete (user-created settings only).
- 3. Touch Done.

#### Fragment analysis settings (size calling)

Setting	Description
Size calling method	• Local Southern—( <i>default</i> ) Determines the fragment sizes using the reciprocal relationship between fragment length and electrophoretic mobility.
	• <b>Global Southern</b> —Compensates for standard fragments with anomalous electrophoretic mobility (similar to least squares methods).
	• <b>2nd LSQ</b> (2nd Order Least Squares)—Uses regression analysis to build a bestfit size calling curve.
	• <b>3rd LSQ</b> (3rd Order Least Squares)—Uses regression analysis to build a bestfit size calling curve.
	• <b>Cubic Spline Interpolation</b> —Forces the sizing curve through all the known points of the selected size standard.

Setting	Description		
Analysis range	• <b>Full Range</b> —( <i>default</i> ) To analyze the entire scan region as collected by the genetic analysis instrument, including the primer peak.		
	<ul> <li>Partial Range—To analyze only data points within a specified range. Enter Start Point in data points after the primer peak and before the first required size standard peak. Enter a Stop Point after the last required size standard fragment. Start and Stop points may vary from instrument to instrument and platform to platform. View raw data to determine the appropriate analysis range. Data points outside the specified analysis range are ignored.</li> </ul>		
	<b>Note:</b> Ensure the <b>Analysis Range</b> contains all size standard fragments included in the <b>Sizing Range</b> .		
Sizing range	The size range (in base pairs) appropriate for the kit you are using:		
	• Full Range for the software to analyze fragments of all sizes in the Analysis Range.		
	• <b>Partial Range</b> for the software to analyze only fragments within a specified range. Enter a <b>Start Size</b> and a <b>Stop Size</b> appropriate for the size standard used.		
Peak amplitudes	The peak height threshold (RFU) for peak detection for each dye color.		
	Peaks below the threshold are not detected. For example, if you use the default values of 175 RFU, peaks with heights equal to or greater than 175 RFU are detected. Peaks with heights below 175 RFU are still displayed in the electropherogram plots but are not detected or labeled.		
	<b>Note:</b> Use the same peak amplitude thresholds in secondary analysis software.		
Primer peak	If the primer peaks in your application obscure peaks of interest, select <b>Present</b> . This instructs the algorithm to ignore primer peaks. Primer peaks are still displayed in the trace.		
	If this setting does not allow detection of the 20- and 40-mer peaks for samples that use the GS600 $LIZ^{M}$ size standard, running samples with the GS600_ $LIZ_{60-600}$ or other size standards that include lower bp starting points may allow detection of the peaks.		
Common settings			
Smoothing	Select an option to smooth the outline of peaks and reduce the number of false peaks detected:		
	• <b>None</b> (default) to apply no smoothing. Best if the data display sharp, narrow peaks of interest.		
	• <b>Light</b> to provide the best results for typical data. Light smoothing slightly reduces peak height.		
	• <b>Heavy</b> for data with very sharp, narrow peaks of interest. Heavy smoothing can significantly reduce peak height.		



Setting	Description		
Baseline Window	Specify a window to adjust the baseline signals of all detected dye colors to the same level for an improved comparison of relative signal intensity. Note the following:		
	• A small baseline window relative to the width of a cluster, or grouping of peaks spatially close to each other, can result in shorter peak heights.		
	• Larger baseline windows relative to the peaks being detected can create an elevated baseline, resulting in peaks that are elevated or not resolved to the baseline.		
Minimum Peak Half Width	Specify the minimum full peak width at half maximum <b>Peak Height</b> required for peak detection. The range is 2 to 99 data points.		
Peak Window Size	Enter a window width in data points for peak detection sensitivity. If more than one peak apex is within the window, all are labeled as a single peak. Note the following:		
	• The maximum value is the number of data points between peaks.		
	The <b>Peak Window Size</b> setting is limited to odd numbers.		
	To increase peak detection sensitivity: Increase polynomial degree, decrease peak window size. To decrease peak detection sensitivity: Decrease polynomial degree, increase peak window size.		
Polynomial Degree	Polynomial Degree cannot be greater than Peak Window Size.		
	Adjust to affect the sensitivity of peak detection. You can adjust this parameter to detect a single base pair difference while minimizing the detection of shoulder effects and/or noise.		
	The peak detector calculates the first derivative of a polynomial curve fitted to the data within a window that is centered on each data point in the analysis range.		
	Using curves with larger polynomial degree values allows the curve to more closely approximate the signal and, therefore, captures more of the peak structure in the electropherogram.		
Slope Thresholds Peak Start and End	• <b>Peak Start</b> —The peak starts when the first derivative (slope of the tangent) in the beginning of the peak signal before the inflection point becomes equal to or exceeds the <b>Peak Start</b> value. This threshold is set to 0 by default, which means that the peak will normally start at the leftmost point where the slope of the tangent is closest to 0° (horizontal line). A value other than 0 moves the peak start point toward its center. The value entered must be non-negative.		
	• <b>Peak End</b> —The peak ends when the first derivative (slope of the tangent) in the end of the peak signal after the inflection point becomes equal to or exceeds the <b>Peak End</b> value. This value is set to 0 by default, which means that the peak will normally end at the rightmost point where the slope of the tangent is closest to 0° (horizontal line). A value other than 0 moves the peak end point toward its center. The value entered in this field must be non-positive.		
	Using curves with larger polynomial degree values allows the curve to more closely approximate the signal and, therefore, captures more of the peak structure in the electropherogram.		

#### Sequencing settings (base calling)

The default settings are optimized for sequencing of PCR amplicons from diploid genes, which are expected to have Mixed bases and which should end At PCR Stop.

For sequencing from plasmid templates, which are of pure sequence (no Mixed bases) and have longer sequence read times than PCR products, create new analysis settings and *disable* (uncheck) **At PCR Stop** and **Mixed base threshold** checkboxes.

Setting	Description
Quality Threshold	<ul> <li>Basecall assignment (ambiguous bases):</li> <li>Do not assign Ns to basecalls</li> </ul>
	<ul> <li>Assign Ns to basecalls with QV&lt;5— Bases with a QV less than the threshold display N instead of the base letter</li> </ul>
	<ul> <li>End base—Last base on which to perform basecalling:</li> <li>At PCR Stop</li> </ul>
	– After X number of bases
	<ul> <li>After X number of Ns in X number of bases</li> </ul>
	<ul> <li>After X number of Ns</li> </ul>
	<b>Note:</b> If you have PCR products with sequences that end while data is still being collected, select the <b>At PCR Stop</b> checkbox.
Mixed bases threshold	When enabled, determines the secondary peak height ratio where the secondary peak is considered a potential mixed base. Reaching the threshold is a necessary but not sufficient condition for the basecalling algorithm to call a mixed base.
Clear range methods	• Use quality values—Sets a window with a specified number of allowed low-quality bases by removing bases until there are <x <y.<="" bases="" number="" of="" per="" qv="" td="" with="" z=""></x>
	• <b>Use base positions</b> —Specifies the first and last base in the range to consider, or trims the specified number of bases from the 3' end.
	• <b>Mask base positions before</b> —Specifies the base position before which to disregard bases.

Modify the default file name	The default file name convention determines how the data files (AB1 or FSA) associated with a plate are named.
convention	The default file name convention is:

Application	Default settings
Fragment analysis	<well>_<sample name="">_<sample type="">_<date and="" timestamp="">.fsa</date></sample></sample></well>
Sequence analysis	<well>_<sample name="">_<date and="" timestamp="">.ab1</date></sample></well>
Sequence analysis with the Sanger variant analysis option selected	<well>_<sample name="">_<amplicon>_<specimen>_<date and="" timestamp="">.ab1</date></specimen></amplicon></sample></well>



1. Access the **File name convention** screen:

From	Action
Plate properties screen	Select the <b>Properties</b> tab, then touch <b>More options &gt; File name convention</b> .
Home screen	Touch

- 2. Touch Attributes.
- **3**. Select the attributes to include in the data file name.

Select Attributes	$\mathbf{x}$
Sequencing Fragment	
Sequencing	
Well number Amplicon Custom Field	1
Sample name Specimen Custom Field	2
Capillary # Instrument name Custom Field	3
Injection number 🖌 Cartridge serial # 📃 Custom Field	4
Custom Field 5	
Reset default Cancel Dor	ie

For information on creating custom fields to include in file name conventions, see "Define custom fields" on page 73.

- 4. Touch Done.
- 5. Touch and drag attributes up or down in the list.
- **6.** Touch **Done**.

## Manage instrument settings

Display instrument hardware and software information In the home screen:

- **1.** Touch ⓐ **Settings** ▶ **Instrument settings** ▶ **About** to access the instrument information:
  - Model name
  - Ethernet IP address
  - Ethernet MAC address
  - Wireless IP address
  - Wireless MAC address

Change the instrument name	<ul> <li>Instrument software release</li> <li>Instrument serial number</li> <li>(Optional) Touch EULA to display the end-user licence agreement or touch Details to display additional instrument information.</li> <li>If the instrument is linked to the Cloud, only a Cloud administrator for the instrument can change the instrument name. The instrument name can be changed by a Cloud administrator on the instrument or in InstrumentConnect.</li> <li>In the home screen: <ol> <li>Touch </li> <li>Settings &gt; Instrument Settings &gt; Instrument name.</li> <li>Touch the Instrument Name field, enter an instrument name, then touch Done.</li> <li>Touch OK.</li> </ol> </li> </ul>
Enable Demo mode	<ul> <li>Demo mode allows you to use all features of the system and provides simulated real-time data and results. You cannot save plates or settings in Demo mode.</li> <li>In the home screen: <ol> <li>Touch</li></ol></li></ul>
Manage date and time settings	<ul> <li>In the home screen:</li> <li>1. Touch (●) Settings &gt; Instrument settings &gt; Date and Time.</li> <li>2. Slide the control to select a time-setting option: <ul> <li>The instrument automatically detects the time via the network.</li> <li>Enter the time manually.</li> </ul> </li> <li>3. Touch Time zone, then select a time zone.</li> <li>4. Touch Date/Format, then select the display order for the month, day, and year.</li> <li>5. Touch Time/Format, then select a 12-Hour or 24-Hour time display format.</li> <li>6. Click OK.</li> </ul>
Manage the network configuration	<b>Note:</b> For a direct connection between the instrument and a computer, set up a wired connection. In the home screen:

- Network Connection

  Wireless

  Status

  Not connected

  Network

  IP address

  10.43.32.82

  Edit

  OK
- **1.** Touch (a) Settings > Insrument settings > Network Configuration.

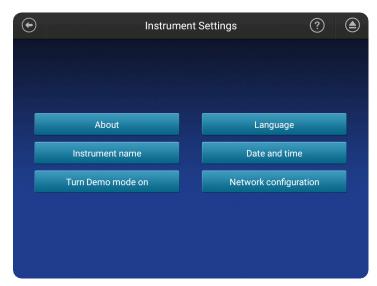
- 2. Touch Edit, or touch one of the network settings fields.
- **3.** (*Optional*) Edit the **Wireless** network settings.
  - a. Select a Network that was automatically detected by instrument.
  - **b.** Enter a password, if prompted.
  - c. Click Join.
  - d. Click OK when the authentication is completed.
- **4.** (*Optional*) Edit the **Wired** network settings.
  - a. Select DHCP or Static IP. An IP address is automatically assigned if DHCP is selected.
  - b. (*Static IP only*) Enter an IP address, Subnet mask, Default gateway, Primary DNS server, and Secondary DNS server.
     For more information, see "Determine IP address for a computer on a network" on page 113.
- 5. Touch OK.

In the home screen:

Check for software updates (administrator only)

Q



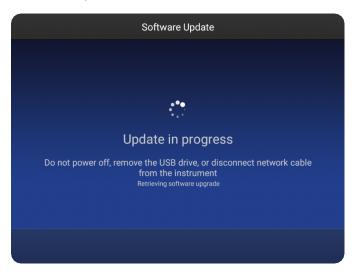


#### 2. Touch Check for updates.

If the software update is located on a USB, it may take 10–15 seconds for the instrument to recognize the USB.

- 3. Touch:
  - **Update** if an update is available.
  - **Cancel** if there is no update available.

A message is displayed during the software updated, then the instrument automatically restarts.





## Manage instrument profiles on the instrument

Instrument profile	Location	Functions allowed
Standard	Local <sup>[1]</sup>	Create, save, open, import, and run plate setups
		<ul><li>Create and modify run settings</li><li>View and export results</li></ul>
Administrator	Local <sup>[1]</sup>	All standard user functions, plus:
		Create or delete an instrument profile
		Change an instrument profile from <b>Standard</b> to <b>Administrator</b> access
		Reset an instrument profile PIN for another user
		Unlock instrument touchscreen
		Backup user data (plates and results)
		• Delete Cloud instrument profile from the instrument, which removes the instrument from the InstrumentConnect. The user can link the instrument to the Cloud again using a local instrument profile. After a user relinks to the Cloud, the Cloud instrument profile is displayed on the home screen and the instrument is listed in the InstrumentConnect.
Guest	Local	All standard user functions, except:
		Cannot link to the Thermo Fisher Cloud
		Cannot modify a public plate setup
		<b>Note:</b> Standard instrument profiles cannot access Guest instrument profile plate setups unless the <b>Plate Setup Security</b> is set to <b>Shared</b> .

<sup>[1]</sup> The first user who signs in to the instrument is assigned a local profile with administrator role.

For more information, see Chapter 3, "Use the instrument with the Thermo Fisher Cloud"  $\,$ 

Create a local<br/>instrument profile<br/>for another user<br/>(administrator<br/>only)An instrument profile can be assigned standard or administrator roles. For more<br/>information, see "Change the role of a local instrument profile (administrator only)"<br/>in page 135.In the home screen:<br/>1. Touch (2).In the home screen:<br/>2. Touch All accounts.3. Touch Add Profile.<br/>4. Touch User name, enter an instrument profile name, then touch Done.

**5.** Touch **PIN (4 digits required)**, enter a four-digit numerical PIN, then touch **Enter**.

	6. Touch <b>Confirm PIN</b> , reenter the PIN, then touch <b>Enter</b> .
	7. Touch Create profile.
	8. Touch Done.
Change the role of	In the home screen:
a local instrument profile	1. Touch <sup>(a)</sup> .
(administrator	2. Touch All accounts.
only)	<b>3.</b> Touch the account of interest.
	<ol> <li>Slide the control from Standard to Administrator or from Administrator to Standard.</li> </ol>
	5. Touch Done.
Delete an instrument profile	Local administrators and Cloud administrators can delete local and Cloud profiles.
from an instrument	<b>IMPORTANT!</b> Before proceeding, back up user data to retain plates and results ("Back up user data (plates and results) (administrator only)" on page 138).
(administrator only)	In the home screen:
	1. Touch 🌒.
	2. Touch All accounts.
	<b>3.</b> Touch the instrument profile to delete.
	4. Touch <b>Delete account</b> .
	5. Touch Yes to confirm.
	6. Touch Done.
Delete the PIN for a local instrument profile (administrator only)	Local administrators and Cloud administrators can delete the PIN for a local instrument profile.
	In the home screen:
	1. Touch <sup>(a)</sup> .
	2. Touch All accounts.
	<b>3.</b> Touch the instrument profile of interest.
	4. Touch <b>Delete PIN</b> .
	The user will be prompted for a new pin upon the next sign in.

- 5. Touch Yes to confirm.
- 6. Touch Done.

Create your own local instrument profile	If you are not signed in to the instrument when you link the instrument to your Cloud account, the software creates a local instrument profile with <b>Standard</b> role using the <i>FirstName LastInitial</i> of your Cloud account.	
	For more information, see "Link the instrument to your Thermo Fisher Cloud account" on page 50.	
Change your own	Sign in to access these features (see "Sign in" on page 33).	
local instrument profile PIN	1. Touch  lefterset.	
	2. Touch Edit.	
	3. Touch Old PIN, enter your current PIN, then touch Enter.	
	Touch the <b>Show PIN</b> checkbox to switch the PIN display on or off.	
	<ol> <li>Touch PIN (4 digits required), enter a new four-digit numerical PIN, then touch Enter.</li> </ol>	
	5. Touch Confirm PIN, reenter your new PIN, then touch Enter.	
	6. Touch Done.	

## Manage storage space

Automatic file Befor cleanup requi

Before starting a run, the instrument calculates the total amount of storage space required to save the run. If the required storage space is not available, the instrument deletes files associated with the oldest exported plates until sufficient space is available.

Note: Only complete plates that have been auto exported (saved to Cloud, network, or USB) or manually exported (using ❀ Settings > Run History > plate name > Export) are deleted.

If the required storage space is not available and no plates have been exported, the instrument displays a notification indicating that there is not enough storage space.

You can export plates and delete plates, then start the run again.

Export or delete a plate setup (PSM file)

1. Touch **Set up run**, then touch **W** instrument.

**2.** Touch **Manage** at the bottom left of the screen.

 $(\bullet)$ Setup Run

Select a plate n	ame from the list
Plate name	Last modified
Plate_20170720_112109_J6	2017/07/20 12:22:41
Plate_20170713_100908	2017/07/13 10:11:20
Plate_20170713_100202	2017/07/13 10:08:50
Plate_20170616_141620000	2017/06/16 14:34:24
Plate_20170616_143344	2017/06/16 14:34:10

- **3.** Touch a plate, then touch **Export** or **Delete**.
- 4. If you touched:
  - **Export**, select a storage location, then touch **Export**.
  - Delete, then touch Yes to delete the plate setup.

Export results from the instrument (sample data files and QC reports)

- In the home screen:
  - 1. Touch ( Settings > Run history.
- 2. Select one or more plates from the **Run History** table.

#### 3. Touch Export.

4. Select a storage location.

The following data is exported for the plate:

- Fragment analysis—FSA file for each sample.
- Sequencing—AB1 file for each sample. ٠
- Plate QC report in CSV and PDF format.

Note: If you select a plate, select View, then select Export, only an FSA or AB1 file for each analyzed sample is exported.

Delete a run history

In the home screen:

- **1.** Touch **③ Settings** ▶ **Run history**.
- 2. Select one or more plates from the Run History table.
- 3. Touch **Delete**, then touch **OK** to confirm.

**Note:** Run histories for the oldest exported plates are automatically deleted if sufficient storage space is not available when you start a run.

#### 1. Select Settings > Maintenance and service > Back up user data.

Back up user data (plates and results) (administrator only)

- 2. Enter a name for the folder that will contain the backup data.
- **3.** Touch the instrument profiles for which you want to back up plate and result information.
- 4. Touch Backup.
- Specify a storage location, then touch Export. A folder containing plate setup and sample data file is backed up to the specified location.

(Optional) After backing up, you can:

- Delete the plates (see "Delete a run history" on page 137).
- Open the CSV files in the folder on another instrument or in the Plate Manager.

## Maintain the instrument



Regular maintenance tasks	139
Manage the cartridge	141
Install cathode buffer	149

## **Regular maintenance tasks**

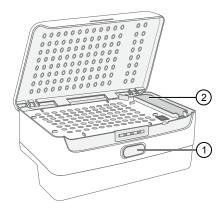
Check the cathode buffer fill level

When a cartridge is installed in the instrument, the volume of buffer in the Cathode Buffer Container (CBC) must be above the fill line.

Check the cathode buffer fill level before each run. It is recommended to check one time each week if the instrument is not in use.

In the home screen:

- 1. Touch (a), touch IIII Eject plate, then open the instrument door when prompted.
- **2.** Press the release button on the autosampler to open the lid, then remove the CBC.



Release button
 CBC

**3.** Ensure that the level of buffer is above the fill line.

If the buffer is at or below the fill line, see "Assemble the SeqStudio<sup>™</sup> Genetic Analyzer Cathode Buffer Container (CBC)" on page 149 and "Insert the Cathode Buffer Container" on page 150.



① Replace if buffer is at or below the fill line



- 4. Reinstall the CBC.
- 5. Close the autosampler lid: Press down on the center of the lid or press down on both sides of the lid with equal pressure until the lid clicks shut.



① New CBC buffer level



6. Touch Retract plate, close the instrument door, then touch Done when the Consumables Status screen is displayed.

Clean the     Power off the instrument before cleaning.       instrument				
exterior and touchscreen	<b>IMPORTANT!</b> The instrument does not maintain the correct temperature conditions for the cartridge when it is powered off. Avoid cartridge exposure to ambient temperature.			
	• Wipe the exterior of the instrument with a lint-free cloth and deionized water.			
	<b>IMPORTANT!</b> Do not allow any moisture to reach the interior of the instrument through the door.			
	• Wipe the touchscreen with a lint-free cloth and glass cleaner.			
Clean the	The autosampler is attached to the instrument.			
autosampler	Clean spills to prevent a build-up of crystallized polymer or dried salt from the buffers.			
	Power off the instrument before cleaning the autosampler.			
	1. Wipe the exterior of the autosampler with a lint-free cloth and deionized water.			
	<b>IMPORTANT!</b> Do not pour liquid directly on to the autosampler. This will damage the heater at the bottom of the cathode buffer reservoir.			
	<b>Note:</b> Do not use detergents or solvents.			

2. Use a lint-free cloth to absorb any liquid spilled in the individual wells of the autosampler.



## Manage the cartridge

### Cartridge storage

		TM	
Table 9	Storage information for the SeqS	Studio <sup>™</sup> Genetic Analyzer	Cartridge (Cat. No. A33671)
	eterage internation for the bege		

Condition	Description
Shipping	Shipped at 2–8°C.
	Store upright at 2–8°C upon receipt.
	Save the white storage box and optical cover for off-instrument cartridge storage.
On-instrument storage	For routine use, can be used and stored on the instrument for up to 4 months. If you store the cartridge on-instrument:
	The instrument must be powered on.
	A Cathode Buffer Container must also be installed.
	The instrument keeps the components under the following conditions when it is powered on and in <b>Cartridge storage mode</b> :
	Optical detection window—Covered
	• <b>Capillary array electrodes</b> —Submerged in cathode buffer (buffer must be above the fill line in the Cathode Buffer Container)
	Polymer—Chilled
	Anode buffer—Ambient temperature
	<b>IMPORTANT!</b> The instrument does not maintain the correct temperature conditions for the cartridge when it is powered off. Avoid cartridge exposure to ambient temperature.
Off-instrument storage	For intermittent use, can be stored off-instrument until the expiry date on the label or up to 4 months after first use. Store upright at 2–8°C, with an integrated capillary protector (ICP) and optical cover installed (see "Store the cartridge" on page 146).
	<b>Note:</b> After you remove the cartridge from the instrument, install an ICP within a few minutes. Avoid cartridge exposure to ambient temperature.
Reuse	Can be removed from an instrument then inserted again on the same instrument or a different instrument, if it was stored properly at 2–8°C and has not expired or exceeded 125 injections.
	Information about the cartridge installation and usage is retained in the cartridge history ( <b>Settings &gt; Cartridge &gt; Instrument-cartridge history</b> ).



Condition	Description
Shipping	Shipped at 2–8°C.
	Store upright at 2–8°C upon receipt.
	Save the white storage box and optical cover for off-instrument cartridge storage.
On-instrument storage	For routine use, can be used and stored on the instrument for up to 6 months. If you store the cartridge on-instrument:
	The instrument must be powered on.
	A Cathode Buffer Container must also be installed.
	The instrument keeps the components under the following conditions when it is powered on and in <b>Cartridge storage mode</b> :
	Optical detection window—Covered
	• <b>Capillary array electrodes</b> —Submerged in cathode buffer (buffer must be above the fill line in the Cathode Buffer Container)
	Polymer—Chilled
	Anode buffer—Ambient temperature
	<b>IMPORTANT!</b> The instrument does not maintain the correct temperature conditions for the cartridge when it is powered off. Avoid cartridge exposure to ambient temperature.
Off-instrument storage	For intermittent use, can be stored off-instrument until the expiry date on the label or up to 6 months after first use. Store upright at 2–8°C, with an integrated capillary protector (ICP) and optical cover installed (see "Store the cartridge" on page 146).
	<b>Note:</b> After you remove the cartridge from the instrument, install an ICP within a few minutes. Avoid cartridge exposure to ambient temperature.
Reuse	Can be removed from an instrument then inserted again on the same instrument or a different instrument, if it was stored properly at 2–8°C and has not expired or exceeded 250 injections.
	Information about the cartridge installation and usage is retained in the cartridge history ( <b>Settings → Cartridge → Instrument–cartridge history</b> ).

	<u> </u>	f → 0		
Table 10	Storage information	tor the SeaStudio	Genetic Analyzer	Cartridge v2 (Cat. No. A41331)
	eter age mier maner		•••••••••••••••••••••••••••••••••••••••	ear

Set cartridge<br/>storage mode<br/>(administrator<br/>only)If a cartridge is installed on the instrument, the instrument will enter Cartridge<br/>storage mode after the time you specify.The instrument keeps the components under the following conditions when it is<br/>powered on and in Cartridge storage mode:<br/>• Optical detection window—Covered

- **Capillary array electrodes**—Submerged in cathode buffer (buffer must be above the fill line in the Cathode Buffer Container)
- **Polymer**—Chilled
- Anode buffer—Ambient temperature

In the home screen:



- 1. Touch (\*) Settings > Cartridge > Cartridge Storage Mode.
- **2.** Select the duration of instrument inactivity before the instrument enters Cartridge Storage Mode.
- 3. Touch OK.

This function lists the cartridges that have been installed on this instrument.

#### Check or export the Instrumentcartridge history

Ins	trument-0	Cartridge History		$(\mathbf{X})$
Serial #	Event	Event time	Injections remaining	
123456789012345678	Insert	2017/07/10 14:11:25		
Export			Close	

1. Touch ( Settings ) Cartridge ) Instrument-cartridge history.

**2.** (*Optional*) Touch **Export**.

#### Check or export the cartridge history

This function lists the instruments on which the cartridge has been installed. Touch (\*) Settings > Consumables status, then touch Cartridge history.

		Cartri	dge History		×
	Instrument	Installed date	Removed date	Injections performed	i
	12345123	2017/05/10 16:33:23	NA	73	
_					
	Export			Close	



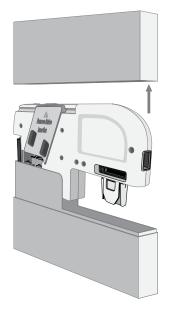
Fill the capillary array and refresh the polymer delivery system	<ul> <li>The functions accessed from Cartridge maintenance are performed automatically during a run. Do not use these commands to manually perform these functions unless instructed to do so in troubleshooting or by Support.</li> <li>In the home screen: <ol> <li>Touch </li> <li>Settings &gt; Cartridge &gt; Cartridge maintenance.</li> </ol> </li> <li>Select Fill array or Refresh PDS.</li> <li>Touch </li> <li>to close the screen when the function ends.</li> </ul>
Remove the cartridge	<ul> <li>3. Touch S to close the screent when the function ends.</li> <li>Perform these steps if you are replacing the cartridge or storing it off-instrument. Removal of the cartridge after each run is not required.</li> <li>For appropriate off-instrument storage conditions, see the following tables: <ul> <li>For the SeqStudio<sup>™</sup> Genetic Analyzer Cartridge, see Table 1 on page 17</li> <li>For the SeqStudio<sup>™</sup> Genetic Analyzer Cartridge v2, see Table 2 on page 18</li> </ul> </li> <li>1. Touch (a), touch (c) Eject cartridge, then open the instrument door when prompted.</li> <li>2. Hold the cartridge at the hand hold above the capillaries, then pull to remove it from the instrument.</li> <li>3. Close the instrument door.</li> </ul>
Insert the cartridge	<ul> <li>For more information, see "Store the cartridge" on page 146.</li> <li>1. If a cartridge is installed: <ul> <li>a. Touch <ul> <li>a. touch <ul> <li>b. Eject cartridge, then open the instrument door when prompted.</li> </ul> </li> </ul></li></ul></li></ul>

**b.** Remove the used cartridge.



- **2.** Prepare the cartridge.
  - **a.** Remove the cartridge from the box.

Note: Save the box for off-instrument cartridge storage.

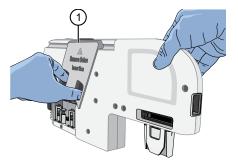


**b.** Remove the optical cover from the cartridge by grasping the finger holds, then pulling toward you.

**Note:** Save the optical cover for offinstrument cartridge storage. Avoid touching the capillaries that are protected by the optical cover.

c. Pinch the clamp on the SeqStudio<sup>™</sup> Integrated Capillary Protector, then pull down.

Discard the SeqStudio<sup>™</sup> Integrated Capillary Protector. The SeqStudio<sup>™</sup> Integrated Capillary Protector is single-use. Use a new SeqStudio<sup>™</sup> Integrated Capillary Protector if you store the cartridge.

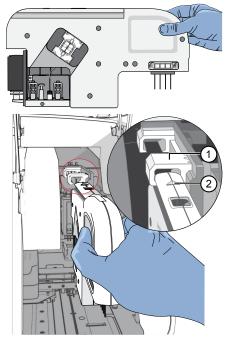








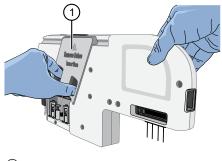
- **3.** Grasp the cartridge above the capillaries.
- **4.** Position the cartridge:
  - **a.** Orient the cartridge with the embossed arrow pointing toward the rear of the instrument.
  - **b.** Align the guides at the top of the cartridge with the insertion rails in the instrument.
- **5.** Slide the cartridge into the instrument until it clicks into place.
- 6. Close the instrument door, then touch Done when the Consumables Status screen is displayed. The cartridge is initialized.



- Insertion rails
- $\textcircled{2} \ \mathsf{Embossed} \ \mathsf{arrow}$

Store the cartridge

- 1. Touch (a), touch (c) **Eject cartridge**, then open the instrument door when prompted.
- **2.** Hold the cartridge at the hand holds above the capillaries, then pull to remove it from the instrument.
- **3.** Close the instrument door.
- Carefully place the optical cover on the cartridge to avoid damage to the capillaries.



Optical cover



- 5. Place a new SeqStudio<sup>™</sup> Integrated Capillary Protector on the cartridge: pinch the clamp on the ICP, then push up on to the capillaries.
- **6.** Place the cartridge in the white storage box.
- **7.** Store upright at 2–8°C.

**Note:** Avoid cartridge exposure to ambient temperature.

**IMPORTANT!** Do not freeze the cartridge. It cannot be used after freezing.

SeqStudio<sup>™</sup> Integrated Capillary Protector The SeqStudio<sup>™</sup> Integrated Capillary Protector (ICP) is a single-use protective cover that clamps onto the SeqStudio<sup>™</sup> Genetic Analyzer Cartridge or the SeqStudio<sup>™</sup> Genetic Analyzer Cartridge v2. The ICP prevents the capillary array from drying out during off-instrument storage of the cartridge, and is removed before insertion of the cartridge into the SeqStudio<sup>™</sup> Genetic Analyzer.

Each cartridge is supplied with an ICP for shipping and one additional ICP for offinstrument cartridge storage.

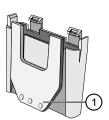


Figure 19 SeqStudio<sup>™</sup> Integrated Capillary Protector ① Clamp mechanism for attaching to cartridge

**IMPORTANT!** Remove the Integrated Capillary Protector before installing the cartridge into the instrument. Installing the cartridge with the ICP in place can damage the capillary array.



#### SeqStudio<sup>™</sup> Integrated Capillary Protector storage

Condition	Description	
Shipping	Is shipped at 2–8°C.	
	Before opening, can be stored until expiry date on label at 2–8°C.	
	Discard the shipping ICP when you insert a new cartridge in the instrument.	
Reuse	Do not reuse.	
	Use a new ICP for off-instrument cartridge storage.	
	<b>IMPORTANT!</b> If an ICP is used for off-instrument cartridge storage more than one time, the capillary array will not be kept fully hydrated.	

#### Long-term oninstrument cartridge storage

You can leave the cartridge installed in the instrument if the instrument is powered on and a CBC is installed. Replace the CBC every 2 weeks.

**IMPORTANT!** If the instrument will be idle for longer than the shelf life of the cartridge components, remove the cartridge and cathode buffer, then power off the instrument.

 Table 11
 Cartridge storage

Cartridge	Storage
SeqStudio <sup>™</sup> Genetic Analyzer Cartridge (Cat. No. A33671)	See Table 1 on page 17
SeqStudio <sup>™</sup> Genetic Analyzer Cartridge v2 (Cat. No. A41331)	See Table 2 on page 18

**IMPORTANT!** The instrument does not maintain the correct temperature conditions for the cartridge when it is powered off. Avoid cartridge exposure to ambient temperature.



## Install cathode buffer

#### SeqStudio<sup>™</sup> Genetic Analyzer Cathode Buffer Container storage

Condition	Description	
Shipping	Is shipped at ambient temperature.	
	Store at 2–8°C upon receipt.	
On-instrument storage	After installation, can be stored for up to 2 weeks when the instrument is powered on and in <b>Cartridge storage mode</b> .	
Off-instrument storage	Before opening, can be stored until expiry date on label at 2–8°C.	
Reuse	Do not remove the CBC from the instrument for storage. Do not reuse.	

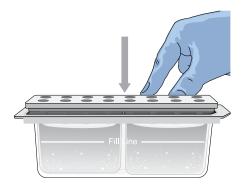
Remove the SeqStudio <sup>™</sup>	Perform these steps if you are replacing the Cathode Buffer Container. It is not necessary to remove the CBC after each run.
Genetic Analyzer Cathode Buffer	1. Touch (a), touch IIII Eject plate, then open the instrument door when prompted.
Container	<b>2.</b> Press the release button on the autosampler to open the lid.
	<b>3.</b> Lift the CBC out of the autosampler.
Assemble the SeqStudio <sup>™</sup>	Equilibrate the CBC to room temperature (15 minutes to overnight) before assembling.
Genetic Analyzer Cathode Buffer	On a clean and level surface:
Container (CBC)	<b>1.</b> Ensure that the buffer is above the fill line.
	<b>2.</b> Carefully peel off the seal.



**3.** Wipe off any buffer on top of the CBC with a lint-free tissue. Ensure that the top of the container is dry.



**4.** Place the reservoir septa on the CBC, then press firmly to seat the septa.



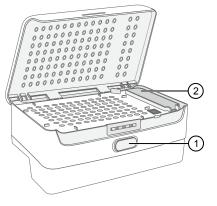
**Note:** The CBC is filled significantly above Fill Line to account for evaporation. Replace the CBC when the fluid level is at or below the fill line.

**Insert the Cathode** Perform these steps if the Cathode Buffer Container has not been installed, if the CBC on the instrument has expired, or if the buffer level is at or below the fill line.

Install the CBC before you install the cartridge.

See Figure 2 on page 13 for the position of the cathode buffer on the instrument.

- 1. Touch (a), touch IIII Eject plate, then open the instrument door when prompted.
- 2. Press the release button on the autosampler to open the lid.



Release button
 Location of CBC

**3.** Insert the cathode buffer in the autosampler with the notch positioned in the back right.

See Figure 8 on page 19.

10

**4.** Press the autosampler lid until it clicks shut.



5. Touch **Retract plate**, then close the instrument door.



# Troubleshooting

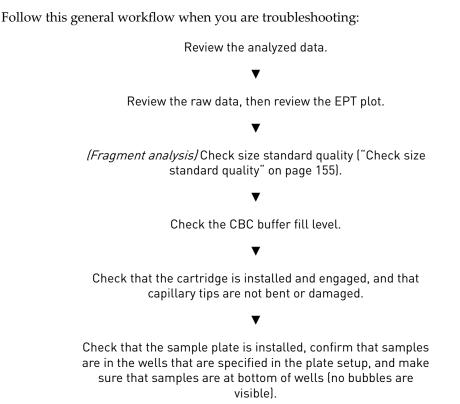
Troubleshooting resources
Troubleshooting workflow 153
Export log files for plates, install runs, injections, and instrument 153
View the raw data and the EPT plot 154
Data quality alerts
Sizecalling and basecalling quality alerts
Check size standard quality 155
Instrument troubleshooting 156
Cartridge troubleshooting 157
Sample and data troubleshooting 158
EPT data
Software troubleshooting 164
Service tools

# Troubleshooting resources

Document	Pub. No.
Troubleshooting Sanger sequencing data User Bulletin	MAN0014435
DNA Sequencing by Capillary Electrophoresis Chemistry Guide Second Edition	4305080
DNA Fragment Analysis by Capillary Electrophoresis User Guide	4474504



#### **Troubleshooting workflow**



#### Export log files for plates, install runs, injections, and instrument

Export log files and provide them to Thermo Fisher Scientific if directed to do so by Technical Support.

In the home screen:

- 1. Touch (\*) Settings > Maintenance and Service > Export logs.
- 2. Select one of the following options.
  - **Export logs for the last injection**—Includes all run types: analysis run, install run, and calibration run.
  - Export logs for the last / current plate—Includes all run types.
  - **Export logs for selected plates**—Includes options to export logs for regular runs and install check runs for the analysis run for the current user.
  - Export recent instrument server logs Includes instrument export logs only.
- 3. Touch Export.
- 4. Select a storage location.

Note: Log files can be large. Select a location with adequate storage space.

A ZIP file containing log information is exported.



## View the raw data and the EPT plot

The EPT view (ElectroPhoresis Telemetry) shows instrument data conditions (currents, temperatures, electrophoresis voltage) as a function of time.

When a run is complete, in the home screen:

- 1. Touch Results.
- 2. Touch List view.
- **3.** Touch an injection group.
- Touch a sample file name.
   If the data triggered any quality alerts, a QC alerts screen is displayed.
   Click View data to display the trace for the sample.
- 5. Touch > or < to scroll to the raw data or EPT Plot.

### Data quality alerts

Quality alert	Description	Action
Offscale peaks. Adjust the injection parameters and/or the sample concentration.	At least 10 scans have saturated the CCD camera.	<ul><li>Reduce the injection voltage.</li><li>Dilute the sample.</li></ul>
No sample was detected.	Poor signal-to-noise ratio with low signal detected.	<ul> <li>Verify that the sample volume follows recommendations in the user manual.</li> <li>Troubleshoot upstream PCR and sequencing steps.</li> </ul>

#### Sizecalling and basecalling quality alerts

 Table 12
 Sizecalling quality alerts

Quality alert	Description	Action
Sizing quality value is low due to poor size standard peak quality. Peak height uniformity is low or the fitting quality in sizing is poor.	Low resolution or poor quality data is present.	<ul> <li>Re-inject the sample.</li> <li>If the problem persists, check the sample quality.</li> </ul>
Sizecaller found broad peak(s) in the size standard peak(s).		
Sizing quality value is in the intermediate range; check size standard data quality.		



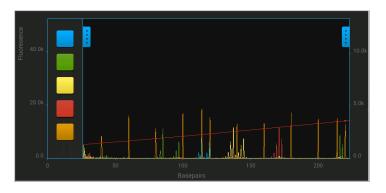
Quality alert	Description	Action
The number of size standard peaks detected is less than what is defined in the size standard.	Size standard definition includes peaks that are not present in the sample. Example: Sample peaks are detected up to 500 bp, but the size standard definition includes peak sizes that are >500 bp.	Use or create a size standard definition with the appropriate number of peaks and peak sizes.
The analysis range is too small. Correct the analysis range in analysis settings and re- analyze in secondary analysis software or re- inject sample.	Various causes.	<ul> <li>Analyze the data in a secondary analysis software with a corrected analysis range.</li> <li>Re-inject the sample.</li> </ul>

 Table 13
 Basecalling quality alerts

Quality alert	Description	Action
Basecalling failed due to poor quality data.	Poor quality data is present.	<ul> <li>Re-inject the sample.</li> <li>If the problem persists, prepare fresh sample.</li> <li>Troubleshoot upstream PCR and sequencing steps.</li> </ul>

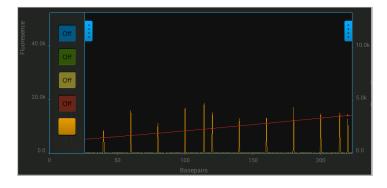
#### Check size standard quality

- **1.** Touch **③ Settings** ▶ **Run history**.
- **2.** Touch a plate name, then touch **View**.
- **3.** Touch a sample file name, then touch **View**. If the data triggered any quality alerts, a QC alerts screen is displayed.
- 4. Touch **View data** to display the trace.





**5.** Touch on the left border of the trace, then deselect all dyes except the size standard dye (red or orange).



**6.** As needed, touch on the right border of the trace to zoom on the trace.

## Instrument troubleshooting

Observation	Possible cause	Recommended action
There was a loss of power to the instrument	There was a power failure.	Restart the instrument. The <b>Sign In</b> screen is displayed. A run that was in progress at the time of the power failure must be restarted.
		Run a control sample to ensure that the consumables have not degraded, especially if the consumables reached room temperature.
		Replace the consumables if the results with a control sample show that the consumables have degraded.
The RFID tags on the cartridge or the cathode buffer are not	The label on the cartridge or the Cathode Buffer Container is	Ensure that the labels are present and not visibly damaged.
read	damaged, not positioned properly, or has been removed.	Ensure that the consumables are installed correctly. The Cathode Buffer Container should be firmly seated in the autosampler. The cartridge will click into place when it is installed correctly.
	The RFID reader is malfunctioning.	Insert consumables that have previously had RFID tags read. Contact technical support if the RFID tags are not read.
The run stopped and <b>Resume</b> is displayed	A user paused the run.	Touch <b>Resume</b> to continue the run.
The amber light is blinking	The run was paused.	Touch <b>Resume</b> to continue the run.
	The instrument door is open.	Close the instrument door.
	There was an instrument error.	Follow the instructions in the error message.
		Restart the instrument.
The electrophoresis failed or <b>Current check failed</b> is displayed	There is insufficient cathode buffer.	Check the fill line on the Cathode Buffer Container. Replace the Cathode Buffer Container if the buffer is at or below the fill line.



Observation	Possible cause	Recommended action
The electrophoresis failed or <b>Current check failed</b> is displayed	The Cathode Buffer Container has been installed on the instrument for more than two weeks or used for more than 125 injections.	Replace the Cathode Buffer Container.
	The septum on either the Cathode Buffer Container or the sample plate is not installed correctly.	Ensure that the septa are fully inserted into both the Cathode Buffer Container and the sample plate.
	Buffer or other liquid was spilled on top of the reservoir septum or on top of the autosampler.	Wipe the spill with a lint-free cloth.
	There is condensation on the Cathode Buffer Container or around the reservoir septum.	Wipe the condensation with a lint-free cloth.
		Ensure that the humidity in the lab is non- condensing.

# Cartridge troubleshooting

Observation	Possible cause	Recommended action	
There are crystals on the cartridge capillary array	Small amounts of leakage around the cartridge capillary array are normal.	No action is required.	
There are crystals on the cartridge polymer delivery	There is a leak in the polymer delivery system.	Run a control sample to determine if the cartridge function is affected.	
system		Replace the cartridge if the leak affects the cartridge function.	
Poor-quality data is observed after prolonged drying of the capillary tips	The capillary tips develop blockages if they are allowed to dry out.	Use an Integrated Capillary Protector when the cartridge is off the instrument to prevent the capillary tips from drying out.	
		Clear the blockage by running polymer through the capillaries.	
		<ol> <li>Touch ☆ Settings ➤ Cartridge ➤ Cartridge maintenance ➤ Fill array.</li> </ol>	
		2. Touch 😢 when the function ends.	
		3. Run a control sample.	
		<ol> <li>Replace the cartridge if there is poor- quality data with the control sample.</li> </ol>	
		Follow the recommendations for cartridge storage to prevent drying of the capillary tips. See "Cartridge storage" on page 17.	



## Sample and data troubleshooting

See also "Data quality alerts" on page 102, "Sizecalling and basecalling quality alerts" on page 102, and *Troubleshooting Sanger sequencing data User Bulletin* (Pub. No. MAN0014435).

Observation	Possible cause	Recommended action
Offscale signal is detected is displayed	The sample concentration is too high.	Dilute the sample, prepare a new plate, and start a new run.
<b>Note:</b> Samples are stable on the instrument for 16–24 hours.		Quantitate the sample prior to adding reagents for capillary electrophoresis.
Determine if samples will be stable if a re-injection is	The injection conditions are too strong for the sample.	Re-inject the samples with adjusted injection conditions.
recommended, then plan the re-injection accordingly.		Injection time, voltage, or a combination of these conditions can be adjusted.
There is no signal or a low	The sample volume was	Use the recommended sample volume.
signal <b>Note:</b> Samples are stable on	insufficient.	See "Sample preparation guidelines" on page 36.
the instrument for 16–24 hours. Determine if samples will be stable if a re-injection is	The sample concentration was too low.	Re-inject the samples with adjusted injection conditions.
recommended, the plan the re- injection accordingly.		Injection time, voltage, or a combination of these conditions can be adjusted.
	There were bubbles in the sample wells.	Centrifuge the sample plate or tubes to remove the bubbles before loading onto the instrument.
		If the samples have been run, centrifuge the plate or tubes, then set up a re-injection.
	The sequence reaction failed.	Review the sequence analysis protocol, the template quality, and the template quantity. Set up a new plate and repeat the reaction.
		See <i>Troubleshooting Sanger sequencing data User Bulletin</i> (Pub. No. MAN0014435).
	The Hi-Di <sup>™</sup> Formamide used to prepare the samples was	Prepare the samples with fresh Hi-Di <sup>™</sup> Formamide and repeat the experiment.
	degraded.	See "Sample preparation guidelines" on page 36.
	The sample was prepared with the BigDye XTerminator <sup>™</sup> Purification Kit but a <b>BDX</b> run module was not selected.	Re-inject the sample with the correct module.
	The sample was degraded.	Prepare the sample according to the protocol provided with the kits for sample preparation. See "Sample preparation guidelines" on page 36 for sample preparation guidelines.
		<b>IMPORTANT!</b> Do not resuspend samples in water.



Observation	Possible cause	Recommended action
There is no signal or a low signal	The sample had a high concentration of salt.	Dilute or desalt the samples.
<b>Note:</b> Samples are stable on the instrument for 16–24 hours. Determine if samples will be stable if a re-injection is recommended, the plan the re- injection accordingly.	There was an excess of unlabeled template competing with the fragments labeled with dye during the injection.	<ul> <li>Dilute or desalt the samples.</li> <li>See: <ul> <li>DNA Sequencing by Capillary</li> <li>Electrophoresis Chemistry Guide Second</li> <li>Edition (Pub. No. 4305080)</li> </ul> </li> <li>DNA Fragment Analysis by Capillary <ul> <li>Electrophoresis User Guide (Pub.</li> <li>No. 4474504)</li> </ul> </li> </ul>
	A capillary array tip is blocked.	<ul> <li>Flush the capillary array.</li> <li>1. Touch (Instable) Settings &gt; Cartridge &gt; Cartridge maintenance.</li> <li>2. Touch Refresh PDS.</li> <li>3. Touch (Instable) to close the screen when the function ends.</li> </ul>
		Replace the cartridge if the problem persists.
	The cartridge is damaged.	Inspect the cartridge for damage. Replace the cartridge if there is damage.
There was poor resolution, poor size quality, or a poor sequencing result	The samples degraded over time while on the instrument.	Analyze a maximum of 48 samples on a plate for long run modules. Additional samples will take more than 24 hours and the samples can degrade. <b>Note:</b> Thermal breakdown of samples is normal. Samples are stable for 16–24 hours on
		the instrument. Use Hi-Di <sup>™</sup> Formamide to prepare the samples. Sample stability is optimal in Hi-Di <sup>™</sup> Formamide.
	The temperature and/or humidity in the lab is too high for optimal sample stability.	Ensure that the conditions in the lab are within the operating range for the instrument (15– 30°C and 20–80% relative humidity).
		Sample stability can be lower near the high end of the operating range of the instrument for temperature and humidity.
		Use 20 $\mu$ L of sample instead of 10 $\mu$ L.
		A larger sample volume can reduce sample breakdown under hot and humid conditions.
	There was a sporadic data quality failure.	Repeat the injection.
	<b>Note:</b> A sporadic data quality failure can happen occasionally.	
Poor resolution in some capillaries	Poor-quality samples were used.	See "Sample preparation guidelines" on page 36.



Observation	Possible cause	Recommended action
Poor resolution in some capillaries		Use a control sample to determine if the poor resolution is due to the samples are another factor.
	A capillary was damaged or is blocked.	Try to clear the capillary, then replace the cartridge if the poor resolution is due to the cartridge and not the sample.
		<ol> <li>Touch Settings ➤ Cartridge ➤ Cartridge maintenance ➤ Fill array.</li> </ol>
		2. Touch 🛿 when the function ends.
		3. Run a control sample.
Poor resolution in all the capillaries	The cartridge has been used for more than the stated number of injections, is past the labeled expiration date, or has degraded polymer from incorrect storage.	Replace the cartridge.
	The Hi-Di <sup>™</sup> Formamide that was used to prepare the samples was degraded.	Prepare the samples with fresh Hi-Di <sup>™</sup> Formamide and repeat the reaction. See:
		<ul> <li>"Sample preparation guidelines" on page 36</li> </ul>
		• <i>DNA Fragment Analysis by Capillary Electrophoresis User Guide</i> (Pub. No. 4474504)
	There was too much sample injected.	Dilute the sample and re-inject it.
Spikes are present in raw and/or analyzed fluorescence data	Trace impurities passed the detector.	For fragment analysis, increasing the <b>Minimum peak half width</b> setting in the analysis settings (under <b>Common settings</b> ) can reduce the identification of spikes as peaks.
		<b>Note:</b> Secondary sequencing and fragment analysis software can recognize and ignore spikes.
		Repeat the injection if necessary.
There is noise in the baseline	Fluorescent contamination has built up in the CBC.	Replace the CBC. Use a new reservoir septum when assembling the new CBC.
Sample carryover from a previous injection	Sample carryover on the SeqStudio <sup>™</sup> Genetic Analyzer can occur in trace amounts when previous injections are off-scale. Very strong peaks, for example, primer peaks in a	Replace the reservoir septum on the CBC before the next injection to minimize the carryover effect.
	fragment analysis sample, are typically visible in subsequent injections.	



Observation	Possible cause	Recommended action
Dye blobs are seen in the sequencing data	Impurities remained in the sample after the sample purification. The impurities cause dye blobs to appear in the sequencing data.	Improve the sample purification method. See "Sample preparation guidelines" on page 36 for guidelines.
Extra peaks are present in the sequencing traces	There was renaturation of the sample.	Heat-denature the samples prepared with fresh Hi-Di <sup>™</sup> Formamide, then immediately place the samples on ice.
	There is low signal. With very low signal, the peaks are barely visible in the baseline noise.	Check the raw data, the raw data signal intensity, and average raw signal-to-noise ratio, then:
		<ul> <li>Increase the injection time and reinject.</li> <li>Or</li> </ul>
		<ul> <li>Remake the sample. Ensure that you are using:</li> </ul>
		<ul> <li>Enough sequencing template</li> </ul>
		<ul> <li>Enough primer and/or a sufficient concentration of primer</li> </ul>
ii t F a r	There is a heterozygous insertion-deletion (het indel) that is causing multiple peaks to appear at the same basecall position. The sequence can appear "clean" for some number of bases until the het	Examine the analyzed trace. A het indel typically has single peaks at the 5' end, then part-way through the trace, two peaks appear in almost every position to the end of the trace. This pattern occurs when one copy of the gene has an insertion or deletion relative to the other copy of the gene.
	indel is encountered.	When aligning your sequence to a reference sequence, a series of bases may have been inserted or deleted in an allele. These indels can be encountered in any number of bases after the gene-specific priming region. To confirm that the het indel is present in both directions of your target, check the sequencing in the opposite direction.



Observation	Possible cause	Recommended action
Extra peaks are present in the sequencing traces	Primer-dimer has occurred.	You can often diagnose primer-dimer by looking at the raw trace data for questionable sequences. When primer-dimer exists, the 5' sequence signal may be significantly higher for a region of bases spanning the length of the forward and reverse gene-specific primers.
		Primer-dimer is the annealing of the 3' end of primers during PCR. The resulting short annealed fragment may amplify more efficiently than fully extended template. Primer-dimer fragments amplified during PCR can display increased 5' signal and extra peaks when multiple PCR products are sequenced simultaneously. In some instances, the secondary or extra peaks can be read as the reverse compliment of the PCR primers in this noisy 5' region. The secondary sequence or multiple PCR product sequences appear as far as 100–200 bp into the sequence, then suddenly disappear.
	The PCR amplification primers do not have specificity and are sequencing two different regions of the genome. The analyzed trace shows extra peaks throughout the entire length of the trace.	Redesign the primers or increase the amplification temperature.
	You accidentally contaminated the DNA and are sequencing two templates at the same time. The analyzed trace shows extra peaks throughout the entire length of the trace.	Repeat the amplification and sequencing reactions with uncontaminated DNA.
	There were impure or contaminated primers.	Primer stocks may have inadvertently had other primer solution introduced. For best results, use HPLC to purify the primers.
	There was a contaminated sample well.	Use a new sample plate and buffer/wash septa whenever possible.
		To avoid getting sample into adjacent wells, centrifuge the plates before you remove the adhesive seal.
There are pull-up or pull-down peaks in the data	The manual dye calibration is not current or is not matched to the samples, or a high-quality sample was not run with the dye set for the first time in the absence of a manual calibration.	If the dye set is a fragment analysis dye set that is being used for the first time, run a manual dye calibration.
		Alternatively, if a high-quality sample is run with an uncalibrated dye set, and the automated spectral calibration successfully generates an optimized matrix, manual calibration is not absolutely required. See "Determine if manual calibration is required" on page 177.



Observation	Possible cause	Recommended action
There are pull-up or pull-down peaks in the data	The incorrect dye set was selected in the <b>Plate setup</b> .	Correct the dye set and repeat the injection.
	<b>Note:</b> This is applicable to both sequence analysis and fragment analysis.	
There are error messages about spectral issues These error messages are seen	A high-quality sample has not been run with the dye set.	If the dye set is a fragment analysis dye set that is being used for the first time, run a manual dye calibration.
more often in fragment analysis data than sequence analysis data.		Alternatively, if a high-quality sample is run with an uncalibrated dye set, and the automated spectral calibration successfully generates an optimized matrix, manual calibration is not absolutely required. See "Determine if manual calibration is required" on page 177.
There is a short read length and uneven peak spacing in sequence data	The incorrect dye set was selected in the <b>Plate setup</b> .	Correct the dye set and repeat the injection. Select from the following: • E_BigDye <sup>™</sup> Terminator v1.1 • Z_BigDye <sup>™</sup> Terminator v3.1 • Z_BigDye <sup>™</sup> Direct Reanalyze the data in the Sequencing Analysis Software, using the correct mobility file.
Fragment analysis peaks are sized differently than previously observed	Aging of the polymer in the cartridge, which can cause small (<0.5 bp) changes in fragment size.	<ul> <li>Use a reference marker (for example, an allelic ladder) for auto bin adjustment.</li> <li>Or</li> <li>Manually adjust the fragment bin positions to account for the size change.</li> </ul>

## EPT data

Observation	Possible cause	Recommended action
There is a fluctuating or unstable electrophoresis current, or <b>Current check</b> failed is displayed	k 1. louch (❀) Settings > Cartridge > Car maintenance. 2. Touch Refresh PDS.	<ol> <li>Touch</li></ol>
		Replace the cartridge if the problem persists.
	The cartridge is damaged.	Inspect the cartridge for damage and replace the cartridge.



# Software troubleshooting

Observation	Possible cause	Recommended action
Strikethrough text is displayed in Save location in Plate properties screen Save location USB; Cloud; Instrument 1 (1) (1) Strikethrough text	The original location to which the plate was saved is no longer accessible by the instrument.	No action.
Import failed message when you select a CSV plate setup on the instrument The message also indicates that a valid run module, dye set, and/or size standard is not present.	The CSV file specifies the name of a run module, dye set, and/or size standard that does not exist on the instrument. IMPORTANT! A CSV file contains only the name of the run module, dye set, and/or size standard, it does not contain the settings.	Add the run module, dye set, and/or size standard to the instrument by selecting a PSM file that contains the items, or by creating the items manually.
Import CSV fails but import PSM with same settings imports with no errors	The size standard, dye set, or run module specified in the CSV file does not exist on the instrument. The CSV file contains the <i>names only</i> of the size standard, dye set, or run module, it does not contain the settings.	Create the size standard, dye set, or run module on the instrument.
When saving a PSM file in Plate Manager (Cloud), "You do not have edit permission to the cloud group" message is displayed	The PSM file contains the names and settings of the size standard, dye set, or run module. The size standard, dye set, or run module are automatically created when the PSM file is imported. You are saving to a Thermo Fisher Cloud group and you do not have edit permissions for the group.	Save to a different location, or request edit permissions from the group administrator.



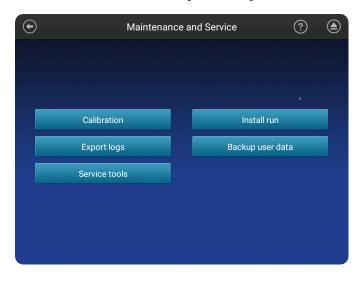
Observation	Possible cause	Recommended action
Analysis settings or run module is not available for selection by some users	Analysis settings and run modules are saved per user.	Change the plate setup security from <b>Hidden</b> to
This situation is seen on the instrument.	If the analysis settings or run modules are associated with a <i>hidden</i> plate setup, the	<b>Shared</b> (see "Hide or share a plate (Plate setup security) " on page 73).
	analysis settings or run modules are not listed for selection unless the user who created the plate setup is signed in.	Create the analysis settings or run modules manually.
Your local instrument profile name is not available for sign in	Your local instrument profile is linked to your Cloud account and is replaced by	See "Local instrument profile roles and functions" on page 134.
	your Cloud profile.	Sign in with your Cloud profile.
Cloud account is already linked to a local profile message	You have a local instrument profile that has previously been linked to Cloud account. You touched <b>Get</b> <b>Started ▶ Connect</b> , then typed in your Cloud account email and password.	Return to <b>Sign in</b> screen, then select your local profile from the <b>Sign in</b> list.
	Another user was signed in to the instrument. You selected Cloud in the Set up run screen or in the Save location field in the Plate properties screen, then typed in your Cloud account email and password.	Return to <b>Sign in</b> screen, touch (2), touch <b>Sign out</b> , then select your local profile from the <b>Sign in</b> list.
The <b>Notifications</b> button is not displayed in <b>Instrument Settings</b>	You are signed in to the instrument with a local instrument profile.	Sign in to the instrument with a Cloud instrument profile.
A button is dimmed	The function is available to users with administrator role only.	None.
Sample data files from the SeqStudio <sup>™</sup> Genetic Analyzer can be opened but not analyzed in secondary analysis software • Sequencing Analysis Software	An updated version of the secondary analysis software is required.	Download and run the latest version of secondary analysis software. See "Secondary analysis software" on page 28.
<ul> <li>GeneMapper<sup>™</sup> Software</li> <li>SeqScape<sup>™</sup> Software</li> <li>Variant Reporter<sup>™</sup> Software</li> </ul>		Note: The SeqStudio <sup>™</sup> Genetic Analyzer instrument model is listed as <b>3200</b> .
Files cannot be imported into Thermo Fisher Cloud applications or cannot be analyzed in	The sample name or file name used a special	Rename the sample name or file name.
Thermo Fisher Cloud sequencing apps	character.	<b>Note:</b> Special characters {/, &, @, %} will impact file import into Thermo Fisher Cloud applications.



Observation	Possible cause	Recommended action
<b>Unexpected error</b> is displayed	There was a software error.	Follow the instructions in the error message.
		Export the log files to determine the potential source of the unexpected error. See "Export log files for plates, install runs, injections, and instrument" on page 153.
		Restart the system using the On/Off switch on the rear panel. See "Power on the instrument" on page 33.

#### Service tools

The Service tools function is password-protected and for service use only.





# Link the instrument to your Cloud account—detailed instructions

Workflow: Set up the instrument for Thermo Fisher Cloud access	167
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Link the instrument from a mobile device	168
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#### Workflow: Set up the instrument for Thermo Fisher Cloud access

Register and obtain a Thermo Fisher Connect account (page 49)

▼

Link the instrument to the Thermo Fisher Cloud in any of the following ways:

- Link the instrument from a mobile device (page 168)
- Link the instrument using a link code (page 169)
- Link the instrument using your Cloud account (page 173)

#### **Network requirements**

The instrument is factory-configured for IPv4 TCP/IP communication and includes a fast Ethernet adapter (10/100 Mbps) with a RJ45-type connector for integrating the device into a local area network (LAN).

By default, the instrument is configured to use the Dynamic Host Configuration Protocol (DHCP) but can use a static IP address.

The instrument should be configured behind a firewall. Contact Support for information on required firewall exceptions.



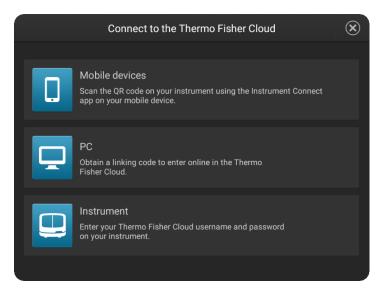
#### Link the instrument from a mobile device

#### Create a Cloud PIN and generate the QR code on the instrument

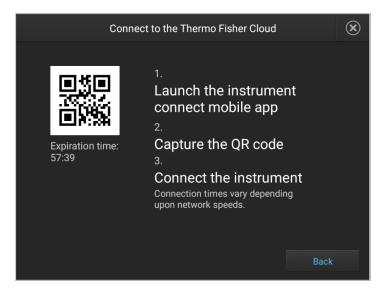
- 1. Sign in to **thermofisher.com/cloud**.
- **2.** Click **a** to access InstrumentConnect.
- **3.** If you have not previously set up a PIN, click **Update PIN**, then enter a PIN that you will use to sign in to the instrument.
- 4. From the instrument **Sign In** screen, navigate to the **Connect to the Thermo Fisher Cloud** screen:

Do you have a local instrument profile?	Description	
Yes	<ol> <li>Touch Sign in, then enter your PIN.</li> <li>In the home screen, touch Setup run.</li> <li>In the Setup run screen, touch Cloud.</li> </ol>	
No	Touch Get started > Connect.	

5. Touch Mobile devices.



The QR code is displayed.



Register the instrument with the Instrument Connect App

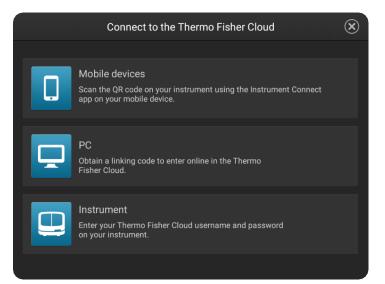
- On your mobile device, download the InstrumentConnect from the Apple Store or from Google<sup>™</sup> Play.
- **2.** Launch, then sign in to the mobile app on your mobile device.
- **3.** Register the instrument:
  - **a.** Touch  $\equiv$ , then touch **Register Instrument**.
  - **b.** Touch **QR code** on your mobile device.
  - **c.** With your mobile device, scan the QR code displayed in the instrument touchscreen.

#### Link the instrument using a link code

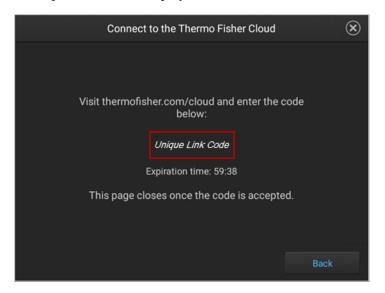
1. From the instrument **Sign In** screen, navigate to the **Connect to the Thermo Fisher Cloud** screen:

Do you have a local instrument profile?	Description	
Yes	<ol> <li>Touch Sign in, then enter your PIN.</li> <li>In the home screen, touch Setup run.</li> <li>In the Setup run screen, touch Cloud.</li> </ol>	
No	Touch Get started > Connect.	

2. Touch PC.

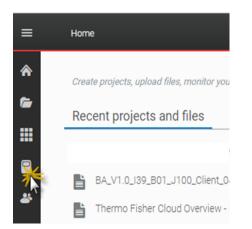


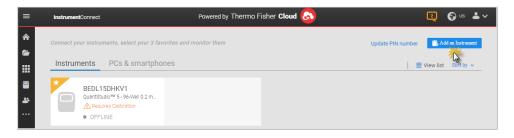
A unique link code is displayed.



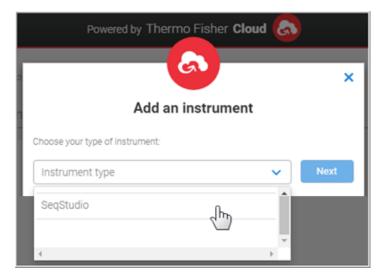
**3.** Sign into your Thermo Fisher Cloud on a separate computer. Go to **thermofisher.com/cloud**.

#### **4.** Click **a**, then click **Add an Instrument**.



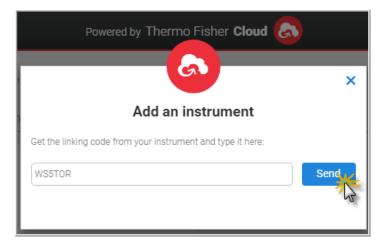


**5.** Select **SeqStudio**<sup>™</sup>, then click **Next**.



B

**6.** Enter the link code from the instrument touchscreen (from step 2), then click **Send**.



**7.** If you have not previously set up a PIN, enter a PIN to use when you sign in to an instrument, then click **Send**.

Powered by Thermo Fisher Cloud			
	6	×	
Set	your pin to proceed		
Insert new pin number			
Confirm new number		Send	
Use only numbers			

A start linking message is displayed.

A confirmation message is displayed on the instrument touchscreen when the instrument is linked and connected to the Thermo Fisher Cloud.

The first time the instrument is linked, the software automatically:

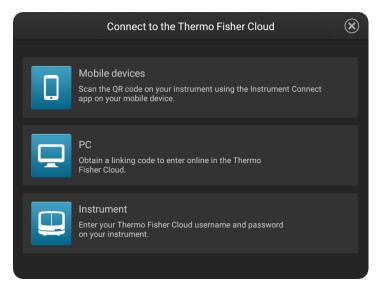
- Creates a Cloud instrument profile with the First Name and Last Name from your Thermo Fisher Cloud account.
- Registers the instrument in the InstrumentConnect software.

## Link the instrument using your Cloud account

1. From the instrument **Sign In** screen, navigate to the **Connect to the Thermo Fisher Cloud** screen:

Do you have a local instrument profile?	Description	
Yes	<ol> <li>Touch Sign in, then enter your PIN.</li> <li>In the home screen, touch Setup run.</li> <li>In the Setup run screen, touch Cloud.</li> </ol>	
No	Touch Get started ► Connect.	

#### 2. Touch Instrument.



**3.** Enter your Thermo Fisher Cloud account username (email address) and password, then touch Link account.

Connect to the Thermo Fisher Cloud		×
test@thermofisher.com		
Cancel	Link acco	unt

**4.** If you have not previously set up a PIN, enter a PIN to use when you sign in to an instrument, then touch **Done**.

Enter PIN	۲
PIN (4 digits required)	
Confirm PIN	
Show PIN	
Cancel	Done

A confirmation message is displayed when the instrument is linked and connected to the Thermo Fisher Cloud.

The first time the instrument is linked, the software automatically:

- Creates a Cloud instrument profile with the First Name and Last Name from your Thermo Fisher Cloud account.
- Registers the instrument in the InstrumentConnect software.

#### Set up email notifications from the instrument

When an instrument is linked to your Cloud account, email notifications are automatically sent to your Cloud account email address.

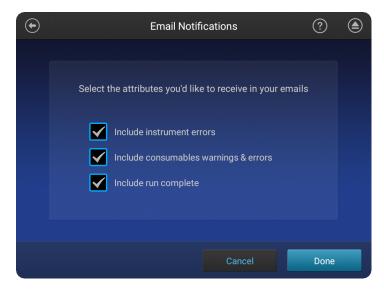
Perform this procedure to disable any of the default notifications.

- 1. Sign in to the instrument with your Cloud instrument profile and PIN.
- 2. In the home screen of the instrument, touch (a) Settings > Instrument settings > Email notifications.

$\odot$	Instrument Settings			
	About	Language		
	Instrument name	Date and tir	ne	
	Turn Demo mode on	Network config	uration	
	Email notifications			

**Note:** If you are signed in with a local instrument profile instead of a Cloud instrument profile, the **Email notifications** button is not displayed on the **Instrument Settings** screen.

**3.** In the **Email notifications** screen, select or deselect the options for which you want to receive email notifications, then touch **Done**.





# Dye calibration and install standard checks

	Calibrate dyes	176
н,	Perform an install run	187

#### **Calibrate dyes**

Overview of system dye set and custom dye set calibration Dye calibration compensates for dye emission spectral overlap.

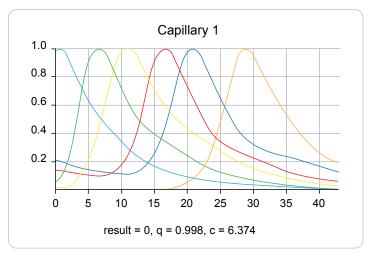


Figure 20 Dye emission spectra for the J6 (DS-36) dye set showing spectral overlap

System dye sets are available from Thermo Fisher Scientific.

# System versus custom dyes

System dye sets				
Fragment analysis dye sets	Sequence analysis dye sets			
<ul> <li>D (DS-30, DS-31)</li> <li>E5 (DS-02)</li> <li>F (DS-32)</li> <li>G5 (DS-33)</li> <li>J6 (DS-36)</li> </ul>	<ul> <li>E_BigDye<sup>™</sup> Terminator v1.1</li> <li>Z_BigDye<sup>™</sup> Terminator v3.1</li> <li>Z_BigDye<sup>™</sup> Direct</li> </ul>			

Custom dye sets are any dyes that are not available from Thermo Fisher Scientific. Custom dye sets require manual calibration.

Three types of calibration can occur on the instrument: Factory, auto, and manual Factory calibration – Default calibration provided with the instrument. It is not calibration optimized for a specific instrument. Manual calibration—Manual procedure performed by the user that provides a ٠ baseline calibration for the instrument on which it is run. Reduces pull-up (false secondary peaks under a true peak). Auto calibration – Automatic adjustment of the baseline calibration to optimally ٠ reduce pull-up (false secondary peaks under a true peak). The instrument performs an auto calibration for system and custom dyes. Determine if Sequence analysis dye sets – Do not typically require manual calibration. ٠ manual Fragment analysis dye sets-Manual calibration is recommended one time before calibration is use. required Note: With high-quality sample data, it is possible for auto calibration to pass using the factory calibration. To determine if a dye set requires manual calibration, review the calibration history for the dye set.

In the home screen:

# 1. Touch (a) Settings > Maintenance and Service > Calibration > Calibration history.

**2.** Examine the entry for the dye set of interest. If the dye set does not list a date and a cartridge serial number for a manual calibration or a date for auto calibration, the dye set requires manual calibration.

•		Calibration His	tory	?	)
Dye set	Chemistry standard	Cartridge serial #	Date of manual calibration	Date of auto calibration	
D (DS-30)™	Matrix		Factory		
E_BigDye <sup>™</sup> Term	Sequence	12345678901234	2017/09/12 16:		_
E5 (DS-02)™	Matrix		Factory	2017/09/12 16:	-
F (DS-32)™	Matrix		Factory		
G5 (DS-33)™	Matrix		Factory		
J6 (DS-36)™	Matrix		Factory	2017/09/13 13:18 Caps 1, 2	_
Z_BigDye™ Ter	Sequence	12345678901234	2017/09/05 11:	2017/09/05 11:	_
Filter	Sho	ow all		Done	

Figure 21 Fragment analysis dye sets example

- (1) Manual calibration is recommended—No auto calibration or manual calibration has been performed.
- (2) Manual calibration is not required—A manual calibration has been performed.
- (3) Manual calibration is not required—The factory calibration has been optimized for the instrument by auto calibration.
- (4) Manual calibration is recommended—Auto calibration has been performed, but only 2 capillaries passed.
- (5) Manual calibration is not required—A manual calibration and an auto calibration have been performed.

Perform a systemA system dye calibration requires ~30 minutes to complete.dye calibrationPrepare the dye set calibration standards and plate as described in the product<br/>information sheet for the dye set.

In the home screen:

1. Touch (\*) Settings > Maintenance and Service > Calibration > Dye Calibration.

2. Touch the injection group for the dye set in the plate, then touch Dye set.

- Dye Calibration
   ?
   ?

   Select your dye sets
- **3.** Touch **Sequence Standard** or **Matrix Standard**, then select a system dye
- calibration standard provided with the instrument.

	Select Dye Set	$\bigotimes$
	Sequence standard Matrix standard	
	F (DS-32)™ ■	
	G5 (DS-33)™ ■	
	J6 (DS-36)™ ■	
Custom dye		

4. Touch Calibrate.

The calibration run starts.

**IMPORTANT!** If the dye calibration fails:

- The results of the calibration are not saved, and the calibration plate is not moved to **Run History**.
- The instrument does not allow you to rerun the plate setup for a failed calibration. Close the calibration screen, then start a new calibration.



#### **Spectral Quality Value**

A spectral Quality Value reflects the confidence that the individual dye emission signals can be separated from the overall measured fluorescence signal. It is a measure of the consistency between the final matrix and the data from which it was computed. A Quality Value of 1.0 indicates high consistency, providing an ideal matrix with no detected pull-up/pull-down peaks.

In rare cases, a high Quality Value can be computed for a poor matrix. This can happen if the matrix standard contains artifacts, leading to the creation of one or more extra peaks. The extra peaks cause the true dye peak to be missed by the algorithm, and can lead to a higher Quality Value than would be computed with the correct peak. Therefore, it is important to visually inspect the spectral calibration profile for each capillary.

#### **Condition number**

A Condition Number indicates the amount of overlap between the dye peaks in the fluorescence emission spectra of the dyes in the dye set.

If there is no overlap in a dye set, the Condition Number is 1.0 (ideal conditions), the lowest possible value. The condition number increases with increasing peak overlap.

Dye Set	Quality Value Minimum	Condition Number Maximum
AnyDye	0.8 (default)	20.0 (default)
D	0.8	8.5
E	0.95	5.5
E5	0.95	6.0
F	0.95	8.5
G5	0.95	13.5
J6	0.95	8.0
Z	0.95	5.5

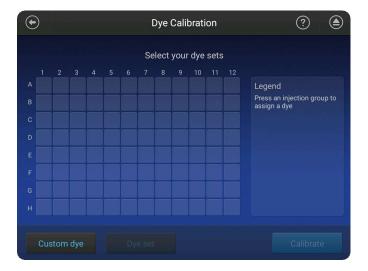
The ranges that the software uses to determine if a capillary passes or fails are:

# Perform a custom dye calibration

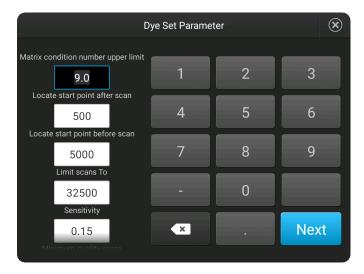
#### Add a custom dye set to the software based on a system dye

In the home screen:

1. Touch (\*) Settings > Maintenance and Service > Calibration > Dye calibration.



- 2. Touch Custom Dye.
- 3. Touch Add.
- 4. Touch the system dye to use as a starting point for the custom dye settings.
- 5. Modify the settings as needed then touch Next.



6. Enter a **Dye set name**, then touch **Done**.

Add a custom dye set to the software using the **Any Dye** template In the home screen:

- Dye Calibration
   ?
   >

   Select your dye sets
- 1. Touch (a) Settings > Maintenance and Service > Calibration > Dye calibration.

- 2. Touch Custom Dye.
- 3. Touch Add.
- **4.** Select **Any Dye** to use as a starting point for the custom dye settings. All dyes are selected by default.
- **5.** Touch a dye color, then manage the dye set arrangement:
  - Touch **Dye selection** to deselect the dye.
  - Touch **Move up** or **Move down** to organize the dyes in the order in which they occur as peaks in the electropherogram of the custom dye set standard.
  - Touch **Reduce selection** to exclude it from the calibration.

- Note: If you are calibrating with a system dye set, but will not run all system dyes in your application:
  - *Enable* (select) the **Dye selection** checkbox (to indicate that the system dye is present in the matrix standard)
  - *Disable* (deselect) the **Reduce selection** checkbox (to indicate that the system dye should not be analyzed, reported, or used for auto calibration)

(	۲ L				
$\odot$		New Dye Se	et Arrangement	?	
6					
3			Move Up		
2	2		Move Down		
1			Dye selection		
5			Reduce selection		
Cancel				Next	

(1) Order of dyes in the custom dye set

- 6. Touch Next, enter dye set parameters, then touch Next.
- 7. Enter a Dye set name, then touch Save.

#### Perform a custom dye calibration

Before you begin:

- Prepare the dye set calibration standards and plate as described in the product information sheet for the dye set.
- Add a custom dye set to the software (see "Add a custom dye set to the software based on a system dye" on page 180).

In the home screen:

- 1. Touch (\*) Settings > Maintenance and Service > Calibration > Dye calibration.
- 2. Touch the injection group for the dye set in the plate, then touch Dye set.

	Select Dye Set	$\bigotimes$
	Sequence standard Matrix standard	
	F (DS-32)™	
	G5 (DS-33)™ ■	
	J6 (DS-36)™ ■	
Custom dye		,

- **3.** Touch a custom dye set.
- 4. Touch Calibrate.
- 5. When the run is complete, touch View results.
- 6. Ensure that all capillaries passed the calibration, then click Done.

**IMPORTANT!** If the dye calibration fails:

- The results of the calibration are not saved, and the calibration plate is not moved to **Run History**.
- The instrument does not allow you to rerun the plate setup for a failed calibration. Close the calibration screen, then start a new calibration.

#### Add a custom dye to the Plate Manager or another instrument

- To add a custom dye set to the Plate Manager:
  - **a**. Open a plate setup that specifies the custom dye set of interest.
  - **b.** Export a plate setup that specifies the custom dye (see "Export or delete a plate setup (PSM file)" on page 121).
  - c. Open the exported plate setup in the Plate Manager.
- To transfer a custom dye set to another instrument:
  - a. Touch Settings > Maintenance and Service > Calibration > Calibration history.
  - b. Touch Custom dye.
  - c. Touch Manage.

- d. Touch a custom dye, then touch Export.
- e. Select a location, then touch Export.
- f. Import the custom dye on another instrument (❀ Settings > Maintenance and Service > Calibration > Dye calibration > Custom dye > Manage > Import).

In the home screen:

#### calibration history

View the dye

#### 1. Touch (\*) Settings > Maintenance and Service > Calibration history.

	Chemistry		Date of manual	Date of auto	
Dye set	standard	Cartridge serial #	calibration	calibration	
D (DS-30)™	Matrix		Factory		
E_BigDye™ Term	Sequence	12345678901234	2017/09/12 16:		_
E5 (DS-02)™	Matrix		Factory	2017/09/12 16:	
F (DS-32)™	Matrix		Factory		
G5 (DS-33)™	Matrix		Factory		
J6 (DS-36)™	Matrix		Factory	2017/09/13 13:18 Caps 1, 2	
Z_BigDye™ Ter	Sequence	12345678901234	2017/09/05 11:	2017/09/05 11:	

(1) Factory calibration (default calibration that is not optimized for the instrument).

- 2 Manual dye calibration performed by a user.
- (3) Factory calibration that has been optimized by auto calibration and does not require manual calibration.
- (4) Automatic adjustment of the baseline calibration to optimally reduce pull-up (false secondary peaks under a true peak).
- (5) A manual dye calibration and an auto dye calibration have been performed.
- 2. Touch a dye set.

A thumbnail of each capillary calibration spectrum is displayed, with the q, and c values (see "Spectral Quality Value" on page 180 and "Condition number" on page 180).

- **3.** Touch **>** and **<** to view a full-screen calibration spectrum for each capillary.
- **4.** Touch ( ) to return to the list of dye sets on the **Calibration History** screen.
- 5. (Optional) Touch Filter to narrow the dye set list down by dye sets.
  - **a**. Select or deselect the dyes listed.



- **b.** (*Optional*) Touch **Deselect All** to clear the dye set selections.
- c. Touch Done.
- 6. Touch OK.

# Import or export a custom dye set In the home screen: • Touch ❀ Settings > Maintenance and Service > Calibration > Dye calibration > Custom dye > Manage.

- **1.** Touch the custom dye set.
- 2. Touch Import or Export.



### Perform an install run

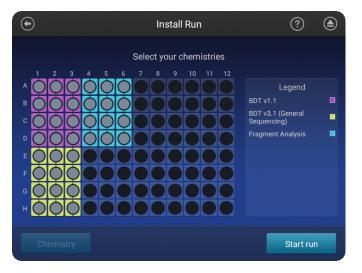
Overview of install checks

Install checks are performed by a Thermo Fisher Scientific Field Service Engineer at the time of installation.

An install check can be run at any time with the following reagents to ensure instrument performance:

Install check type	Reagent
Sequencing	<ul> <li>3500/3500xL Sequencing Standards, BigDye<sup>™</sup> Terminator v3.1 (Cat. No. 4404312)</li> </ul>
	<ul> <li>3500/3500xL Sequencing Standards, BigDye<sup>™</sup> Terminator v1.1 (Cat. No. 4404314)</li> </ul>
Fragment	DS-33 GeneScan <sup>™</sup> Installation Standards with GeneScan <sup>™</sup> 600 LIZ <sup>™</sup> Size Standard v2.0 (Cat. No. 4376911)

You can include multiple injection groups, multiple applications, and/or multiple chemistries on an install run plate. For example, you can prepare an install run plate that contains replictate injections of the fragment install standard and both sequencing standards.



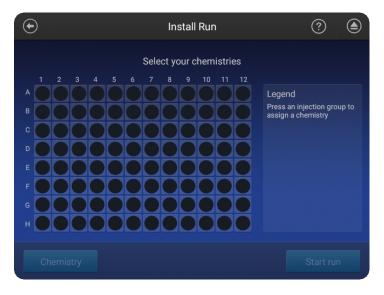


Perform an installAn install run requires ~45 minutes to complete each injection group.runPrepare the installation standard and plate as described in the product information<br/>sheet for the installation standard.

In the home screen:



- 1. Touch ( Settings > Maintenance and service > Install run > Install run.
- **2.** Touch an injection group on the plate to select a location for the install standards.



A plate can be set up with different injection groups for one install run.

**3.** Touch **Chemistry**, then select the type of install run. Select **None** to cancel the selection.

	Choose a Chemistry		$(\mathbf{x})$
[	None		
	BDT v1.1		
	BDT v3.1 (General Sequencing)		
	Fragment Analysis		

- **4.** Repeat the steps above for additional injection groups and/or multiple chemistries.
- 5. Touch Start Run.

#### View install run results

- **1.** Open the install run results summary:
  - To view results immediately after the install run completes, touch **Results**.

Туре	Run date	Status	Cartridge serial number
BDT v3.1 (General Se	2017/09/01 19:42:49	Passed	123456789012345678
BDT v1.1	2017/09/01 19:40:50	Passed	123456789012345678
Fragment Analysis	2017/09/01 19:39:04	Passed	123456789012345678
Fragment Analysis	2017/08/31 16:06:41	Failed	123456789012345678
BDT v3.1 (General Se	2017/08/30 16:40:24	Passed	123456789012345678
BDT v1.1	2017/08/30 16:38:30	Passed	123456789012345678
Fragment Analysis	2017/08/30 16:36:51	Passed	123456789012345678





2. Touch an install run to display the results, including the pass/fail results.

$\odot$				Install Run		?	۲
				Ilysis - 2017083( : 15; Expected # Size st			
	Cap #	# All	ele peaks	# Size standard peal	ks QC	Pass/Fail	
				34	0	Pass	
				34	0	Pass	
				34	0	Pass	
					0	Pass	
Exp	ort report		Detail				
$\odot$				Install Run		?	
•	BD.	T v3.		Install Run equencing) - 20' when QV30 CRL >= 500		Ŭ	
•		T v3.	Pass v	equencing) - 20´ when QV30 CRL >= 500		_164024	٢
•	BD <sup>-</sup> Cap # 1	T v3.		equencing) - 20´ when QV30 CRL >= 500		Ŭ	
•	Cap #	T v3.	Pass v QV30 CRL (ran	equencing) - 20´ when QV30 CRL >= 500	QC	_164024 Pass/Fail	
•	<b>Cap #</b> 1	T v3.	Pass v QV30 CRL (ran 533 (15-547)	equencing) - 20´ when QV30 CRL >= 500	QC	_164024 Pass/Fail Pass	
	Cap # 1 2	T v3.	Pass v QV30 CRL (rang 533 (15-547) 537 (15-551)	equencing) - 20´ when QV30 CRL >= 500	QC O	_164024 Pass/Fail Pass Pass	
	Cap # 1 2 3	Τ ν3.	Pass v QV30 CRL (rang 533 (15-547) 537 (15-551) 535 (17-551)	equencing) - 20´ when QV30 CRL >= 500		_164024 Pass/Fail Pass Pass Pass	
	Cap # 1 2 3 4	Τν3.	Pass v QV30 CRL (rang 533 (15-547) 537 (15-551) 535 (17-551) 542 (17-558)	equencing) - 20´ when QV30 CRL >= 500		_164024 Pass/Fail Pass Pass Pass	
	Cap # 1 2 3 4 Mean	Τν3.	Pass v QV30 CRL (rang 533 (15-547) 537 (15-551) 535 (17-551) 542 (17-558) 536.8	equencing) - 20´ when QV30 CRL >= 500		_164024 Pass/Fail Pass Pass Pass	

Each injection group displays a QC color for each capillary:

- – All QC tests passed.
- — At least 1 warning quality alert was triggered.
- At least 1 failing quality alert was triggered.

For information on quality alerts, see:

- "Data quality alerts" on page 102
- "Sizecalling and basecalling quality alerts" on page 102

**3.** Touch **Detail** to see allele-specific details for fragment analysis and basecall accuracy for sequencing.

€				Install Run		?	
		Fragment Analysis - 20170830_163651 Expected # Allele peaks: 15; Expected # Size standard peaks: 34					
		Dye	Allele	Mean size	Sizing precision	Pass/Fail	
			D5S644[VIC][1]	85.13	0.04	Pass	
			D5S644[VIC][2]	96.95	0.03	Pass	
			D20S119[FAM][	113.68	0.00	Pass	
			D20S119[FAM][	119.61	0.05	Pass	
			D9S288[NED][1]	137.52	0.04	Pass	
				145.27			
	xport i	report	Summary				

$\odot$			Install Run	?	۲
	BC		equencing) - 201709 when QV30 CRL >= 500	01_194249	
	Cap #	% QV30 CRL Basecall Accuracy	Alignment Read Length (range)	% RL Basecall Accuracy	
		100.0	553 (8-560)	99.1	
		100.0	562 (8-569)	99.1	
		99.6	555 (7-561)	99.1	
		98.7	553 (7-559)	99.1	
Exp	oort repoi	rt Summary			

4. Touch Export report (see "Export the install run report" on page 192).

#### Fragment analysis install run pass/fail criteria

Result	Acceptance criteria
# Allele peaks	15 for all four capillaries
# Size standard peaks	34 for all four capillaries
Standard deviation of the observed allele fragment sizes for all 4 capillaries	<0.15 base pairs (bp)

```
C
```



#### Sequence analysis install run pass/fail criteria

Result	Acceptance criteria
<b>QV30 CRL</b> (contiguous read length) <b>Note:</b> The contiguous read length passing criteria for install checks is an uninterrupted segment of bases with an average Quality Value (QV) of 30.	BigDye <sup>™</sup> Terminator v3.1 Capillaries with QV30 CRL ≥500 bp pass. BigDye <sup>™</sup> Terminator v1.1 Capillaries with a QV30 CRL ≥500 bp pass.

The remaining results on the screen are for information only.

Export the install run report

- 1. Touch (a) Settings > Maintenance and Service > Install run > Install run history.
  - **2.** Touch the install run of interest.
  - 3. Touch Export report.
  - **4.** Select a destination for the report.
  - 5. Touch OK.

Date: 2017/04/11

#### Example fragment analysis run report

applied biosystems

Install run report

|--|

#### System Information

Instrument Name:	seqstudio-SVT2	Run Date:	2017/04/10
Instrument Serial Number:	132000002	Chemistry Type:	Fragment Analysis
Instrument Software Version:	SVT-1.4.21-1306-r12832	Cartridge Serial Number:	2852889004
Buffer Installation Date:	2017/03/04	Cartridge Installation Date:	2017/04/10
Buffer Expiration Date:	2017/03/18	Cartridge Expiration Date:	2017/11/16

motan Recourt				
Capillary	1	2	3	4
# of Allele Peaks	15	15	15	15
# of Size Standard Peaks	34	34	34	34

Legend: Passed Failed

#### Running info

	Dye	Allele	Nominal Size	Mean	Average peak height	Peak height% > Min	Sizing precision	Sizing accuracy	Pass/Fail
1		D5S644[VIC][1]	85.06	85.1	1851.75	100.0	0.02	0.04	Pass
2		D5S644[VIC][2]	96.89	96.92	1184.0	100.0	0.03	0.03	Pass
3		D20S119[FAM][1]	113.7	113.67	1393.25	100.0	0.0	-0.03	Pass
4		D20S119[FAM][2]	119.6	119.56	1308.5	100.0	0.05	-0.04	Pass
5		D9S288[NED][1]	137.5	137.38	1545.0	100.0	0.02	-0.12	Pass
6		D9S288[NED][2]	145.24	145.09	1044.75	100.0	0.04	-0.15	Pass
7		D6S289[PET][1]	171.49	171.35	1677.0	100.0	0.05	-0.14	Pass
8		D6S289[PET][2]	173.41	173.25	1054.0	100.0	0.03	-0.16	Pass
9		D5S424[VIC][1]	216.39	216.26	1457.5	100.0	0.04	-0.13	Pass
10		D5S424[VIC][2]	218.27	218.14	955.75	100.0	0.03	-0.13	Pass
11		D9S1690[FAM][1]	236.74	236.48	2030.0	100.0	0.06	-0.26	Pass
12		D9S1690[FAM][2]	238.67	238.38	1300.5	100.0	0.04	-0.29	Pass
13		D18S462[NED][1]	303.06	302.86	1718.25	100.0	0.06	-0.2	Pass
14		D15S117[PET][1]	336.81	336.71	1331.0	100.0	0.02	-0.1	Pass
15		D15S117[PET][2]	338.73	338.63	914.25	100.0	0.01	-0.1	Pass

Capillary (Raman) Uniformity 0.4

The spectral dye matrix has not been updated.



#### Example sequencing run report

eport name: Sequencing instal	I Run_20170424_13	1753					Date: 2017
System Informat	ion						
Instrument Name:	seqstudio-SVT1		Run		2017/04/24		
Instrument Serial Number:	132000001			nistry Type:		neral Sequencing	)
Instrument Software Version: Buffer Installation Date:	0.6.10 2016/12/05			ridge Serial Numbe	er: 2883720037 ate: 2017/04/23	,	
Buffer Expiration Date:	2016/12/05				ate: 2017/04/23		
Install Run Result	C						
		1	2				
Capillary			_	3	4	Mean	SD
Contiguous Read Length	(CRL) (range)	530 (26-555)	535 (29-563)	3 531 (29-559)	4 530 (32-561)	Mean 531.5	SD 2.1
Contiguous Read Length CRL Pass/Fail			_	-			
Contiguous Read Length CRL Pass/Fail Comparison with Referen	nce Sequence	530 (26-555)	535 (29-563)	531 (29-559)	530 (32-561)		
Contiguous Read Length CRL Pass/Fail Comparison with Referen %CRL Basecall Accuracy	nce Sequence	530 (26-555) 99.8	535 (29-563) 99.6	531 (29-559) 99.8	530 (32-561) 100.0		
Contiguous Read Length CRL Pass/Fail Comparison with Referen %CRL Basecall Accuracy %RL Basecall Accuracy	nce Sequence	530 (26-555) 99.8 99.1	535 (29-563) 99.6 99.1	531 (29-559) 99.8 99.1	530 (32-561) 100.0 99.1		
Contiguous Read Length CRL Pass/Fail Comparison with Referen %CRL Basecall Accuracy %RL Basecall Accuracy Alignment Read Length (i	nce Sequence ( range)	530 (26-555) 99.8	535 (29-563) 99.6	531 (29-559) 99.8	530 (32-561) 100.0		
Contiguous Read Length CRL Pass/Fail Comparison with Referen %CRL Basecall Accuracy %RL Basecall Accuracy Alignment Read Length (r	nce Sequence ( range) d Failed	530 (26-555) 99.8 99.1 562 (5-566)	535 (29-563) 99.6 99.1	531 (29-559) 99.8 99.1	530 (32-561) 100.0 99.1		

gnal Strengt					
Capillary #	Sample file name	G	А	т	С
1	A2_A2_20170424_104040.ab1	792	1275	968	917
2	B2_B2_20170424_104041.ab1	1016	1625	1193	1108
3	C2_C2_20170424_104042.ab1	751	1336	1126	878
4	D2_D2_20170424_104043.ab1	635	1087	901	742

## View the install run history

In the home screen:

1. Touch (a) Settings > Maintenance and Service > Install run history.

Туре	Run date	Status	Cartridge serial number
BDT v3.1 (General Se	2017/09/01 19:42:49	Passed	123456789012345678
BDT v1.1	2017/09/01 19:40:50	Passed	123456789012345678
Fragment Analysis	2017/09/01 19:39:04	Passed	123456789012345678
Fragment Analysis	2017/08/31 16:06:41	Failed	123456789012345678
BDT v3.1 (General Se	2017/08/30 16:40:24	Passed	123456789012345678
BDT v1.1	2017/08/30 16:38:30	Passed	123456789012345678
Fragment Analysis	2017/08/30 16:36:51	Passed	123456789012345678

2. (*Optional*) Touch Filter, select the install run type, then touch Done.

	Install Run His		$\mathbf{X}$	
	Filter by cher	mistry		
	BDT v1.1			
	BDT v3.1 (General Sec	quencing)		
	Fragment Analy	ysis		
Deselect all		Cancel	Done	

**Note:** If the screen is blank when you click **Done**, no install runs are present for the chemistry you selected.





## Run modules, read lengths, size ranges, and run times

Run module	Contiguous read length (CRL) <sup>[1]</sup>	QV threshold	Approximate run time
ShortSeq	≥350	QV30	30 minutes
ShortSeq_BDX			
MediumSeq	≥500	QV30	45 minutes
MediumSeq_BDX			
LongSeq	≥800	QV20	~ 2 hours
LongSeq_BDX			

 Table 14
 Sequencing run modules for standard sequencing

[1] CRL was determined using the Long Read Sequencing standard. A minimum of 90% of analyzed sequences with an average QV >QV threshold were observed.

**IMPORTANT!** Use BDX run modules only if you prepare samples with BigDye XTerminator<sup>™</sup> Purification Kit. Use non-BDX run modules for samples purified with other methods.

Table 15	Fragment	analysis run	modules
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Run module	Resolution range	Approximate run time	Sizing precision	Compatible size standards
SNaPshot	40–120 bp	25 minutes	40–120: <0.5	GeneScan <sup>™</sup> 120 LIZ <sup>™</sup> Size Standard
FragAnalysis	60–460 bp <sup>[1]</sup>	45 minutes	60-460: <0.15	All except GeneScan <sup>™</sup> 1200 LIZ <sup>™</sup> Size Standard
LongFragAnalysis <sup>[2]</sup>	60-600 bp <sup>[1]</sup>	< 2 hours	60-460: <0.15 461-600: <0.3 601-800: >0.45	<ul> <li>GeneScan<sup>™</sup> 600 LIZ<sup>™</sup> Size Standard v2.0</li> <li>GeneScan<sup>™</sup> 1200 LIZ<sup>™</sup> Size Standard</li> </ul>

[1] Resolution Range: The range of bases over which the resolution (peak spacing interval divided by the peak width at half-max in a GS600 or GS1200 LIZ size standard sample sized with a third order fit) is >1. The table shows the resolution range in >90% of samples.

<sup>[2]</sup> Load a maximum of 48 samples per plate if you use a long run module.

Note: The following size standards have not been validated for use with the instrument. A default size standard definition is not provided in the software.

- GeneScan<sup>™</sup> 500 LIZ<sup>™</sup> Size Standard
  GeneScan<sup>™</sup> 350 ROX<sup>™</sup> Size Standard
  GeneScan<sup>™</sup> 400HD ROX<sup>™</sup> dye Size Standard



## Parts and materials

## Required materials not supplied

Unless otherwise indicated, all materials are available through **thermofisher.com**. MLS: Fisher Scientific (**fisherscientific.com**) or other major laboratory supplier.

Item	Source
Software	
<i>(Optional)</i> SeqStudio <sup>™</sup> Plate Manager and App	Available on the Thermo Fisher Cloud or for download at <b>thermofisher.com</b>
<i>(Optional)</i> SeqStudio <sup>™</sup> Remote Monitoring App	Available on the Thermo
(Optional) InstrumentConnect	Fisher Cloud
Equipment	
(Optional) Handheld Barcode Scanner	4488442
Consumables for SeqStudio <sup>™</sup> Genetic Analyzer	
SeqStudio <sup>™</sup> Genetic Analyzer Cartridge	A33671
SeqStudio <sup>™</sup> Genetic Analyzer Cartridge v2	A41331
SeqStudio <sup>™</sup> Genetic Analyzer Cathode Buffer Container	A33401
Reservoir Septa (for Cathode Buffer Container)	A35640
SeqStudio <sup>™</sup> Integrated Capillary Protector	A31923
Tubes, plates, and other consumables	
MicroAmp <sup>™</sup> Optical 96-Well Reaction Plate	4316813
MicroAmp <sup>™</sup> Optical 96-Well Reaction Plate with Barcode	4326659
MicroAmp <sup>™</sup> Optical 8-Tube Strip, 0.2 mL	4316567
Septa for SeqStudio <sup>™</sup> Genetic Analyzer, 96 well	A36541
Septa for SeqStudio <sup>™</sup> Genetic Analyzer, 8 strip	A36543
MicroAmp <sup>™</sup> 96-Well Tray/Retainer Set	403081
(Adapter for 8-Tube Strip)	405001
Reagents	
Hi-Di <sup>™</sup> Formamide	4401457

Item	Source			
BigDye XTerminator <sup>™</sup> Purification Kit	4376486			
Sequencing kits				
BigDye <sup>™</sup> Terminator v1.1 Cycle Sequencing Kit	4337449			
BigDye <sup>™</sup> Terminator v3.1 Cycle Sequencing Kit	4337454			
BigDye <sup>™</sup> Direct Cycle Sequencing Kit	4458689			
Fragment analysis dye calibration standards and installation standard				
GeneScan <sup>™</sup> 600 LIZ <sup>™</sup> Size Standard v2.0	4408399			
GeneScan <sup>™</sup> 120 LIZ <sup>™</sup> Size Standard	4324287			
GeneScan <sup>™</sup> 1200 LIZ <sup>™</sup> Size Standard	4379950			
GeneScan <sup>™</sup> 500 ROX <sup>™</sup> Size Standard	401734			
GeneScan <sup>™</sup> 500 LIZ <sup>™</sup> Size Standard <sup>[1]</sup>	4322682			
GeneScan <sup>™</sup> 350 ROX <sup>™</sup> Size Standard <sup>[1]</sup>	401735			
GeneScan <sup>™</sup> 400HD ROX <sup>™</sup> dye Size Standard <sup>[1]</sup>	402985			
DS-33 GeneScan <sup>™</sup> Installation Standards with GeneScan <sup>™</sup> 600 LIZ <sup>™</sup> Size Standard v2.0	4376911			

[1] This size standard has not been validated for use with the instrument. A default size standard definition is not provided in the software.



## Instrument specifications and layout

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### Instrument dimensions

Ensure that the installation site bench space can accommodate the dimensions and support the weight.

Configuration	Height	Length (depth)	Width	Weight
(Door Closed)	44.2 cm (17.4 in.)	64.8 cm (25.5 in.)	49.5 cm (19.5 in.)	53.5 kg (118 lbs)
(Door Open)	56.9 cm (22.4 in.)			



WARNING! PHYSICAL INJURY HAZARD. Do not attempt to lift or move the instrument without professional assistance. The crated instrument is heavy. Any incorrect lifting or moving of the crated instrument can cause serious injury.

### Instrument clearances

During instrument setup and maintenance, it is necessary to access the back and sides of the instrument. If the back of the instrument faces a wall, it will be necessary to have enough space to rotate the instrument on the bench for access.

**IMPORTANT!** For safety, the power outlet used for powering the instrument must be accessible at all times.

Component	Тор	Front	Left	Right	Back
SeqStudio <sup>™</sup> Genetic	30.5 cm	30.5 cm	10.0 cm	20.0 cm	10.0 cm
Analyzer	(12.0 in)	(12.0 in)	(4.0 in)	(8.0 in)	(4.0 in)

## **Environmental requirements**

Ensure that the installation room is maintained under correct environmental conditions.

Condition	Acceptable range			
Installation site	Indoor use only			
Electromagnetic interference	Do not use this device in close proximity to sources of strong electromagnetic radiation (for example, unshielded intentional RF sources). Strong electromagnetic radiation may interfere with the proper operation of the device.			
	This equipment has been designed and tested to CISPR 11 Class A. In a domestic environment it may cause radio interference. You may need to take measures to mitigate the interference.			
Altitude	Located between sea level and 2000 m (6500 ft.) above sea level			
Humidity (instrument and computer)	<ul> <li>Operation: 20%-80% (noncondensing)</li> <li>Transport and storage: 15%-80% (noncondensing)</li> </ul>			
Temperature (instrument and computer)• Operation: 15°C to 30°C (60°F to 85°F) Note: The room temperature must not fluctuate more than 2°C over a 2-hour per 				
Transient category	Installation categories II			
Overvoltage category	Installation categories II			
Vibration	Ensure that the instrument is not adjacent to strong vibration sources, such as a centrifuge, pump, or compressor. Excessive vibration will affect instrument performance.			
Pollution degree	II Install the instrument in an environment designated pollution degree II (only non-conductive pollution (e.g., dust) occurs except that occasionally a temporary conductivity caused by condensation is to be expected). Typical pollution degree II environments are laboratories and office spaces.			
Liquid waste collection	Dispose of the polymer, buffer, reagents and any liquid waste as hazardous waste in compliance with local and national regulations.			
Other conditions	Ensure the installation site is away from any vents that could expel particulate material on the system components.			
	Avoid placing the instrument and computer adjacent to heaters, cooling ducts, or in direct sunlight.			



## **Electrical requirements**

**CAUTION!** Do not unpack or plug in any components until the Field Service Engineers (FSEs) have configured the system for the proper operating voltage.



**WARNING!** For safety, the power outlet used for powering the instrument must be accessible at all times. See "Instrument clearances" on page 200 for information about the space needed between the wall and the instrument. In case of emergency, you must be able to immediately disconnect the main power supply to all the equipment. Allow adequate space between the wall and the equipment so that the power cords can be disconnected in case of emergency.

- Electric receptacle required: 2-prong with ground pin
- Maximum power dissipation: 380 W (approximately, not including computer and monitor)
- Mains AC line voltage tolerances must be up to ±10 percent of nominal voltage

Rated voltage	Circuit required	Rated frequency	Rated power
100-240 ±10% VAC <sup>[1]</sup>	10 A	50–60 Hz	400 W

<sup>[1]</sup> If the supplied power fluctuates beyond the rated voltage, a power line regulator may be required. High or low voltages can adversely affect the electronic components of the instrument.

## **Electrical protective devices**

We recommend several protective devices to protect the system in environments with large voltage and power fluctuations.

Device	Description		
Power line regulator	We recommend the use of a 1.5-kVA power line regulator in areas where the supplied power fluctuates in excess of $\pm$ 10% of the normal voltage. Power fluctuations can adversely affect the function of the instrument and computer.		
	<b>Note:</b> A power line regulator monitors the input current and adjusts the power supplied to the instrument or computer. It does not protect against a power surge or failure.		

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- 1				

Device	Description
Uninterruptible power supply (UPS)	We recommend the use of a 1.5-kVA uninterruptible power supply (UPS), especially in areas prone to power failure. Power failures and other events that abruptly terminate the function of the instrument and computer can corrupt data and possibly damage the system.
	WARNING! PHYSICAL INJURY HAZARD. Do not attempt to lift the UPS unit without assistance of at least two people. Improper lifting can cause painful and permanent back injury. Refer to the UPS manufacturer user guide for more information.
	<b>IMPORTANT!</b> UPSs provide power for a limited time. They are meant to delay the effects of a power outage, not to serve as replacement power sources. In the event of a power loss, power off the instrument and computer unless you expect to regain power within the battery life of the UPS.
Surge protector	We recommend the use of a 10-kVA surge protector (line conditioner) in areas with frequent electrical storms or near devices that are electrically noisy, such as refrigerators, air conditioners, or centrifuges. Short-duration, high-voltage power fluctuations can abruptly terminate the function of, and thereby damage the components of, the computer and the instrument.
	<b>Note:</b> A dedicated line and ground between the instrument, computer, and the building's main electrical service can also prevent problems caused by power fluctuations.

### **Network requirements**

The instrument is factory-configured for IPv4 TCP/IP communication and includes a fast Ethernet adapter (10/100 Mbps) with a RJ45-type connector for integrating the device into a local area network (LAN). If the instrument will be connected to a LAN, an active, tested network jack must be in place before the scheduled installation date. Also, a representative from your information technologies department must complete and return the *SeqStudio*<sup>TM</sup> *Genetic Analyzer IT Checklist* (Pub. No. MAN0016055) before installation, and be available during the installation to help connect the instrument to your network.

### Safety requirements

#### Safety practices

A safety representative from your facility must ensure that:

- Personnel establish and follow all applicable safety practices and policies to protect laboratory personnel from potential hazards.
- All applicable safety devices and equipment are available at all times.



## Required safety equipment

Your laboratory has specific safety practices and policies designed to protect laboratory personnel from potential hazards that are present. Follow all applicable safety-related procedures at all times.

The following safety equipment and protection from hazards must be available at the installation site:

- Protection from any sources of hazardous chemicals, radiation (for example, lasers, radioisotopes, radioactive wastes, and contaminated equipment), and potentially infectious biological material that may be present in the area where the service representative will work.
- Appropriate fire extinguisher:
  - You are responsible for providing an appropriate fire extinguisher for use on or near the equipment.
  - The types and sizes of fire extinguishers shall be suitable for use on electrical and chemical fires as specified in current codes, regulations, and/or standards, and with approval of the Fire Marshall or other authority having jurisdiction.
  - The installation of appropriate fire extinguishers shall be in addition to other fire-protection systems and not as a substitute or alternative to them.
- Eyewash
- Safety shower
- Eye and hand protection
- Adequate ventilation, including vent line/fume hood, if applicable
- Biohazard waste container, if applicable
- First-aid equipment
- Spill cleanup equipment
- Applicable Safety Data Sheets (SDSs)

## Safety





**WARNING!** GENERAL SAFETY. Using this product in a manner not specified in the user documentation may result in personal injury or damage to the instrument or device. Ensure that anyone using this product has received instructions in general safety practices for laboratories and the safety information provided in this document.

- Before using an instrument or device, read and understand the safety information provided in the user documentation provided by the manufacturer of the instrument or device.
- Before handling chemicals, read and understand all applicable Safety Data Sheets (SDSs) and use appropriate personal protective equipment (gloves, gowns, eye protection, and so on). To obtain SDSs, see the "Documentation and Support" section in this document.

#### Symbols on this instrument

Symbols may be found on the instrument to warn against potential hazards or convey important safety information. In this document, the hazard symbol is used along with one of the following user attention words:

- **CAUTION!**—Indicates a potentially hazardous situation that, if not avoided, may result in minor or moderate injury. It may also be used to alert against unsafe practices.
- **WARNING!**—Indicates a potentially hazardous situation that, if not avoided, could result in death or serious injury.
- **DANGER!**—Indicates an imminently hazardous situation that, if not avoided, will result in death or serious injury.

Symbol	English	Français
	Caution, risk of danger	Attention, risque de danger
	Consult the manual for further safety information.	Consulter le manuel pour d'autres renseignements de sécurité.
	Caution, risk of electrical shock	Attention, risque de choc électrique
	Caution, piercing hazard	Attention, danger de perforation

Symbol	English	Français
	Caution, hot surface	Attention, surface chaude
Ś	Potential biohazard	Danger biologique potentiel
	On	On (marche)
$\bigcirc$	Off	Off (arrêt)
Φ	On/Off	On/Off (marche/arrêt)
	Protective conductor terminal (main ground)	Borne de conducteur de protection (mise à la terre principale)
$\sim$	Terminal that can receive or supply alternating current or voltage	Borne pouvant recevoir ou envoyer une tension ou un courant de type alternatif
	Do not dispose of this product in unsorted municipal waste CAUTION! To minimize negative environmental impact from disposal of electronic waste, do not dispose of electronic waste in unsorted municipal waste. Follow local municipal waste ordinances for proper disposal provision and contact customer service for information about responsible disposal options.	Ne pas éliminer ce produit avec les déchets usuels non soumis au tri sélectif. MISE EN GARDE ! Pour minimiser les conséquences négatives sur l'environnement à la suite de l'élimination de déchets électroniques, ne pas éliminer ce déchet électronique avec les déchets usuels non soumis au tri sélectif. Se conformer aux ordonnances locales sur les déchets municipaux pour les dispositions d'élimination et communiquer avec le service à la clientèle pour des renseignements sur les options d'élimination responsable.

#### Conformity symbols

Conformity mark	Description
	Indicates conformity with safety requirements for Canada and U.S.A.

Conformity mark	Description
CE	Indicates conformity with European Union requirements.
	Indicates conformity with Australian standards for electromagnetic compatibility.

## Safety alerts on this instrument

Additional text may be used with one of the symbols described above when more specific information is needed to avoid exposure to a hazard. See the following table for safety alerts found on the instrument.

English		Français	
	<b>CAUTION! Hazardous chemicals.</b> Read the Safety Data Sheets (SDSs) before handling.		MISE EN GARDE ! Produits chimiques dangereux. Lire les fiches signalétiques (FS) avant de manipuler les produits.
	<b>CAUTION! Hazardous waste.</b> Refer to SDS(s) and local regulations for handling and disposal.		MISE EN GARDE ! Déchets dangereux. Lire les fiches signalétiques (FS) et la réglementation locale associées à la manipulation et à l'élimination des déchets.
	<b>DANGER!</b> Class 3B (III) visible and/or invisible laser radiation present when open and interlocks defeated. Avoid exposure to beam.		<b>DANGER !</b> Rayonnement laser ou DEL visible ou invisible de classe 3B (III) présent en position ouverte et avec les dispositifs de sécurité non enclenchés. Éviter toute exposition au faisceau.



## Safety information for instruments not manufactured by Thermo Fisher Scientific

Some of the accessories provided as part of the instrument system are not designed or built by Thermo Fisher Scientific. Consult the manufacturer's documentation for the information needed for the safe use of these products.

### **Instrument safety**

General

**CAUTION!** Do not remove instrument protective covers. If you remove the protective instrument panels or disable interlock devices, you may be exposed to serious hazards including, but not limited to, severe electrical shock, laser exposure, crushing, or chemical exposure.

Physical injury

**CAUTION!** Moving Parts. Moving parts can crush, pinch and cut. Keep hands clear of moving parts while operating the instrument. Disconnect power before servicing.

**Electrical safety** 

**WARNING!** Ensure appropriate electrical supply. For safe operation of the instrument:

- Plug the system into a properly grounded receptacle with adequate current capacity.
- Ensure the electrical supply is of suitable voltage.
- Never operate the instrument with the ground disconnected. Grounding continuity is required for safe operation of the instrument.



**WARNING!** Power Supply Line Cords. Use properly configured and approved line cords for the power supply in your facility.



**WARNING!** Disconnecting Power. To fully disconnect power either detach or unplug the power cord, positioning the instrument such that the power cord is accessible.

## Cleaning and decontamination



**CAUTION!** Cleaning and Decontamination. Use only the cleaning and decontamination methods specified in the manufacturer's user documentation. It is the responsibility of the operator (or other responsible person) to ensure the following requirements are met:

- No decontamination or cleaning agents are used that could cause a HAZARD as a result of a reaction with parts of the equipment or with material contained in the equipment.
- The instrument is properly decontaminated a) if hazardous material is spilled onto or into the equipment, and/or b) prior to having the instrument serviced at your facility or sending the instrument for repair, maintenance, trade-in, disposal, or termination of a loan (decontamination forms may be requested from customer service).
- Before using any cleaning or decontamination methods (except those recommended by the manufacturer), users should confirm with the manufacturer that the proposed method will not damage the equipment.

Instrument component and accessory disposal

Laser

To minimize negative environmental impact from disposal of electronic waste, do not dispose of electronic waste in unsorted municipal waste. Follow local municipal waste ordinances for proper disposal provision and contact customer service for information about responsible disposal options.

WARNING! LASER HAZARD. Under normal operating conditions, the SeqStudio<sup>™</sup> Genetic Analyzer is categorized as a Class 1 laser product. However, removing the protective covers and defeating the interlock(s) may result in exposure to the internal Class 3B laser. Lasers can burn the retina, causing permanent blind spots. To ensure safe laser operation:

- Never look directly into the laser beam.
- Do not remove safety labels, instrument protective panels, or defeat safety interlocks.
- The system must be installed and maintained by a Thermo Fisher Scientific Technical Representative.
- Remove jewelry and other items that can reflect a laser beam into your eyes or those of others
- Wear proper eye protection and post a laser warning sign at the entrance to the laboratory if the laser protection is defeated for servicing
- DO NOT operate the laser when it cannot be cooled by its cooling fan; an overheated laser can cause severe burns on contact.



## Safety and electromagnetic compatibility (EMC) standards

The instrument design and manufacture complies with the following standards and requirements for safety and electromagnetic compatibility.

#### Safety compliance

Reference	Description	
EU Directive 2014/35/EU	European Union "Low Voltage directive"	
EN 61010-1	<i>Safety requirements for electrical equipment for measurement, control, and laboratory use – Part 1: General requirements</i>	
UL 61010-1		
CSA C22.2 No. 61010-1		
EN 61010-2-010	<i>Safety requirements for electrical equipment for measurement, control and laboratory use – Part 2-010: Particular requirements for laboratory equipment for the heating of materials</i>	
EN 61010-2-081	Safety requirements for electrical equipment for measurement, control, and laboratory use – Part 2-010: Particular requirements for automatic and semi-automatic laboratory equipment for analysis and other purposes	
EN 60825-1	Safety of lasers products – Part 1: Equipment classification and requirements	

#### EMC

Reference	Description	
EU Directive 2014/30/EU	European Union "EMC Directive"	
EN 61326-1	Electrical equipment for measurement, control and laboratory use – EMC requirements – Part 1: General requirements	
AS/NZS CISPR 22	Limits and Methods of Measurement of Electromagnetic Disturbance Characteristics of Industrial, Scientific, and Medical (ISM) Radiofrequency Equipment	
ICES-003, Issue 5	Industrial, Scientific and Medical (ISM) Radio Frequency Generators	
FCC Part 15 Subpart B (47 CFR)	U.S. Standard Radio Frequency Devices	

Reference	Description
RFID	FCC Notice (for U.S. Customers):
	This device complies with Part 15 of the FCC Rules:
	Operation is subject to the following conditions:
	1. This device many not cause harmful interference, and
	2. This device must accept any interference received, Including interference that may cause undesired operation
	Changes and Modifications not expressly approved by Thermo Fisher Scientific can void your authority to operate this equipment under Federal Communications Commissions rules.
	Canada (English):
	This device complies with Industry Canada licence-exempt RSS standard(s). Operation is subject to the following two conditions:
	(1) this device may not cause interference, and (2) this device must accept any interference, including interference that may cause undesired operation of the device.
	Canada (Français):
	Le présent appareil est conforme aux CNR d'Industrie Canada applicables aux appareils radio exempts de licence. L'exploitation est autorisée aux deux conditions suivantes :
	(1) l'appareil ne doit pas produire de brouillage, et (2) l'utilisateur de l'appareil doit accepter tout brouillage adioélectrique subi, même si le brouillage est susceptible d'en compromettre le fonctionnement.

#### Environmental design

Reference	Description	
Directive 2012/19/EU	European Union "WEEE Directive"—Waste electrical and electronic equipment	
Directive 2011/65/EU	European Union "RoHS Directive"—Restriction of hazardous substances in electrical and electronic equipment	
SJ/T 11364-2014	"China RoHS" Standard—Marking for the Restricted Use of Hazardous Substances in Electronic and Electrical Products	

#### Radio compliance

Reference	Description
Directive 2014/53/EU (as of June 12, 2017)	European Union "RE Directive"—Radio equipment

G

## **Chemical safety**



**WARNING!** GENERAL CHEMICAL HANDLING. To minimize hazards, ensure laboratory personnel read and practice the general safety guidelines for chemical usage, storage, and waste provided below. Consult the relevant SDS for specific precautions and instructions:

- Read and understand the Safety Data Sheets (SDSs) provided by the chemical manufacturer before you store, handle, or work with any chemicals or hazardous materials. To obtain SDSs, see the "Documentation and Support" section in this document.
- Minimize contact with chemicals. Wear appropriate personal protective equipment when handling chemicals (for example, safety glasses, gloves, or protective clothing).
- Minimize the inhalation of chemicals. Do not leave chemical containers open. Use only with adequate ventilation (for example, fume hood).
- Check regularly for chemical leaks or spills. If a leak or spill occurs, follow the manufacturer's cleanup procedures as recommended in the SDS.
- Handle chemical wastes in a fume hood.
- Ensure use of primary and secondary waste containers. (A primary waste container holds the immediate waste. A secondary container contains spills or leaks from the primary container. Both containers must be compatible with the waste material and meet federal, state, and local requirements for container storage.)
- After emptying a waste container, seal it with the cap provided.
- Characterize (by analysis if necessary) the waste generated by the particular applications, reagents, and substrates used in your laboratory.
- Ensure that the waste is stored, transferred, transported, and disposed of according to all local, state/provincial, and/or national regulations.
- **IMPORTANT!** Radioactive or biohazardous materials may require special handling, and disposal limitations may apply.

### **Biological hazard safety**



**WARNING!** BIOHAZARD. Biological samples such as tissues, body fluids, infectious agents, and blood of humans and other animals have the potential to transmit infectious diseases. Conduct all work in properly equipped facilities with the appropriate safety equipment (for example, physical containment devices). Safety equipment can also include items for personal protection, such as gloves, coats, gowns, shoe covers, boots, respirators, face shields, safety glasses, or goggles. Individuals should be trained according to applicable regulatory and company/ institution requirements before working with potentially biohazardous materials. Follow all applicable local, state/provincial, and/or national regulations. The following references provide general guidelines when handling biological samples in laboratory environment.

• U.S. Department of Health and Human Services, *Biosafety in Microbiological and Biomedical Laboratories (BMBL)*, 5th Edition, HHS Publication No. (CDC) 21-1112, Revised December 2009; found at:

https://www.cdc.gov/labs/pdf/ CDC-BiosafetymicrobiologicalBiomedicalLaboratories-2009-P.pdf

• World Health Organization, *Laboratory Biosafety Manual*, 3rd Edition, WHO/CDS/CSR/LYO/2004.11; found at:

www.who.int/csr/resources/publications/biosafety/Biosafety7.pdf

## **Documentation and support**

### **Related documentation**

Document	Pub. No.
SeqStudio <sup>™</sup> Genetic Analyzer Instrument and Software User Guide	MAN0016138
SeqStudio <sup>™</sup> Genetic Analyzer with Data Collection Software v 1.1 Getting Started Guide	MAN0017464
DNA Fragment Analysis by Capillary Electrophoresis User Guide	4474504
DNA Sequencing by Capillary Electrophoresis Chemistry Guide Second Edition	4305080
Troubleshooting Sanger sequencing data	MAN0014435
SeqStudio <sup>™</sup> Genetic Analyzer Site Preparation Guide	MAN0016143

## **Customer and technical support**

Visit **thermofisher.com/support** for the latest service and support information.

- Worldwide contact telephone numbers
- Product support information
  - Product FAQs
  - Software, patches, and updates
  - Training for many applications and instruments
- Order and web support
- Product documentation
  - User guides, manuals, and protocols
  - Certificates of Analysis
  - Safety Data Sheets (SDSs; also known as MSDSs)
    - **Note:** For SDSs for reagents and chemicals from other manufacturers, contact the manufacturer.

### Limited product warranty

Life Technologies Corporation and/or its affiliate(s) warrant their products as set forth in the Life Technologies' General Terms and Conditions of Sale at **www.thermofisher.com/us/en/home/global/terms-and-conditions.html**. If you have any questions, please contact Life Technologies at **www.thermofisher.com/support**.

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