
QX600™ Droplet Reader and QX Manager Software Premium Edition

User Guide



BIO-RAD

QX600 Droplet Reader and

QX Manager Software Premium Edition

User Guide

Version 2.1



Bio-Rad Technical Support Department

The Bio-Rad Technical Support department is open Monday through Friday, 5:00 AM to 5:00 PM, Pacific time.

Phone: 1-800-424-6723, option 2

Email: Support@bio-rad.com (U.S./Canada Only)

For technical assistance outside the U.S. and Canada, contact your local technical support office.

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Safety and Regulatory Compliance

This section cites regulatory requirements for laboratory and electrical equipment, as well as requirements for working with chemicals and hazardous substances, and also explains safety precautions and recommendations.

Important: Only trained personnel should use this instrument.




Regulatory Compliance

The instrument has been tested and found to be in compliance with all applicable requirements of the following safety and electromagnetic standards:

- IEC 61010-1:2010 (3rd:2016, EN61010-1:2010 (3rd ed.). Electrical equipment for measurement, control, and laboratory use - Part 1: General requirements
- IEC 61326-1:2012 (Class A), EN 61326-1:2013 (Class A). Electrical equipment for measurement, control, and laboratory use. EMC requirements, Part 1: General requirements
- IEC 61010-2-081:2015, 3.0 edition, UL 61010-2-081:2015, CAN/CSA C22.2 No. 61010-2-081:19. Safety requirements for electrical equipment for measurement, control, and laboratory use. Part 2-081: Particular requirements for automatic and semi-automatic laboratory equipment for analysis and other purposes (includes Amendment 1)
- CAN/CSA 22.2 No 61010-1-04, Safety requirements for electrical, equipment for measurement, control, and laboratory use, Part 1: General requirements
- CAN/CSA C22.2 No. 61010-2-101:2019 IEC 61010-2-101: 2018 (3rd ed.)
- Restriction of hazardous substances (ROHS) directive (European Union)
- Registration, evaluation, authorization and restriction of chemicals (REACH). European Chemicals Agency (ECHA) June 1, 2007
- Waste electrical and electronic equipment (WEEE) directive

This equipment has been tested and found to comply with the limits for a Class A digital device pursuant to Part 15 of FCC rules. These limits are designed to provide reasonable protection against harmful interference when the equipment is operated in a commercial environment.




Table 1. Regulatory symbols

Symbol	Definition
	The CE symbol indicates that the manufacturer ensures the product conforms with the essential requirements of the applicable EN directives.
	<p>The CSA symbol indicates that a project has been tested to Canadian and U.S. standards, and it meets the requirements of those applicable standards.</p> <p>This equipment has been tested and found to comply with the limits for a Class A digital device pursuant to Part 15 of the FCC Rules. These limits are designed to provide reasonable protection against harmful interference when the equipment is operated in a commercial environment.</p>
	The Waste Electrical and Electronic Equipment (WEEE) Directive symbol indicates that when the end-user wishes to discard this product, it must be sent to separate collection facilities for recovery and recycling.

Safety Warning Labels

Warning labels alert you about sources of injury or harm. [Table 2](#) defines each safety warning label.

Table 2. Meaning of safety warning labels

Icon	Meaning
	Warning about risk of harm to body or equipment Operating the system before reading this manual can constitute a personal injury hazard. For safe use, do not operate this instrument in any manner unspecified in this manual. Only qualified laboratory personnel trained in the safe use of electrical equipment should operate this instrument. Always handle all components of the system with care and with clean, dry hands.
	Warning about handling biohazardous materials When handling biohazardous samples, adhere to the recommended precautions and guidelines and comply with any local guidelines specific to your laboratory and location.
	Warning about risk of electric shock In order to prevent electric shock, use caution when plugging and unplugging the instrument. Always turn off and unplug the instrument when performing maintenance procedures.

Safe Use Specifications

For safe operation of the instrument, Bio-Rad strongly recommends that you comply with instructions listed in this section and in [Appendix B, Instrument Maintenance](#).

This instrument is intended for laboratory use only. Bio-Rad is not responsible for any injury or damage caused by use of this instrument for purposes other than those for which it is intended, or by modifications to the instrument not performed by Bio-Rad or an authorized agent.

- This instrument is for use only by trained personnel.
- Use only the power cord, power switch, and USB port supplied with the instrument, and the plug adapter corresponding to the electrical outlets in your region.
- Position the instrument on a solid, stable surface, with adequate room at the back and on each side so that users can easily reach the power cord and USB port.
- This equipment generates, uses, and can radiate radio frequency energy and, if not installed and used in accordance with the provided instructional documentation, may cause harmful interference to radio communications. Operation of the systems in a residential area is likely to cause harmful interference, in which case users will be required to correct the interference at their own expense.

Note: Bio-Rad recommends maintaining a backup power source in case of power outages. A universal power supply (UPS) can protect from brown outs and power surges, while a regular backup generator does not.

For information on environmental requirements, see [Environmental Requirements on page 24](#).

General Precautions

- Always wear laboratory coat, laboratory gloves, and safety glasses with side shields or goggles.
- Keep your hands away from your mouth, nose, and eyes.
- Completely protect any cut or abrasion before working with potentially infectious materials.
- Wash your hands thoroughly with soap and water after working with any potentially infectious material before leaving the laboratory.
- Store all infectious or potentially infectious material in unbreakable leak-proof containers.
- Before leaving the laboratory, remove protective clothing.
- Change gloves frequently. Do not use a gloved hand to write, answer the telephone, turn on a light switch, or touch anything that other people may touch without gloves. Remove gloves immediately when they are visibly contaminated.
- Do not expose materials that cannot be properly decontaminated to potentially infectious material.
- Upon completion of an operation involving biohazardous material, decontaminate the work area with an appropriate disinfectant (for example, a 1:10 dilution of household bleach).

Personal Protective Equipment

Proper use of gloves is recommended with use of oils and sample plates. OSHA requirements for PPE are set forth in the Code of Federal Regulations (CFR) at 29 CFR 1910.132 (General requirements); 29 CFR 1910.138 (Hand protection); 29 CFR 1926.95 (Criteria for standard personal protective equipment). Any gloves with impaired protective ability should be discarded and replaced. Consider the toxicity of the chemicals and factors such as duration of exposure, storage, and temperature when deciding to reuse chemically exposed gloves.

Listed below are features to aid glove selection for handling of machines, assays, oils, and cleaning solvents:

- Butyl gloves are made of a synthetic rubber and protect against peroxide, hydrofluoric acid, strong bases, alcohols, aldehydes, and ketones
- Natural (latex) rubber gloves are comfortable to wear and feature outstanding tensile strength, elasticity, and temperature resistance
- Neoprene gloves are made of synthetic rubber and offer good pliability, finger dexterity, high density, and tear resistance; they protect against alcohols, organic acids, and alkalis
- Nitrile gloves are made of copolymer and provide protection from chlorinated solvents such as trichloroethylene and tetrachloroethene; they offer protection when working with oils, greases, acids, and caustic substances

Hazards

The instrument is designed to operate safely when used in the manner prescribed by the manufacturer. If the instrument or any of its associated components is used in a manner not specified by the manufacturer, the inherent protection provided by the instrument may be impaired.

Bio-Rad Laboratories, Inc. is not liable for any injury or damage caused by the use of this equipment in any unspecified manner, or by modifications to the instrument not performed by Bio-Rad or an authorized agent. Only trained Bio-Rad personnel should perform service on the system.

Biohazards

The instrument is a laboratory product. However, if biohazardous samples are present, adhere to the following guidelines and comply with any local guidelines specific to your laboratory and location.

Note: No biohazardous substances are expended during normal operations of this instrument.

Chemical Hazards

The instrument contains no potentially hazardous chemical materials.

Explosive or Flammability Hazards

The instrument poses no uncommon hazard related to flammability or explosion when used in a proper manner as specified by Bio-Rad.

Electrical Hazards

The instrument poses no uncommon electrical hazard to operators if installed and operated properly without physical modification and connected to a power source of proper specification.

Surface Decontamination



WARNING! To prevent electrical shock, always turn off and unplug the instrument before performing decontamination procedures.

Important: Do not use abrasive or corrosive detergents or strong alkaline solutions. These agents can scratch surfaces and damage the system.

The following areas can be cleaned with 10% bleach solution:

- Outer area and chassis
- Inner plate holders
- Droplet reading surfaces

To prepare and apply the disinfectant, refer to the instructions provided by the product manufacturer. For questions regarding the use of other cleaning agents, contact Bio-Rad Technical Support.

For more information on surface cleaning, see [Appendix B, Instrument Maintenance](#).

Important: Do not clean the handler Y-axis rail when the front door is open. This is a lubricated surface, and failures will occur if the lubrication is removed.

Decommissioning and Disposal

The instrument contains electrical materials that should be disposed of as unsorted waste and must be collected separately, according to European Union Directive 2012/19/EU on waste electrical and electronic equipment — WEEE Directive. The purpose of decommissioning is to make sure that the equipment is electrically and environmentally safe for disposal. Before disposal, contact your local Bio-Rad representative for country-specific instructions.

Transport

You must perform the specified decontamination procedures before moving or shipping the system. Always move or ship the instrument with the supplied packaging materials, which will protect the instrument from damage. If appropriate containers cannot be found, contact your local Bio-Rad office.

Warranty

Important: The instrument is for research use only, and not for use in diagnostic procedures.

The instrument and its associated accessories are covered by a standard Bio-Rad warranty. Contact your local Bio-Rad office for the details of the warranty. Follow the safety specifications listed in this chapter and throughout this guide.

The instrument is intended for laboratory use only. Bio-Rad Laboratories, Inc. is not responsible for any injury or damage caused by use of this instrument for purposes other than those for which it is intended, or by modifications of the instrument not performed by Bio-Rad Laboratories or an authorized agent. Alteration of this instrument voids the warranty and safety certification, as it creates a potential safety hazard.

Use of unapproved supermixes or additives may harm the instrument and voids the warranty.

Use only the power cord supplied with the instrument, using only the plug adaptor that corresponds to the electrical outlets in your region.

Chapter 1 Introduction to Droplet Digital PCR

Droplet Digital™ PCR (ddPCR™) is a digital polymerase chain reaction method based on water-oil emulsion droplet technology. ddPCR uses a combination of microfluidics and proprietary surfactant chemistries to divide each sample into water-in-oil droplets. The technology uses reagents and workflows similar to those used for most standard TaqMan probe-based assays, and provides absolute quantification of nucleic acid target sequences by counting nucleic acid molecules encapsulated in discrete, volumetrically defined water-in-oil droplet partitions.

ddPCR is highly effective in the following areas:

- **Absolute quantification** — ddPCR provides a concentration of target DNA copies per input sample without the need for running standard curves, making this technique ideal for target DNA measurements, viral load analysis, and microbial quantification.
- **Genomic alterations such as gene copy number variation (CNV)** — CNVs result in too few or too many dosage-sensitive genes responsible for phenotypic variability, complex behavioral traits, and disease. ddPCR enables measurement of 1.2x differences in gene copy number.
- **Detection of rare sequences** — researchers must amplify single genes in a complex sample, such as a few tumor cells in a wild-type background. ddPCR is sensitive enough to detect rare mutations or sequences.
- **Gene expression and microRNA analysis** — ddPCR provides stand-alone absolute quantification of expression levels, especially low-abundance microRNAs, with sensitivity and precision.
- **Next-generation sequencing (NGS)** — ddPCR quantifies NGS sample library preparations to increase sequencing accuracy and reduce run repeats. Validate sequencing results such as single nucleotide polymorphisms or copy number variations with absolute quantification.
- **Single cell analysis** — the high degree (10-fold to 100-fold) of cell-cell variation in gene expression and genomic content among homogeneous post-mitotic, progenitor, and stem cell populations drives a need for analysis from single cells. ddPCR enables low copy number quantification and gene expression of individual cells.
- **Genome edit detection** — ddPCR enables fast, precise, and cost-effective assessment of HDR (Homology directed repair) and NHEJ (non-homologous end joining) generated by CRISPR-Cas9 or other genome editing tools.

ddPCR has the following benefits for nucleic acid quantification:

- **Unparalleled precision** — The massive sample partitioning afforded by ddPCR enables small fold differences in target DNA sequence between samples to be reliably measured.
- **Increased signal-to-noise ratio** — High-copy templates and background are diluted, effectively enriching template concentration in target-positive partitions. This allows for the sensitive detection of rare targets and enables a $\pm 10\%$ precision in quantification.
- **Removal of PCR efficiency bias** — Error rates are reduced by removing the amplification efficiency reliance of qPCR, enabling accurate quantification of targets to near zero.
- **Simplified quantification** — There is no requirement for a standard curve for absolute quantification.


ddPCR Workflow

The ddPCR process adheres to the following workflow:

- You prepare your samples for PCR by combining DNA or RNA with primers, probes dye, and Bio-Rad ddPCR supermix.
- A droplet generator fractionates a sample into ~20,000 nanoliter-sized droplets with target and background DNA distributed randomly into the droplets during the partitioning process.
- Following droplet generation, the droplets are run through a thermal cycler, which performs PCR amplification of the nucleic acid target in each individual droplet.
- A droplet reader reads each droplet to determine the fraction of positive droplets in the original sample and uses Poisson statistical formulas to determine the absolute quantity.

Note: Positive droplets containing at least one copy of the target DNA molecule exhibit increased fluorescence compared to negative droplets.

Finding Out More

Click the Help tab () on the left, and then click the Bio-Rad Website to access links to technical notes, manuals, videos, product information, and technical support. The website also provides many technical resources on a wide variety of methods and applications related to PCR, Droplet Digital PCR, and gene expression.

Chapter 2 Using the QX600 Droplet Reader

Using the QX600™ Droplet Reader, you can detect rare DNA target copies, determine copy number variation, and precisely measure gene expression levels. Applications and uses include:

- Copy number variation
- Rare sequence detection
- Next generation sequencing (NGS) library quantification
- Viral load determination, including SARS-CoV-2 assays
- mRNA analysis
- Gene expression analysis and single cell gene expression analysis
- Absolute quantification
- Rare mutant detection
- NGS sample preparation
- GMO detection
- Contaminant testing
- High-order multiplexing

Following the droplet generation and thermal cycling processes, the QX600 Droplet Reader collects binary data for analysis as it singulates the ~20,000 nanoliter-sized droplets in each sample, and then streams them in single-file past a six-color detector to determine which droplets contain target molecules (positive reading) and which do not (negative reading).

The QX600 Droplet Reader detects multiple targets in six fluorescent dyes using three interrogation points across defined spectrums. The first interrogation point detects targets in FAM or EvaGreen® and HEX or VIC, the second in Cy5 and Cy5.5, and the third in ROX and ATTO 590.

Important: Use only the dyes listed above (FAM or EvaGreen®, HEX/VIC, Cy5, Cy5.5, ROX, and ATTO 590).

Important: Your QX600 Droplet Reader performs the post-PCR droplet reading function only. A droplet generator and a thermal cycler are required to perform a full ddPCR cycle. You can purchase a droplet generator and thermal cycler, as well as associated accessories and consumables, from Bio-Rad. For information, see [Appendix D, Ordering Information](#).

The following graphic and legend illustrate the functional areas on the instrument exterior:



LEGEND

- | | |
|---|---|
| 1 | Button to open and close lid |
| 2 | Status indicator lights (see Status Indicator Lights on page 29) |
| 3 | On the back of the instrument, power and USB outlets |
| 4 | Oil and waste compartment (see Fluid Maintenance and Waste Management on page 209) |

Instrument Specifications

This section contains information on the instrument size and weight of the instrument. Safety information is cited in [Safe Use Specifications on page 12](#) and the proper environmental maintenance is cited in [Environmental Requirements on page 24](#).

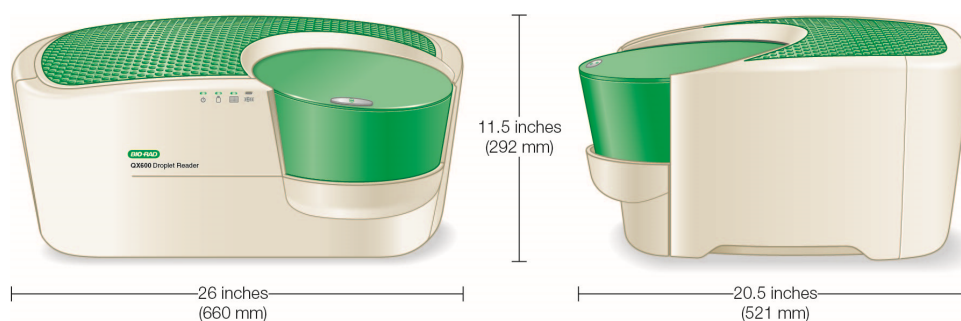


Table 3. Instrument specifications

Element	Specifications
Weight	56.6 lbs (26 kg)
Size (W x D x H)	26 x 20.5 x 11.5" (660 x 521 x 292 cm)

Environmental Requirements

Table 4 lists the environmental requirements for the QX600 Droplet Reader.

Important: You must use the supplied shielded cables with your instrument to ensure compliance with the Class A FCC limits.

Table 4. Conditions for safe use

Usage aspect	Requirements
Rated input power	100–240 V, 50–60 Hz, 2A (plugs into standard AC receptacle)
Fuses	T5AH, 250 V
Voltage fluctuations	± 10% for the included external power supply Note: Use only the power cord supplied with the equipment.
Pollution degree	2
Usage temperature	18–30° C
Relative humidity	85% max non-condensing
Altitude	0 to 8,200 ft (0 to 2,500 m) above sea level
Installation category	II (external power supply plugs into a standard AC receptacle) Indoor use only
Ventilation requirements	The following distances should be unobstructed for proper ventilation: <ul style="list-style-type: none"> ■ 5 in (12 cm) on the left and right sides of the instrument ■ 10 in (25 cm) behind the instrument

Included Items

This section identifies the functional items included with your droplet reader purchase, as well as additional components and consumables. For information on items you may need for a full ddPCR cycle, see [Appendix D, Ordering Information](#).

[Table 5](#) explains the components that ship with the QX600 Droplet Reader.

Table 5. QX600 Droplet Reader and Included Components

Component	Description	Catalog Number
QX600 Droplet Reader	Droplet reading instrument	12013328
USB cable and power cord	USB cable that connects the computer to the instrument and power cord that connects the instrument to the power source Note: To obtain cables or power cords, contact Bio-Rad technical support.	Included
Droplet reader plate holders (2)	Positions and secures the 96-well plate in the droplet reader plate compartment	12006834

Additional Components

The following tables identify additional components you can purchase for use with your system or instrument.

Table 6. Instrument components

Component	Description	Catalog Number
Computer	Computer that connects to the QX600 Droplet Reader for data collection and analysis	12017458
Software purchase	Bio-RadQX Manager Software, Premium Edition Note: Standard Edition is included with your droplet reader purchase.	12012172
ddPCR 96-Well Plates	Clear well/clear shell semi-skirted plates	12001925 (pkg of 25)

Component	Description	Catalog Number
Droplet Reader Oil	ddPCR droplet reader oil for QX600 and QX200 instruments and systems	1863004
Rainin Pipets	■ 20 µl pipet for sample loading	Rainin L-20
	■ 50 µl pipet for droplet transfer	Rainin L-50, L8-50
	■ 8-channel 200 µl pipet for oil	Rainin L8-200
Rainin Pipet Tips	Filtered	Rainin GP-L10F Rainin GP-L200F

Table 7. Thermal cycler and plate sealer

Product	Description	Catalog Number
C1000™ Touch Thermal Cycler with 96–Deepwell Reaction Module	Includes the C1000 Touch Thermal Cycler chassis, 96–deepwell reaction module, USB flash drive	1851197
PTC Tempo Deepwell Thermal Cycler	Includes the thermal cycler, power cord, USB cable, and Ethernet cable	12015392
PX1 PCR Plate Sealer	PCR plate sealer, includes heat sealing instrument, plate support block that holds 96-well and 384-well plates, sealing frame, power cord	1814000
Piercable Foil Heat Seal	Heat seal for the PX1 PCR Plate Sealer	1814040

Table 8. ddPCR supermixes

Supermix	Description	Catalog number	
ddPCR EvaGreen® Supermix	2x supermix, for use in nucleic acid sample preparation with the QX600 and QX200 ddPCR systems	1864033	2ml (2 x 1ml)
		1864034	5ml (5 x 1ml)
		1864035	25ml (25 x 1ml)
		1864036	50ml (50 x 1ml)
ddPCR Supermix for Probes (no dUTP)	2x supermix, for use in nucleic acid sample preparation with the QX600 and QX200 ddPCR systems	1863023	2ml (2 x 1ml)
		1863024	5ml (5 x 1ml)
		1863025	25ml (25 x 1)

Table 8. ddPCR supermixes, continued

Supermix	Description	Catalog number	
ddPCR Supermix for Probes	2x supermix, for use in sample preparation with the QX600 and QX200 ddPCR systems	1863026	2ml (2 x 1ml)
		1863010	5ml (5 x 1ml)
		1863027	25ml (25 x 1ml)
		1863028	50ml (50 x 1ml)
ddPCR Multiplex Supermix	4x supermix especially suited for probe-based detection of multiple targets in DNA samples using the QX600 and QX200 ddPCR systems	12005909	1.2ml (2 x 0.6ml)
		12005910	3 ml (5 x 0.6 ml)
		12005911	12.5 ml (5 x 2.5 ml),
ddPCR Supermix for Residual DNA Quantification	2x supermix, for use in residual DNA detection with the QX600 and QX200 ddPCR systems	1864037	2ml (2 x 1ml)
		1864038	5ml (5 x 1ml)
		1864039	25ml (25 x 1ml)
		1864040	50ml (50 x 1ml)
One-step RT-ddPCR Advanced Kit for Probes	200 or 500x 20 µl reaction kit, for absolute quantification of target RNA in a one-step format with the QX600 and QX200 ddPCR systems	1864021	2ml (2 x 1ml)
		1864022	5ml (5 x 1ml)

Table 9. Buffer controls

Buffer	Catalog Number
ddPCR Buffer Control Kit for Probes	1863052
QX200 Buffer Control Kit for EvaGreen®	1864052

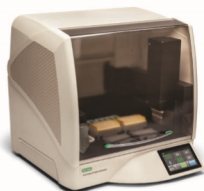
Compatible ddPCR Process Instruments

Along with the QX600 Droplet Reader, you can purchase the droplet generators shown below from Bio-Rad.

QX200 Droplet Generator



Automated Droplet Generator (left)



QX600 Droplet Reader



Important: A thermal cycler, which can be purchased separately, is required to perform a full ddPCR cycle. For information on additional instruments, accessories, and consumables, see [Appendix D, Ordering Information](#).

Connecting and Starting the Instrument

To execute runs on the QX600 Droplet Reader, you must connect the instrument to a computer on which the QX Manager Software is installed and configured. Bio-Rad recommends purchasing at least one configured computer that can remain connected to the QX600 Droplet Reader and be used for adding and running plates in QX Manager Software. For information on additional items you can purchase for your instrument, see [Additional Components on page 25](#).

To connect and start the instrument

1. Connect the USB cable provided by Bio-Rad to the USB port on the back of the QX600 Droplet Reader, and then to the computer.
2. Press the Power button on the instrument.

The Power indicator light turns solid green to indicate that power is on.

3. Turn on the connected computer and log in with a Windows account. For information, see your system administrator.
4. Open and log into QX Manager Software.

Important: The first time you log into the software, the Instrument window appears and you must select the instrument you are using. For information on selecting the correct instrument in QX Manager Software, see [Selecting the Instrument on page 36](#).









Status Indicator Lights

This section describes the indicator lights on the front of the QX600 Droplet Reader.



Before you start a run, check the indicator lights to ensure the instrument is ready. For information, see [Table 10](#).

Table 10. Droplet Reader Status Indicator Lights

Indicator	 (Power)	 (Fluids)	 (Plate)	 (Run Status)
Solid Green 	Power is on	Fluid levels OK	Plate loaded properly	Complete
Flashing Green 	N/A	<div><div>■ Less than 40% oil is available</div><div>■ More than 70% waste in container</div></div>	N/A	In progress
Flashing Amber 	N/A	<div><div>■ Less than 10% oil available</div><div>■ More than 90% waste in container</div></div>	N/A	Error during run
No color 	Power is off	N/A	No plate loaded	Instrument is idle

If the bottle indicator light is solid green or flashing green, there is enough oil in the bottle to complete a run on 96 wells. You can configure the full plate in QX Manager Software for your run. Once the low level sensor is triggered, the Add Plate button is disabled and you cannot continue to run plates (less than 10%). You must replace the oil and waste bottles with new ones before the instrument can start the run.

Chapter 3 About QX Manager Software

QX Manager Software, when connected to your QX600 Droplet Reader, provides the necessary functionality to create, run, and analyze droplet reading experiments on your samples.

QX Manager Software, Premium Edition, provides features to permit operation in compliance with Title 21 of the U.S. Code of Federal Regulations Part 11 (21 CFR Part 11) within a closed system, which is defined as “an environment in which system access is controlled by the persons who are responsible for the content of electronic records that are on the system” (Section 11.3(b) (4)).

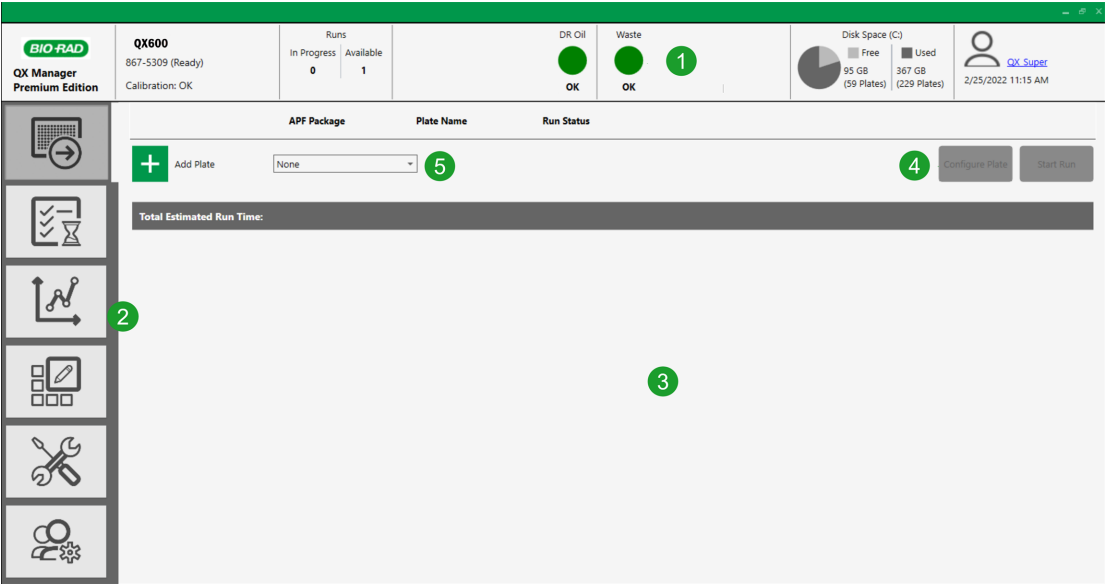
Note the following:

- The security controls built into QX Manager Software, Premium Edition, must be properly configured and administered by the software administrators in your organization in order to be secure and in compliance with 21 CFR Part 11.
- Bio-Rad makes no claim that QX Manager Software, Premium Edition, is CFR-compliant in and of itself, nor does the company guarantee compliance for the user. Your organization must establish policies and Standard Operating Procedures that work in conjunction with the tools provided by Bio-Rad to ensure compliance with 21 CFR Part 11.

Using the instrument and software, you can

- Set up customized, real-time ddPCR experiments for plate runs
- Read standard droplets
- Choose between two droplet volumes
- See the available disk space and number of plates you can process
- Use the live analysis function during the droplet reading process
- Analyze your data files in a variety of charts and tables in the Analysis module
- Reprocess data with updated color calibration values
- Create and store plate and report templates, and produce reports on your data
- Use Assay Protocol Files (APFs) to automatically configure your run with a range of predefined settings
- Manage lots and assays
- View audit logs and produce an audit log report on old and new data values after reprocessing a file

The following image highlights functional areas.



LEGEND

- 1 The status bar displays information about the instrument and user. For information on specific functionality, see [Instrument Status Bar on page 41](#).
- 2 Tabs provide access to the main functional windows.
- 3 The main pane displays the details of the selected tab.
- 4 The buttons allow you to configure the plate and start the run.
- 5 The dropdown list allows you to select an Assay Protocol File (APF).

Chapter 4 Getting Started

Use the information in this chapter to

- Sign into the software or change users
- Select the instrument
- Become familiar with the instrument status bar and main functional windows
- Understand the compatible file types for your software edition

Tip: If QX Manager Software goes into standby mode after being idle, click in the lockout screen to log into the software again.

Signing In, Viewing Privileges, and Changing Preferences

Typically, a system or software administrator sets up users and privileges for QX Manager Software, and communicates the information to you.

Note the following:

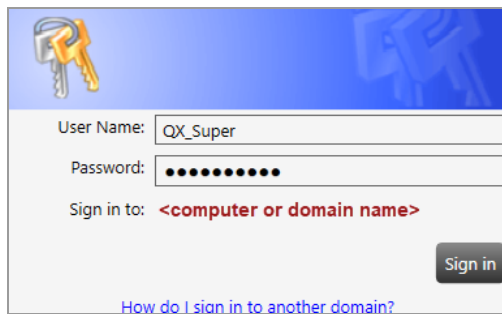
- When you sign in the first time after the software is installed, you must select the instrument to which the computer is connected. See [Selecting the Instrument on page 36](#) for information.
- You cannot change your user privileges unless you are assigned the Add/Manage Users privilege. For information, see [Viewing User Privileges on page 37](#).
- You can change your user preferences, which are set to defaults. For information, see [Managing Your Preferences on page 39](#).

Signing into the Software

Important: The Premium Edition includes audit capabilities. For auditable actions, you are prompted to sign in again before continuing.

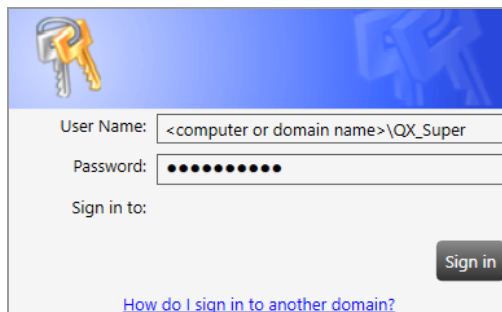
To sign in

1. Right-click the software icon on the desktop and select Open to display the Sign in dialog box.
2. Do one of the following to enter your user name:
 - If the Sign in to: label shows the correct domain name, enter only the user name and password.



The screenshot shows a 'Sign in' dialog box with a blue header and a light gray body. In the top left corner is an icon of two keys. The 'User Name:' field contains 'QX_Super'. The 'Password:' field is filled with ten black dots. The 'Sign in to:' label is followed by the text '<computer or domain name>' in red. A 'Sign in' button is located at the bottom right. At the bottom of the dialog is a blue hyperlink that reads 'How do I sign in to another domain?'.

- If the Sign in to: label is blank or shows a different domain, then enter it as **<computer or domain name>**, followed by the password.



The screenshot shows the same 'Sign in' dialog box. The 'User Name:' field now contains '<computer or domain name>\QX_Super'. The 'Password:' field remains filled with ten black dots. The 'Sign in to:' label is now blank. The 'Sign in' button and the bottom hyperlink are still present.

For more information, click the How do I sign into another domain? link.



A sign-in dialog box with a light gray background. In the top right corner is a dark gray button labeled "Sign in". Below it is a blue hyperlink that reads "How do I sign in to another domain?". Under the link, there are two lines of text: "Type domain name\domain user name to sign in to another domain." and "Type <computer name> \local user name to sign in to this PC only (not a domain).".

Note: If you are logging into a network domain, but don't know the name, contact your system administrator.

3. Ensure the Do not show EULA on sign in checkbox is selected and tap I Agree.



A horizontal bar containing a checkbox with a green checkmark, followed by the text "Do not show EULA on sign in". To the right of this bar are two dark gray buttons: "I Disagree" and "I Agree".

Each user must agree to the end user license agreement on initial login in order to use the software. If you clear the checkbox, the End User License Agreement appears the next time you log into the software, and you must agree again to the terms before the software opens. If you do not agree to the EULA, the application closes immediately.

When you agree to the EULA, the dialog box closes and the application opens. Your user name appears in the Instrument Status bar.

If you are connected to the QX600 Droplet Reader, the Add Plate window opens.

If you are opening the software on a computer not connected to an instrument, the Data Analysis window opens.

Selecting the Instrument

When you open the software and sign in for the first time, the instrument connection window appears and you must select the connected instrument and a droplet volume.

Note: This procedure assumes the instrument is connected to the computer and QX Manager Software is installed. You can use QX Manager Software with the QX200 Droplet Reader and the QX600 Droplet Reader. If you are using both instruments, you can change instruments at any time.

To select the instrument

1. Select QX600, and then click Save.

System Settings	Datafile and Template Storage	Instrument
	Instrument Connection	
Event Log	Instrument chosen here will be used to set parameters at the application level. When connecting to a new instrument, please unplug previous instrument, and connect and power on new instrument so the appropriate application settings can be applied.	
Maintenance Log	<input type="radio"/> QX200	
Maintenance Reports	<input checked="" type="radio"/> QX600	
System Data	<input type="button" value="Save"/>	
Reprocess Data		

Viewing User Privileges

In the User Management window, your privileges are identified by the selected checkboxes.

To view your assigned privileges

- Select the User Management tab.



Unless you are assigned the Add/Manage Users privilege, checkboxes for your assigned privileges are selected but not enabled, as shown in the following graphic. To request changes to your privileges, contact your software administrator.

User Name: (computer or domain name\QX_User1) Check Name

Full Name: QX_User1 (full name)

Privileges

☐ Add/Manage Users
☐ Create New Templates
☒ View Datafiles (created by other users)
☒ Overwrite Existing Datafile name
☐ System Settings
☐ Maintenance
☒ System Data
☒ Manage Plate

APF Privileges

☒ Manage APFs
☒ Run APFs
☐ Only Run APFs

[Table 11](#) lists the available privileges, which can be assigned in any combination.

Table 11. User privileges

Privilege	Purpose
Add and manage users	<p>Add or remove users, set privileges, and change preferences.</p> <p>Important: Users who are assigned the Add/Manage Users privilege can create users and update privileges and preferences for any user. Only the superuser can remove this privilege from user accounts.</p>
Create new templates	<p>Save a design for a plate or analysis report as a template.</p> <p>Create inventories of kits, consumables, and reagents.</p> <p>Import APF files.</p>
View data files (created by other users)	<p>See run files that were created from runs executed by other users.</p>
Overwrite existing data file name	<p>Save the file with a new file name. Files are always saved with the extension for the application you are using. For information, see Compatible File Types on page 43.</p> <p>Note: Users without this permission can open files and perform analysis but cannot create a new file.</p>
System settings	<p>View logs, reprocess data with new calibration values, and change file locations. All users can view the file locations.</p> <p>Important: If your administrator selected the Preferred Locations checkbox in System Settings, individual user location preferences are overridden.</p>
Maintenance	<p>View the event log and the system data settings.</p> <p>Note: All users can reprocess data, view system folder settings, as well as the maintenance log and maintenance reports.</p>
System Data	<p>Move raw data from the computer to free up disk space.</p> <p>Access troubleshooting data.</p>
Manage Plate	<p>Cancel a run while it is in process on the QX600 Droplet Reader.</p>
APF Privileges	<p>To use a preconfigured APF for runs, you can be assigned the following:</p> <ul style="list-style-type: none"> ■ Manage APFs, which provides all APF privileges when selected. ■ Run APFs, which gives you the option to execute a run configured with an APF. ■ Only Run APFs, which allows you to only execute runs configured with an APF.

Managing Your Preferences

Use the User Management window to modify your personal user preferences. Default folder locations appear in your preferences, but you can change them. You can also choose to keep all data files and templates private. The software prompts you to choose either your personal folder or the shared folder each time you save a file or template.

The screenshot shows the 'User Preferences' window. The sidebar on the left has three icons: a folder, a wrench and screwdriver, and a person with a gear. The main area is titled 'User Preferences' and contains the following sections:

- My Datafiles:** A text input field containing 'C:\Users\Public\Documents\Bio-Rad\QXMGrRegulatory\Users\QX_Super_USPLEC0TKYF2\Data' and a 'Browse' button. Below it is a checkbox labeled 'Do Not Share My Data'.
- My Templates:** A text input field containing 'C:\Users\Public\Documents\Bio-Rad\QXMGrRegulatory\Users\QX_Super_USPLEC0TKYF2\Data' and a 'Browse' button. Below it is a checkbox labeled 'Do Not Share My Templates'.
- System Time Out (in minutes):** A numeric input field containing '60'.
- Number of plates in the completed section on the status screen:** A numeric input field containing '7'.
- At the bottom is a checkbox labeled 'Acquire All Wells By Default'.
- At the bottom right are 'Add', 'Save', and 'Remove' buttons.

To change your preferences

1. Change any of the following preferences:
 - Enter a different file path for your data files and templates.
 - Select or clear the check boxes to change your data file and template privacy settings.
 - **Important:** The paths and checkboxes are disabled if your administrator has set Preferred Locations in System Settings.
 - Enter a different system timeout period.
 - Enter a different total of completed plates to show in the Run Status window, up to a maximum of 100.
 - Select the Acquire all Wells By Default checkbox. If you do not select the checkbox, all wells are excluded by default and you must manually include the wells for each droplet reading run.
2. Do one of the following:
 - If you are setting preferences for a new user, click Add.
 - If you are updating preferences, click Save.
3. When the confirmation message appears, click Yes, and then click OK.

Signing Out or Changing Users

While the instrument is idle, one user can log out and another user can log in.

Important: If User Management was not enabled during the software installation, the sign in, sign out, and change users functions are not available.

Note the following:

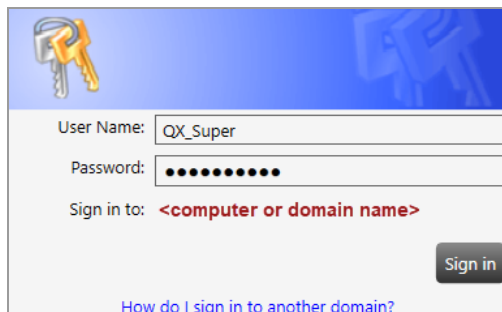
- Do not change users while a run is in progress or data might be affected.
- If the screen is locked and the initial user left unsaved changes, an advisory prompt appears. Click Save before proceeding or the previous user's work is lost.

To sign out

1. Click the user name link in the upper-right corner and select Sign Out.
2. Click Yes to confirm.

To change users

1. Click anywhere in the lockout screen to display the Sign in window.
2. Enter a user name.
 - The domain name appears below the user login fields. If you are on the same domain as the previous user, you can enter your user name only.



- If the user is on a different domain, enter the domain name followed by a backslash and the user name.

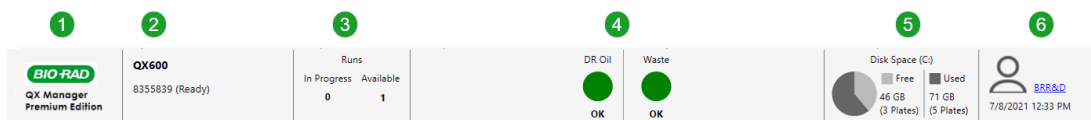
<domain name>\<user name> (for example, global\john_smith)

3. Enter the password and click Sign in.

Instrument Status Bar

An instrument status bar appears above all windows except the analysis displays, which open in a separate module.

When a computer is connected to the instrument, the status bar appears as shown in the following graphics. If a computer is not connected to the instrument, the status bar shows only the software, disk space, and user information.




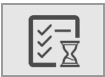





LEGEND

1	Software name and edition
2	Information and status of the connected instrument
3	Instrument availability
4	Oil and waste levels
5	Used and free disk space Available storage is displayed in GB, and also in number of plates that you can run before running out of space on the computer. Note: When QX Manager Software detects low disk space, an advisory prompt to archive data appears.
6	Name of the current user, along with the current date and time.

Functional Windows

Table 12 describes the functional areas and primary windows in QX Manager Software.

Table 12. Window Tabs

Tab	Name	Purpose
	Add Plate	<ul style="list-style-type: none"> ■ Add and configure a plate for droplet reading ■ Apply Assay Protocol Files (APFs) which contain preconfigured settings
	Run Status	<ul style="list-style-type: none"> ■ In the top pane, view the run in progress ■ In the bottom pane, view up to 100 completed runs
	Data Analysis	<ul style="list-style-type: none"> ■ Access the Data Analysis and Gene Study modules
	Templates	<ul style="list-style-type: none"> ■ Set up templates for plates and reports, and search for existing template files ■ Import and manage APFs, and manage lots of consumables and reagents
	System	<ul style="list-style-type: none"> ■ View storage locations, logs, and maintenance reports ■ Archive data, reprocess data with new calibration values, access troubleshooting information <p>Important: Available functionality depends on your assigned user privileges. Instrument calibration is available only to the Bio-Rad service engineer.</p>
	Users	<ul style="list-style-type: none"> ■ View the user privileges you have been assigned, and view and modify your personal preferences ■ Create, edit, or remove QX Manager Software users (if you are assigned the Add/Manage Users privilege)
	Help	<ul style="list-style-type: none"> ■ Access software version information, current calibration values, the End User License Agreement, the company website, and open source software license information <p>Tip: You can also link the user and instrument guides to the labeled buttons.</p>

Compatible File Types

This section describes the file types you can open in QX Manager Software.

The following applies if your organization is using more than one edition of the software:

- You can open files created using Premium Edition only in Premium Edition.
- If you open a .qlps file in QX Manager, Premium Edition, the file opens and is saved as a .ddpcrs file.

Table 13. Compatible file types

File Type	Extension	Details
Plate template	.ddplt	Plate template file containing setup details to perform experiments; this file type opens in the Plate Editor.
Plate template	.csv	Importable plate template file containing setup details in CSV format; this file type opens in the Plate Editor.
Plate	.ddpltapf	Plate template containing the configuration specified in the APF. This file type opens in the Plate Editor, but you can add wells only and they automatically assume the APF configuration.
Plate	.apfpack	APF file containing a static configuration, and which is imported into a targeted storage area for use when setting up runs
Data	.ddpcrs	Contains the results of an experiment run performed on the droplet reading instrument and opened in QX Manager Software, Premium Edition.
Data	.qlps	File type from earlier QuantaSoft™ regulatory versions, for two-channel experiment runs and data analysis; this file type opens in the Analysis module.

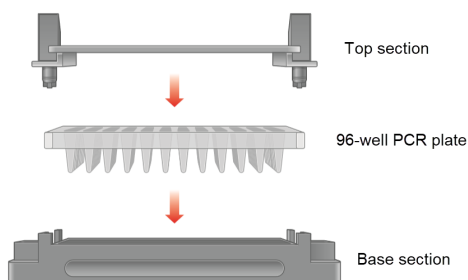
Chapter 5 Running Experiments

When the Start Run button is enabled, you can start the run. You can also cancel a run after it has started if you are assigned the Manage Plate user privilege. For information, see [Canceling a Run on page 65](#).

Assembling a Plate

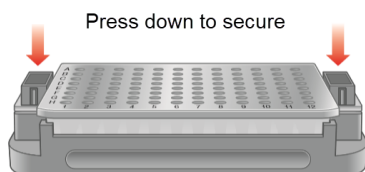
The QX600 Droplet Reader accepts a solid 96-well plate that fits into a secure holder. You must assemble the plate components before you can insert the plate into the QX600 Droplet Reader. Three separate components comprise the plate:

- A base section to hold the plate
- The plate itself
- A top section to secure the plate



To assemble the plate

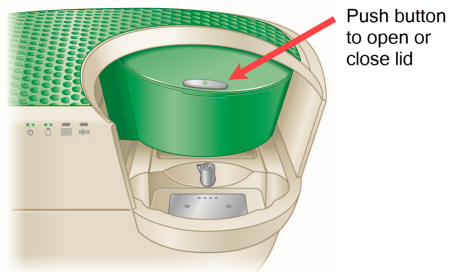
1. Place the plate into the base section, and then raise the release tabs on the top section.
2. Place the top section over the plate, and then lower the release tabs to secure the plate in the holder.



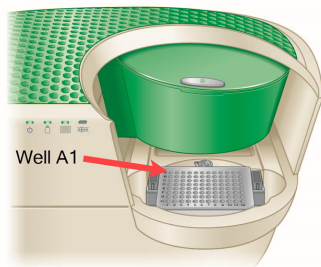
Inserting an Assembled Plate into the Instrument

To insert a plate into the QX600 Droplet Reader

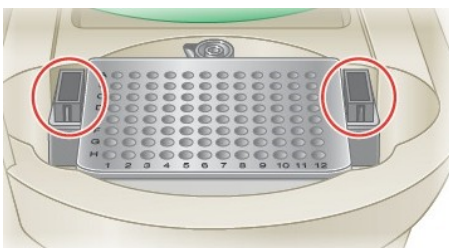
1. Press the button to open the compartment.



2. Fit the assembled plate into the compartment. Well A1 must be in the top-left position.



Important: Make sure the tabs on the plate holder are securely clamped and the plate lays flat. Otherwise, the sampling needle can strike the lid and damage the instrument.



3. Press the button again to close the lid.

Adding the Plate in QX Manager Software

This section explains how to add the plate in QX Manager Software, Premium Edition when using the QX600 Droplet Reader for your droplet reading run. You can insert an assembled plate into the instrument before or after you add the plate in the software.

In Premium Edition, you can use either of the following workflows:

- Standard, where you manually configure the plate.
- APF, where you select a file containing predefined settings to automatically configure the plate. For information on APFs, see [Managing Assay Protocol Files on page 158](#).


To add a plate in QX Manager Software

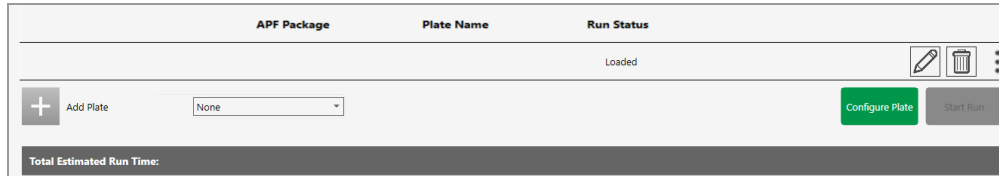
1. Click the Add Plate tab.




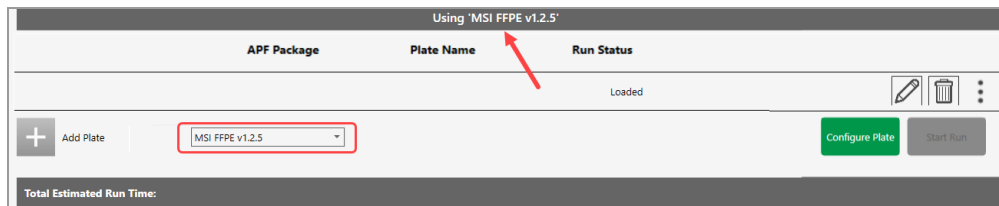
If the Runs Available number in the Instrument Status Bar is 1, the Add Plate button is enabled.

2. To add the plate in the software, do one of the following:

- To follow the standard workflow, ensure that None appears in the APF field, and then click .



- To use an Assay Protocol File (APF), select one from the dropdown list and then click the  button. The APF name appears in the title bar, as shown in the following graphic.



Important: APFs are available for use after they have been imported. For information, see [Importing Assay Protocol Files on page 160](#).

When QX Manager Software recognizes the plate holder in the compartment, the software enables the Configure Plate button.

Note: If the software does not recognize the plate holder, the software displays an advisory message. Remove and reinsert the plate. If necessary, contact Bio-Rad Technical Support.

3. Click Configure Plate. The Plate Configuration window opens.

Configuring the Plate

After you click Configure Plate, the Plate Configuration window opens to the Plate Information display. The window features the following tabs:

- Plate Information
- Well Selection
- Well Information
- Lot Selector (for information, see [Lot and APF Management on page 153](#))

All users can start runs on selected plates using the initial configuration, an altered configuration, or just the plate and data file names and the supermix. Users with the Create New Template user privilege can save the configuration as a template before the droplet reading run. Users with APF privileges can select and run APF configurations. For information on APFs, see [Managing Assay Protocol Files on page 158](#).

Plate Information Tab

The Plate Information tab is selected by default when the Plate Configuration window opens, and provides functionality allowing you to quickly set up a plate for a run.

You can

- Select, modify (optional), and run an existing template

Note: CSV template files appear in the dropdown list on the Plate Information tab, but not in the Templates window. For information, see [Creating and Storing Plate Templates from CSV Files on page 83](#).
- Create and run a plate design using a manual configuration or a template

Important: If you selected an APF in the Add Plate window, you can select only APF templates (.ddpltpf file extension) or .csv templates for your run.
- With the Create New Templates user privilege, save plate designs as templates for reuse
- For an expedited run, identify a plate name and a supermix, and then start the run

Note: The data file name field is automatically filled with the plate name, but you can to change it. If you selected an APF, the fields defined in the APF are grayed out throughout the plate setup. You can identify the remainder of your experiment parameters either in the Plate Editor before you start the run, or in the plate editor layout in the Analysis module after the run has concluded.
- Select to acquire wells by columns or rows

Identifying Plate Information

When you click Configure Plate, the following window appears.

To identify the plate

1. Do one of the following:

Use an existing template — click the dropdown arrow under Choose Existing and select a template file. The software automatically populates the fields under Create New Plate.

Create a new template — click Create New, and see [Creating or Editing Templates on page 67](#).

- Templates created with an APF have the .ddpltapf file extension, and can only be used when an APF is selected.

Create a new plate — enter a plate name and select a supermix.

- When you enter the plate name, the Save data file as field is automatically populated, but you can change it.
- If you selected an APF in the Add Plate window, the APF identifier appears in the banner and the Supermix field is grayed out. The supermix defined in the APF is automatically selected and cannot be modified.

2. (Optional) Under Acquire Wells By, select the alternative well acquisition method. The default is Column.

Well Selection Tab

The wells in the plate layout are automatically excluded and you must manually include the wells to be processed. Use this window to select wells to include wells for droplet reading.

Tip: If you typically run full plates, you can change the setting in User Preferences to acquire all wells by default. For information, see [Managing Your Preferences on page 39](#).

Note: The Well Selection window is identical to the Exclude window in Template Setup.

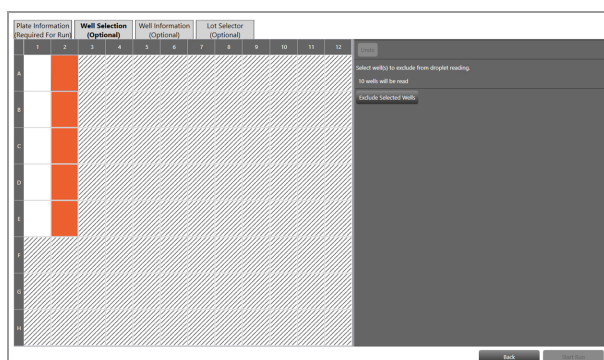
Including or Excluding Wells in the Plate Layout

All wells are excluded by default when the plate editor appears.

Important: If you selected an APF, you can add wells if the plate is not full, but they assume the settings from the APF. You cannot exclude the wells defined by an APF.

To include wells in the run

1. Select the Well Selection tab.
2. In the Plate Editor layout, select the wells containing sample and then click the Include Selected Wells toggle button.



3. On the right, QX Manager Software specifies the number of wells that will be included in the droplet reading run and the button text changes to Exclude Selected Wells.



Well Information Tab

When you select the Well Information tab, the Plate Editor opens. You can configure your experiment parameters for selected wells before or after the run.

Note: QX Manager Software can calculate automatic thresholds for positive control wells, but you must select Pos Ctrl from the Sample Type drop-down list. You can make the selection before or after the run. For information, see [Sample Types on page 75](#).

Important: If you selected an APF in the Add Plate window, fields defined in the APF are grayed out in the Well Information window. The APF can also enable fields that you must complete before running the plate. If you added wells in the Well Selection window, fields that are defined in the APF are automatically populated.

Using 'MSI FFPE v1.9.9'

Plate Information (Required For Run)		Well Selection (Required For Run)		Well Information (Optional)		Lot Selector (Optional)					
1	2	3	4	5	6	7	8	9	10	11	12
A	DOF BAT26 BAT25 WT_BA	DOF BAT26 BAT25 WT_BA	NR24 NR24 NR21 WT_NH	NR24 NR24 NR21 WT_NH	DOF Mono2 WT_M	DOF Mono2 WT_M					
B	DOF BAT26 BAT25 WT_BA	DOF BAT26 BAT25 WT_BA	NR24 NR24 NR21 WT_NH	NR24 NR24 NR21 WT_NH	DOF Mono2 WT_M	DOF Mono2 WT_M					
C											
D											
E											
F											
G											
H											

Clear Selected Wells

Well Type
Assay 1

Experiment Type
Setup Off (DOF)

Sample ID

Sample Description1 **Sample Description2**

Sample Description3 **Sample Description4**

Sample Type
NTC

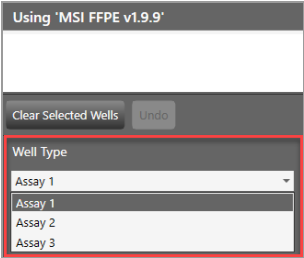
Assay Type
Double Drop Off

Target Info

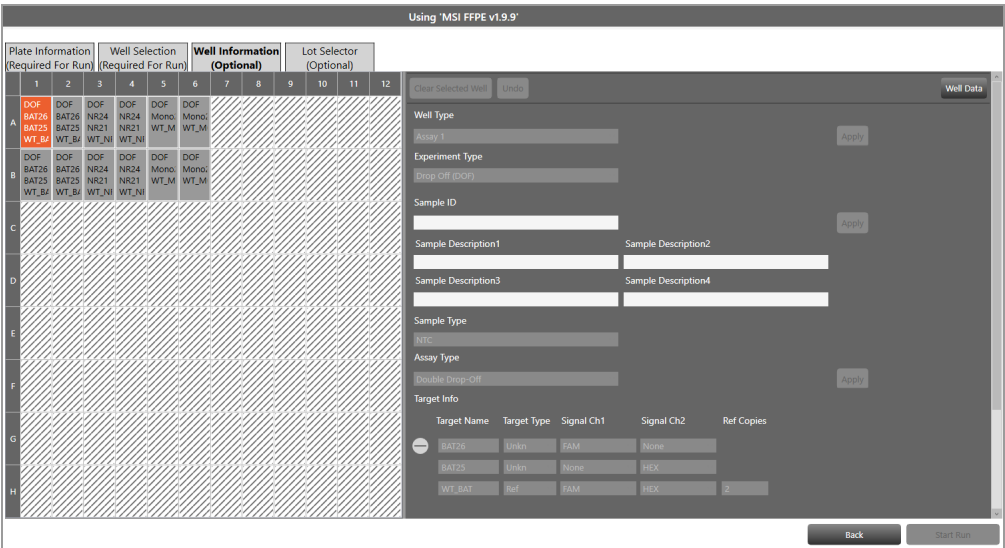
Target Name	Target Type	Signal Ch1	Signal Ch2	Ref Copies
BAT26	Linkin	BAM	None	
BAT25	Linkin	None	HEX	
WT_BAT	Ref	BAM	HEX	2

Defining Well Information When Using an APF

If you selected an APF to define your plate layout, wells are configured according to the well type selected. APF well types contain predefined settings, and can be applied to one or more wells in the plate layout. In the examples shown in this section, the experiment type, sample type, assay type, and target information fields are all predefined in three well types, with each well type containing a different set of targets.



The predefined fields are not editable, but you can enter values in undefined fields and in some cases, APF rules might require that you enter values in certain undefined fields. For example, a unique sample ID might be required for each well before the Start Run button is enabled.



If there is room on the plate, you can also include wells on the Well Selection tab and assign well types.

Using 'MSI FFPE v1.9.9'

Plate Information (Required For Run)		Well Selection (Required For Run)		Well Information (Optional)		Lot Selector (Optional)	
		1	2	3	4	5	6
A	DOF	DOF	DOF	DOF	DOF	DOF	DOF
B	BAT25	BAT25	BAT25	NR21	NR21	WT_M	WT_M
C	WT_B4	WT_B4	WT_B4	WT_N	WT_N	WT_N	WT_N
D	DOF	DOF	DOF	DOF	DOF	DOF	DOF
E	BAT25	BAT25	BAT25	NR21	NR21	WT_M	WT_M
F	WT_B4	WT_B4	WT_B4	WT_N	WT_N	WT_N	WT_N
G							
H							
I							
J							
K							
L							
M							

Clear Selected Wells

Well Type:

Experiment Type:

Sample ID:

Sample Description1: Sample Description2:

Sample Description3: Sample Description4:

Sample Type:

Assay Type:

Target Info

Target Name	Target Type	Signal Ch1	Signal Ch2	Ref Copies
BAT25	Unknown	RAW	None	
BAT25	Unknown	None	RAW	
WT_B4	Ref	RAW	RAW	2

Note: The graphics included herein are examples only; the array of predefined and undefined fields might be different in your APF.

To add wells and assign well types

1. Select the Well Selection tab and select one or more wells.
2. Click Include Selected Wells.
3. Select the Well Information tab.
4. Select one or more of the included wells.
5. From the Well Type drop-down list, select a well type and click Apply.
6. Repeat as applicable.

To enter values in undefined fields for existing or new wells

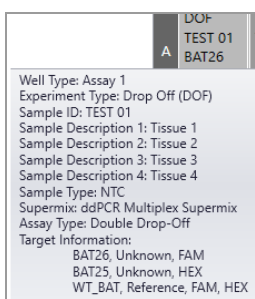
1. Do one of the following:
 - If the undefined fields must contain a different value for each well, select one well.
 - If the undefined fields must contain the same value, select multiple wells with the same well type.

Important: If you select a group of wells with different well types, a message appears in the applicable fields. If your entry contradicts a requirement or quality rule, you must select different wells.

2. Enter applicable values into the fields and click Apply.

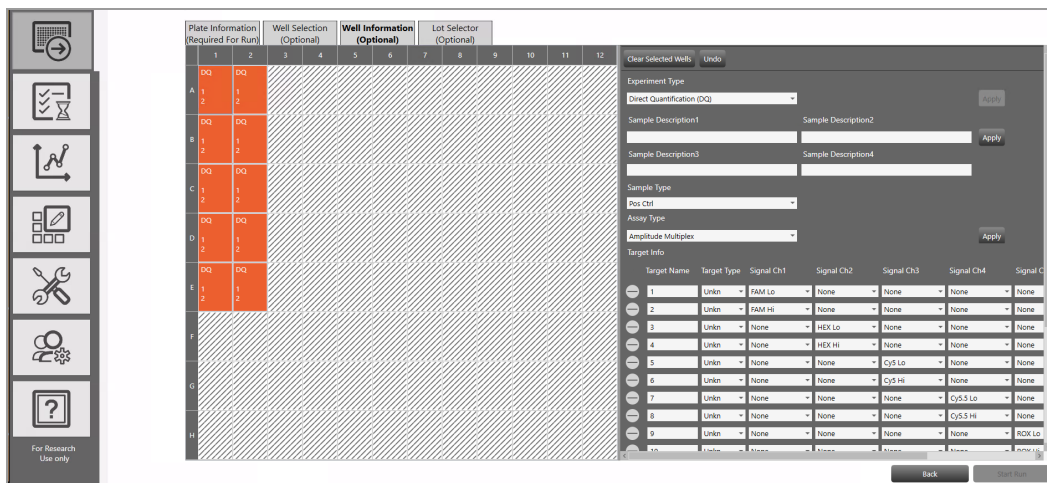
Important: You must click Apply for the software to recognize your new or edited information. If you enter information in a field and need to apply earlier changes first, an advisory message appears.

3. Repeat as applicable.
4. (Optional) To clear one or more wells at any time, select the wells and click Clear Selected Wells.
5. After you have applied your entries and selections, you can pause the cursor on a well to see the well information.



Defining or Editing Well Information

You can assign different experiment parameters to each well or groups of wells.



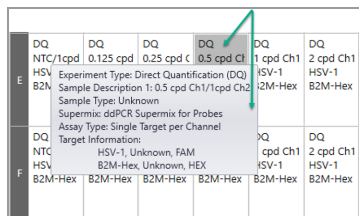
You can apply your entries at any time. You must click Apply for the software to recognize your new or edited information. If you enter information in a field and need to apply earlier changes first, QX Manager Software displays an advisory message.

To define the configuration for the wells

1. Select a well or group of wells.
2. Select an experiment type; see [Experiment Types on page 73](#).
3. In the Sample Description fields, enter up to four words or phrases that describe the sample; see [Sample Descriptions on page 75](#).
4. Select a sample type; see [Sample Types on page 75](#).

Note: For automatic thresholding of positive control wells, you must select Pos Ctrl.

5. Select a supermix; see [Supermixes on page 76](#).
6. Select an assay type. Available assay types vary, depending on the selected experiment type; see the fluorophore options in [Assay Types and Fluorophores on page 77](#).
7. (Optional) Under Target Info, change the fluorophore assigned to a channel. The fluorophore information populates automatically for each target, with the maximum number of rows allowed for the assay method. However, you can modify the default fluorophore assignments, if applicable.
 - a. Click the dropdown arrow and select an alternative.
 - b. To delete a row, click the minus (-) icon next to the target.
 - c. To add a row, click the plus (+) icon.
8. (Optional) To clear one or more wells at any time, select the wells and click Clear Selected Wells.
9. Click Apply. After you have applied your entries and selections, you can pause the cursor on a well to see the well information.



	DQ	DQ	DQ	DQ	DQ	DQ
	NTC/1cpd	0.125 cpd	0.25 cpd	0.5 cpd	1 cpd Ch1	2 cpd Ch1
E	HSV	Experiment Type: Direct Quantification (DQ)	1SV-1	2M-Hex	B2M-Hex	
	B2M	Sample Description 1: 0.5 cpd Ch1/1cpd Ch2				
		Sample Type: Unknown				
		Supermix: ddPCR Supermix for Probes				
		Assay Type: Single Target per Channel				
		Target Information:				
		HSV-1, Unknown, FAM				
		B2M-Hex, Unknown, HEX				
F	B2M-Hex	B2M-Hex	B2M-Hex	B2M-Hex	B2M-Hex	B2M-Hex

10. When you finish configuring the plate, click Save.
11. If you are assigning lots, select the Lot Selector tab and see [Lot and APF Management on page 153](#) for information.

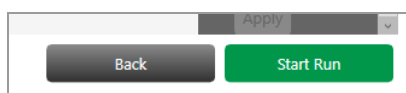
Starting the Run

When all required elements to set up a plate for a run are satisfied, QX Manager Software enables the Start Run button. You can configure the elements of the plate before you start the run, or you can run the plate with just the plate name and supermix, and define the configuration from the Analysis module.

If you selected an APF in the Add Plate window, part or all of the configuration has already been defined. You can add wells, and enter or select information in fields not already configured by the APF, but the APF configuration cannot be altered.

To start the droplet reading run

1. Tap Start Run.

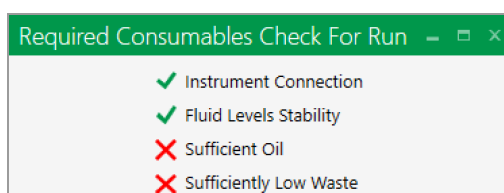


Important: The Premium Edition prompts you to log in again and reauthenticate your user credentials. Enter your user name and password, and then click Sign in.

The software performs a mandatory prerequisites check to ensure that the instrument is ready.

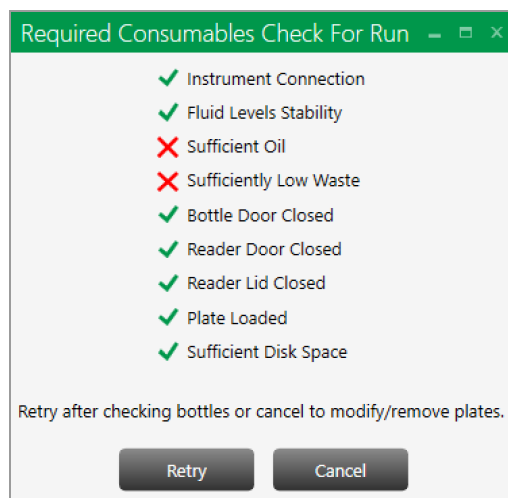
One of the following occurs:

- If the instrument passes the check, the run starts normally. Continue to [Running the Experiment on page 61](#).
- If the check finds issues with the instrument, the software displays a dialog box in which the issues are identified. The instrument cannot start the run until all issues are resolved. For information, see [Run Prerequisites Checklist on page 58](#).



Run Prerequisites Checklist

If the prerequisites check on the instrument status finds one or more problems with instrument readiness, QX Manager Software identifies them with an **✗** in the Check for Run dialog box. You must resolve all issues before the instrument can start the run.



Note: If you encounter a problem with the instrument or software that is not described here, see [Appendix C, Troubleshooting](#).

For information on each issue that can prevent the instrument from starting the run, see [Table 14 on page 59](#).

Table 14. Pre-Run Error Indicators

Status Check Item	Issue	Suggested Resolution
Instrument Connection	Power connection not recognized.	Ensure the power outlet is working. Disengage the power cord from the outlet, and then reinsert it into the outlet.
	USB cable connection not recognized.	Disconnect, and then reconnect, the USB cable between the instrument and the computer.
	Instrument connection not recognized.	Ensure the correct instrument is selected in the System Settings tab > Instrument tab.
Fluid Levels Stability	The oil or waste is moving within the bottle so the level cannot be accurately determined.	Wait until the liquid is settled, then try again. Ensure the instrument rests on a solid, stable surface.
Sufficient Oil	Oil level is below the acceptable level for a run.	Remove the oil bottle and replace it with a full one.
Sufficiently Low Waste	Waste level is above the acceptable level for a run.	<ol style="list-style-type: none"> 1. Empty any remaining oil from the bottle and then place a Waste label (provided by Bio-Rad) over the Oil label. 2. Insert the bottle into the Waste slot.
Bottle Door Closed	Bottle door is not closed properly.	Open the bottle door and ensure there are no blockages, and then close the door again.
Reader Door Closed	Reader door is not closed properly.	Open the reader door and ensure there are no blockages, and then close the door again.
Reader Lid Closed	Reader lid is not closed properly.	Open the reader lid and ensure there are no blockages, and then close the lid again.

Table 14. Pre-Run Error Indicators, continued

Status Check Item	Issue	Suggested Resolution
Plate Loaded	Plate is not loaded properly.	<ol style="list-style-type: none">1. Open the reader door and remove the plate.2. Ensure the tabs of the plate cover are securely clamped.3. Insert the plate again and ensure it lays flat.
Sufficient Disk Space	There is not enough storage space available for the run data.	Archive older data files to clear space.

Running the Experiment

The instrument allows you to do the following:

- During the run, perform real-time analysis on the droplets in each well after the well has been read
- Cancel a run (with the Manage Plate permission).
- When the run concludes, QX Manager Software moves the run to the Completed list in the Run Status window, and saves the analysis data file.

Data files are always saved to your personal file location or a preferred location, and can also be saved to a shared location.

Important: if your system administrator has designated preferred locations in System Settings, they override any file paths indicated in your preferences.

- During and after the run, QX Manager Software displays run information in the Run Status window.

During and after the run, the software displays run information in the Run Status window.

- During the run, you can perform real-time analysis on the droplets in each well after the well has been read.
- You can cancel the run at any time.

Note: Canceling a run also shuts down the instrument.

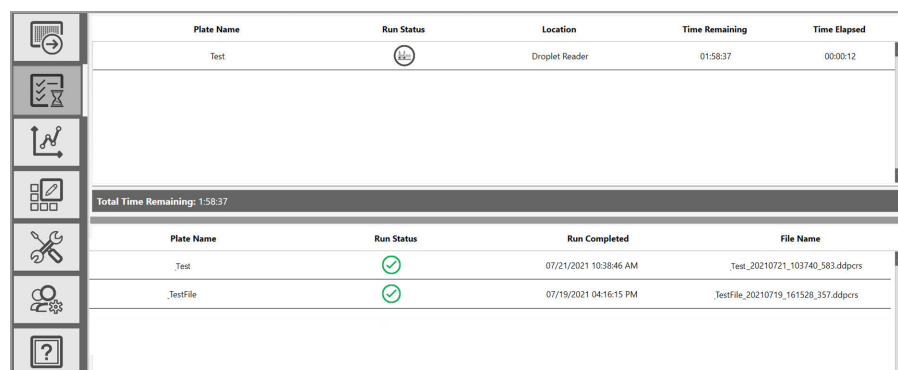





Plate Name	Run Status	Location	Time Remaining	Time Elapsed
Test		Droplet Reader	01:58:37	00:00:12

Total Time Remaining: 1:58:37

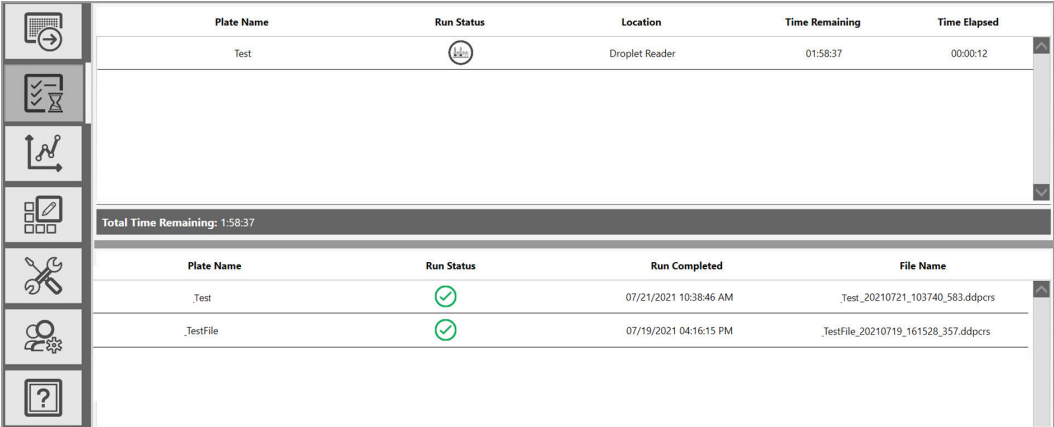
Plate Name	Run Status	Run Completed	File Name
.Test		07/21/2021 10:38:46 AM	Test_20210721_103740_583.ddpcrs
.TestFile		07/19/2021 04:16:15 PM	TestFile_20210719_161528_357.ddpcrs

- When the run concludes, the software moves the run to the Completed list in the Run Status window, and saves the analysis data file either to your personal file location or to the shared location.


Note: The default location is the shared folder shown in the System Settings window, but you can opt to keep your files private in your personal preferences. For information, see [Managing Your Preferences on page 39](#).

Tracking the Run Status



You can track each run from the Run Status window.



The screenshot shows the Run Status window with a sidebar on the left containing icons for various functions. The main area displays two tables. The top table shows a run in progress for 'Test' at 'Droplet Reader' with 01:58:37 remaining and 00:00:12 elapsed. The bottom table shows two completed runs: 'Test' completed on 07/21/2021 at 10:38:46 AM, and 'TestFile' completed on 07/19/2021 at 04:16:15 PM. A 'Total Time Remaining: 1:58:37' bar is also visible.

Plate Name	Run Status	Location	Time Remaining	Time Elapsed
Test		Droplet Reader	01:58:37	00:00:12






Total Time Remaining: 1:58:37

Plate Name	Run Status	Run Completed	File Name
Test		07/21/2021 10:38:46 AM	Test_20210721_103740_583.ddpcrs
TestFile		07/19/2021 04:16:15 PM	TestFile_20210719_161528_357.ddpcrs

The top pane shows the run in progress. The run remains in the top pane for the duration of the processing.

Icons displayed in the Run Status column indicate each process point, as shown in [Table 15](#).

Table 15. Run status indicators

Status Icon	Description
	Ready to begin
	Droplet reading run
	Run completed
	Run completed, plate removed
	Run stopped or canceled by the system, or canceled by the user before it began

When the run is finished, the software moves the run to the bottom pane, where up to 100 completed runs are visible.

Tip: You set the number of completed runs to display in your user preferences. See [Managing Your Preferences on page 39](#).

The content of each column is explained in [Table 16](#).

Table 16. Run Information Columns

Column Name	Location	Description
Plate name	Top and bottom panes	Name entered under Create New Plate Note: If you selected an existing template, the template name appears.
Run status	Top and bottom panes	Icons for each process step (see Table 15 on page 62)
Time remaining	Top pane only	Hours and minutes until the run will be finished
Time elapsed	Top pane only	Time, in hours and minutes the run has been in progress
Total time remaining	Below the top pane	Total hours and minutes remaining for all in-progress runs
Run completed	Bottom pane only	Date and time stamp showing when the instrument completed the run
File name	Bottom pane only	Plate name and system-generated identifier


After run completion, the software saves the analysis data file in a location designated by you or a system administrator.

Using Live Analysis

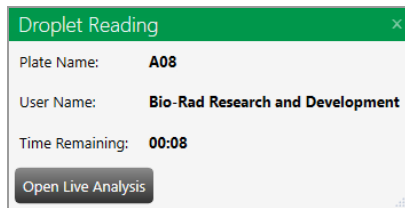
When the QX600 Droplet Reader initiates droplet reading, QX Manager Software provides an option for real-time analysis of the wells that have been read, while the process as a whole is still active.

QX Manager Software initially shows each well in the Plate Editor as disabled. As the software confirms that the instrument has acquired a well and read the droplets, the software enables the well for live analysis in read-only mode. The process continues until all wells are enabled and the process is finished.

To use live analysis

1. Tap the droplet reading icon () for the run in progress.

The Droplet Reading dialog box opens.



2. Click Open Live Analysis.
3. Select an analysis window.
4. In the well selector, select one or more wells that are enabled.

Important: QX Manager Software displays read-only data in the selected analysis window. You can view the data but to make changes, you must wait for the run to complete, and then change the data in the Analysis module.

Canceling a Run

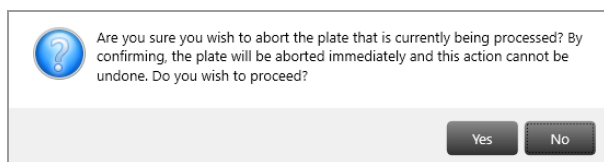
You can cancel a run if you are assigned the Manage Plate user privilege.

1. In the Run Status window, click the X on the right to cancel the run.

Location	Time Remaining	Time Elapsed	
Droplet Reader	00:03	00:00	

You can also click the icon under Run Status before the run begins. The run cannot be canceled after droplet reading has commenced.

2. When the Are you sure message appears, click Yes.



The instrument finishes reading the current well, stops the process and reverts to its pre-run state.

Note: If you click No, or if you do not respond to the confirmation message within 30 sec, the process continues without interruption.

The icon appears in the Run Status column.

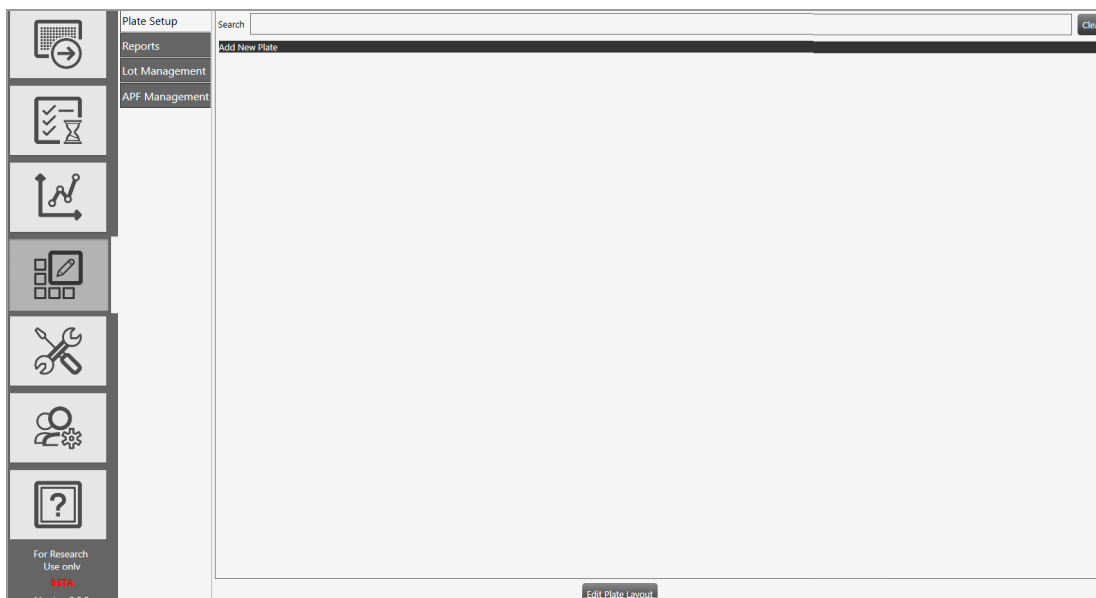
QX Manager Software creates the data file for the wells that were processed on the canceled plate and the status in the Event Log is specified as “run aborted by user.”

Chapter 6 Creating or Editing Templates

If you have been assigned the Create New Templates user privilege, you can create, edit, and save plate and report templates for your common experiments. Templates that are available to you appear in the dropdown list in the Plate Information window, and also in the default view of the Template Setup tab. If the list is long, you can use the Search field to narrow the results.

For information on report templates, see [Creating or Editing Report Templates on page 80](#).

All users can configure a plate template in a CSV file, and then import the template into QX Manager Software. For information, see [Creating and Storing Plate Templates from CSV Files on page 83](#). If you have been assigned APF privileges, you can select an APF in the Add Plate window. All users can select or create a template with an APF selected. For information on APFs, see [Managing Assay Protocol Files on page 158](#).



Templates you created are saved to one of the following folders:

- The path specified in your user preferences; these appear under My Templates.
- The paths specified by your administrator in the shared or preferred location fields in System Settings.

Important: If a shared location is specified in System Settings, you can save your templates there or in your personal storage location. Preferred locations override personal folder settings so that templates from all users are saved there.

Template Plate Editor Window

Use the Template Plate Editor to

- Create new or edit existing plate template files, and save new or modified templates for reuse
- Add up to four sample descriptions per well
- Exclude wells from droplet reading

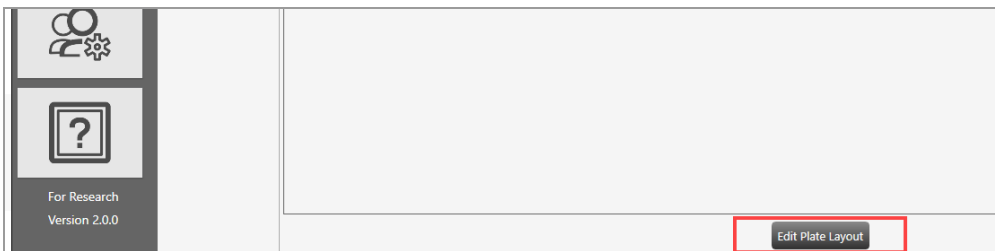
Important: When you create an experiment from a template with wells excluded in the plate layout, the instrument pierces the foil for all wells in the physical plate, but reads droplets for active wells only.

To open the template plate editor

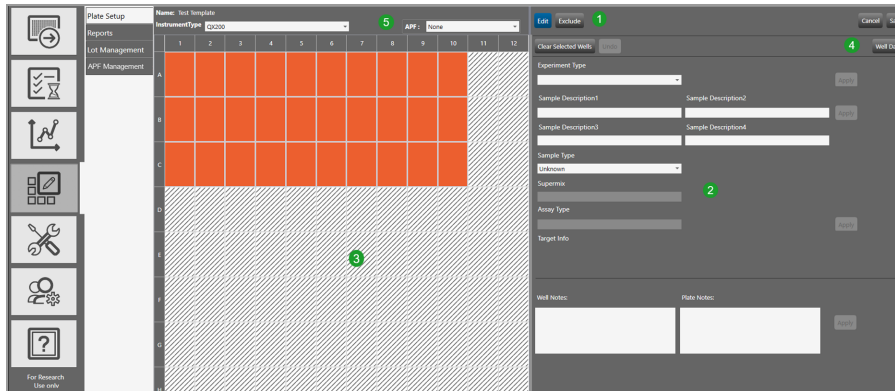
1. Click the Templates tab on the left.



2. In the Templates window, click Edit Plate Layout

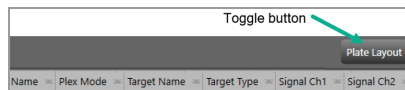


You can also open the template plate editor from the Add Plate window.



LEGEND

- 1 The Edit and Exclude buttons are accessible from the top of the screen.
- 2 The right pane contains the interface for defining your experiment parameters.
- 3 The left pane displays the plate grid and configuration information in each well.
- 4 The Well Data button toggles to the Well Data table, where you can see your plate setup in a tabular format, and then back to the Plate Editor. When the Well Data table appears, the button name changes to Plate Layout.



- 5 From the dropdown list, you can select an APF to associate with the template.

Important: If you selected an APF, you can add wells to the plate layout if there is room on the plate, but the wells inherit the APF configuration. You can update fields that are not predefined by the APF.

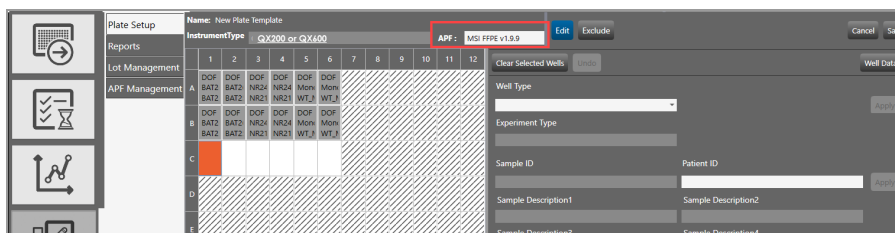


Plate Editor Tools

Use the buttons described in [Table 17](#) when you are creating a template or defining an experiment in the Plate Template Editor.

Table 17. Plate Editor Buttons

Button	Description
Edit	<p>Allows you to</p> <ul style="list-style-type: none"> ■ Set the experiment type, sample name and type, supermix, and assay type ■ Set the target name, type, and fluorophore for each channel ■ Add well or plate notes
Exclude	<p>Toggle button that allows you to include or exclude wells from droplet reading</p> <p>Note: Wells in the plate editor are initially excluded by default.</p>
Cancel	Allows you to cancel and return to the Template Setup window
Save	Opens a dialog box where you can enter a file name and storage location
Clear Selected Wells	Allows you to delete information from selected wells
Undo	<p>Allows you to reverse your last action</p> <p>Note: This button is enabled only after you have applied at least one experiment parameter (for example, the selection for Experiment Type).</p>
Well Data or Plate Layout	Toggle button that allows you to access the Well Data table and return to Plate Layout view
Apply	Applies your entries and choices for selected wells

Creating or Editing Plate Templates

Important: You must be assigned the Create New Template user privilege to create or edit, and then save, a template.

To create or edit a template

1. Do one of the following:

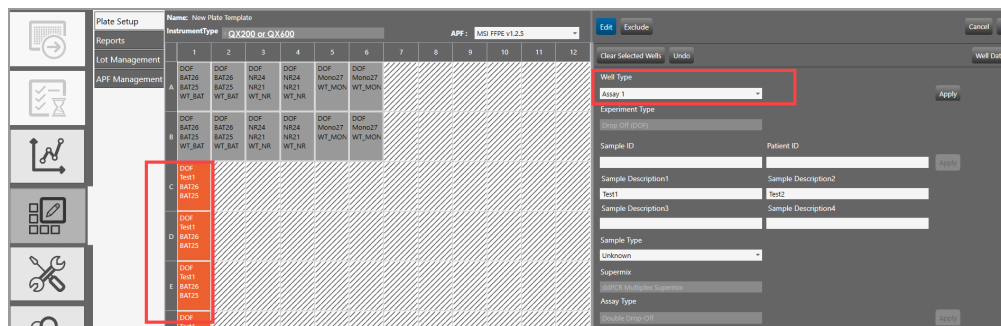
- Click Create New under Plate Template in the Plate information window, or you can select the Templates tab.
- Click Edit Plate Layout in the main Templates window.
- Search for and select a template under Plate Template in the Plate Information window.
- Select the Templates tab, and then double-click an existing template under My Templates.

Note: Templates imported from a CSV file are available from the dropdown list only.

The Plate Editor opens in Edit mode. The Instrument Type field is grayed out.

2. (Optional) Select an APF from the APF dropdown list.

Important: If you select an APF, you can enter or select information only for fields that have not been defined with static APF entries, as shown in the following graphic. All other fields conform to the configuration of the APF. You can add wells, but you cannot delete wells set in the APF. Wells that you add assume the APF settings automatically.



3. Click the Exclude button, and select the wells to include in the plate layout, and then click Include selected wells.

By default, all wells in the Plate Editor are automatically excluded. You must select and include new wells in the template. If you typically run full plates, you can change the setting to Acquire all wells by default in your user preferences. For information, see [Managing Your Preferences on page 39](#).

4. Click the Edit button to display the layout for experiment and sample identification.

The fields are enabled when you include wells.

5. For one or more wells at a time, complete any or all of the following substeps to include the selections and entries that should be static. Leave blank any field that should be completed at the time of the experiment run.

- a. Select an experiment type, sample type, supermix, and assay type, and enter sample descriptions. For information, see

- [Experiment Types on page 73](#)
- [Sample Types on page 75](#)
- [Sample Descriptions on page 75](#)
- [Supermixes on page 76](#)
- [Assay Types and Fluorophores on page 77](#)

Note: For automatic thresholding of positive control wells, you must select the Pos Ctrl sample type.

Note: If you selected an APF on the Add Plate screen, fields that are configured in the APF for one or more wells, are grayed out.

- b. Enter a sample ID and a subject ID.
 - c. If you selected Residual DNA Quantification as the experiment type, enter a conversion factor.
 - d. Enter up to four sample descriptions, and any relevant well or plate notes.
 - e. Edit the Target Name and Target Type fields.
6. Click Apply.
 7. Click Save and select one of the following:
 - **Shared Templates.** The template is saved in the folder specified in the System Settings.
 - **My Templates** (saves the file to the path in your user preferences) or **System Templates** (saves the file to the preferred location specified by your system administrator).
 - If your system administrator has specified preferred locations for all users, templates are automatically saved to that location.

Experiment Types

QX Manager Software offers the experiment types described in [Table 18](#), each enabling different assay options and analysis tools.

Tip: You can assign multiple experiment types across the plate, but you can assign only one experiment type to each well.

Table 18. Experiment types

Type	Description
Direct quantification (DQ)	<p>DQ uses absolute quantification to determine the concentration (copies/μl) of target DNA copies in a sample. DQ measures the number of positive and negative droplets for each target in a well based on their fluorescence amplitude, and uses a Poisson algorithm to calculate the starting concentration of each target DNA molecule.</p> <p>Note: The ABS experiment type created in QuantaSoft Software versions 1.4 through 1.7 is automatically mapped into the software as a DQ experiment type.</p>
Copy number variation (CNV)	CNV determines concentration and calculates the copy number of an unknown (CNV) target relative to a known reference or references within the same well.
Mutation detection (MUT)	MUT determines concentration and calculates the fractional abundance of an unknown mutant present at a low frequency in a wild-type background.
Rare event detection (RED)	RED determines the concentration of a known mutant or a rare target species relative to a given reference species within a large pool of background DNA.
Drop-off (DOF)	DOF determines absolute quantification of targets for assays designed to detect non-wild type sequences, such as indels and genome edits; DOF is designed to support an assay strategy where one probe counts all alleles and one “drop-off” probe sits on top of a predicted cut site or mutation site.
Gene expression (GEX)	GEX determines concentration (as with DQ) and calculates relative expression levels of an unknown target relative to a known reference or references within the same well.

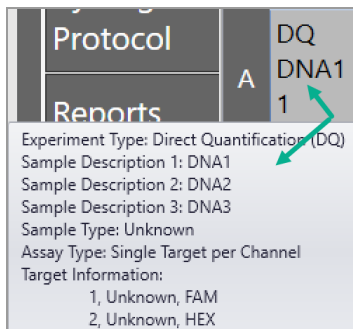
Table 18. Experiment types, continued

Type	Description
Residual DNA quantification (RDQ)	<p>RDQ provides a precise method for residual host cell DNA quantification.</p> <p>Note: Users can enter a conversion factor (1/C-value) for a given genome (human, mouse, etc.) to calculate the mass for a well based on the number of copies measured, up to three decimals. The software displays the result in pg/ul of the ddPCR well in the Molecular Weight column of the Data Table. Starting sample concentration can then be back-calculated based on dilution factor.</p>

Sample Descriptions

QX Manager Software allows you to enter descriptive words or phrases for your sample in up to four fields per well. Descriptors can include information such as research type, dilution factors, and so forth.

Tip: Only the information from the first field appears in the Plate Editor layout. However, you can see entries in the second, third, and fourth fields in the Well Data table, or you can pause on a well to display a tool tip, as shown in the following graphic.



Sample Types

You can assign multiple sample types within the plate, but you can assign only one sample type to each well.

QX Manager Software provides four sample types for selection:

- Unknown
- NTC (no template control)
- Pos Ctrl (positive control) where one or more targets are expected
- Neg Ctrl (negative control), where no response is expected

Important: Beginning in v1.2, QX Manager Software can automatically calculate thresholds for positive control wells, but you must select Pos Ctrl from the Sample Type dropdown list. You can select Pos Ctrl during plate setup or in the Analysis module after the run is finished.

Supermixes

QX Manager Software offers five PCR supermixes. Each supermix is optimized to deliver maximum PCR efficiency and sensitivity for the amplification and detection of DNA and RNA targets.

Important: Use of unapproved supermixes can harm the instrument and void the warranty.

ddPCR Supermix	Description
Multiplex Supermix	For use in nucleic acid sample preparations to amplify and detect DNA targets in probe-based experiments involving multiple targets. Note: Bio-Rad recommends using ddPCR Multiplex Supermix for Cy5 and Cy5.5 probes assays.
Supermix for Probes (No dUTP)	For use in nucleic acid sample preparations, with sensitivity for the amplification and detection of DNA targets using hydrolysis probe-based assays.
Supermix for Probes	For use in sample preparation with uracil N-glucosylase (UNG) decontamination protocols to prevent the reamplification of carryover PCR products between experiments.
Supermix for Residual DNA Quantification	For use in residual DNA detection.
One-Step RT Advanced Kit for Probe	For use in absolute quantification of target RNA molecules.
EvaGreen® Supermix	For use in gene expression.

Assay Types and Fluorophores

QX Manager Software offers the assay methods explained in [Table 19](#).

Table 19. Assay types

Assay type	Description
Single target per channel	<p>Method assuming up to six probe colors (FAM/EvaGreen®, HEX/VIC, Cy5, Cy5.5, ROX, and ATTO 590) and up to six targets per channel.</p> <p>This is the default assay type, with the maximum six rows displayed. You can delete up to three rows.</p>
Amplitude multiplex	<p>Method to increase multiplexing up to twelve targets per well, with one or two targets detected per channel. The maximum of eight rows are displayed. You can delete up to seven rows.</p>
Probe mix triplex	<p>Triplex mode that allows three groups of three targets each.</p> <p>Group 1 has one target in FAM, one target in HEX/VIC, and one target in both channels. Group 2 has one target in Cy5, one target in Cy5.5, and one target in both. Group 3 has 1 target in ROX, 1 target in ATTO 590, and one target in both groups. You can delete a group, but not the individual targets within the group.</p> <p>Note: The channels within each group are not interchangeable therefore FAM is always paired with HEX/VIC and so forth.</p>
Advanced classification	<p>Method to increase multiplexing rate in any combination of targets detected per channel. This method requires manual classification of droplets.</p>
Basic drop-off	<p>This mode allows two groups of two targets each. The first group appears by default, with one target in FAM <u>and</u> HEX/VIC and one target in FAM <u>or</u> HEX/VIC.</p> <p>This assay type is available only for the DOF experiment type.</p> <p>You can add one more group of two targets, with one target in Cy5 <u>and</u> Cy5.5, one target in Cy5 <u>or</u> Cy5.5. You can delete a group, but not the individual targets in the group.</p> <p>Note: The channels within each group are not interchangeable therefore FAM is always paired with HEX/VIC and Cy5 with Cy5.5.</p>

Table 19. Assay types, continued

Assay type	Description
Double drop-off	<p>This assay type is available only for the DOF experiment type, and allows three groups of three targets each.</p> <p>The first group appears by default, and looks for one target in FAM <u>and</u> in HEX/VIC (target type reference) one target in FAM (target type unknown), and one target in HEX/VIC (target type unknown).</p> <p>You can add the second group to look for one target in Cy5 <u>and</u> Cy5.5 (target type reference, with two reference copies), one target in Cy5 (target type unknown), and one target in Cy5.5 (target type unknown).</p> <p>You can also add a third group to look for one target in ROX <u>and</u> ATTO 590 (target type reference), one target in ROX (target type unknown), and one target in ATTO 590 (target type unknown).</p> <p>By default, all reference copies are set to 2.</p> <p>The channels are static for each group. (Channel 1 FAM is always paired with Channel 2 HEX/VIC, Channel 3 Cy5 is always paired with Channel 4 Cy5.5, and Channel 5 ROX is always paired with Channel 6 ATTO 590.</p>

The experiment type you select determines the available assay methods. [Table 20](#) displays the sample type associated with each assay method.

Table 20. Assay types by experiment type

Assay type	Experiment type						
	DQ	CNV	MUT	DOF	GEX	RDQ	RED
Single target per channel	✓	✓	✓		✓	✓	✓
Amplitude multiplex	✓	✓	✓		✓	✓	✓
Probe mix triplex	✓	✓	✓		✓	✓	✓
Advanced classification	✓						✓
Basic drop-off				✓			
Double drop-off				✓			

QX Manager Software automatically populates the fluorophore in each channel for each target but you can change the default selections.

Use [Table 21](#) to select from the fluorophore options for each assay method listed.

Table 21. Fluorophore options

Assay method	Signal Ch1	Signal Ch2	Signal Ch3	Signal Ch4	Signal Ch5	Signal Ch6
Single target per channel, for 1 to 6 targets	FAM	HEX	Cy5	Cy5.5	ROX	ATTIO590
	EvaGreen®	VIC	None	None	None	None
	None	None				
Amplitude multiplex, for 1 to 12 targets	FAM Hi	HEX Hi	Cy5 Hi	Cy5.5 Hi	ROX Hi	ATTO 590 Hi
	FAM Lo	HEX Lo	Cy 5 Lo	Cy 5.5 Lo	ROX Lo	ATTO 590 Lo
	EvaGreen® Hi	VIC Hi	None	None	None	None
	EvaGreen® Lo	VIC Lo				
	None	None				
Probe mix triplex, for 9 targets	FAM	HEX	Cy5	Cy5.5	ROX	ATTO 590
	None	VIC	None	None	None	None
		None				
Advanced classification	FAM	HEX	Cy5	Cy5.5	ROX	ATTO 590
	+1-10	1 to 10	1 to 10	1 to 10	1 to 10	1 to 10
	EvaGreen®	VIC	None	None	None	None
	1 to 10	1 to 10				
	None	None				

Creating or Editing Report Templates

If you are assigned the Create New Templates user privilege, you can create and save a report template by editing the default report structure or by opening and editing an existing report.

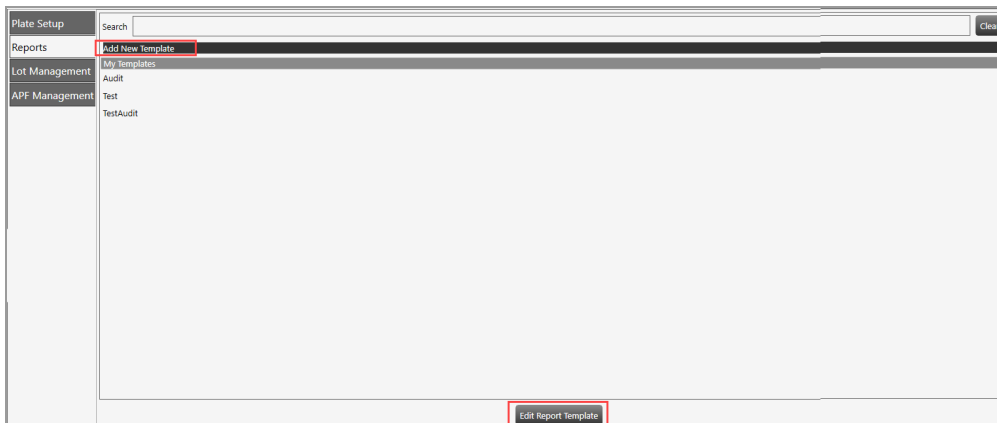
To create or edit a report template

1. Select the Template Setup tab.



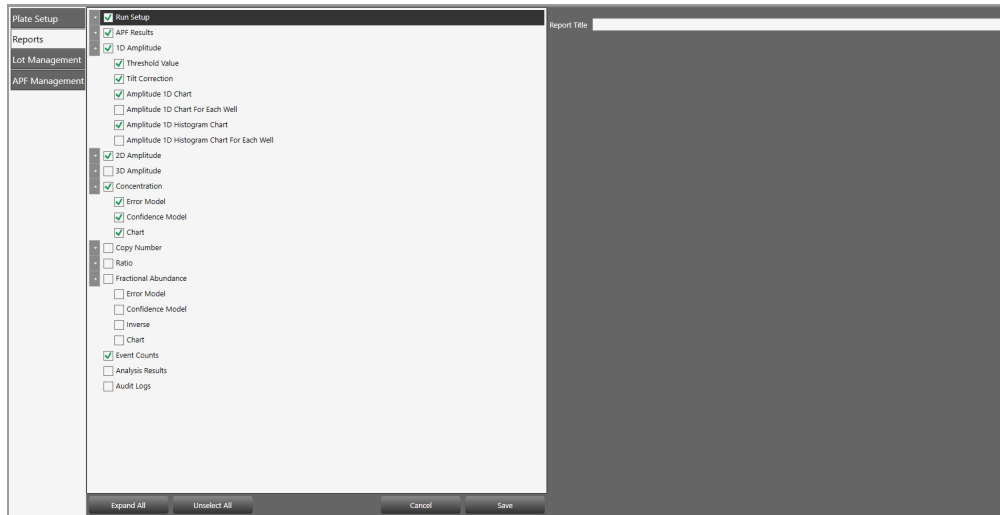
2. Click Reports and do one of the following:

- To create a new template, click Add New Template. and click Edit Report Template.
- To open an existing template, select the template and click Edit Report Template.



- Click the arrow to the left of each category to expand the list of reportable items.

Important: The Audit Log checkbox is enabled in the Premium Edition only.

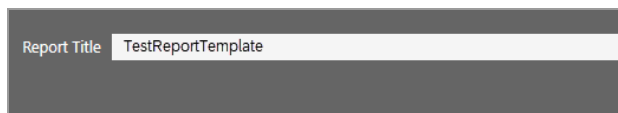


- Select and clear checkboxes to create the report structure.

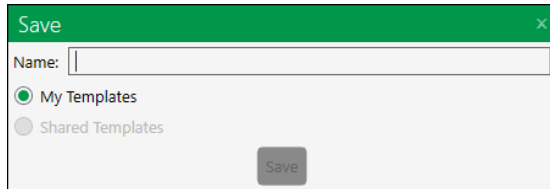
Note: Use the buttons at the bottom of the screen to expand or collapse all categories at once, or to select or clear all checkboxes at once. You can also drag and drop the section to rearrange the order of appearance in the report, and drag and drop items in the section to reorder them.



- Enter a name for your report in the Report Title field and click Save.



6. When the Save dialog box appears, enter a file name.



7. Select a storage location. Choose from the following:
 - System Templates (preferred location, if specified by the administrator)
 - My Templates (personal storage location specified in user preferences)
 - Shared Templates (files saved to this location are available to all users)
8. Click Save again to save the report format as a template.

Creating and Storing Plate Templates from CSV Files

You can create a plate template in a spreadsheet application, save it in CSV format, and then store it in a personal or shared folder for use in your droplet reading runs. Note the following:

- The headings for the columns containing the experiment information match the fields in the Plate Editor.
- Once you specify the information in row 1, the field entries must remain static until you install a new software version or edition.
- CSV template files appear in the Plate Setup dropdown list only.

To set up and save a CSV template

1. Open a spreadsheet program and enter the run settings into the template in the format shown below.

Fig. 1: Template CSV Layout Example, Columns A through H

	A	B	C	D	E	F	G	H
1	ddplate - DO NOT MODIFY THIS LINE	Version=1	ApplicationName=QX Manager Standard Edition	ApplicationVersion=2.0.0.11098	ApplicationEdition=ResearchEmbedded	User=QLDR-XXXX\QXManager_user	CreatedDate=7/24/2022 1:54:15 PM	
2	PlateSize=GCR96							
3	PlateNotes=Plate note text							
4								
5	Well	Perform Droplet Reading	ExperimentType	Sample description 1	Sample description 2	Sample description 3	Sample description 4	SampleType
6	A01	Yes	Direct Quantification (DQ)	Description text 1	Description text 2	Description text 3	Description text 4	Unknown
7	A01	Yes	Direct Quantification (DQ)	Description text 1	Description text 2	Description text 3	Description text 4	Unknown
8	A01	Yes	Direct Quantification (DQ)	Description text 1	Description text 2	Description text 3	Description text 4	Unknown
9	A01	Yes	Direct Quantification (DQ)	Description text 1	Description text 2	Description text 3	Description text 4	Unknown
10	A01	Yes	Direct Quantification (DQ)	Description text 1	Description text 2	Description text 3	Description text 4	Unknown
11	A01	Yes	Direct Quantification (DQ)	Description text 1	Description text 2	Description text 3	Description text 4	Unknown
12	A02	Yes	Direct Quantification (DQ)	Description text 1	Description text 2	Description text 3	Description text 4	Unknown
13	A02	Yes	Direct Quantification (DQ)	Description text 1	Description text 2	Description text 3	Description text 4	Unknown
14	A02	Yes	Direct Quantification (DQ)	Description text 1	Description text 2	Description text 3	Description text 4	Unknown
15	A02	Yes	Direct Quantification (DQ)	Description text 1	Description text 2	Description text 3	Description text 4	Unknown
16	A02	Yes	Direct Quantification (DQ)	Description text 1	Description text 2	Description text 3	Description text 4	Unknown
17	A02	Yes	Direct Quantification (DQ)	Description text 1	Description text 2	Description text 3	Description text 4	Unknown

Fig. 2: Template CSV Layout Example, Columns I through R

[illegible]

2. When the configuration is finished, navigate to one of the following locations and save the .csv file to ensure it is available for your runs:
 - The Templates folder location specified in your user preferences. For information on your personal storage locations, see [Managing Your Preferences on page 39](#).
 - The shared or preferred Templates location determined by your system administrator. For information on shared folders, including preferred locations, see [Shared Settings on page 180](#).

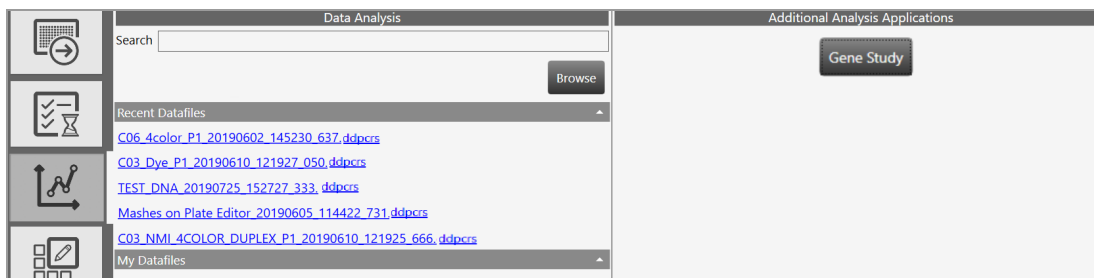
Important: The .csv file must be in one of these locations to be available for selection from the dropdown list in the Plate Information window. CSV templates are not listed with standard templates in the Plate Templates window.

Chapter 7 Data Analysis Module Overview

As the instrument reads droplets in each well, the QX600 Droplet Reader collects raw data. At the end of each run, QX Manager Software uses the raw data to produce a data file, which contains analysis information based on the number of positive and negative droplets identified for each target in each sample. The fraction of positive droplets is fit to a Poisson algorithm to determine the starting concentration of the target DNA molecule in units of copies/ μ L input.

This section summarizes the analysis functionality, and also describes the general options governing your chart displays. More detailed information, including specific information and uses for each analysis window, as well as information on data calculations, is contained in [Chapter 8, Data Analysis Methodology](#).

When you select the Data Analysis tab, you can access the Analysis module and the Gene Study module.



When you open the data file in the Analysis module, you can view the data with the automatic analysis thresholds and calculations, or you can choose different options to recalculate, replot, or recolor the results.

The Data Analysis module functionality allows you to

- View your droplet data in 1D, 2D, and 3D plots
- Analyze multiple targets within a well using amplitude multiplexing and probe mixing strategies
- Detect genomic editing events and non-wild type events with a “drop-off assay” analysis option
- Visualize and export data with improved flexibility
- Generate reports for different analysis types
- The Gene Study module allows you to
- Collect data by adding one or more data files
- View gene expression calculations across multiple experiments

Important: The Gene Study module is accessible from a standalone computer only. The maximum number of samples you can analyze in a gene study is limited by the size of the RAM and virtual memory of the standalone computer.

Opening a Data File for Analysis

You can open the following file types in the Data Analysis module:

- .qlps
- .ddpcrs

To open data files, you must be on a computer on which QX Manager Software is installed and have access to the storage locations.

To open a data file for analysis

1. In QX Manager Software, select the Data Analysis tab.



Available data files are displayed under the following headings.

- **Recent Datafiles**

Displays a list of files you have recently opened.

- **System Datafiles or My Datafiles**

Files are always saved to one of the following locations:

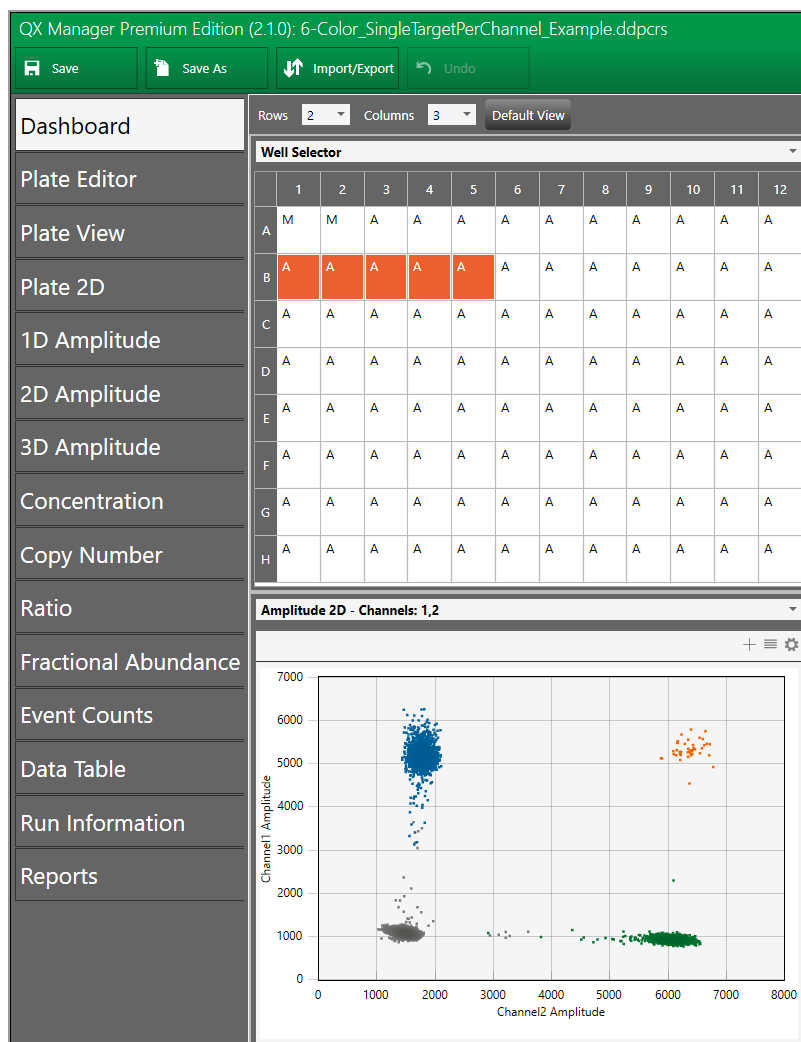
- ☐ If preferred locations are enabled, then files for all users are saved there and they appear under System Datafiles.
- ☐ If preferred locations are not enabled for all users, your files are saved to your personal folder and appear under My Datafiles.

- **Shared Datafiles**

You can also save your data files to a shared location; these appear under Shared Datafiles.

- Click a file to open it in the Analysis module.

Tabs for each analysis and data window appear on the left of the Analysis window.



Tip: From the computer attached to the QX600 Droplet Reader, you can also open the data file from the Run Status window. In the completed files section, click the completed run icon (✓) for the file, and then click Launch Analysis.

Saving a Data File

You can save data files with or without modifications, with the same or different file names, and in the same or different locations.



Important: When you save a .qlp.qlps file type in QX Manager Software, the software automatically saves the file as a .ddpcrs file. This creates a new file and does not overwrite the original.

To save a data file

1. Do one of the following:
 - To save the file under the same file name and in the same location, click Save.
 - To save the file under a different file name or in a different location, click Save as, and then do one of the following:
 - If you are saving the file under the same name and in the same location, QX Manager Software saves the file immediately.
 - If you are saving the file under a different file name or in a different location, navigate to the new location (if applicable) and enter the new file name. You must be assigned the Overwrite existing file name user privilege to change the file name.
2. Click Save.

Analysis Display and Output Options

QX Manager Software provides the following option that you can choose or set:

- **Universal Options** — available from the primary toolbar to save, import, or export data files
Note: The APF Tools button appears only when an APF was used for a droplet reading run.
- **Chart Display Preferences** — to change how your chart appears in the analysis window
- **Chart Scale Options** — to change the scale represented on an axis
- **Chart Menu Options** — available in chart views (from the  icon) to copy, save, and print chart images. In Amplitude charts showing data points, you can show or hide
- **Table Menu Options** — available in table views (from the  icon) to copy data, export data to Excel or CSV files, and show or hide columns

You can also expand the chart display to full-screen size and then return to the original display.

Universal Options

Universal options appear on the top menu bar, and are available from all windows, although functionality within the main option might differ according to the analysis window that appears.



Note: If you open a run that was executed using an APF, the APF Tools button appears in the menu bar.



Universal options are explained in [Table 22](#), and are available in all Analysis module windows.

Table 22. Universal options

Option	Description
Save	Saves changes to the analysis file under its current file name or file extension
Save as	Saves the file with a different name If you open a file in the latest application that was created in an earlier application, the file assumes the file extension of the new application. See Saving a Data File on page 89 for information.
Import/export	<ul style="list-style-type: none">■ Imports or exports plate setup data to a CSV file■ Imports or exports plate setup data to a plate template file■ Exports visible data to a CSV file■ Exports amplitude, cluster, and linkage data■ Exports and prints the visible charts for the 2D plate layout
Undo	Enabled in the Plate Editor only, after you have made a change
APF Tools	Enabled only when an APF was used for the run For information, see Reanalyzing the Run File on page 108 .
User Settings	Allows you to restore your user preferences to the default settings and to show or hide color matrix change messages

To import or export setup or analysis data

- Click Import/Export.

Table 23 describes the available import/export options.

Note: If there is unsaved data, the software prompts you to save the file before its contents can be imported or exported.

Table 23. Import/export options

Import/export option	Description
Import plate setup CSV	Import plate layout data from a CSV file to the Plate Editor Note: This option is only available in the Plate Editor, and is not available for files that were run using an APF. The new plate layout overwrites the existing layout.
Export plate setup CSV	Export a configured plate layout to a CSV file Note: This option is only available in the Plate Editor and is not available for files that were run using an APF.
Import plate setup	Import a plate layout from a plate template (.ddplt) file Note: This option is only available in the Plate Editor.
Export plate setup	Export the plate layout to a plate template file Note: This option is only available in the Plate Editor.
Export visible data to CSV	Export only the data from the wells you selected in the Well Selector to a CSV file
Export amplitude data	Export the data to an Excel spreadsheet, and indicates whether or not the target was found (1 = yes, 0 = no)
Export cluster data	Export the data to an Excel spreadsheet Note: Includes (for each well) whether or not the target was found (1 = yes, 0 = no), the droplet count, and the mean and standard deviation for each dye, and the cluster ID.
Export linkage data	Exports to a CSV file the number of copies of a target that are present in overabundance, versus. the expected value in copies per μL
Export visible charts	Exports all charts that are displayed
Print visible charts	Prints all charts that are displayed

Table 24 shows where the options are available.

Table 24. Option availability

Import/expo rt option	APF results	Plate editor	Plate view	Plate 2D	Ampl plots	Data visual charts	Event counts	Data table
Import plate setup from CSV		✓						
Export plate setup to CSV		✓						
Import plate setup		✓						
Export plate setup		✓						
Export visible data to CSV					✓	✓	✓	✓
Export amplitude data	✓	✓	✓	✓	✓	✓	✓	✓
Export cluster data	✓	✓	✓	✓	✓	✓	✓	✓
Export linkage data	✓	✓	✓	✓	✓	✓	✓	✓
Export visible charts				✓	✓	✓	✓	
Print visible charts				✓	✓	✓	✓	

Note: Amplitude plots are available in 1D, 2D and 3D, and Data Visualization charts are Concentration, Copy Number, Ratio, and Fractional Abundance. In the Dashboard view corresponding options are available for each plot or chart displayed.



Expanding or Reducing the Chart

You can expand or reduce the chart size in the following windows:

- Dashboard
- 1D Amplitude
- 2D Amplitude
- 3D Amplitude
- Concentration
- Copy Number
- Ratio
- Fractional Abundance
- Event Counts

If you expand an amplitude or data visualization chart to full size, the Well Selector and the Well Data table are hidden from view until you collapse the chart again.

To expand or reduce the chart size

1. To increase the chart size to the full window, click the  icon in the upper-right corner of the chart.
2. To reduce the window to its original size, click the  icon.

Selecting Chart Display Preferences

The Chart Display preferences primarily control font sizes and positioning. [Table 25](#) explains each preference, as well as the windows in which it is available.

Table 25. Chart display preferences

Preference	Window	Description
Channel or Dye	Plate 2D	Select Channel or Dye to identify your chart accordingly ■ If you select Channel, the channel number appears as the chart title ■ If you select Dye, FAM, HEX, Cy5, C5.5, ROX, or ATTO 590 appears as the chart title
	1D Amplitude	
	2D Amplitude	
	3D Amplitude	
Axis Data Label Size	1D Amplitude	Select a different font size
	2D Amplitude	
	3D Amplitude	
	Concentration	
	Copy Number	
	Ratio	
	Fractional Abundance	
	Event Counts	
Chart Data Label Size	Concentration	Select a different font size
	Copy Number	
	Ratio	
	Fractional Abundance	
	Event Counts	
Chart Marker Size	Concentration	Select a different marker size
	Copy Number	
	Ratio	
	Fractional Abundance	
Chart Marker Label Position	Concentration	Reposition the marker label
	Copy Number	
	Ratio	
	Fractional Abundance	

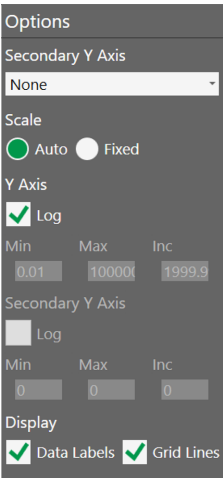
To select chart display preferences

1. Click the Chart Display button.
2. Select your options.

The display changes immediately to reflect your choices.
3. Do one of the following:
 - To save your choices as your default settings, click Save as my default. When the success message appears, click OK to close the dialog.
 - To close the menu without saving the settings, click anywhere in the chart display.

Selecting Chart Scale and Label Options

Chart scale options primarily control chart scale and label displays.




Each preference, as well as the windows in which it is available, is explained in [Table 26 on page 97](#).

Table 26. Chart scale options

Option	Description
Scale	<p>Appears in options for all charts</p> <p>The Scale option determines the numerical range for your chart data:</p> <p>Auto — Selected by default; QX Manager Software automatically displays a scale to match the data distribution.</p> <p>Fixed — Selecting Fixed enables the Axis fields, and allows you to manually adjust the range and increment.</p>
Axis	<p>Appears in options for all charts</p> <p>When the Axis fields are enabled, you can manually adjust the minimum (Min) and maximum (Max) points for the data display on each axis, as well as the increments (Incr).</p> <p>X — Horizontal axis</p> <p>Y — Vertical axis</p> <p>Note: You can modify the x axis scale in the 2D Amplitude chart only.</p>
Secondary Y Axis	<p>Appears in options for Concentration only</p> <p>From the dropdown list, you can select from Channel 2 concentration, ratio, copy number, or fractional abundance to display the corresponding data in a secondary row on the y axis.</p>
Log	<p>Appears in options for all Data Visualization charts (Concentration, Copy Number, Ratio, and Fractional Abundance)</p> <p>You can select the Log checkbox to display the y axis in base 10 logarithmic format.</p> <p>Note: In the Concentration chart, you can also select Log for the secondary y axis.</p>
Inverse	<p>Appears in options for Ratio and Fractional Abundance only</p> <p>The Inverse option equals the opposing number of the Ratio data point to a total of 2, and the opposing number of the Fractional Abundance data point to a total of 100.</p>
Data labels	<p>Appears in options for all Data Visualization charts</p> <p>A data label appears by default for each point in the chart, but you can clear the checkbox to remove the labels.</p>
Grid Lines	<p>Appears in options for all charts</p> <p>Gridlines appear by default. You can clear the check box to remove them.</p>

To select chart scale and label options

- 1. Click the  icon in the upper-left corner of the chart to display the corresponding chart options.
- 2. To manually define the chart scale, select Fixed.
Note: Auto is the default setting.
- 3. In the Min, Max, and Inc fields, enter your lower and upper limits, and the increment, for the y axis. For 2D Amplitude, you can also change the scale on the x axis.
- 4. For Concentration, Copy Number, Ratio, Fractional Abundance, and Event Counts, select an option for a secondary y axis display. To change to a base 10 logarithmic scale on the y axis, select the Log checkbox.
- 5. For Ratio and Fractional Abundance, to view an inverse calculation, select the Inverse checkbox.
- 6. To remove the data labels or grid lines from the display, clear the Data Labels or Grid Lines checkbox.

Using Chart Menu Options

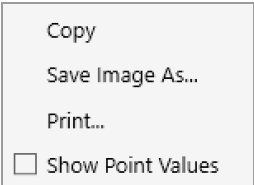


Chart menu options are explained in [Table 27](#).

Table 27. Chart menu options

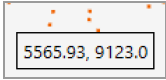
Option	Description
Copy	Copies the chart to the clipboard, which you can paste into a separate document, such as an MS Word file
Save Image as	Saves the chart as an image file
Print	Prints the image
Show Point Values	Shows corresponding point values when you pause on a data point  Tip: Select the Show Point Values checkbox to see the data. Data is available for 1D and 2D amplitude only.

Chart menu options are available in the following windows:

- 1D Amplitude
- 2D Amplitude
- 3D Amplitude
- Concentration
- Copy Number
- Ratio
- Fractional Abundance
- Event Counts

To use the chart menu options

1. Select one or more wells in the Well Selector.

	1	2	3	4	5	6	7	8	9	10	11	12
A												
B												
C												
D												
E	A	A	A	A	A	A						
F	A	A	A	A	A	A						
G	A	A	A	A	A	A						
H	A	A	A	A	A	A						

Corresponding data appears in the Well Data table and the chart.

Well Data						
Well	Target	Conc(copies/μL)	Status	Experiment	SampleType	
F01	1	534	OK	DQ	Unknown	
F01	2	531	OK	DQ	Unknown	
F01	3	539	OK	DQ	Unknown	
F01	4	551	OK	DQ	Unknown	
F02	1	551	OK	DQ	Unknown	
F02	2	551	OK	DQ	Unknown	
F02	3	550	OK	DQ	Unknown	

2. In the upper-right corner of the chart, click the menu icon (☰) and select from the dropdown list.
- Select Copy to copy the chart to the Clipboard. To paste the data, navigate to the location (such as a Word file), right-click and select Paste.
 - Select Save Image As and follow the prompts to save the file.
 - Select Print and follow the prompts to print the image.
3. Click anywhere in the display to close the dialog box.

Using Table Menu Options

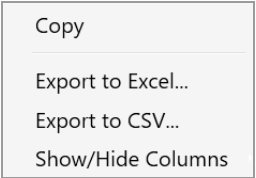


Table menu options are explained in [Table 28](#).

Table 28. Table options

Option	Description
Copy	Copies the information from the selected wells to the clipboard
Export to Excel	Exports the data to an .xlsx file
Export to CSV	Exports the data to a .csv file
Show/Hide Columns	<div> <div>Add or remove columns from a table display</div> <div>When you select Show/Hide Columns, the following dialog box opens. Select or clear check boxes to show or hide columns in the data table.</div> <div> <div><input checked="" type="checkbox"/> Select All</div> <div><input checked="" type="checkbox"/> Well</div> <div><input checked="" type="checkbox"/> Sample description 1</div> <div><input checked="" type="checkbox"/> Sample description 2</div> <div><input type="checkbox"/> Sample description 3</div> <div><input type="checkbox"/> Sample description 4</div> <div><input checked="" type="checkbox"/> Target</div> <div><input checked="" type="checkbox"/> Conc(copies/μL)</div> <div><input checked="" type="checkbox"/> Status</div> <div><input checked="" type="checkbox"/> Experiment</div> <div><input checked="" type="checkbox"/> SampleType</div> <div><input checked="" type="checkbox"/> TargetType</div> <div><input checked="" type="checkbox"/> Supermix</div> </div> </div>

Table menu options are available in the following windows:

- 1D Amplitude
- 2D Amplitude
- 3D Amplitude
- Concentration
- Copy Number
- Ratio
- Fractional Abundance
- Event Counts

Tip: You can access the menu icon from the individual windows, or from the windows in the Dashboard display.

To use the table menu options

If you are using these options in the Data Table, skip to Step 3.

1. Select wells in the Well Selector.

	1	2	3	4	5	6	7	8	9	10	11	12
A												
B												
C												
D												
E	A	A	A	A	A	A						
F	A	A	A	A	A	A						
G	A	A	A	A	A	A						
H	A	A	A	A	A	A						

Corresponding data appears in the Well Data table.

Well Data							
	Well	Target	Conc(copies/μL)	Status	Experiment	SampleType	
▸	F01	1	534	OK	DQ	Unknown	⬆
	F01	2	531	OK	DQ	Unknown	
	F01	3	539	OK	DQ	Unknown	
	F01	4	551	OK	DQ	Unknown	
	F02	1	551	OK	DQ	Unknown	
	F02	2	551	OK	DQ	Unknown	
	F02	2	550	OK	DQ	Unknown	⬇

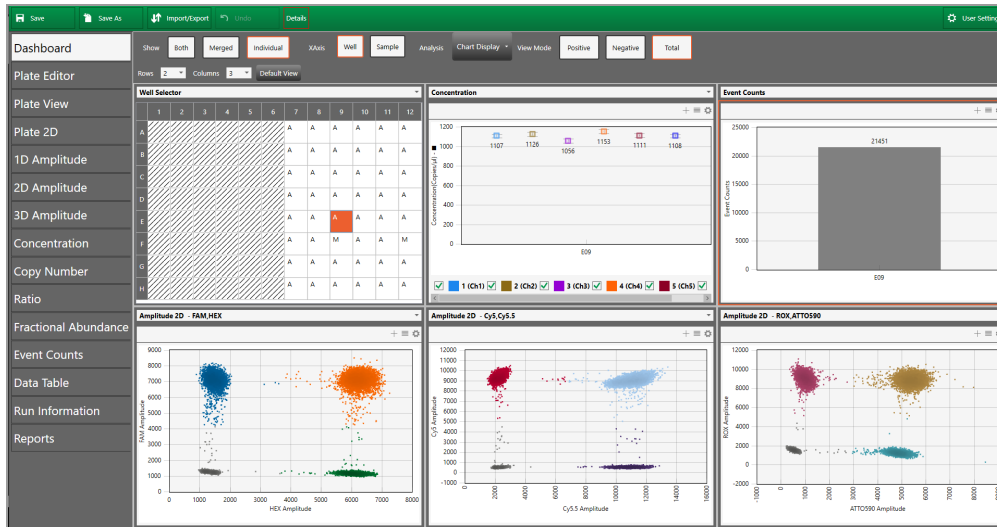
2. In the upper-right corner of the table, click the menu icon (☰) and select from the dropdown list.
 - To copy the data from the selected wells to the Clipboard, select Copy.

Note: To paste the data, navigate to the location (for example, an Excel file) and then right-click and select Paste.
 - To export your data, select Export to Excel or Export to CSV, and then follow the prompts to save the file.

Note: Exporting to Excel exports the data in spreadsheet format, while Exporting to CSV exports the data in comma-delimited format.
 - To show or hide the corresponding column in the table, select Show/Hide Columns, and then select or clear checkboxes.
3. Click anywhere in the display to close the dialog box.

Analysis Dashboard

When you open a data file, the Analysis module opens to a Dashboard window containing default analysis and data panes in a summary view. You can customize the view to change panes, or increase or decrease the number of panes.



Note: If you open a .qlps file, the Plate Editor opens as the default view and you must select the Dashboard tab to open it.

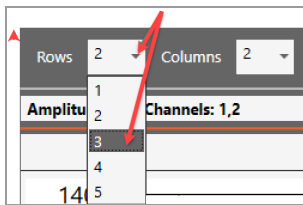
From the tabs on the left, you can access plate views, amplitude charts, data visualization distribution charts, droplet counts, and a complete data table. You can also view data specific to the run in the Run Information window, and run reports on any of the analysis charts from the Reports window.

Modifying the Dashboard Display

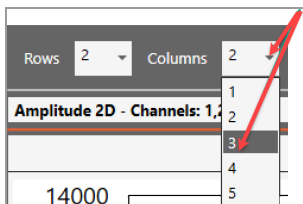
You can modify the column number and row number to display any or all of the analysis windows simultaneously.

To change the number of windows in the display

1. Click the dropdown arrow for Rows and then select a different number.



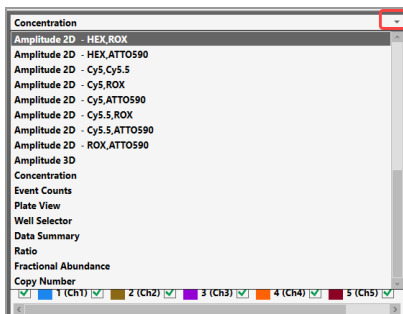
2. Click the dropdown arrow for Columns, and then select a different number.



You can also use the dropdown menus to change the windows that appear in the Dashboard.

To display a different window in a particular Dashboard slot

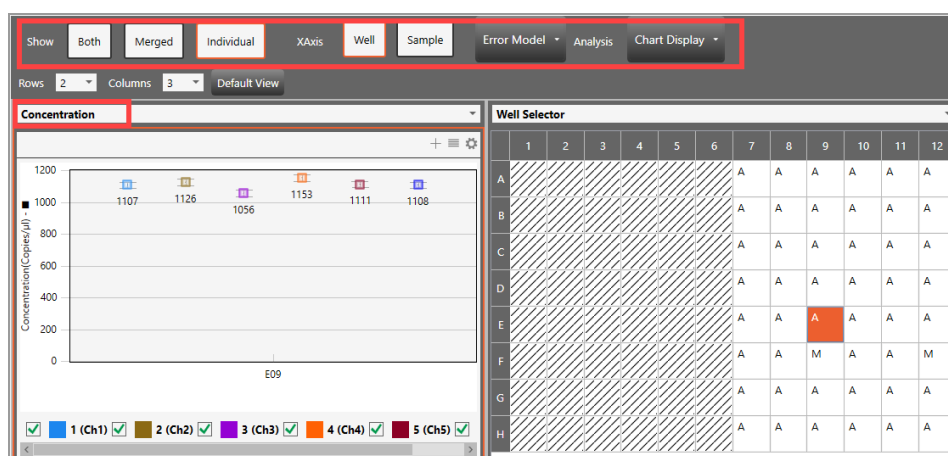
1. Click the dropdown arrow in the upper-right corner to display the list of available windows, and then select a window. The dropdown list closes and the window changes immediately.



Dashboard Options

From the Dashboard window, depending on the individual dashboard window you have selected, you can use a range of options as described below:

- When you select a window, the options that appear in the menu bar correspond to the options you see if you open the same window from the side menu. For example, if you select the Concentration chart in the Dashboard, the Concentration menu options appear.



- You can reset the entire dashboard display by adding or removing rows or columns to display more or fewer individual windows. For information, see [Modifying the Dashboard Display on page 104](#).



Click Default View to return to the default display of two rows and two columns.

- From any window you can
 - ☐ Expand or collapse the window
 - ☐ Copy, save, or print the image
 - ☐ Select display, scale, axis, and logarithmic options
 - ☐ Use the dropdown list to replace the current display with a different display
 - ☐ Use the menu bar options for the particular chart type

Table 29 describes the option sets identified in the following graphic.

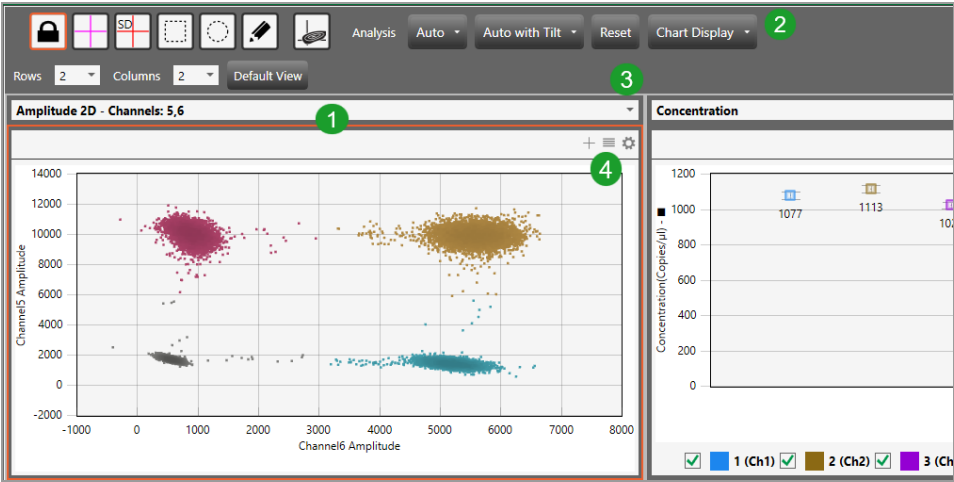
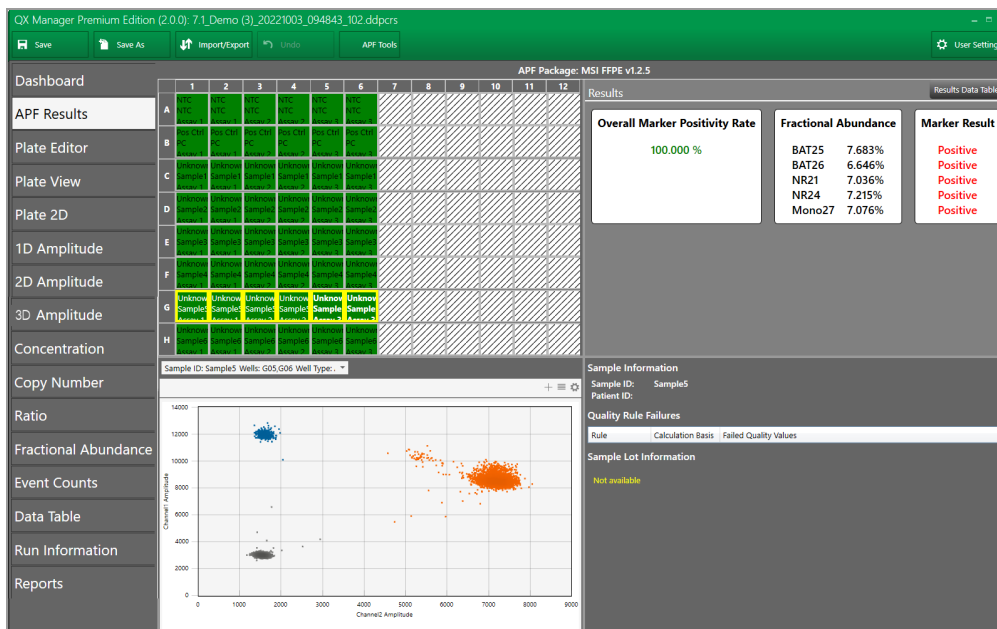


Table 29. Dashboard options

Item	Description
1	Border to identify selected window
2	Toolbar corresponding to the selected window type in the Dashboard
3	Dropdown arrow to replace the current display with a new display
4	Chart options

APF Results Dashboard

The APF Results tab appears in the Analysis module tab layout if the experiment was run using an APF. When you select APF Results, the corresponding window opens in a dashboard layout. In the following example, the plate is color-coded in accordance with the visual setup in the APF. For information on setting up an APF, see the QX Designer User Guide.



The dashboard displays the following windows:

- Plate layout (upper-left) showing processed wells.
- Results (upper-right) for the selected wells.
- Toggle button to the Results Data Table, and then back to APF Results (top-right).

APF Package: MSI FFPE v1.2.5

APF Results

Sample	Failed Quality Rule	Assay 1.BAT26 Cp/	Assay 1.BAT26 Drc	Assay 1.BAT26 Pos	Assay 1.BAT26 Neg	Assay 1.BAT25 Cp/	Assay 1.BAT25 Drc	Assay 1.BAT25 Pos	Assay 1.BAT25 Neg	Assay 1.WT_BAT C	A
NTC	None	0	20212	0	20212	0	20212	0	20212	0	2
PC	None	14.8184925166979	23169	290	22879	13.9549532866742	23152	273	22879	365.94158419495	3
Sample1	None	0	26597	0	26597	0	26597	0	26597	0	2
Sample2	None	0	30861	0	30861	0	30861	0	30861	0	3
Sample3	None	0	33977	0	33977	0	33977	0	33977	0	3
Sample4	None	12.8757420030017	22968	250.0000000000001	22718	13.5414416972776	22981	263	22718	326.666997161202	3
Sample5	None	12.4961797084357	22810	240.99999999999999	22569	14.6089370167792	22851	282.00000000000001	22569	351.054808119437	3
Sample6	None	13.8445028090805	21797	255	21542	26.5138952015679	22033	491	21542	386.948220014509	3

- 2D Amplitude display for the selected wells
- Other information specified in the APF, including sample ID, quality rule failures, and lot information recorded for the plate run

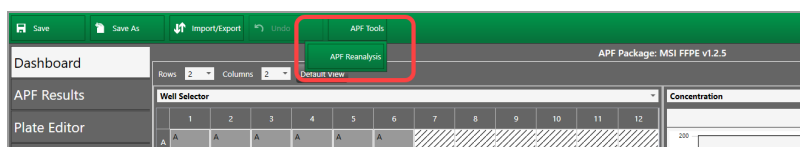
The APF Results dashboard is a static display and the windows cannot be changed. In the Universal Options toolbar, you can also select APF Tools and choose to reanalyze the file. See [Reanalyzing the Run File](#) for more information.

Reanalyzing the Run File

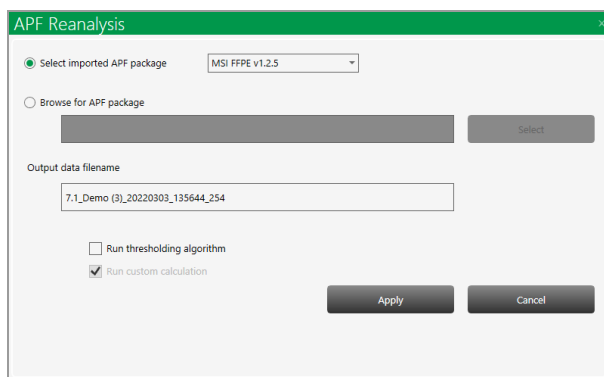
You can use the reanalysis tool if an APF was selected for the run.

To reanalyze the run file

1. In the Analysis module, click APF Tools > APF Reanalysis.



The APF Reanalysis window appears, with the APF package used in the run selected by default.



Note: If you select the Browse for APF package, you must select a package with the same plate configuration or you will receive an error message.

2. (Optional) Under Output and filename, enter a new file name.
3. (Optional) Select the run thresholding algorithm and click Apply.

Analysis Windows

In its Analysis module, QX Manager Software provides amplitude displays and data visualization charts in the Analysis module. For more information on each chart, see [Chapter 8, Data Analysis Methodology](#).

Amplitude Charts

Amplitude charts illustrate the droplets by degree of dye fluorescence. QX Manager Software displays fluorescence for positive and negative targets identified with FAM/EvaGreen®, HEX/VIC, Cy5, Cy5.5, ROX, and ATTO 590 fluorophores in the following analysis windows:

- 1D Amplitude
- 2D Amplitude
- 3D Amplitude

Data Visualization Charts

Data visualization charts show calculations based on positive droplet concentrations. QX Manager Software counts the number of positive and negative droplets for each fluorophore in each sample, and then fits the fraction of positive droplets to a Poisson algorithm to determine the starting concentration of the target DNA molecule in units of copies/μL input in the following analysis windows:

- Concentration
- Copy Number
- Ratio
- Fractional Abundance

When you open the analysis data file in QX Manager Software and select one or more wells in the grid, the software plots each calculated Poisson distribution average as a data point in the grid. You can pause on the data point to see the high, median, and low numbers.

Note: You can display the data using a confidence level of either 68% or 95% (default setting).

Event Counts

Event counts record the number of droplets in which the target was found (positive) and was not found (negative). The Event Counts tab shows a bar chart view of the number of positive, negative, and total droplets for each of the targets in each of the selected wells.

Plate View Windows

In the QX Manager Software Analysis module you can open the following windows that contain data on all wells in the run:

- **Plate Editor (analysis view)** — Displays the experiment type, sample names, sample types, supermixes, and assay type for each well in the run (similar to the Template Setup Plate Editor)
- **Plate View** — Displays concentration results in a text format for each processed well
- **2D Plate View** — Displays a two-dimensional amplitude chart for each processed well

Plate Editor — Analysis View

You can use the Plate Editor tab in the Analysis module to select wells, and then view or change the experiment parameters. Similar to other Plate Editor displays, the Analysis Plate Editor displays the experiment information for each well in the run.

Wells you select in the Analysis Plate Editor determine the data initially displayed in other analysis windows.

The screenshot displays the 'Plate Editor (analysis view)' window. On the left is a sidebar with navigation options: Dashboard, Plate Editor (selected), Plate View, Plate 2D, 1D Amplitude, 2D Amplitude, 3D Amplitude, Concentration, Copy Number, Ratio, Fractional Abundance, Event Counts, Data Table, Run Information, and Reports. The main area shows a 96-well plate grid. Columns 1-6 are hatched, columns 7-12 contain numerical data. Row C, column 8 is highlighted in orange. To the right of the grid is a configuration panel. It includes a 'Clear Selected Wells' button, an 'Experiment Type' dropdown (set to 'Direct Quantification (DQ)'), and four 'Sample Description' fields. Below these are 'Sample Type' (set to 'Unknown') and 'Assay Type' (set to 'Single Target per Channel') dropdowns. The 'Target Info' section contains a table with 6 rows and 6 columns (Target Name, Target Type, Signal Ch1, Signal Ch2, Signal Ch3, Signal Ch4, Signal Ch5). The bottom of the panel has 'Well Notes' and 'Plate Notes' text areas with an 'Apply' button.

Note: If an APF was selected for the run, fields that were automatically populated from the APF are grayed out.

Plate View

The Plate View tab provides, within the plate layout grid, a text view of the calculated concentration for each target in each well processed in the run. For information on the concentration results in chart format, see [Concentration on page 142](#).

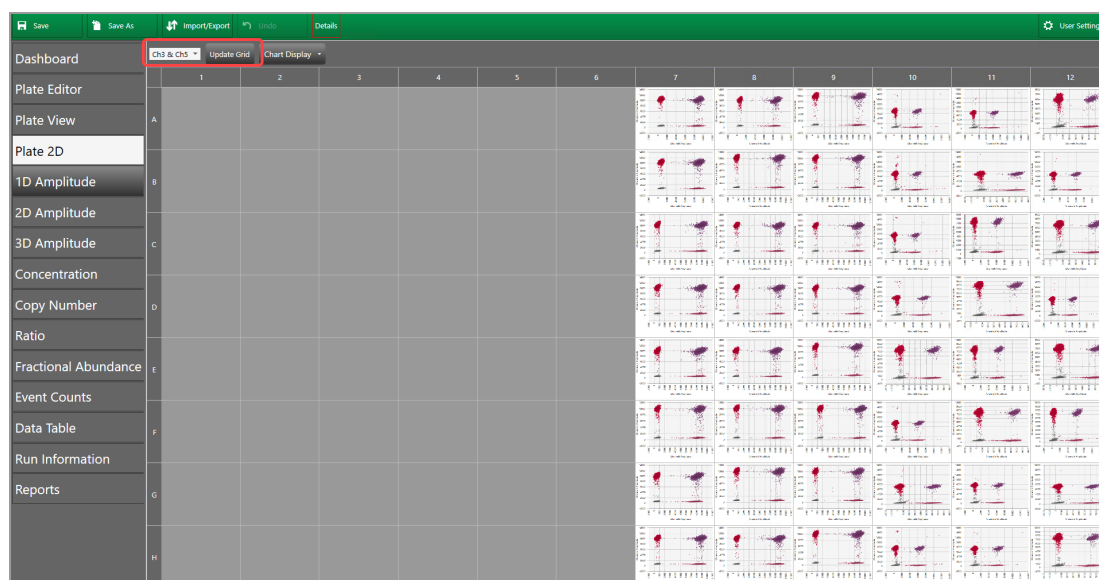
Save Save As Import/Export Undo Details User Settings												
	1	2	3	4	5	6	7	8	9	10	11	12
A	DQ						DQ		DQ		DQ	
	6 Color Check						6 Color Check		6 Color Check		6 Color Check	
	871.01	951.07	951.07	951.07	951.07	951.07	951.07	951.07	951.07	951.07	951.07	951.07
	1015.5	1015.5	1015.5	1015.5	1015.5	1015.5	1015.5	1015.5	1015.5	1015.5	1015.5	1015.5
B	DQ						DQ		DQ		DQ	
	6 Color Check						6 Color Check		6 Color Check		6 Color Check	
	1020.5	1020.5	1020.5	1020.5	1020.5	1020.5	1020.5	1020.5	1020.5	1020.5	1020.5	1020.5
	1020.5	1020.5	1020.5	1020.5	1020.5	1020.5	1020.5	1020.5	1020.5	1020.5	1020.5	1020.5
C	DQ						DQ		DQ		DQ	
	6 Color Check						6 Color Check		6 Color Check		6 Color Check	
	1020.5	1020.5	1020.5	1020.5	1020.5	1020.5	1020.5	1020.5	1020.5	1020.5	1020.5	1020.5
	1020.5	1020.5	1020.5	1020.5	1020.5	1020.5	1020.5	1020.5	1020.5	1020.5	1020.5	1020.5
D	DQ						DQ		DQ		DQ	
	6 Color Check						6 Color Check		6 Color Check		6 Color Check	
	1020.5	1020.5	1020.5	1020.5	1020.5	1020.5	1020.5	1020.5	1020.5	1020.5	1020.5	1020.5
	1020.5	1020.5	1020.5	1020.5	1020.5	1020.5	1020.5	1020.5	1020.5	1020.5	1020.5	1020.5
E	DQ						DQ		DQ		DQ	
	6 Color Check						6 Color Check		6 Color Check		6 Color Check	
	1020.5	1020.5	1020.5	1020.5	1020.5	1020.5	1020.5	1020.5	1020.5	1020.5	1020.5	1020.5
	1020.5	1020.5	1020.5	1020.5	1020.5	1020.5	1020.5	1020.5	1020.5	1020.5	1020.5	1020.5
F	DQ						DQ		DQ		DQ	
	6 Color Check						6 Color Check		6 Color Check		6 Color Check	
	1020.5	1020.5	1020.5	1020.5	1020.5	1020.5	1020.5	1020.5	1020.5	1020.5	1020.5	1020.5
	1020.5	1020.5	1020.5	1020.5	1020.5	1020.5	1020.5	1020.5	1020.5	1020.5	1020.5	1020.5

2D Plate View

To facilitate comparisons of amplitude in the processed wells, the Plate 2D tab provides a two-dimensional amplitude view in a separate chart for each well in a single display. To see a larger representation of a well, select the well in the 2D Amplitude view.

To view different color arrays

- Select the channel combination from the dropdown on the left, and then click Update Grid.



Data Windows

Important: For information on the APF Results table, see [APF Results Dashboard on page 107](#).

QX Manager Software provides the following table views for your well and run data:

- **Data Table** — available from the main tabs on the left, shows your configured experiment parameters, plus all calculations from the run
Note: You can display the data in individual well format, merged format, or both.
- **Plate Well Data Table** — available in plate setup windows (including the Analysis module Plate Editor), shows only your configuration information for each individual well
- **Analysis Well Data Table** — available in the Analysis module, shows a subset of configuration and run data for the wells selected in the Well Selector

Data Table Tab

The Data Table window contains columns of data generated for each target in each well, as droplet clusters are identified through thresholding or manual clustering tools.

Save Save As Import/Export Undo User Settings												
Dashboard	Show Both Merged Individual											
Plate Editor	Well Data											
Plate View	Well	SampleDesc1	Target	Conc(copies/ μ L)	Status	Status Reason	Experiment	SampleType	TargetType	Supermix	DyeName(s)	Copies/20 μ LWell
Plate 2D	E01	NTC/1cpd Ch2	HSV-1	No Call	CHECK	Automatic Analysis Unsuccessful	DQ	Unknown	Unknown	ddPCR S...	FAM	
1D Amplitude	E01	NTC/1cpd Ch2	B2M-...	1050	OK		DQ	Unknown	Unknown	ddPCR S...	HEX	21005
2D Amplitude	E02	0.125 cpd Ch...	HSV-1	121	OK		DQ	Unknown	Unknown	ddPCR S...	FAM	2420
Concentration	E02	0.125 cpd Ch...	B2M-...	1008	OK		DQ	Unknown	Unknown	ddPCR S...	HEX	20158
Copy Number	E03	0.25 cpd Ch1...	HSV-1	238	OK		DQ	Unknown	Unknown	ddPCR S...	FAM	4768
Ratio	E03	0.25 cpd Ch1...	B2M-...	1046	OK		DQ	Unknown	Unknown	ddPCR S...	HEX	20920
Event Counts	E04	0.5 cpd Ch1/...	HSV-1	482	OK		DQ	Unknown	Unknown	ddPCR S...	FAM	9636
Data Table	E04	0.5 cpd Ch1/...	B2M-...	1018	OK		DQ	Unknown	Unknown	ddPCR S...	HEX	20357
Run Information	E05	1 cpd Ch1/1c...	HSV-1	956	OK		DQ	Unknown	Unknown	ddPCR S...	FAM	19127
Reports	E05	1 cpd Ch1/1c...	B2M-...	1020	OK		DQ	Unknown	Unknown	ddPCR S...	HEX	20400
Fractional Abundance	E06	2 cpd Ch1/1c...	HSV-1	1939	OK		DQ	Unknown	Unknown	ddPCR S...	FAM	38786
	E06	2 cpd Ch1/1c...	B2M-...	997	OK		DQ	Unknown	Unknown	ddPCR S...	HEX	19938
	F01	NTC/1cpd Ch2	HSV-1	No Call	CHECK	Automatic Analysis Unsuccessful	DQ	Unknown	Unknown	ddPCR S...	FAM	
	F01	NTC/1cpd Ch2	B2M-...	1022	OK		DQ	Unknown	Unknown	ddPCR S...	HEX	20435
	F02	0.125 cpd Ch...	HSV-1	126	OK		DQ	Unknown	Unknown	ddPCR S...	FAM	2529
	F02	0.125 cpd Ch...	B2M-...	995	OK		DQ	Unknown	Unknown	ddPCR S...	HEX	19890
	F03	0.25 cpd Ch1...	HSV-1	250	OK		DQ	Unknown	Unknown	ddPCR S...	FAM	4991
	F03	0.25 cpd Ch1...	B2M-...	1008	OK		DQ	Unknown	Unknown	ddPCR S...	HEX	20156
	F04	0.5 cpd Ch1/...	HSV-1	475	OK		DQ	Unknown	Unknown	ddPCR S...	FAM	9492
	F04	0.5 cpd Ch1/...	B2M-...	1031	OK		DQ	Unknown	Unknown	ddPCR S...	HEX	20621
	F05	1 cpd Ch1/1c...	HSV-1	943	OK		DQ	Unknown	Unknown	ddPCR S...	FAM	18865
	F05	1 cpd Ch1/1c...	B2M-...	1048	OK		DQ	Unknown	Unknown	ddPCR S...	HEX	20962
	F06	2 cpd Ch1/1c...	HSV-1	No Call	CHECK	Droplet Count < 10000	DQ	Unknown	Unknown	ddPCR S...	FAM	
	F06	2 cpd Ch1/1c...	B2M-...	No Call	CHECK	Droplet Count < 10000	DQ	Unknown	Unknown	ddPCR S...	HEX	
	G01	NTC/1cpd Ch2	HSV-1	No Call	CHECK	Automatic Analysis Unsuccessful	DQ	Unknown	Unknown	ddPCR S...	FAM	
	G01	NTC/1cpd Ch2	B2M-...	1012	OK		DQ	Unknown	Unknown	ddPCR S...	HEX	20245

Data Table Column Definitions

Table 30 defines each field in the Data Table.

Table 30. Data Table columns

Column name	Content description
Well	The well location in the plate Note: The instrument accepts 96-well plates only.
Well Type	APF only — user can select Plate, Well, or Sample, a calculation basis for analysis data
Sample ID	APF only — a unique ID for a particular sample
Patient ID	APF only — a secondary ID to align with a first-level ID
Sample description 1 – 4	Words or phrases to describe the sample in each well Note: Only the sample description 1 data appears in plate layout views but you can pause on a well in the Plate Editor to display a tool tip containing the entries in all completed Sample description fields.
Target	Target name from Plate Editor tab
Conc(copies/ μ L)	Concentration of the target molecules recorded as copies per microliter
pg/ μ L	Mass concentration of DNA in the well in pg/ μ L
Status	Contains either Check, OK, Multi, or Manual <ul style="list-style-type: none"> ■ If Check, either the automatic analysis for the well failed or fewer than 10,000 droplets were counted in the well ■ If OK, enough droplets were counted to trigger automatic analysis ■ If Multi, the data was automatically analyzed as part of a multi-well selection ■ If Manual, the droplets were analyzed manually
Status Reason	Identifies the reason for the status (unsuccessful automatic analysis, not enough droplets counted, or droplets analyzed manually)

Table 30. Data Table columns, continued

Column name	Content description
Experiment	Experiment type selected for the well in the Plate Editor
SampleType	Sample type selected for the well in the Plate Editor
TargetType	Target type selected for each dye (FAM or EvaGreen®, HEX or VIC, Cy5, Cy5.5, ROX, and ATTO 590) in the Plate Editor
Supermix	Supermix selected for the well in Plate Editor
DyeName(s)	Dye assigned to channels 1, 2, 3, 4, 5, and 6, respectively, for the well in the Plate Editor
Copies/20µLWell	Concentration of the target normalized to a volume of 20 µL
TotalConfMax	For merged wells the high error bar for the target concentration of the combined wells at a 95% confidence interval
TotalConfMin	For merged wells the low error bar for the target concentration of the combined wells at a 95% confidence interval
PoissonConfMax	Maximum target concentration normalized for the high error bar of the droplet Poisson distribution for the 95% confidence interval
PoissonConfMin	Minimum target concentration normalized for the low error bar of the droplet Poisson distribution for the 95% confidence interval
Accepted Droplets	Total number of droplets accepted by quality algorithm
Positives	Number of droplets that contain the target
Negatives	Number of droplets that do not contain the target
Copies/µL linked molecules	Shows number of copies of a target that are present in overabundance versus the expected value in copies per µL
CNV	Copy number calculated for the target relative to the reference
TotalCNVMax	For merged wells, the high error bar for the copy number of the combined wells at a 95% confidence interval
TotalCNVMin	For merged wells, the low error bar for the copy number of the combined wells at a 95% confidence interval

Table 30. Data Table columns, continued

Column name	Content description
PoissonCNVMax	Maximum copy number normalized for the high error bar of the droplet Poisson distribution for the 95% confidence interval
PoissonCNVMin	Minimum copy number normalized for the low error bar of the droplet Poisson distribution for the 95% confidence interval
ReferenceCopies	Copy number identified for the reference target in the Plate Editor Default is 2, indicating 2 copies per diploid genome.
UnknownCopies	This field is currently unused in QX Manager Software
Threshold1	Threshold value of first threshold line from left to right and bottom to top
Threshold2	Threshold value of second threshold line from left to right and bottom to top
Threshold3	Threshold value of third threshold line from left to right and bottom to top
ThresholdSigmaAbove	Distance of the threshold from the mean of the negative cluster, as a multiple of the negative cluster standard deviation
ThresholdSigmaBelow	Distance of the threshold from the mean of the positive cluster, as a multiple of the positive cluster standard deviation
ReferenceUsed	Identifies which target was used as a reference
Ratio	The ratio of the target against the identified reference
TotalRatioMax	For merged wells the high error bar for the ratio of the unknown against the reference of the combined wells at a 95% confidence interval
TotalRatioMin	For merged wells the low error bar for the ratio of the unknown against the reference of the combined wells at a 95% confidence interval
PoissonRatioMax	Maximum ratio of the unknown against the reference normalized for the high error bar of the droplet Poisson distribution for the 95% confidence interval

Table 30. Data Table columns, continued

Column name	Content description
PoissonRatioMin	Minimum ratio of the unknown against the reference normalized for the low error bar of the droplet Poisson distribution for the 95% confidence interval
FractionalAbundance	Calculation of fractional abundance of this unknown target vs. the reference target
TotalFractionalAbundanceMax	For merged wells the high error bar for the fractional abundance of the combined wells at a 95% confidence interval
TotalFractionalAbundanceMin	For merged wells the low error bar for the fractional abundance of the combined wells at a 95% confidence interval
PoissonFractionalAbundanceMax	Maximum fractional abundance normalized for the high error bar of the droplet Poisson distribution for the 95% confidence interval
PoissonFractionalAbundanceMin	Minimum fractional abundance normalized for the low error bar of the droplet Poisson distribution for the 95% confidence interval
MeanAmplitudeOfPositives	Mean amplitude value of all droplets that contain the target
MeanAmplitudeOfNegatives	Mean amplitude value of all droplets that contain no target
MeanAmplitudeTotal	Mean amplitude value of all droplets
ExperimentComments	Data populates this field only from files created in QuantaSoft™ versions 1.4 – 1.7, if comments were added.
MergedWells	Identifies which wells were merged together
TotalConfidenceMax68	For merged wells the high error bar for the target concentration of the combined wells at a 68% confidence interval
TotalConfidenceMin68	For merged wells the low error bar for the target concentration of the combined wells at a 68% confidence interval
PoissonConfidenceMax68	Maximum target concentration normalized for the high error bar of the droplet Poisson distribution for the 68% confidence interval

Table 30. Data Table columns, continued

Column name	Content description
PoissonConfidenceMin68	Minimum target concentration normalized for the low error bar of the droplet Poisson distribution for the 68% confidence interval
TotalCNVMax68	For merged wells the high error bar for the copy number of the combined wells at a 68% confidence interval
TotalCNVMin68	For merged wells the low error bar for the copy number of the combined wells at a 68% confidence interval
PoissonCNVMax68	Maximum target concentration normalized for the high error bar of the droplet Poisson distribution for the 68% confidence interval
PoissonCNVMin68	Minimum target concentration normalized for the low error bar of the droplet Poisson distribution for the 68% confidence interval
TotalRatioMax68	For merged wells the high error bar for the ratio of the unknown against the reference of the combined wells at a 68% confidence interval
TotalRatioMin68	For merged wells the low error bar for the ratio of the unknown against the reference of the combined wells at a 68% confidence interval
PoissonRatioMax68	Maximum ratio of the unknown against the reference normalized for the high error bar of the droplet Poisson distribution for the 68% confidence interval
PoissonRatioMin68	Minimum ratio of the unknown against the reference normalized for the low error bar of the droplet Poisson distribution for the 68% confidence interval
TotalFractionalAbundanceMax68	For merged wells the high error bar for the fractional abundance of the combined wells at a 68% confidence interval
TotalFractionalAbundanceMin68	For merged wells the low error bar for the fractional abundance of the combined wells at a 68% confidence interval
PoissonFractionalAbundanceMax68	Maximum fractional abundance normalized for the high error bar of the droplet Poisson distribution for the 68% confidence interval

Table 30. Data Table columns, continued

Column name	Content description
PoissonFractionalAbundanceMin68	Minimum fractional abundance normalized for the low error bar of the droplet Poisson distribution for the 68% confidence interval plate list after correcting the issue
TiltCorrected	Indicates if tilt correction was applied to the well Note: Tilt correction rotates the angles from the double-negative cluster centroid to each droplet, such that the two single positive cluster centroids are orthogonal with the double-negative cluster centroid.
Ch1+Ch2+	Number of droplets that contain both channel 1 and channel 2 targets
Ch1+Ch2-	Number of droplets that contain just the channel 1 target
Ch1-Ch2+	Number of droplets that contain just the channel 2 target
Ch1-Ch2-	Number of droplets that contain neither channel 1 nor channel 2 targets
Ch3+Ch4+	Number of droplets that contain both channel 3 and channel 4 targets
Ch3+Ch4-	Number of droplets that contain just the channel 3 target
Ch3-Ch4+	Number of droplets that contain just the channel 4 target
Ch3-Ch4-	Number of droplets that contain neither channel 3 nor channel 4 targets
Ch5+Ch6-	Number of droplets that contain just the channel 5 target
Ch5-Ch6+	Number of droplets that contain just the channel 6 target
Ch5-Ch6-	Number of droplets that contain neither channel 5 nor channel 6 targets
Ch5+Ch6+	Number of droplets that contain both channel 5 and channel 6 targets

Plate Editor Well Data Table

A well data table containing both configuration and run data appears in the analysis windows, next to the Well Selector. This table displays a subset of data columns for only the wells selected in the Plate Editor or Well Selector. [Table 31](#) defines each column in the consolidated table.

Table 31. Fields in the Analysis Well Data table

Column name	Content description
Well	The well location in the plate the sample is from
Perform Droplet Reading	Yes or No
Experiment Type	Experiment type from the Plate Editor
Sample description 1 – 4	The sample name used to identify the well in the Plate Editor tab
SampleType	Sample type from the Plate Editor
Supermix Name	Supermix from the Plate Editor
Assay type	Assay type selected in the Plate Editor
Target Name	Target Name from Plate Editor tab
TargetType	Target type assigned to the target in the Plate Editor
Signal Ch1	Selected fluorophore in Channel 1
Signal Ch2	Selected fluorophore in Channel 2
Signal Ch3	Selected fluorophore in Channel 3
Signal Ch4	Selected fluorophore in Channel 4
Signal Ch5	Selected fluorophore in Channel 5
Signal Ch6	Selected fluorophore in Channel 6
Reference Copies	Copy number identified for the reference target in the Plate Editor tab
Well Notes	Entries by the user
Plot?	If selected, target will be used to calculate the copy number for the well
RDQ Conversion Factor	The Residual DNA Quantification conversion factor for the well
	Note: RDQ is only available in Residual DNA Quantification experiments.

Viewing Run, Lot, and Audit Information

Use the Run Information window to view the information about the plate and run. You can also view or add post-run notes regarding the plate. In Premium Edition, you can also view lots used, the audit logs, and APF information..

Run Details	
Instrument Type:	QX200
Instrument Serial Number:	771801413
Plate Name:	N/A
Date of Run:	N/A
Run Started by:	globalstaylor2
Supermix Type:	dPCR Supermix for Probes (No dUTP)
Droplet Size:	Standard (0.85nL)
Number Of Wells Read:	56
Bottle Configuration:	Oil and Waste
Oil Name:	Probes
DR Elapsed Time:	N/A
Run Status:	N/A
Software Version:	1.7.4.0917
MCU Version:	N/A
DLL Version:	2.1.0.51
FPGA Version:	N/A
Fluidics Version:	N/A
Motion Version:	N/A
System Version:	65535
Calibration State:	Required

Lot Information				
Well	Lot Type	Catalog Number	Lot Number	Lot Expiry Date

If an APF was used in the run, then the Run Information windows displays the APF information on the right.

Run Details	
Instrument Type:	QX200
Instrument Serial Number:	867-5309
Plate Name:	71_Demo
Date of Run:	7/1/2021 11:36:07 AM
Run Started by:	BR860
Supermix Type:	dPCR Multiplex Supermix
Droplet Size:	Standard (0.85nL)
Number Of Wells Read:	48
Oil Name:	Probes
DR Elapsed Time:	9h 6m 1s
Run Status:	Success
Software Version:	2.0.0
MCU Version:	8
DLL Version:	2.0.0.462
FPGA Version:	16
Fluidics Version:	3
Motion Version:	10
System Version:	7
Calibration State:	OK

APF Information	
APF Name:	MSI FFPE v1.2.5
APF Version:	1.2.5
Custom Variables:	BAT25LGB-2.11 BAT25LGB-0.34 NR24LGB-2.38 NR24LGB-1.51 Mono27LGB-0.56 DoubleDropOffWFactor-0.5

Lot Information				
Lot Type	Catalog Number	Lot Number	Lot Expiry Date	Well

For information on displaying your used lots in the Run Information window, see [Showing Lots in Run Information Reports on page 122](#).

You can also view the Audit Log. For information, see [Viewing the Audit Log on page 123](#).

Showing Lots in Run Information Reports

After you have set up your lots, you can ensure they appear in the Run Information screen when you open a file for analysis.

To show the lots in Run Information

1. Click Add Plate, then click Configure Plate.
2. Select the Lot Selector tab.
3. For kit lots, consumables, and reagents
 - a. Select a tab.
 - a. Select a lot for the run.
 - b. Click Select Lots.
4. Continue setting up the plate until the Start Run button is enabled.
5. Click Start Run.
6. When the run is completed, open the analysis file.
7. Click the Run Information tab to see the used lots.

To show the lots in a report

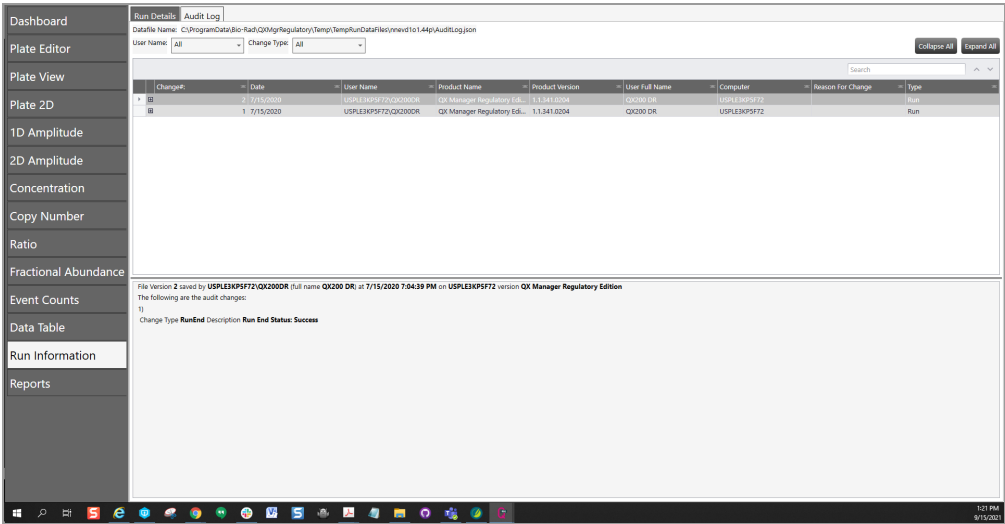
- Ensure the Lot Information check box is selected in the report elements before running the report.

Viewing the Audit Log

You can view the audit log entries for the data file using the Audit Log viewer found in the Run Information tab in the Analysis module.

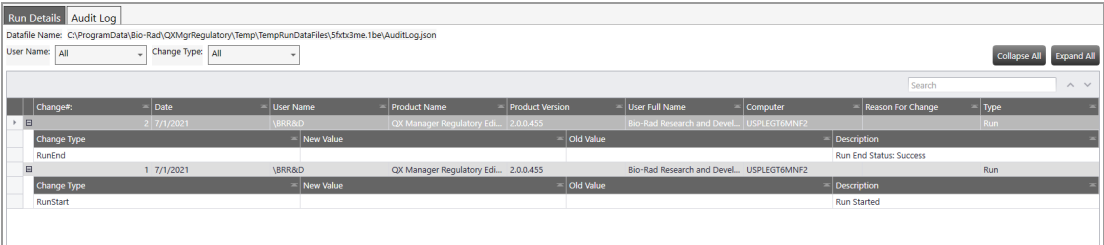
To view the Audit Log

- In the Analysis module, select the Run Information tab, and then select the Audit Log tab to display the audit log entries.



If the run used an APF, the APF package identifier appears in the gray banner above the window, but the columns are the same as the display for a standard run.

Tip: To expand the table to display more rows, click Expand All.



Chapter 8 Data Analysis Methodology

As summarized in [Chapter 7, Data Analysis Module Overview](#), the software presents analysis data in several different formats. When you open the data file in the Analysis module, you can view the data with the automatic analysis thresholds and calculations or you can choose different options to recalculate, replot, or recolor the results.

Data is shown in the following formats:

- Amplitude, which shows each droplet in each well in a scatter plot, in views
- Data visualizations, which show estimated target molecule concentration, ratio, and fractional abundance of one target against another target based on selected error model and confidence interval

Note: Copy number variation is also calculated if the user ran a CNV experiment.

- Event counts, which are precise counts of positive and negative droplets for each target in each well

Amplitude Plot Analysis Options

QX Manager Software provides three amplitude plot displays in which you can view the data with automatic thresholding where applicable, or choose different options to replot or recolor the results.

In the 1D amplitude and 2D amplitude plots, you can

- Show well calculations

Note: The 1D amplitude plot shows the data for each selected well in its own section, while the 2D amplitude plot shows all selected wells together.

- Modify automatic thresholds calculated by QX Manager Software

Important: If the data file contains calculations defined in an APF, they are displayed in the top-right table of the APF Results dashboard. If you manually change thresholds in the amplitude displays, those calculations are affected and the original data in the table is overwritten.

- View data in a heat map
- View histograms (1D amplitude tab only)
- Re-apply automatic analysis
- Apply tilt correction to automatic analysis

In the 3D amplitude plot, you can

- Rotate the plot to gain a better view of the droplets in 3D orientation
- View thresholds previously set in 1D or 2D amplitude display
- View 2D data behind 3D droplets in clusters or heat maps

Table 32 explains the analysis options that are available in both 1D amplitude and 2D amplitude plots.

Table 32. 1D and 2D amplitude tool options




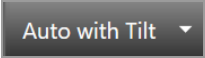

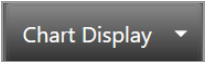
Icon	Mode	Description
	View mode	Data appears with the thresholds or clusters assigned by QX Manager Software. When this icon is selected, the view is locked and you cannot modify the thresholds.
	Heat map	Shows the distribution of droplets, with higher density of droplets shown as hot.
	Automatic analysis	Disables tilt correction and automatically reanalyzes the data.
	Automatic analysis with tilt correction	Enables tilt correction and automatically reanalyzes the data. Note: Tilt correction rotates the angles from the double-negative cluster centroid to each droplet, such that the two single positive cluster centroids are orthogonal with the double-negative cluster centroid.
	Reset	Resets the data to the original automatically analyzed data.
	Chart display options	Choose whether to display axis with channel number or fluorophore, and change the size of axis markers, data labels, and axis labels.

Table 33 explains the analysis options available only for 1D amplitude plots.

Table 33. 1D amplitude tool options











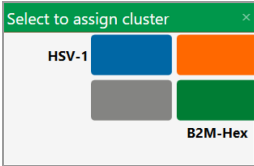
Icon	Mode	Description
	Automatic thresholding	<p>Individual Wells: Clusters are generated for each selected well separately.</p> <p>Combined Wells: Droplets from all selected wells are combined, and clusters are generated on that combined set.</p> <p>Positive Control Wells: Thresholds are generated for each channel based on any positive controls currently selected, and applies those thresholds to all currently selected wells.</p> <p>Note: If you change the thresholds manually, you can click Reset to return to the automatic thresholds.</p>
	Threshold, single well	For a single well or multiple wells, the user can manually input thresholding values in the 1D amplitude plot.
	Threshold, multiple wells	<p>You can click anywhere in the plot to apply threshold lines to the data.</p> <p>Important: Thresholding options are disabled if an APF was used in the run, and was configured to disallow manual data file analysis.</p> <p>To adjust the threshold values</p> <ul style="list-style-type: none"> ■ Drag the lines horizontally in the plot area ■ Enter a new threshold value in the boxes at the end of each threshold line ■ Click the up and down arrows to incrementally adjust the numbers
	Threshold, single well SD	For a single well or multiple wells, QX Manager Software automatically calculates and populates thresholding values with standard deviations (SD).
	Threshold, multiple wells SD	<p>Important: SD thresholds are not recommended for RMD, since these clusters are not typically located at right angles to each other.</p> <p>Note: The SD thresholds are set at a limit that is less than the mean number of positive targets, and more than the mean number of negative targets. Starting from where the user clicks in the plot, SD locates the threshold between the positive and negative clusters such that the classification error is minimized.</p>
	Show histogram	Hides or displays histogram plots for 1D amplitude

Table 34 explains the analysis options available only for 2D amplitude plots.

Table 34. 2D amplitude tool options

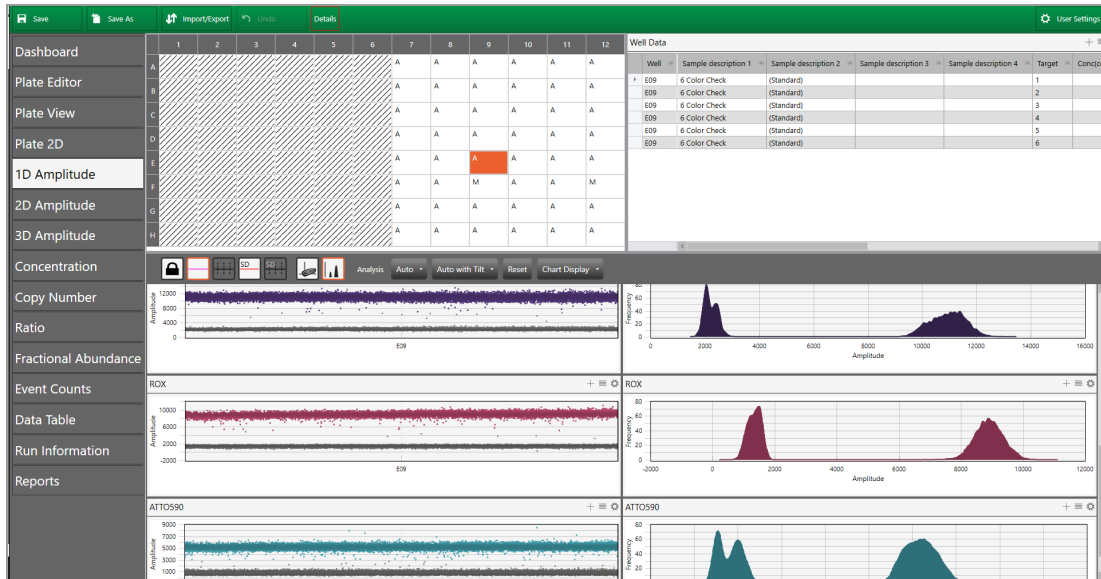
Icon	Mode	Description
	Automatic thresholding	<p>Individual Wells: Clusters are generated for each selected well separately.</p> <p>Combined Wells: Droplets from all selected wells are combined, and clusters are generated on that combined set.</p> <p>Positive Control Wells: The software generates thresholds for each channel based on any positive controls currently selected, and applies those thresholds to all currently selected wells.</p> <p>Note: If you change the thresholds manually, you can click Reset to return to the automatic thresholds.</p>
	Threshold line	<p>Use the Threshold line icons to establish thresholds across a pair of channels, dividing droplets into distinct clusters separated by different colors.</p> <p>You can click anywhere in the plot to apply threshold lines to the data.</p> <p>To adjust the threshold values, you can</p> <ul style="list-style-type: none"> ■ Drag the lines horizontally or vertically in the plot area ■ Enter a new threshold value in the boxes at the end of each threshold line ■ Click the up and down arrows to incrementally adjust the numbers
	Threshold line SD	<p>QX Manager Software automatically calculates and populates threshold line values with standard deviations (SD).</p> <p>Important: SD thresholds are not recommended for RMD, since these clusters are not typically located at right angles to each other.</p> <p>Note: The SD thresholds are set at a limit that is less than the mean number of positive targets, and more than the mean number of negative targets. Starting from where the user clicks in the plot, SD locates the threshold between the positive and negative clusters such that the classification error is minimized.</p>

Icon	Mode	Description
	Threshold cluster square	Use the Threshold Cluster icons to establish thresholds across a pair of channels, dividing droplets into distinct clusters separated by different colors.
	Threshold cluster circle	To classify your droplets, you can
	Threshold cluster freeform	<div><div>■ Click any of the three Threshold Cluster Mode buttons (square, circle, or free form)</div><div>■ Draw the chosen shape around the cluster of droplets</div></div> <p>Note: When using the cluster mode tools to draw a shape around a cluster of droplets, a dialog box with color selections by target appears in the window to help you select the correct target combination.</p> <div></div> <p>Target names are shown along the two axes based on the signal values assigned during plate setup. Click the colored box that best represents the location of the cluster of droplets selected to automatically classify the droplets as positive or negative for each given target.</p> <p>Note: The Cluster dialog box display varies according to the plex mode.</p>

1D Amplitude

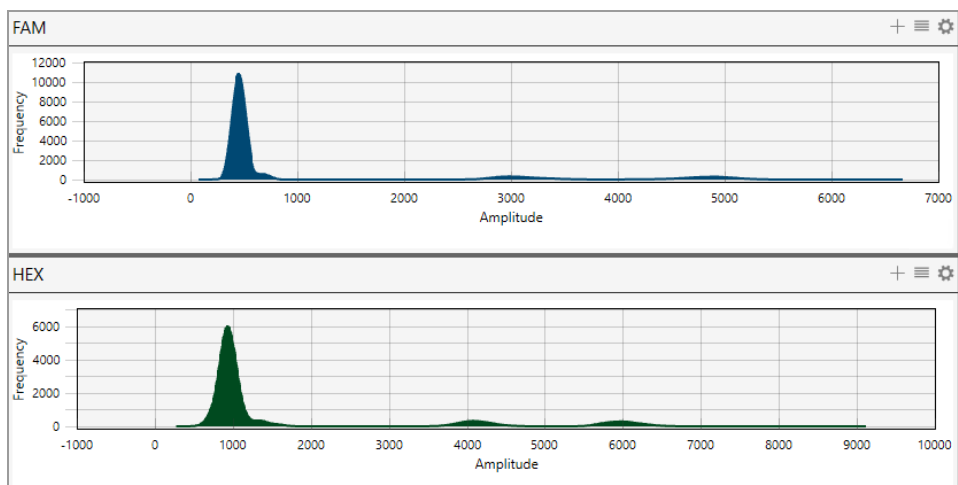
Use the 1D Amplitude analysis window to view droplet amplitudes for each channel, in selection order, for each well selected in the Well Selector.

The following graphic shows six colors in standard-size droplets.



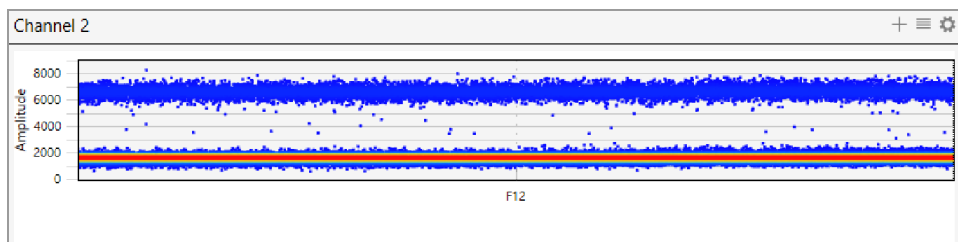
The 1D Amplitude plots illustrate

- All accepted droplets, with a visual representation of the positive and negative droplets in each well
 - Positive droplets appear in color, while negative droplets appear in gray.
 - The Well Selector displays an A in each well where QX Manager Software automatically thresholded the droplet data. An M appears in any well in which you manually changed the threshold.
- Amplitude in a histogram plot

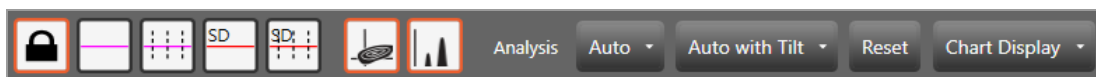


Note: The Histogram icon is enabled by default so a Histogram plot appears automatically for each fluorophore (channel).

- Amplitude in a Heat Map view



When you select the 1D Amplitude tab, the toolbar options that allow you to change thresholds and show or hide the heat map and histogram appear above the plots.



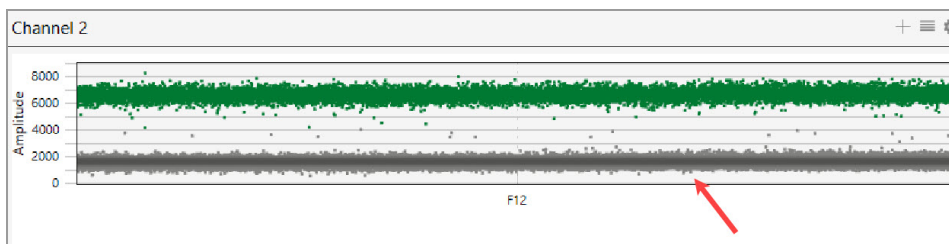
Note: For detailed information on each toolbar option, see [Amplitude Plot Analysis Options on page 126](#).

Default color assignments for 1D amplitude plots are

Positive FAM	=	Blue
Positive HEX	=	Green
Positive Cy5	=	Red
Positive Cy5.5	=	Violet
Positive ROX	=	Maroon
Positive ATTO 590	=	Aqua
Negative	=	Gray


The images in this section display data from a direct quantification experiment involving six targets:

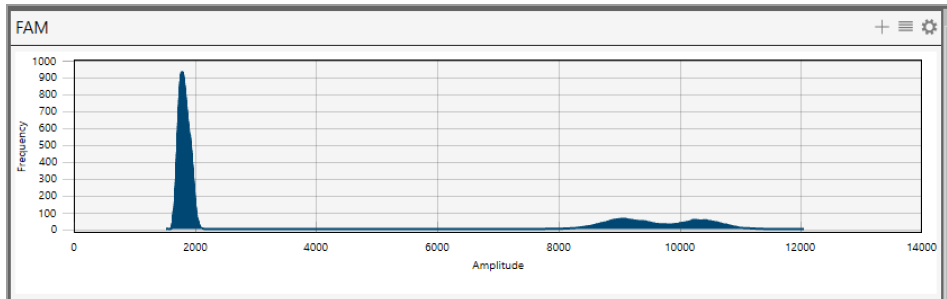
Negative droplets appear in grayscale in each plot.




For experiments where enough droplets are recognized, QX Manager Software automatically configures an amplitude threshold, which distinguishes between positive and negative droplets.

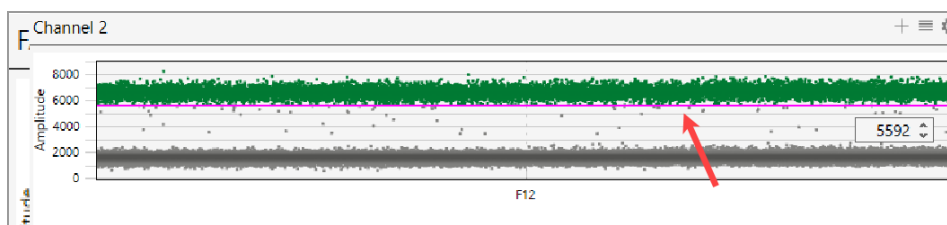
Note: Well data must meet certain quality metrics (minimum of 10,000 droplets) before the software will automatically calculate a threshold above which droplets are considered positive. You can use the manual thresholding tools to ensure correct quantification of positive and negative droplets.

By default, the Histogram icon () is enabled so the Histogram plot automatically appears with the amplitude plot.



To manually adjust the thresholds

1. Select one or more wells.
2. Click any of the threshold icons () to set manual thresholds:
 - Regular threshold for a single well; when multiple wells are selected, you can set a different threshold for each well
 - Regular threshold for multiple wells, where you can set one threshold for all selected wells
 - Standard deviation threshold for a single well; when multiple wells are selected, you can set a different threshold for each well
 - Standard deviation for multiple wells, where you can set one threshold for all selected wells
3. Click on the location in the plot where you want to draw the new threshold.
4. When the line appears, use the up or down arrows, or click and drag the threshold, to adjust the location.



QX Manager Software displays the manual threshold and changes the color of the droplets that are now above or below the threshold.

Note: You can also manually enter a threshold number.

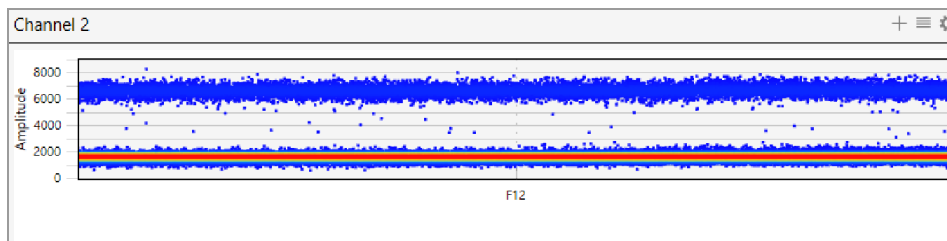
When you manually modify a threshold, the well selector displays an M for manual instead of the A for Automatic.

	A	A	A	A	A	A
	A	A	A	A	A	A
	A	A	A	A	A	M
	A	A	A	A	A	A

5. (Optional) To reset to the automatic threshold, click Auto.
6. (Optional) To adjust the automatic threshold to consider tilt correction, click Auto with Tilt.
7. (Optional) To reset everything to the original display, click Reset.

To view a heat map representation of the plot data

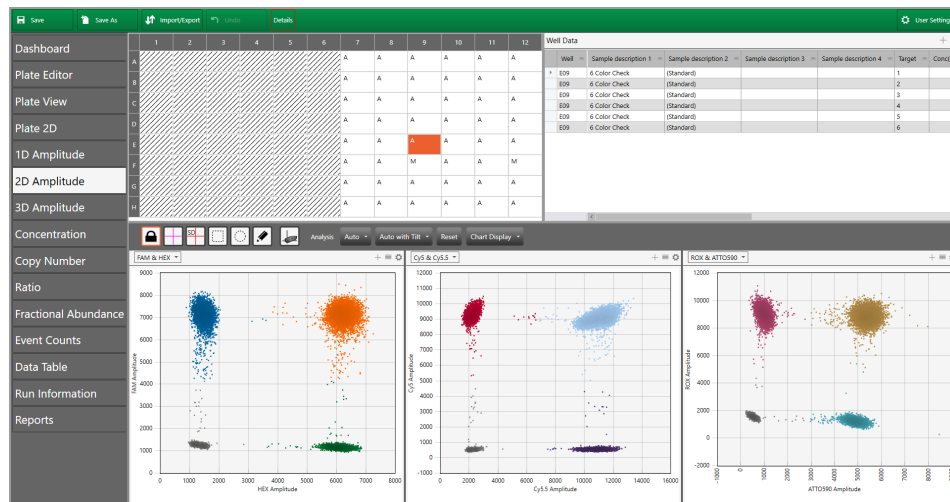
- Click the heat map () icon.



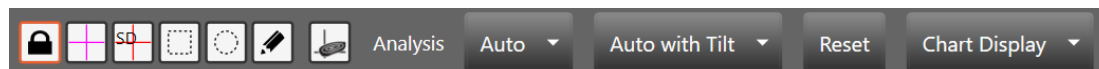
2D Amplitude

For the wells you select in the analysis view of the Plate Editor, the 2D Amplitude window illustrates fluorescence for six targets. The images in this section display data from an experiment involving six targets. Each plot shows individual cluster representations for first and second target positive droplets, droplets containing both targets, and droplets containing neither target.

The following graphic shows six colors in standard-size droplets.



When you select the 2D Amplitude tab, the toolbar options allow you to change thresholds, manually cluster wells, and show or hide the heat map above the plots.



For detailed information on each toolbar option, see [Amplitude Plot Analysis Options on page 126](#).

Default color assignments for 2D amplitude plots are:


Positive FAM	=	Blue
Positive HEX/VIC	=	Green
Positive FAM and HEX/VIC	=	Orange (may appear brown on some monitors)
Positive Cy5	=	Red
Positive Cy5.5	=	Violet
Positive Cy5 and Cy5.5	=	Light Blue
Positive ROX	=	Maroon
Positive ATTO 590	=	Gold
Positive ROX and ATTO 590	=	Aqua
Double negative	=	Gray

Note: Well data must meet certain quality metrics (minimum of 10,000 droplets) before QX Manager Software will automatically calculate a threshold above which droplets are considered positive. You can use the manual thresholding tools to ensure correct quantification of positive and negative droplets.

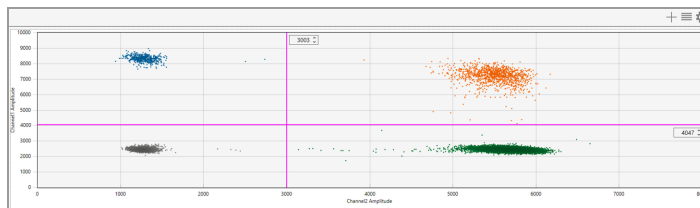
To use the 2D amplitude tools




1. Open your data file in the Analysis module and select the 2D Amplitude tab.
2. Choose a threshold line or cluster option.

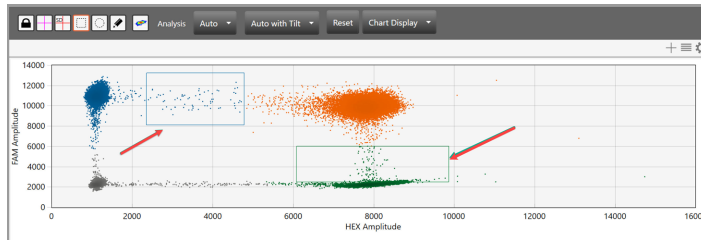
Note: Use the cluster option when the data is not orthogonal and the threshold lines do not adequately divide the populations of droplets.

- To adjust threshold lines, select a threshold line type () and then click in the plot to apply crosshair lines. Use any of the following methods:

- ☐ Drag the lines horizontally or vertically in the plot area.
- ☐ Enter a new threshold value in the boxes at the end of each threshold line.
- ☐ Tap the up and down arrows to incrementally adjust the numbers.



- To adjust thresholds using the cluster option, select a cluster type (  ) and draw the chosen shape around the cluster of droplets to classify.



3. (Optional) To reset to the automatic threshold, click Auto.
4. (Optional) To adjust the automatic threshold to consider tilt correction, click Auto with Tilt.
5. (Optional) To reset everything to the original display, click Reset.

Data Visualization Chart Analysis Options

QX Manager Software also provides chart displays where you can view the data from the original calculations, or choose different options to recalculate the results.



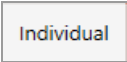

In charts reflecting Data Visualization distributions, you can

- Show individual or merged well data
- Change the error model and confidence level

Note: For event counts, you can also alter the view mode to see positive, negative, or all droplet counts.

[Table 35](#) explains the available options.

Table 35. Chart tool options for Data Visualization distribution charts

Icon	Mode	Description
	Show both	Shows the calculated total or percentage as a merged calculation for all selected wells and as an individual calculation for each selected well
	Show merged	Shows the calculated total or percentage as a combined number for all selected wells with the same parameters Note: Well setup must match for each well before the software can merge them.
	Show individual	Shows the calculated total or percentage as an individual calculation for each selected well
	X-axis well	Shows the well number on the x axis


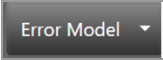
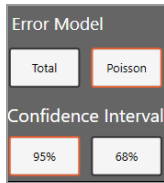
Icon	Mode	Description
	X-axis sample	<p>Shows a partial sample name on the x axis</p> <p>Note: You can select Well and Sample simultaneously.</p>
	Error model and confidence interval	<p>Select an error model and confidence level</p> <p>Note: The default choices are Poisson and 95%, to yield the highest confidence in the results.</p>  <p>Important: The software uses the following multipliers in its calculations:</p> <ul style="list-style-type: none">■ For 95% confidence interval, the sigma multiplier is 1.96■ For 68% confidence interval, the sigma multiplier is 1.0

Table 36 identifies the additional options available in the Event Count window. These options are available for event counts only.

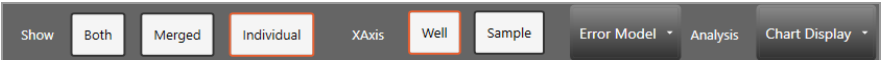
Table 36. Additional options for event counts

Icon	Mode	Description
<div>Positive</div>	View mode positive	Displays in a bar chart the number of positive droplets for each target in each well
<div>Negative</div>	View mode negative	Displays in a bar chart the number of negative droplets for each target in each well
<div>Total</div>	View mode total	Displays in a bar chart the number of total droplets in each well

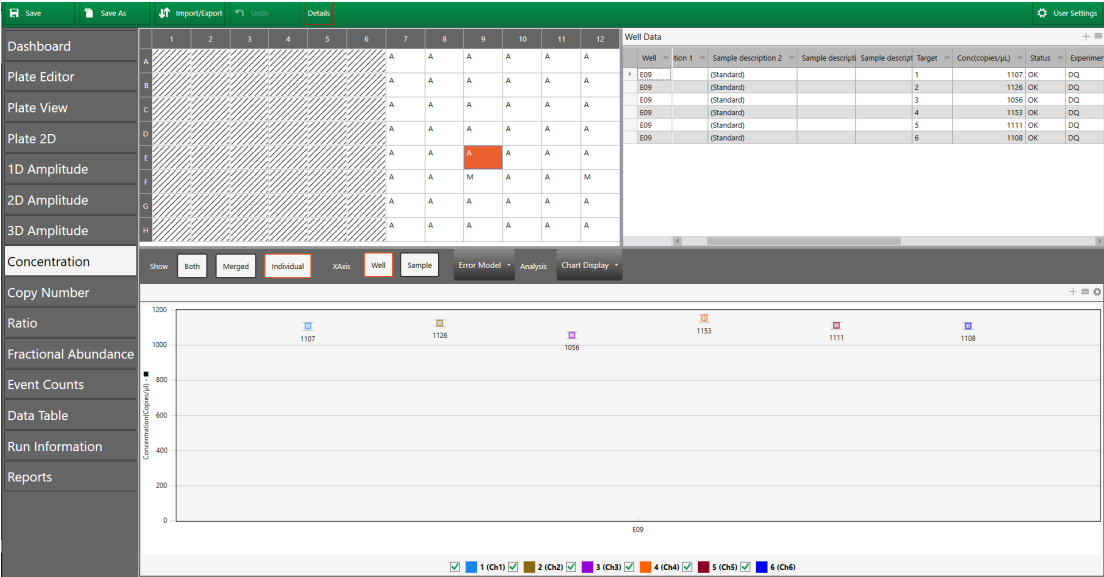
Concentration

Concentration measures the number of copies of the target molecules in each μl of sample based on the default settings. Use the toolbar to merge or separate the calculated results for each well, and to recalculate your results based on a different error model and confidence interval.

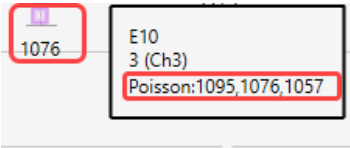
For information on toolbar options, see [Data Visualization Chart Analysis Options on page 139](#).



The following graphic shows the concentration for six colors in standard-size droplets.



QX Manager Software plots the averaged concentration for each target as a data point. Pause on a data point to see the high, median, and low numbers.



Concentration Calculation

The software uses the formula and variables shown below.

$$c = -\ln\left(\frac{N_{neg}}{N}\right) / V_{droplet}$$

Where:

-ln	Negative natural logarithm
$V_{droplet}$	Volume of droplet
N_{neg}	Number of negative droplets
N	Total number of droplets

Confidence Interval Calculation

Note: If you select a 68% confidence interval, the multiplier is changed from 1.96 to 1.00.

$$p = \frac{N_{neg}}{N}$$

$$\partial_{N_{neg}} = \sqrt{Np(1-p)}$$

$$N_{neg,min} = N_{neg} - 1.96 * \partial_{N_{neg}}$$

$$N_{neg,max} = N_{neg} + 1.96 * \partial_{N_{neg}}$$

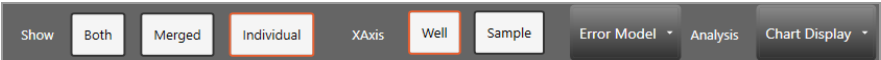
Where:

$V_{droplet}$	Volume of droplet
N_{neg}	Number of negative droplets
N	Total number of droplets
p	p-value (probability of finding the target in the sample)
∂	Delta

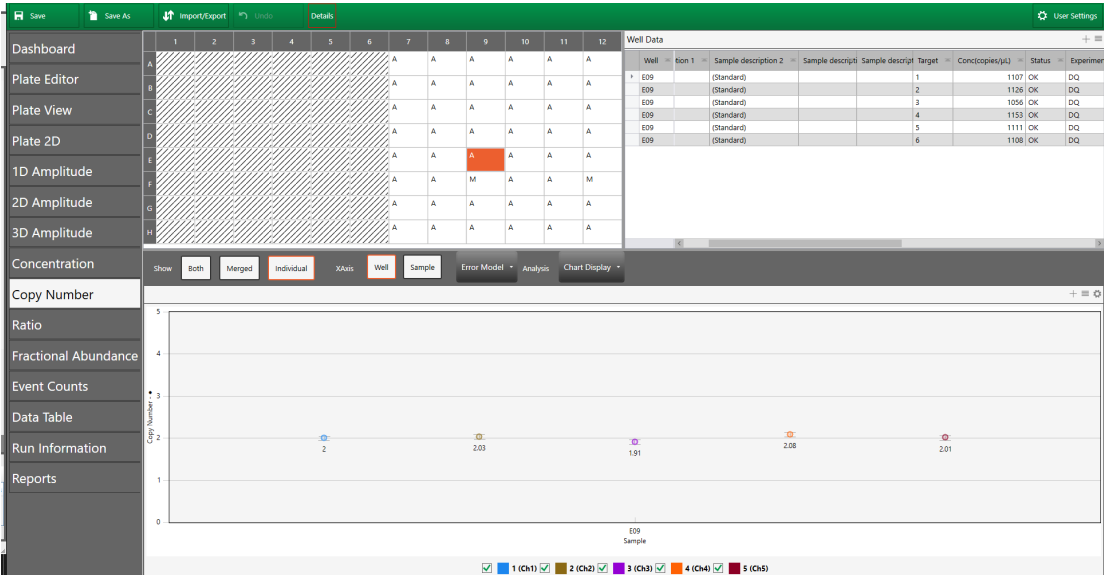
Copy Number

Important: To see copy number variation data, you must run a copy number variation (CNV) ddPCR experiment.

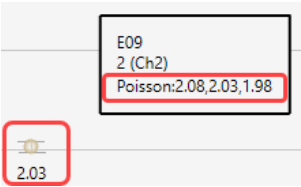
Copy number measures the count of the target gene, relative to the reference gene, to calculate the number gene targets per genome. Use the toolbar to merge or separate the calculated results for each well, and to recalculate your results based on a different error model and confidence interval. For information on toolbar options, see [Data Visualization Chart Analysis Options on page 139](#).



The following graphic shows copy number data for six colors in standard-size droplets.



QX Manager Software plots each copy number variation as a data point. Pause on a data point to see the high, median, and low numbers.



Copy Number Calculation

The software uses the formula and variables shown below.

$$CNV = \frac{A}{B} N_B$$

Where:

A	Target concentration
B	Reference concentration
N_B	Number of reference copies in genome (usually 2)

Confidence Interval Calculations

Note: If you specify the lesser confidence interval when you define your error model, the software uses 68% in its calculations instead of 95%.

$$CI_{CNV} = N_B \cdot \frac{A}{B} \sqrt{\frac{CI_A^2}{A^2} + \frac{CI_B^2}{B^2}}$$

$$(CNV_{low_95\%}, CNV_{hi_95\%}) = (CNV - (1/2)CI_{CNV}, CNV + (1/2)CI_{CNV})$$

Where:

A	Target concentration
B	Reference concentration
N_B	Number of reference copies in genome (usually 2)
CI_A	Poisson confidence interval of target (95% max - 95% min)
CI_B	Poisson confidence interval of reference (95% max - 95% min)

Notes:

Copy Number analysis involves determining the number of copies of a given target DNA sequence as compared to an invariant reference DNA sequence. Use the CNV analysis to determine structural variations, such as ploidy changes (addition or deletion of chromosomes), deletions (missing partial chromosomes) or duplications (repeated target sequences) in the genome.

The ability to discriminate between consecutive CN states has been a technical challenge in copy number assessment. Fundamentally, as CN state increases, the percentage difference in target genomic material between states decreases. For example, for a given target locus, a CN of 3 is 50% more abundant in concentration per genome than a CN of 2, while a CN of 5 is only 25% more abundant per genome than a CN of 4, and so on.

Due to the massive partitioning of the reaction across up to 20,000 droplets per well, and the absolute nature and precision of the ddPCR concentration measurement, ddPCR CNV analysis enables the quantitative discrimination required to resolve consecutive copy number states beyond CN 3. For example, a 20% difference in target concentration between a CN 5 versus CN 6 sample can be reproducibly resolved using ddPCR. This discrimination is both a function of concentration differences between consecutive CN states, as well as the amount of DNA assayed per well.

Ratio

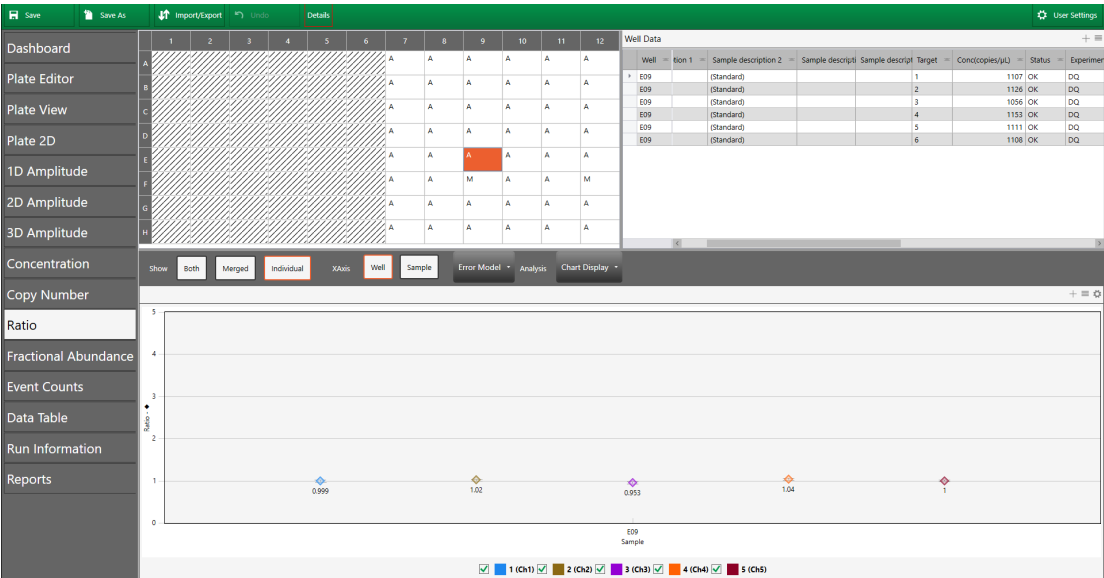
Ratio determines the quotient of two target molecules in each µl of sample based on the default settings.

T1:T2

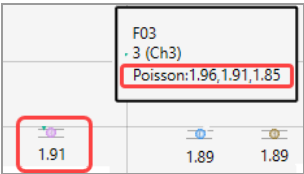
Use the toolbar to merge or separate the calculated results for each well, and to recalculate your results based on a different error model and confidence interval. For information on toolbar options, see [Data Visualization Chart Analysis Options on page 139](#).



The following graphic shows the ratio for standard-size droplets.



QX Manager Software plots each ratio as a data point. To see the confidence level numbers, pause on the data point.



Ratio Calculation

The software uses the formula and variables shown below.

$$r = \frac{A}{B}$$

Where:

A	Target concentration
B	Reference concentration

Note: If no reference is selected, the target with the highest dye concentration in the highest channel will automatically be considered as the reference. If multiple references are selected, the geometric mean is used.

Confidence Interval Calculations

Note: If you specify the lesser confidence interval when you define your error model, the software uses 68% in its calculations instead of 95%.

$$CI_r = \frac{A}{B} \sqrt{\frac{CI_A^2}{A^2} + \frac{CI_B^2}{B^2}}$$

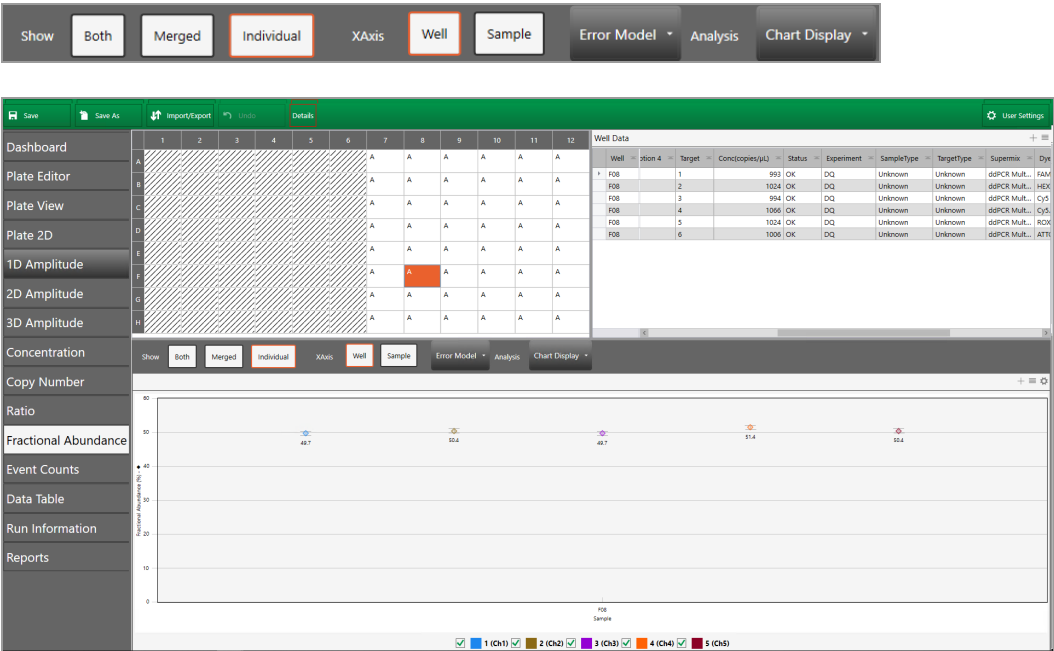
$$(r_{low_95\%}, r_{hi_95\%}) = (r - (1/2)CI_r, r + (1/2)CI_r)$$

Where:

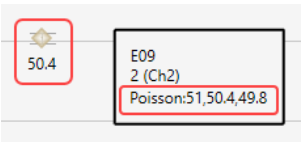
A	Channel 1 target concentration
B	Channel 2 target concentration
CI_A	Confidence interval of Channel 1 target (95% max - 95% min)
CI_B	Confidence interval of Channel 2 target (95% max - 95% min)

Fractional Abundance

Fractional Abundance determines the relative abundance of your targets in each µl of sample based on the default settings. Use the toolbar to merge or separate the calculated results for each well, and to recalculate your results based on a different error model and confidence interval. For information on toolbar options, see [Data Visualization Chart Analysis Options on page 139](#).



QX Manager Software plots the fractional abundance as a data point. To see the high and low confidence interval numbers, pause on the data point.



Fractional Abundance Calculation

The software uses the formula and variables shown below.

$$f = \frac{A}{A + B}$$

Where:

A	Target concentration
B	Reference concentration

Note: If no reference is selected, the target with the highest dye concentration in the highest channel will automatically be considered as the reference. If multiple references are selected, the geometric mean is used.

Confidence Interval Calculations

Note: If you specify the lesser confidence interval when you define your error model, the software uses 68% in its calculations instead of 95%.

$$CI_f = \frac{1}{(A + B)^2} \sqrt{B^2 CI_A^2 + A^2 CI_B^2}$$

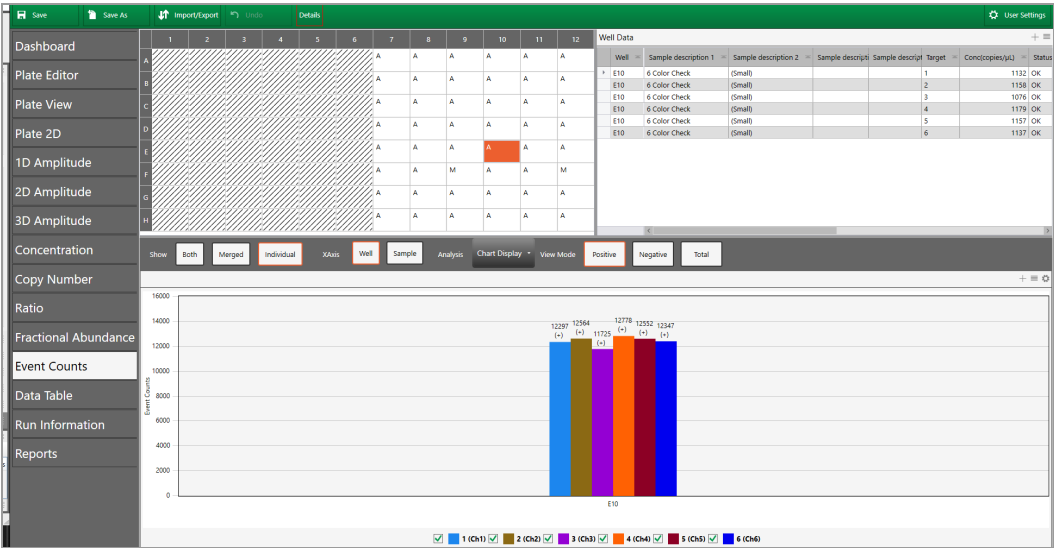
$$(f_{low_95\%}, f_{hi_95\%}) = (f - (1/2)CI_f, f + (1/2)CI_f)$$

Where:

A	Channel 1 target concentration
B	Channel 2 target concentration
CI_A	Confidence interval of Channel 1 target (95% max - 95% min)
CI_B	Confidence interval of Channel 2 target (95% max - 95% min)

Event Counts

The Event Counts tab shows a bar chart view of the number of positive and negative droplets for each of the targets in each of the selected wells.



Use the toolbar to change the display by combining or separating data for each well. You can also show data by well name or by sample name, and you can show any combination of positive, negative, or combined droplet counts.



Linkage Analysis

In ddPCR, physical linkage of two targets can occur in one of the following patterns:

- Tandem repeats of the same target sequence
- Proximity of two target sequences physically linked on the same piece of DNA

QX Manager Software uses the formulas and variables shown below.

- The following formula presents the linkage in copies per µL:

$$linkage = \ln(N) - \ln\left(n_{00} + n_{01} + n_{10} + \frac{n_{01}n_{10}}{n_{00}}\right) \left(\frac{1000}{droplet\ volume}\right)$$

- The following formula presents the linkage in copies per droplet.

$$linkage = \ln(N) - \ln\left(n_{00} + n_{01} + n_{10} + \frac{n_{01}n_{10}}{n_{00}}\right)$$

Where:

N	Total number of droplets
n ₀₀	Number of negative droplets for both channels
n ₀₁	Number of negative droplets in channel 1 and positive droplets in channel 2
n ₁₀	Number of positive droplets in channel 1 and negative droplets in channel 2
p	p-value (calculated probability)
∂	Delta

Where:

Confidence Interval Calculation

$$CI_{linkage} = (linkage - (1.96)\sigma_{linkage}, linkage + (1.96)\sigma_{linkage})$$

Where:

σ	Standard deviation
---	--------------------

Chapter 9 Lot and APF Management

From the Templates tab in the Premium Edition, you can add lots by lot number, catalog number, and expiration date to track your usage, and import preconfigured assay protocol files (APFs) that you can use to expedite configuration of your droplet reading runs.

Kit Lot	Consumables	Reagents	Lot Number	Catalog Number	Lot Expiry Date	Used
Supermix			1212121	1863027	10/31/2021	<input type="checkbox"/>
ddPCR Supermix for Probes			2323234	12005910	1/1/2022	<input type="checkbox"/>
ddPCR Multiplex Supermix			4545333	1863027	2/2/2022	<input type="checkbox"/>
ddPCR Supermix for Probes						<input type="checkbox"/>
ddPCR Supermix for Probes (No dUTP)						<input type="checkbox"/>

The following options are available:

- From the Lot Management tab, you can manage your inventory and track use of lots for test-specific kits, consumables, and reagents. You can select lots on a per well basis. If you selected an APF, lot fields are defined if they are required before you can start the run.
- From the APF Management tab, if you are assigned the Manage APFs user privilege, you can import assay packages that contain preconfigured plate and run settings. You can also modify variables in an APF and delete APFs.

Important: If you select the Show Expired and Uses Lots checkbox in the lower-left corner, lots that are expired or empty are displayed.

Managing Consumable Lots

If you are using a Bio-Rad droplet generation instrument, you can manage your inventory of consumables packs, cartridge, and pipet tips, as well as your plate and seal inventory.

To enter a consumables lot

1. Select the Template Setup icon.



2. Select Lot Management, and then select the Consumables tab.

The screenshot displays the QX Manager Premium Edition software interface. The top status bar shows the instrument model (QX200), IP address (867-5309), and calibration status (OK). It also includes indicators for runs in progress and available, and status for oil and waste (both OK). A disk space usage pie chart and a user profile icon are also visible.

The left sidebar contains a vertical menu of icons. The 'Template Setup' icon, which is a grid with a pencil, is highlighted. Below it are icons for Reports, Lot Management, APF Management, and a help/question mark icon.

The main window is divided into several tabs: Plate Setup, Consumables (selected), Reagents, Reports, QXDs Consumables Pack, Cartridges, Plates and Seals, and Pipet Tips. The 'Consumables' tab is active, showing a table with columns for Lot Number, Catalog Number, Lot Expiry Date, and Used. The table contains one entry with Lot Number 451327, Catalog Number 1814040, and Lot Expiry Date 12/23/2024. The 'Used' column has a checkbox that is currently unchecked.

At the bottom left of the main window, there is a checkbox labeled 'Show Expired & Used Lots' which is currently unchecked. The bottom right corner shows the version number 'Version 2.1.0'.

From the Consumables window, the following tabs appear above the lot information grid:

- QXDx Consumables Pack
- Cartridges
- Plates and Seals
- Pipet Tips

3. Select a tab.
4. (Optional) To display expired or used lots, select the checkbox at the bottom-left side of the window.
5. Click in each field and enter the corresponding information as follows:
 - Lot Number
 - Catalog Number
 - Lot Expiry Date
6. If the consumable was used in a run, select the Used checkbox.
7. Repeat until all lots are added, and then click Save.

Managing Reagent Lots

You can manage your inventory of supermixes, droplet generation oil, droplet reader oil, buffers and assays. You can purchase ready-to-use assays, such as the SARS-CoV-ddPCR kit, or, if you know your assay sequences, you can request custom ddPCR assays from Bio-Rad.

To enter a reagent lot

1. Select the Template Setup icon.



2. Select Lot Management, and then select the Reagents tab.

Lot Number	Catalog Number	Lot Expiry Date	Used
885855	1863005	12/21/2023	<input type="checkbox"/>

3. From the Reagents window, select one of the following tabs in the Reagents window:
 - Supermix (appears by default)
 - Droplet Generator Oil
 - Droplet Reader Oil
 - Buffer Controls
 - Assays
4. (Optional) To display expired or used lots, select the checkbox in the lower-left corner.
5. Click in each of the following fields and enter the corresponding information:
 - Supermix (in Supermix grid only)
 - Lot Number
 - Catalog Number
 - Lot Expiry Date
6. If the consumable was used in a run, select the Used checkbox.
7. Repeat until all lots are added, and then click Save.

Managing Assay Protocol Files

An Assay Protocol File (APF) is a package containing preconfigured settings applied to a plate run. Along with allowing you to streamline the configurations of your droplet reading runs, APFs can be set to apply more rigor to quality controls and positive versus negative determinations in each well. APFs can specify any combination of settings for different wells.

Fields configured in the APF are not editable in the configuration windows. You can add wells if there is room on the plate, but they automatically inherit the settings specified in the APF. If the APF contains rules for validation that must be met before you start the run, then QX Manager Software, Premium Edition validates information or patterns you enter into enabled fields. If they do not match, an error message appears.

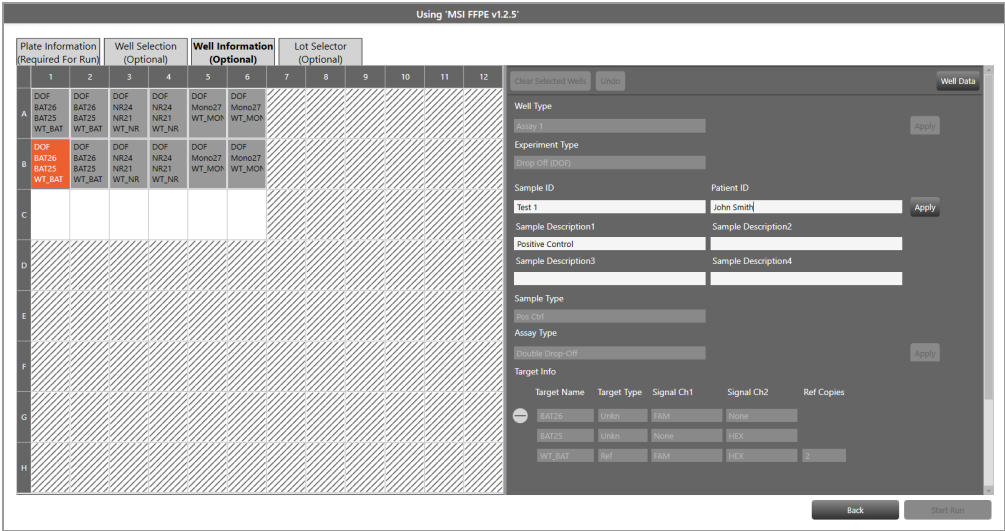
From the APF Management tab in the Templates window, you can

- Import APFs into QX Manager Software
- Modify variables defined in the APF
- Delete APFs

Important: You must be assigned the Manage APFs user privilege to upload, modify, or delete an APF. To create your own APF configurations, you can purchase the QX Designer companion application from Bio-Rad Laboratories, Inc.

Note the following:

- After an APF is imported, it is available for selection in the Add Plate window by users with Manager APFs, Run APFs or Only Run APFs user privileges.
- When an APF is selected in the Add Plate window, the APF name appears in the gray banner at the top of the window.
- When you click Configure Plate, the wells configured in the APF appear as shown in the following example.



For analysis information, see [APF Results Dashboard on page 107](#).

Importing Assay Protocol Files

APFs must be imported into the APF Packages window before they are available to use for a run.

Important: Only a user with the Manage APFs user privilege can import APFs.

To import an APF

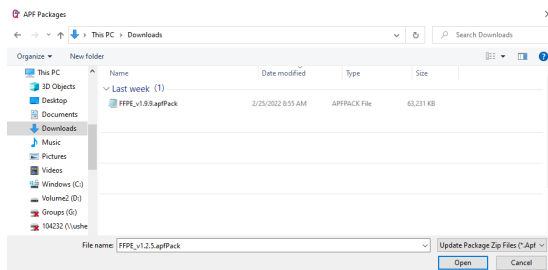
1. Select the Template Setup icon.



2. Select APF Management, and then click Import Package.

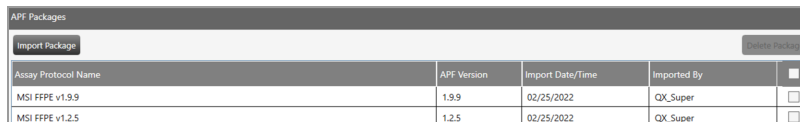


3. When the APF Packages dialog box appears, locate the APF to import.



4. Select the file and click Open.

QX Manager Software, Premium Edition imports the APF into the APF Packages window.



Important: You cannot import multiple versions of the same APF. Each APF is assigned a version number. When you import a newer version of the same APF, QX Manager Software will ask if you would like to upgrade your existing APF. If yes, the earlier version is overwritten.

Modifying Variables in an Assay Protocol File

When you select an APF under Assay Protocol name, the APF package details appear in the bottom-left corner. You can modify any or all variables for your specified targets, or you can restore the variables to their APF predefined values.

To modify a variable

1. Select the Template Setup icon.



2. Select APF Management, and then select the row for the applicable APF package to display the Variables grid.

Assay Protocol Name	APF Version	Import Date/Time	Imported By
MSI FFPE v1.9.9	1.9.9	02/25/2022	QX_Super
MSI FFPE v1.2.5	1.2.5	02/25/2022	QX_Super

Name	Original	Modified
BAT25LOB	2.11	2.11
BAT26LOB	0.34	0.34
NR21LOB	2.38	2.38
NR24LOB	1.51	1.51
Mono27LOB	0.56	0.56
DoubleDropoffWTFactor	0.5	0.5

3. Click in a field under Modified, and then enter the new variable value.
4. (Optional) To cancel your entries and restore the default variable values, click Reset.
5. To save your changes, click Save.

The modified values are saved, and will apply for all runs using this APF.

Deleting Assay Protocol Files

To delete an APF from the Assay Management window

1. Select the Template Setup icon.



2. Select APF Management, and then select the checkbox on the right for the APF to be deleted.

Plate Setup	APF Packages				
Reports	Import Package				
Lot Management					
APF Management	Assay Protocol Name	APF Version	Import Date/Time	Imported By	
	MSI FFPE v1.9.9	1.9.9	02/25/2022	QX_Super	<input type="checkbox"/>
	MSI FFPE v1.2.5	1.2.5	02/25/2022	QX_Super	<input checked="" type="checkbox"/>

3. Click Delete Package to remove the APF package from APF Management.

Chapter 10 Analysis Module Reporting

Use the Reporting function to create a variety of reports in the QX Manager Software Analysis module.

You can create reports on the fly from any analysis file or, if you need to generate specific reports on a regular basis, you can set up report templates for reuse.

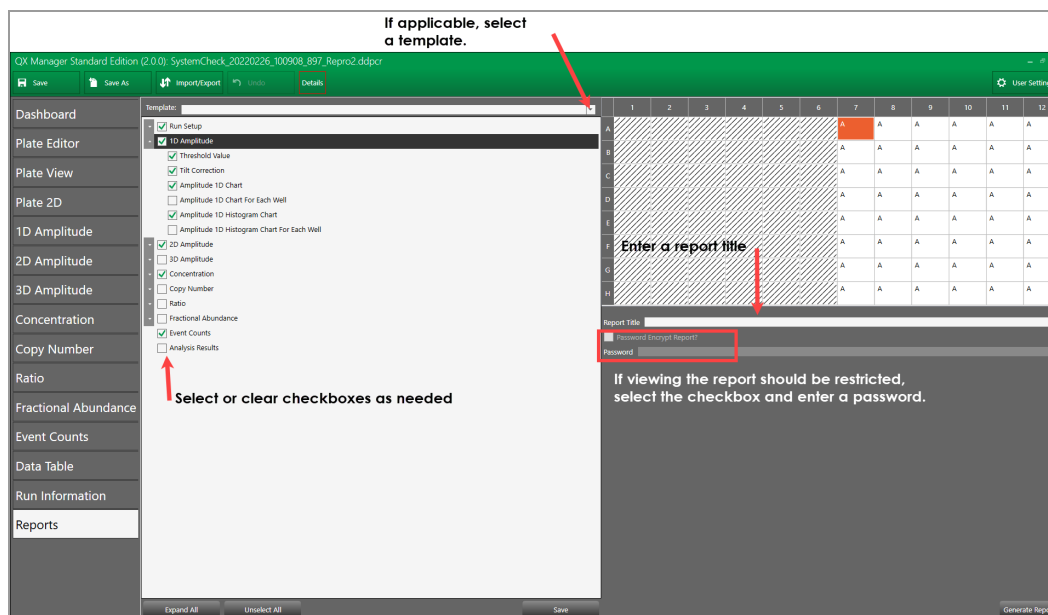
This section contains information on how to create analysis reports.

Creating Analysis Reports

You can create reports with or without a template. For information on creating templates for reports you generate often, see [Creating or Editing Report Templates on page 80](#).

To create a report

1. Select the Reports tab to display the Reports window.



2. Do one of the following:

- To create a report from a template, click the Template dropdown arrow and select a template from the list
- To create a custom report, click the arrows to expand reporting categories, and then select the checkbox for each report element to include, and clear checkboxes for elements to eliminate.

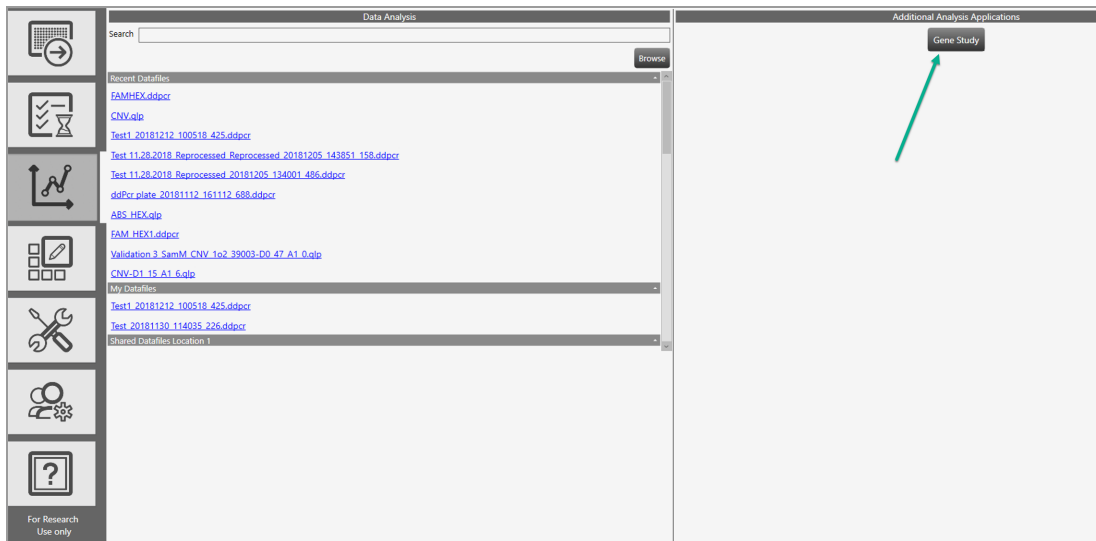
Note: Use the buttons at the bottom of the screen to expand or collapse all categories at once, or to select or clear all checkboxes at once. You can also drag and drop the section to rearrange the order of appearance in the report, and drag and drop items in the section to reorder them.



3. Enter a report title.
4. (Optional) To restrict viewing, select the Password Encrypt Report checkbox and then enter a password.
5. Click Generate Report to generate a PDF report.
6. To save the report, click Save.

Chapter 11 Gene Study Module

You access the Gene Study module from the Data Analysis window.

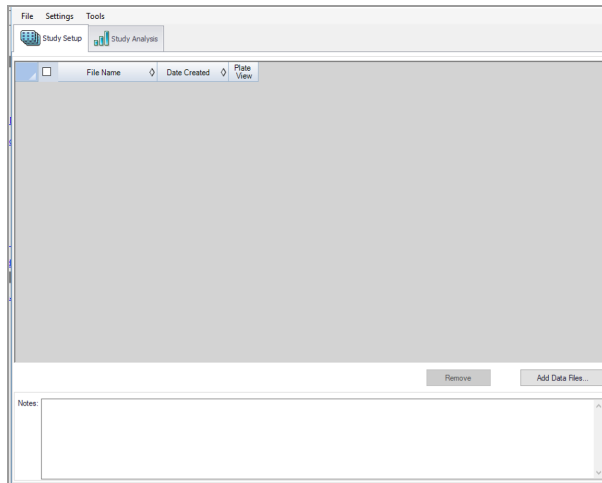


To create a gene study, you add one or more data files and then compare gene expression data from one or more ddPCR experiments.

Note: The maximum number of samples you can analyze in a gene study is limited by the size of the computer's RAM and virtual memory.

To open the Gene Study module

- Click Gene Study.



The Gene Study dialog box includes two tabs:

- **Study Setup** — manages the run files in the gene study

Important: Adding or removing data files in a gene study does not change the data in the original file.

- **Study Analysis** — displays the gene expression data for the combined runs.

From these tabs you can add files to set up a gene study, analyze your data in charts and plots, and run reports on the gene study data.

Gene Study Options

The Gene Study module provides the menus and corresponding options described below.

- **File** — Open, save, and close the gene study, and add files
- **Settings** — Restore the default layout
- **Tools** — Create a report

To open a gene study file

1. Click File → Open.
2. In the Open dialog box, navigate to and select the data file, and then click Open.

To save a gene study file

1. Click File and select Save or Save As.
2. If prompted, enter a file name and click Save.
3. If prompted to overwrite an existing file, click Yes or No.

To close a gene study file

- ▶ Click File → Close.

Note: If you have not saved the file, an advisory prompt appears.

To add data files

1. Click File → Add Data Files.
2. In the Open dialog box, navigate to the data files to be added to the study.
3. Select one or more files and then click Open to add the files to the Gene Study window.

To restore default settings

- ▶ Click Settings → Restore default settings.

Note: At least one modified data file must be open.

To generate a report

- ▶ See [Gene Study Reports on page 175](#).

Setting up the Gene Study

Use the Study Setup window to

- Add data files to include in the study
- Remove data files from the study
- Select data files for viewing and analysis
- Show the plate view for a data file

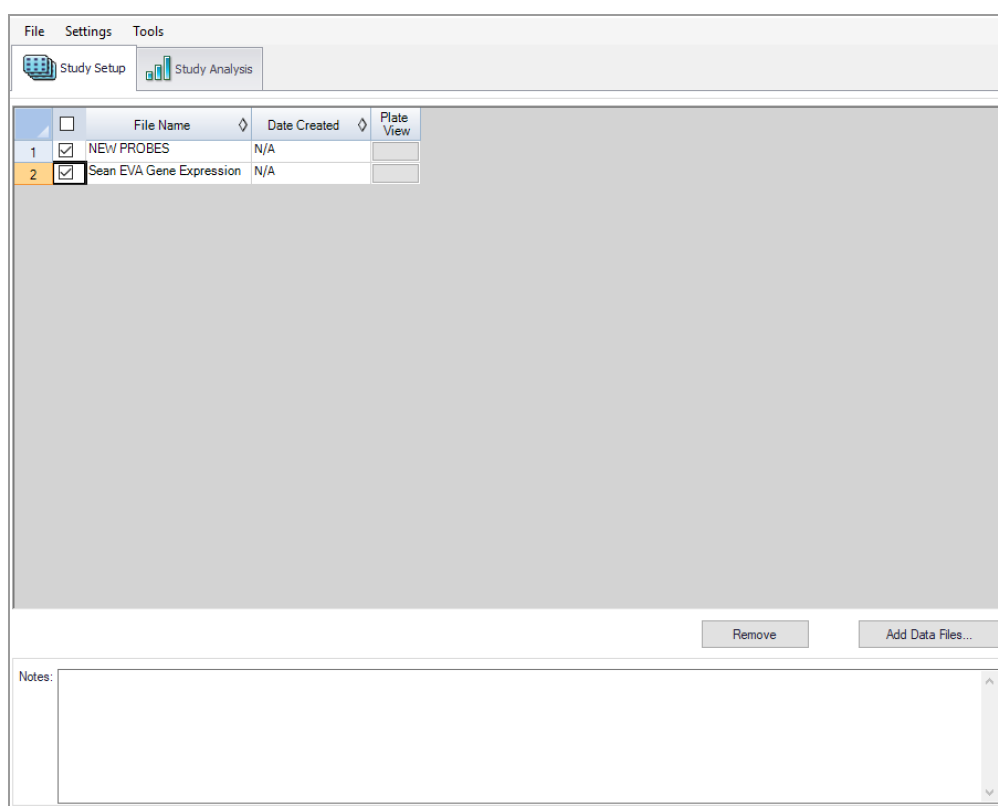


Table 37 defines each column in Study Setup.

Table 37. Gene Study Setup Fields

Column Title	Description
File Name	Name of the data file
Date Created	Date the file was imported into the Gene Study module
Plate View	Opens a map of the plate with the data for each dye in each well included in the Gene Study

Gene Study Plate View

	1	2	3	4	5	6	7	8	9	10	11	12
A	FAM 8.014	FAM 7.889	FAM 6.696	FAM 9.344	FAM 7.082	FAM 9.086	FAM 0.000					
	1	1	1	1	1	1	NTC					
	Unk	Unk	Unk	Unk	Unk	Unk	Unk					
B	FAM 4.705	FAM 3.815	FAM 3.995	FAM 4.353	FAM 4.447	FAM 4.647	FAM 0.000					
	2	2	2	2	2	2	NTC					
	Unk	Unk	Unk	Unk	Unk	Unk	Unk					
C	FAM 2.369	FAM 2.169	FAM 2.223	FAM 1.885	FAM 2.357	FAM 1.747	FAM 0.000					
	3	3	3	3	3	3	NTC					
	Unk	Unk	Unk	Unk	Unk	Unk	Unk					
D	FAM 1.000	FAM 1.226	FAM 1.011	FAM 1.298	FAM 1.175	FAM 0.929	FAM 0.000					
	4	4	4	4	4	4	NTC					
	Unk	Unk	Unk	Unk	Unk	Unk	Unk					
E	FAM-1 32.904	FAM-1 31.401	FAM-1 29.440	FAM-1 32.486	FAM-1 31.268	FAM-1 33.993	FAM-1 0.000					
	1	1	1	1	1	1	NTC					
	Unk	Unk	Unk	Unk	Unk	Unk	Unk					
F	FAM-1 33.789	FAM-1 33.305	FAM-1 33.518	FAM-1 32.269	FAM-1 30.299	FAM-1 32.104	FAM-1 0.000					
	2	2	2	2	2	2	NTC					
	Unk	Unk	Unk	Unk	Unk	Unk	Unk					
	FAM-1	FAM-1	FAM-1	FAM-1	FAM-1	FAM-1	FAM-1					

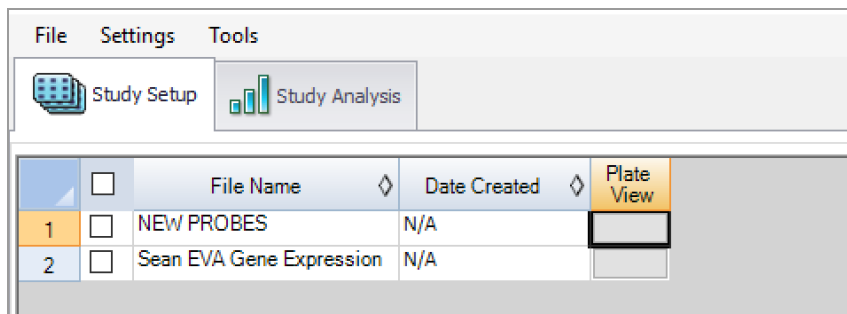
Close

Adding Data Files

To add data files

1. On the Study Setup tab, click Add Data Files.
2. In the Open dialog box, navigate to the data files to be added to the study.
3. Select one or more files and then click Open.

The files are added to the Study Setup window.



Removing Data Files

To remove data files

- Select the checkbox next to the file and then click Remove.

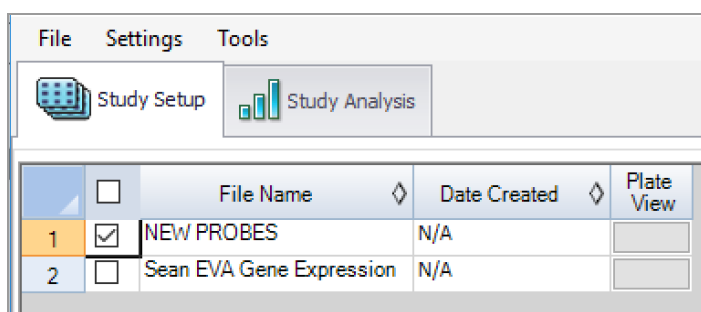
The file is removed from the Study Setup window.

Note: The Remove button is disabled until you select a file.

Selecting Data Files for Analysis

To select data files for analysis

1. Select the checkboxes next to the files to be included in the analysis.

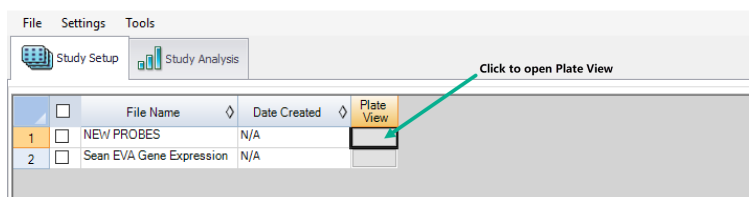


2. Select the Study Analysis tab to see the analysis results. For information, see [Using the Study Analysis Features on page 173](#).

Showing the Plate View

To show the plate view

1. In the Plate View column, click the button for the plate view to be displayed.



The Plate view appears.

Gene Study Plate View

	1	2	3	4	5	6	7	8	9	10	11	12
A	FAM	FAM	FAM	FAM	FAM	FAM	FAM					
	8.014	7.889	6.696	9.344	7.082	9.086	0.000					
	1	1	1	1	1	1	NTC					
B	Unk	Unk	Unk	Unk	Unk	Unk	Unk					
	FAM	FAM	FAM	FAM	FAM	FAM	FAM					
	4.705	3.816	3.995	4.353	4.447	4.647	0.000					
C	2	2	2	2	2	2	NTC					
	Unk	Unk	Unk	Unk	Unk	Unk	Unk					
	FAM	FAM	FAM	FAM	FAM	FAM	FAM					
D	2.369	2.169	2.223	1.885	2.357	1.747	0.000					
	3	3	3	3	3	3	NTC					
	Unk	Unk	Unk	Unk	Unk	Unk	Unk					
E	FAM	FAM	FAM	FAM	FAM	FAM	FAM					
	1.000	1.226	1.011	1.298	1.175	0.929	0.000					
	4	4	4	4	4	4	NTC					
F	Unk	Unk	Unk	Unk	Unk	Unk	Unk					
	FAM-1	FAM-1	FAM-1	FAM-1	FAM-1	FAM-1	FAM-1					
	32.904	31.401	29.440	32.486	31.268	33.993	0.000					
	1	1	1	1	1	1	NTC					
	Unk	Unk	Unk	Unk	Unk	Unk	Unk					
	FAM-1	FAM-1	FAM-1	FAM-1	FAM-1	FAM-1	FAM-1					
	33.789	33.305	33.518	32.269	30.299	32.104	0.000					
	2	2	2	2	2	2	NTC					
	Unk	Unk	Unk	Unk	Unk	Unk	Unk					
	FAM-1	FAM-1	FAM-1	FAM-1	FAM-1	FAM-1	FAM-1					

Close

- 2. Click Close to close the view.

Using the Study Analysis Features

The Study Analysis tab displays the data from all runs in the gene study in the views described in Table 38.

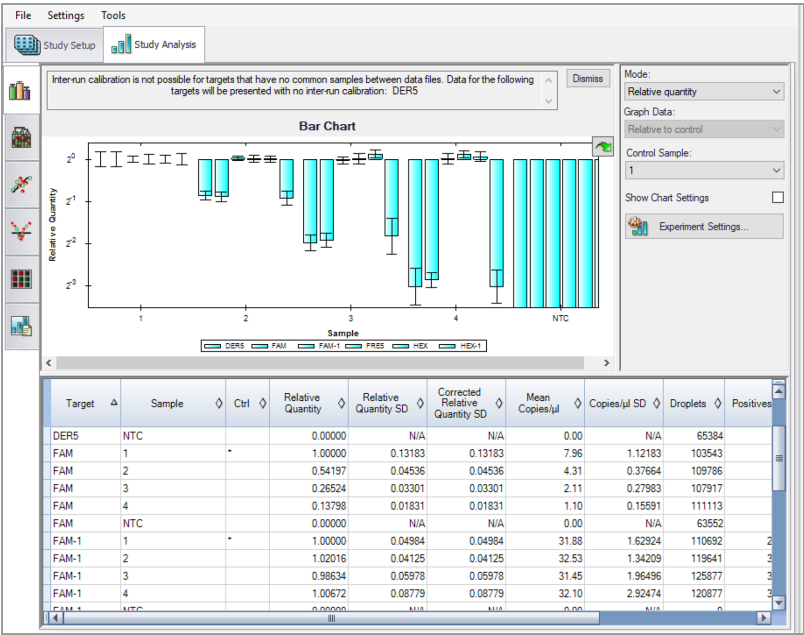


Table 38. Analysis Toolbar







Button	Description
<div>Bar Chart</div> <div></div>	Size of bars correspond to normalized expression or relative quantity
<div>Clustergram</div> <div></div>	Data is arranged in a hierarchy based on the degree of similarity of expression for different targets and samples
<div>Scatter Plot</div> <div></div>	Dot plot of normalized expression of targets for a control versus an experimental sample

Table 38. Analysis Toolbar, continued

Button	Description
Volcano Plot 	Dot plot of change in expression (regulation) of a target for an experimental sample compared to a control
Heat Map 	Plate layout grid showing target regulation for an experimental compared to a control sample based on relative normalized expression Note: If the same targets are in the same location on multiple plates but with different samples, use the dropdown menu to select a particular plate for analysis.
Results Table 	Provides results in a tabular format

Gene Study Reports

Use the Gene Study Report dialog box to arrange the gene study data into a report. Table 34 lists all the options available for a gene study report.

Table 39. Gene Study Report Categories

Category	Option	Description
Header		
	Report Information	Date, user name, data file name, data file path, and the selected well group
	Gene Study File List	List of all the data files in the gene study
	Notes	Notes about the data report
Study Analysis Bar Chart		
	Analysis Settings	List of the selected parameters
	Chart	Gene expression bar chart showing the data
	Target Names	List of targets in the gene study
	Sample Names	List of samples in the gene study
	Data	Spreadsheet that shows the data
	Target Stability	Target stability data
	Inter-run Calibration	Inter-run calibration data
Study Analysis Clustergram, Scatter Plot, Volcano Plot, and Heat Map		
	Analysis Settings	Settings for each chart type
	Chart	Gene expression chart showing the data
	Data	Spreadsheet listing the data in each target

Creating a Gene Study Report

Before creating a report, make any needed adjustments to the gene study data and charts.

To create a gene study report

1. Select Tools → Reports in the Gene Study menu to open the Report dialog box.

2. Select the options you want to include in the report.

The report opens with default options selected. Select or clear the checkboxes to change whole categories or individual options within a category.

For information on the available options to display, see [Gene Study Reports on page 175](#).

3. Change the order of categories and items in a report.

Drag the options to the required position. Note that Items can be reordered only within the categories to which they belong.

4. Click Update Report to update the Report Preview window with any changes.

5. Print or save the report:

- a. Click the Print Report button on the toolbar to print the current report.
- b. Select File → Save to save the report in PDF, MHT or MHTML file format and select a location to save the file.

Note: MHT and MHTML are Microsoft formats.

- c. Select File → Save As to save the report with a new name or in a new location.

6. (Optional) To save the current report settings as a template, select Template → Save or Save As.

Chapter 12 System Utilities

From the System tab, you can access the following tabs for managing tasks and data related to the instrument, software, data, and storage:

■ Datafile and Template Storage

- **System Settings** — View shared template and data file locations
- **Event Log** — View logged data about all software activities
- **Maintenance Log** — Record maintenance activities
- **Maintenance Reports** — PDF maintenance records from Bio-Rad maintenance
- **System Data** — archive data to clear up space in primary storage areas, access troubleshooting data, and use the data recovery function
- **Reprocess Data** — Reprocess a .data file using new calibration values
- **Tools** — Flush or prime the instrument

■ Instrument

- Change the instrument

Note: The Instrument Calibration tab is enabled only for the Bio-Rad service engineer user account.

System Settings Tab

When you select the System tab, the System Settings window opens by default.



The window contains the following data file and template storage options set by your software administrator:

- **Preferred Locations** — if the checkbox selected, storage for all users is limited to a single folder for data files and a single folder for templates.
- **Shared Settings** — if the Preferred Locations checkbox remains cleared, then shared locations can be defined, but remain optional. Users can choose to save their files to their personal folders specified in User Preferences or to the shared location.

Note: The software administrator can also define a backup location for data file storage if the priority folder is full.

Preferred Location

If the administrator sets Preferred Location file paths, then data files and templates for all users are automatically saved to the specified folders. Preferred Locations override selections in individual user preferences.

Important: Only users assigned the System Settings user privilege can set preferred locations.

System Settings	Datafile and Template Storage	Instrument
	Preferred Location ⓘ	
Event Log	<input checked="" type="checkbox"/> Use for all users	
Maintenance Log	Datafile Storage Location: <input type="text" value="C:\Users\Public\Documents\Bio-Rad\QXMgrStandard\AppData\Data"/>	
Maintenance Reports	<input type="button" value="Browse"/>	
System Data	Template Storage Location: <input type="text" value="C:\Users\Public\Documents\Bio-Rad\QXMgrStandard\AppData\Templates"/>	
Reprocess Data	<input type="button" value="Browse"/>	

To specify preferred locations

1. Select the Use for all users checkbox.
2. (Optional) Keep the default paths or modify them to alternative secure storage locations.
3. Click Save.
4. When the confirmation message appears, click OK.

Shared Settings

Under Shared Settings, you can choose to save your files to locations that are available to all users.

The screenshot shows the 'System Settings' window with a sidebar on the left containing the following menu items: System Settings, Event Log, Maintenance Log, Maintenance Reports, System Data, Reprocess Data, Instrument Calibration, Instrument Check, and Tools. The main panel is titled 'Datafile and Template Storage' and has a sub-tab 'Instrument'. It contains several sections: 'Preferred Location' with a 'Use for all users' checkbox, 'Datafile Storage Location' with a text field and 'Browse' button, 'Template Storage Location' with a text field and 'Browse' button, 'Shared Settings' (highlighted with a red border) containing 'Datafile Storage Location' and 'Template Storage Location' fields with 'Browse' buttons, and a 'Save' button at the bottom.

The software allows the software administrator to set a primary storage folder and a backup storage folder. Bio-Rad recommends routinely checking available storage space and if necessary, using the Archive function.

Important: Only users assigned the System Settings user privilege (typically your software administrator) can change storage locations in the System Settings window.

To specify shared folder locations

1. In each of the storage path fields, enter a file path or click Browse to search for the folder.
2. Click Save.
3. When the confirmation message appears, click OK.

Changing Instruments

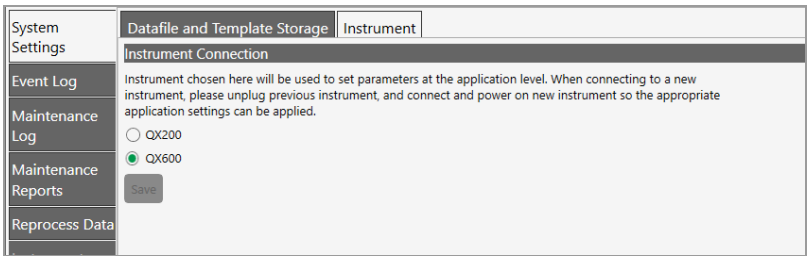
When you open QX Manager Software for the first time after installation, the Instrument connection window appears and you must select the instrument you will be using. If both the QX200 and QX600 droplet readers are used in your laboratory, you can select the alternate instrument on the Instrument tab in System Settings.

To change the instrument in the software

1. Select the System Settings tab.



2. In the System Settings window (default), select the Instrument tab.



3. Select the instrument and then click Save.

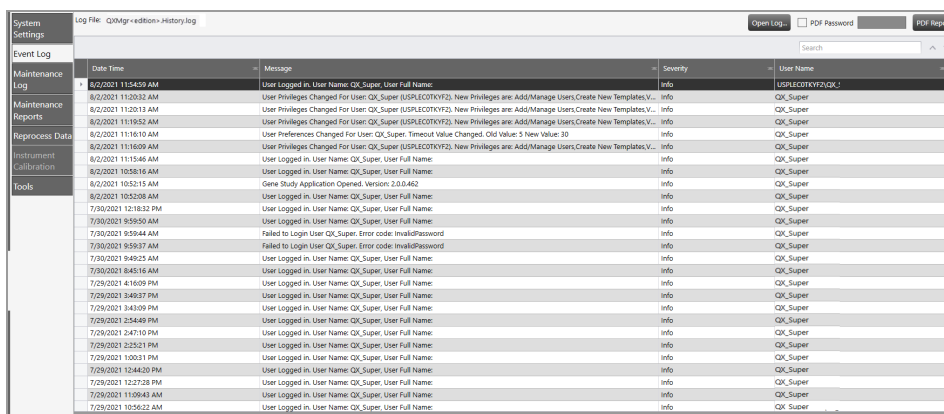
Features associated with the selected instrument become available when the instrument is connected.

System Log Files

From the System tab, you can access log files for system and software events (event log), and maintenance activities (maintenance log).

Event Log

Users who are assigned the Maintenance user privilege can view the system event log, which contains timestamped information on all software activities.



Date Time	Message	Severity	User Name
8/2/2021 11:54:59 AM	User Logged in, User Name: QX_Super, User Full Name:	Info	US\$LECCOTY2\QX_S
8/2/2021 11:55:32 AM	User Privileges Changed For User: QX_Super (US\$LECCOTY2). New Privileges are: Add/Manage Users, Create New Templates, V...	Info	QX_Super
8/2/2021 11:55:32 AM	User Privileges Changed For User: QX_Super (US\$LECCOTY2). New Privileges are: Add/Manage Users, Create New Templates, V...	Info	QX_Super
8/2/2021 11:55:32 AM	User Privileges Changed For User: QX_Super (US\$LECCOTY2). New Privileges are: Add/Manage Users, Create New Templates, V...	Info	QX_Super
8/2/2021 11:56:10 AM	User Preferences Changed For User: QX_Super. Timeout Value Changed. Old value: 5 New Value: 30	Info	QX_Super
8/2/2021 11:56:09 AM	User Privileges Changed For User: QX_Super (US\$LECCOTY2). New Privileges are: Add/Manage Users, Create New Templates, V...	Info	QX_Super
8/2/2021 11:56:46 AM	User Logged in, User Name: QX_Super, User Full Name:	Info	QX_Super
8/2/2021 10:58:16 AM	User Logged in, User Name: QX_Super, User Full Name:	Info	QX_Super
8/2/2021 10:52:15 AM	Gene Study Application Opened. Version: 2.0.0.462	Info	QX_Super
8/2/2021 10:52:09 AM	User Logged in, User Name: QX_Super, User Full Name:	Info	QX_Super
7/29/2021 12:18:32 PM	User Logged in, User Name: QX_Super, User Full Name:	Info	QX_Super
7/29/2021 9:59:50 AM	User Logged in, User Name: QX_Super, User Full Name:	Info	QX_Super
7/29/2021 9:59:44 AM	Failed to Login User QX_Super. Error code: InvalidPassword	Info	QX_Super
7/29/2021 9:59:37 AM	Failed to Login User QX_Super. Error code: InvalidPassword	Info	QX_Super
7/29/2021 9:45:25 AM	User Logged in, User Name: QX_Super, User Full Name:	Info	QX_Super
7/29/2021 8:45:16 AM	User Logged in, User Name: QX_Super, User Full Name:	Info	QX_Super
7/29/2021 4:16:09 PM	User Logged in, User Name: QX_Super, User Full Name:	Info	QX_Super
7/29/2021 3:49:37 PM	User Logged in, User Name: QX_Super, User Full Name:	Info	QX_Super
7/29/2021 3:43:09 PM	User Logged in, User Name: QX_Super, User Full Name:	Info	QX_Super
7/29/2021 2:54:49 PM	User Logged in, User Name: QX_Super, User Full Name:	Info	QX_Super
7/29/2021 2:47:10 PM	User Logged in, User Name: QX_Super, User Full Name:	Info	QX_Super
7/29/2021 2:23:21 PM	User Logged in, User Name: QX_Super, User Full Name:	Info	QX_Super
7/29/2021 1:00:21 PM	User Logged in, User Name: QX_Super, User Full Name:	Info	QX_Super
7/29/2021 12:44:30 PM	User Logged in, User Name: QX_Super, User Full Name:	Info	QX_Super
7/29/2021 12:27:28 PM	User Logged in, User Name: QX_Super, User Full Name:	Info	QX_Super
7/29/2021 11:09:43 AM	User Logged in, User Name: QX_Super, User Full Name:	Info	QX_Super
7/29/2021 10:56:22 AM	User Logged in, User Name: QX_Super, User Full Name:	Info	QX_Super

To view the Event Log

1. Select the System Utilities tab.



2. Select Event Log.
3. Click Open Log to choose either the debug log or the history log.
4. Review the log entries.
5. (Optional) To automatically generate and display a PDF document, click PDF Report.

Tip: To restrict access to the file, you can assign a password. Select the PDF Password check box and then enter a password in the field. The file is saved in the Log Reports folder and can be opened only by entering the password.

Maintenance Log

All users can view the maintenance log, which contains a list of maintenance records. Users who are assigned the Maintenance user privilege can set up maintenance records, specify associated activities, and generate PDF reports. This guide also contains a form on which you can document completion of maintenance activities. For information, see [Instrument Maintenance Log on page 215](#).

Date Time	User	Product Version	Firmware Version	Activity
12/10/2018	USERABC	1.0.00	1.0.00	

To add or view an activity

1. Select the System Utilities tab.



2. Select Maintenance Log. Note that activities that are already scheduled appear in the grid, sorted by newest activity first.
3. Do one of the following:
 - Scroll to view a particular activity.
 - **Important:** You can view or add static activities; you cannot modify them.
 - Add an activity; click the + icon next to New Activity.

A line item appears in the bottom section, with all fields except Activity completed automatically.

4. In the Activity field in the bottom section, type the activity into the Activity field, and click Save.
5. Repeat to add another activity.
6. (Optional) Click PDF Report to create a report of the added activities.

Maintenance Reports

Reports generated from the Bio-Rad service engineer when performing maintenance on your instrument are stored on the Maintenance Reports tab.

- To open a report, click the list item.



Following is an example of a Color Calibration report:

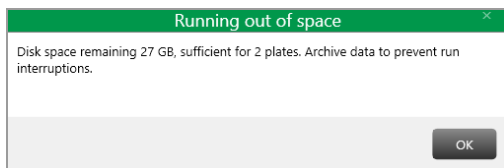
BIO-RAD																				
Color Calibration and Carryover Report																				
Username of the Person who did the Run: BERRAD																				
Report Generation Date and Time: 7/20/2019 09:03:29 PM																				
Instrument Serial Number: CHARLIE-1																				
Firmware Version: 1.2760																				
Software Version: 1.0.774.0628																				
Software: QX ONE Software Standard Edition																				
Kit Information																				
GC300 Cartridge: 10250																				
Droplet Generation: DR262DR101, DR262DR101																				
Droplet Reader: DR262DR101, DR262DR101																				
Calibration Effective Date: 2/7/2019 03:20:11 PM																				
<table><tr><th>Color Calibration Result</th><th>Carryover Test Result</th><th># of carryover events</th></tr><tr><td>Channel 1 (FAM)</td><td>Pass</td><td>0</td></tr><tr><td>Channel 2 (BIBX/VIC)</td><td>Pass</td><td></td></tr><tr><td>Channel 3 (IC-51)</td><td>Pass</td><td></td></tr><tr><td>Channel 4 (IC-25.5)</td><td>Pass</td><td></td></tr><tr><td>Low Dns</td><td>Fail</td><td></td></tr></table>			Color Calibration Result	Carryover Test Result	# of carryover events	Channel 1 (FAM)	Pass	0	Channel 2 (BIBX/VIC)	Pass		Channel 3 (IC-51)	Pass		Channel 4 (IC-25.5)	Pass		Low Dns	Fail	
Color Calibration Result	Carryover Test Result	# of carryover events																		
Channel 1 (FAM)	Pass	0																		
Channel 2 (BIBX/VIC)	Pass																			
Channel 3 (IC-51)	Pass																			
Channel 4 (IC-25.5)	Pass																			
Low Dns	Fail																			
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System Data

Select the System Data tab to archive raw data and access troubleshooting data.

Archiving Raw Data

When the storage space on the computer connected to the QX600 Droplet Reader approaches the limit displayed under Disk Space, you can archive raw data to an archive storage folder in a different location . The software also displays an advisory message, as shown in the following graphic:



Important: You must be assigned the System Data user privilege to archive data.

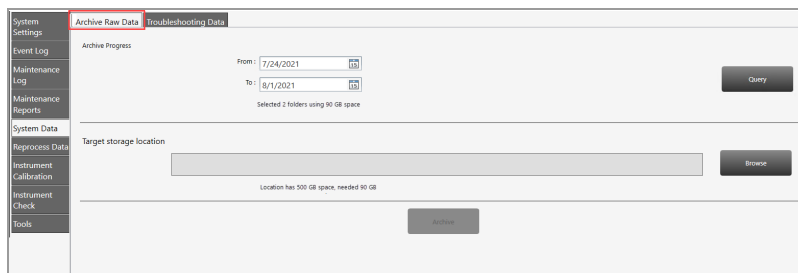
To archive files

1. Select the System Utilities tab.



2. Select System Data.

The window opens to the Archive Raw Data screen by default.



3. In the Archive Progress section, click the calendar icons and select a From date and a To date.
4. Click Query to search within the defined date span.
5. Click Browse to locate the archive location on your network, and then click Archive.

Troubleshooting Data

If issues occur with the instrument or software, you can access data to assist in troubleshooting the problem.

Important: You must be assigned the System Data user privilege to access troubleshooting data.

To collect data for troubleshooting

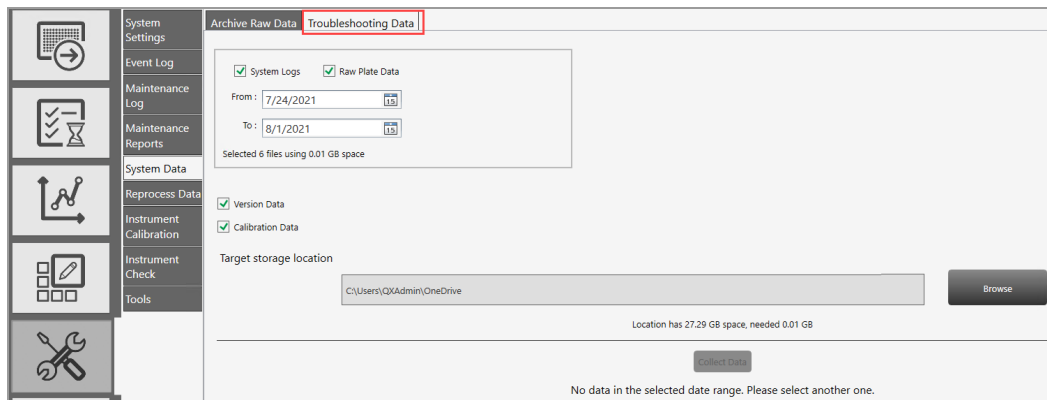
1. Select the System Settings tab.



2. Select System Data.

The window opens to the Archive Raw Data screen by default.

3. Select the Troubleshooting Tab.



4. Click the From and To calendar icons to select a date range.

5. Select any of the following checkboxes:

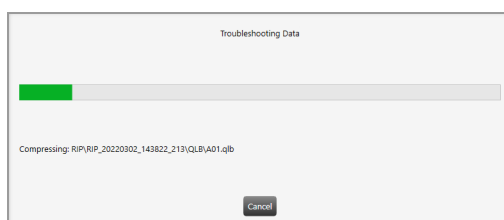
- System Logs
- Raw Plate Data
- Version Data
- Calibration Data

- Click Browse to navigate to a folder where the data will be stored.

QX Manager Software advises how much space you need and how much is available, and also advises if there is no data from your specified date range.

- Click Collect Data.

A progress bar appears.



QX Manager Software creates a subfolder containing the data you specified and places it in the target location.

Reprocessing Data

After your instrument has been recalibrated, you can reprocess a data file from a run using different color calibration values.

Note: The instrument must be in an idle state before you can reprocess the data in QX Manager Software. If the file was created in an older software version, the software cannot reprocess the file and displays an advisory message.

To reprocess a data file

- Select the System Utilities tab.



- Click Reprocess Data.
- Under Datafile, click Browse and navigate to the analysis data file.

4. In the Select Datafile dialog box, select the file and click Open.

The calibration values appear in the first Color Calibration table.

The screenshot shows the 'Instrument Calibration' section of the software. The 'Color Calibration Table' is displayed, showing a table with columns for Dye, Channel 1, Channel 2, Channel 3, Channel 4, Channel 5, and Channel 6. The table contains data for various dyes including FAM, SYBRGREEN, HEX, VIC, CY5, CY5.5, ROX, and ATTO590. The 'Reprocess Data' button is visible at the bottom right of the table.

Dye	Channel 1	Channel 2	Channel 3	Channel 4	Channel 5	Channel 6
FAM	10000/10000	0/0	0/0	0/0	0/0	0/0
SYBRGREEN	10000/10000	0/0	0/0	0/0	0/0	0/0
HEX	0/0	10000/10000	0/0	0/0	0/0	0/0
VIC	0/0	10000/10000	0/0	0/0	0/0	0/0
CY5	0/0	0/0	10000/10000	0/0	0/0	0/0
CY5.5	0/0	0/0	0/0	10000/10000	0/0	0/0
ROX	0/0	0/0	0/0	0/0	10000/10000	0/0
ATTO590	0/0	0/0	0/0	0/0	0/0	10000/10000

5. In the table, enter new values for the fluorophores.
6. Click Reprocess Data.

Note: When you reprocess a data file, the software creates an audit record containing the old and new calibration values.

System Calibration and Tools

You cannot execute runs on the QX600 Droplet Reader unless the instrument is properly calibrated.

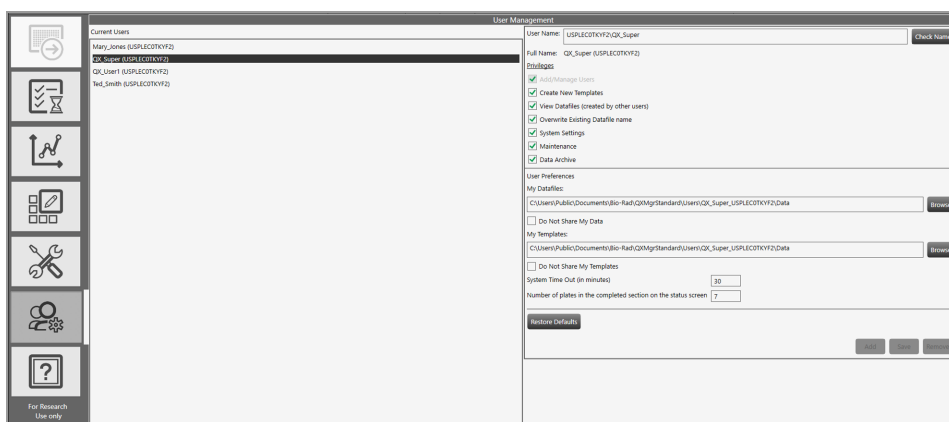
Only a Bio-Rad service engineer can calibrate your instrument and use the Instrument Calibration options in QX Manager Software.

If your instrument requires service, contact Bio-Rad Technical Support.

The Tools functions are currently applicable only to instrument maintenance. For information, see [Appendix B, Instrument Maintenance](#).

Chapter 13 Managing Users

Use the User Management window to add, change, or remove QX Manager Software users, and set or change user privileges and preferences.



An administrative user account, with all privileges, should be set up first. You will use this user account to create additional users and assign user privileges. If you need assistance, contact Bio-Rad Technical Support. Users who are not assigned any of the specified privileges can still execute runs and use the Analysis module.

You can set up local or domain accounts. If the computer connected to the QX600 Droplet Reader

- Cannot access your company's network, your IT department or system administrator can set up user names as local Windows accounts.
- Can access your company's network, QX Manager Software can recognize the Active Directory user names.

You can create user accounts with any user name convention and password character structure, as long as it complies with your organizational requirements. The software is designed to validate each user against the name it finds in either the Active Directory or local domain, so users must log in with the proper domain and user name.

Adding Users

Before you can add users to QX Manager Software, you must be logged in with the initial superuser account, or another account that is assigned the Add/Manage Users privilege.

Important: Before you begin, ensure that all users either have local Windows user accounts on the local computer or user accounts in Active Directory (network domain).

To add a user

1. Open QX Manager Software and sign into the application.
2. Click the User Management tab on the left



3. In the User Name field on the right, enter the user name as follows:
 - If the computer is connected to a network and you are using Active Directory accounts, enter <domain name>/<user name>. For example, **global/johnsmith**.
 - If the computer is not connected to a network, enter <local computer name>/<user name>. For example, **analysiscomputer/johnsmith**.

Important: Active Directory users must log in every time with the domain name preceding the user name.

Important: To locate the computer name, right-click on the yellow folder in the lower menu bar and select File Explorer. When the dialog box appears, the computer name is located at the top of the left panel. Local users are not required to reenter the computer name after the initial login.

4. Click Check Name.

One of the following occurs:

- If the software recognizes the network domain or computer name and the user name, the software displays a validation message.
- If the user is not recognized, an error message appears. Do one of the following:
 - For network users, ensure there is a working connection to the network and then verify your entry is accurate.
 - For local users, verify the entry is accurate. If the user is not set up locally or on the network, contact your system administrator for assistance.

5. When the This user is valid message appears, click OK, and then click Add at the bottom of the pane.

An Are you sure you want to add this user? message appears.

6. Click Yes.

A confirmation message appears.

7. Click OK.

8. Continue until all users are added.

Adding or Removing User Privileges

Only a user assigned the Add/Manage users privilege can add or remove user privileges. By default, individual users without privileges can set up and execute a run, open files, view and analyze results.

For descriptions of all user privileges, see [Viewing User Privileges on page 37](#).

User Name: (computer or domain name\QX_User1) Check Name

Full Name: QX_User1 (full name)

Privileges

☐ Add/Manage Users

☐ Create New Templates

☒ View Datafiles (created by other users)

☒ Overwrite Existing Datafile name

☐ System Settings

☐ Maintenance

☒ System Data

☒ Manage Plate

APF Privileges

☒ Manage APFs

☒ Run APFs

☐ Only Run APFs

[Table 40](#) contains a sample set of privileges assigned by user roles common to a laboratory. User roles can be defined and used in any scenario, but might be required in regulatory environments.

Table 40. Example User Roles

Privilege	Superuser	Lab Manager	Group Lead	Technician/ Student Intern
Add/manage users	✓	✓		
Create new templates	✓	✓	✓	
View data files created by other users	✓	✓	✓	✓
Override existing ddPCR file name	✓	✓	✓	
System settings	✓			
Maintenance	✓			

Table 40. Example User Roles, continued

Privilege	Superuser	Lab Manager	Group Lead	Technician/ Student Intern
Archive data files	✓	✓	✓	✓
Access the module recovery tool	✓	✓	✓	✓
APF Privileges:				
Manage APFs	✓	✓	✓	
Run APFs	✓	✓	✓	✓
Only Run APFs				✓
Manage Plates	✓	✓	✓	

You can assign the Create new templates privilege to non-administrative roles if anyone can create and save a template. If template formats are restricted, then you should assign this permission to administrative or supervisory roles only.

Note: Bio-Rad recommends defining at least one additional superuser as a backup, as well as other administrative and standard roles based on your needs. Guest users can open shared templates, execute runs, and perform analysis but are not typically assigned other privileges.

To add or modify privileges

1. Click the Add/Manage Users tab to open the User Management window.
2. Select a user from the Current Users list. The name appears in the User Name field. A network or computer name and backslash might appear before the user name.
3. Select or clear the checkbox for each privilege in accordance with the user's role in using the software. You can assign user privileges in any combination.
4. When the confirmation message appears, click Yes to save the changes, and then click OK to close the pop-up.

Modifying User Preferences

Users assigned the Add/Manage Users privilege can change the preferences for other users. Individual users can also change their own preferences.

To modify a user's preferences

1. Click the Add/Manage Users tab, and then enter the user name.
2. Change any of the following preferences for the user:
 - Enter a different file path for user data files and templates, and choose to keep them private.
Important: You can also specify Preferred Locations in System Settings, to override all file paths specified in individual preferences. For information, see [System Settings Tab on page 178](#).
 - Enter a different default system timeout period.
 - Enter a different number of completed plates to show in the Status section, up to a maximum of 100.
3. Click Save. QX Manager Software displays a confirmation message.
4. Click Yes to save the changes, and then click OK to close the dialog box.

Removing Users

Only a user assigned the Add/Manage Users privilege can remove users. Removing a user from the software does not remove the user from other systems, such as Windows, or databases

1. Click the Add/Manage Users tab to open the User Management window.
2. In the Current Users pane, select the user and click Remove. QX Manager Software displays a confirmation message.
3. Click Yes to remove the user, and then click OK to close the dialog box.

Appendix A Experiment Examples

This section contains setup and analysis information for the following sample experiments:

- Direct Quantification, using the Amplitude Multiplex assay type
- Copy Number Variation, using the Probe Mix Triplex assay type
- Drop-Off, using the Basic Drop-off Assay type for genome edit detection
- Direct Quantification, using the Advanced Classification Method assay type

Direct Quantification Multiplexed

You can increase the number of targets to be evaluated by using the Amplitude Multiplex option. Optionally, to expedite plate setup you can copy and paste data from one well to another, and you can click Undo to undo an applied well setting. You cannot change the supermix that was used during data collection.

Setting Up Experiment Parameters

To set experiment parameters

1. Open the analysis data file and select the Plate Editor tab.
2. Select one or more wells to be assigned well analysis attributes.
3. Change the experiment type to Direct Quantification (DQ), and then click Apply.
4. (Optional) Change the sample description identifiers.
5. (Optional) Change the sample type. The default type is Unknown.

Tip: For automatic thresholding of positive control wells, you must select Pos Ctrl.

6. Under Assay Type, select Amplitude Multiplex and click Apply. Twelve target rows appear.
7. Change the target name and type (unknown or reference) for each target in the well.

The software defaults to FAM Lo, FAM Hi, HEX Lo, HEX Hi, Cy5 Lo, Cy5 Hi, Cy5.5 Lo, Cy5.5 Hi, ROX Lo, ROX Hi, ATTO 590 Lo, and ATTO 590 Hi, respectively, for each target.

8. (Optional) Change the high and low target identifiers.

Amplitude Multiplex allows for up to two assays in each channel, indicated by Lo and Hi labels. In this multiplex mode, each target may have a signal for only one channel. You can

- Select a different dye, if the channel supports two dyes (for example change HEX to VIC or VIC to HEX)
- Select a different channel for the target (for example, change FAM to HEX)
- Change Hi to Lo or Lo to Hi

9. Click Apply.

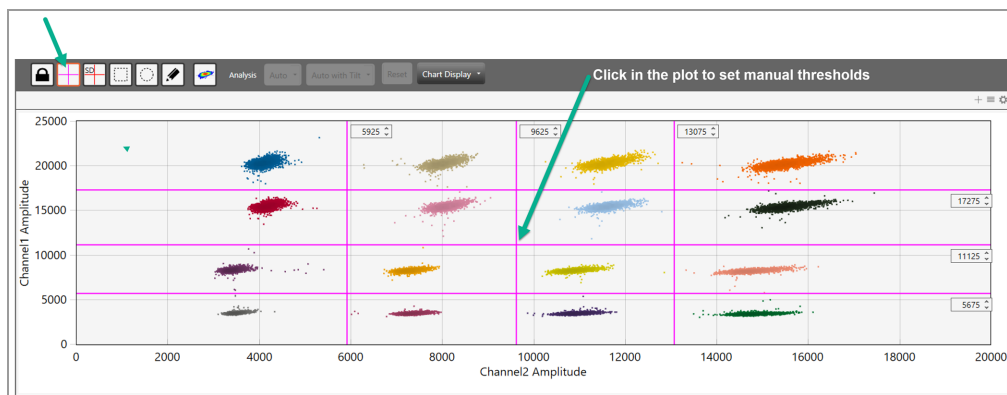
Viewing and Adjusting Results

Automatic thresholding is not available for Amplitude Multiplex experiments.

For clusters that are easily identifiable across the chart, use Threshold Lines mode to set manual thresholds. If the data is not orthogonal and the threshold lines do not adequately divide the populations of droplets, use Threshold Clusters mode to set manual thresholds.

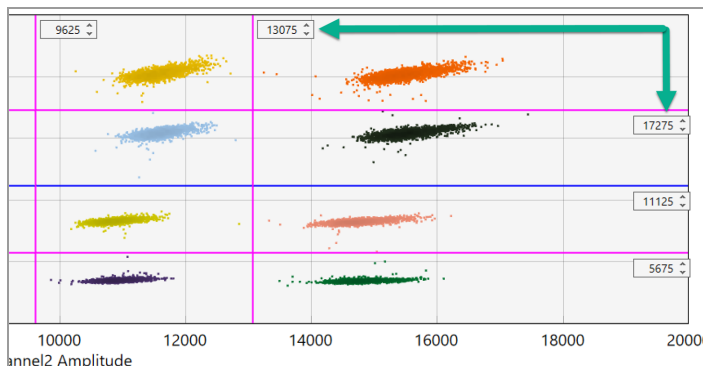
To set thresholds in the 2D Amplitude window

1. Select the 2D Amplitude tab.



2. On the toolbar, click the Threshold Line Mode button.
3. Click anywhere on the plot to apply “best fit” threshold lines to the data.
4. To adjust the thresholds, drag the lines horizontally or vertically in the plot area.

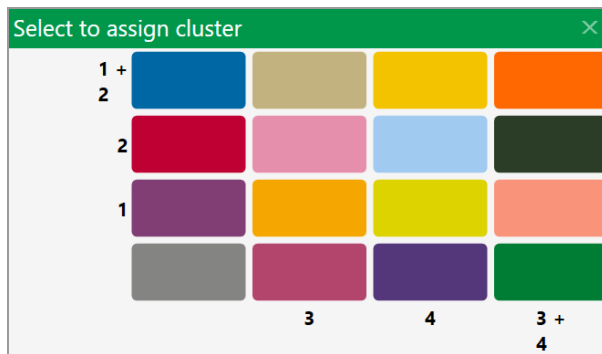
Tip: You can also enter a specific threshold value in the boxes at the end of each threshold line.



To apply and adjust manual clusters

1. Click any of the three cluster mode buttons (square, circle, or freehand).
2. Press the left mouse button and draw the chosen shape around the cluster of droplets to be classified first.

Tip: When you release the mouse button, a pop-up tool appears to help you select the correct target combination. Target names are shown along the two axis based on the signal values assigned during plate setup.



3. To classify droplets as positive or negative for each given target, select the colored box that best represents the location of the cluster of droplets.

Copy Number Variation Triplexed

A CNV experiment with probe mix triplexing uses two targets and a reference in three groups of three to produce analysis data. Optionally, to expedite plate setup you can copy and paste data from one well to another, and you can click Undo to undo an applied well setting.

Setting Up Experiment Parameters

To set experiment parameters

1. Open the analysis data file and select the Plate Editor tab.
2. Select one or more wells to be assigned well analysis attributes.
3. Set the experiment type to Copy Number Variation (CNV), and click Apply.
4. (Optional) Change the sample description identifiers.
5. (Optional) Change the sample type. The default type is Unknown.

Tip: For automatic thresholding of positive control wells, you must select Pos Ctrl.

Note: You cannot change the supermix that was used during data collection.

6. Click Apply.
7. From the Assay Type dropdown list, select Probe Mix Triplex and click Apply.

QX Manager Software displays three targets.

Important: You cannot add or delete triplex targets.

8. Enter target names and select the target type (unknown or reference) for each target in the well.

This creates three groups of three targets each.

9. Assign the targets as follows:

Group 1

Target	Target Type	Signal Ch1	Signal Ch2	Ref Copies	Plot?
Target 1	Unknown	FAM	None	N/A	N/A
Target 2	Unknown	None	HEX/VIC	N/A	N/A
Target 3	Reference	FAM	HEX/VIC	2	Yes

Group 2

Target	Target Type	Signal Ch3	Signal Ch4	Ref Copies	Plot?
Target 4	Unknown	Cy5	None	N/A	N/A
Target 5	Unknown	None	Cy5.5	N/A	N/A
Target 6	Reference	Cy5	Cy5.5	2	Yes

Group 3

Target	Target Type	Signal Ch3	Signal Ch4	Ref Copies	Plot?
Target 7	Unknown	ROX	None	N/A	N/A
Target 8	Unknown	None	ATTO 590	N/A	N/A
Target 9	Reference	ROX	ATTO 590	2	Yes

10. Click Apply.

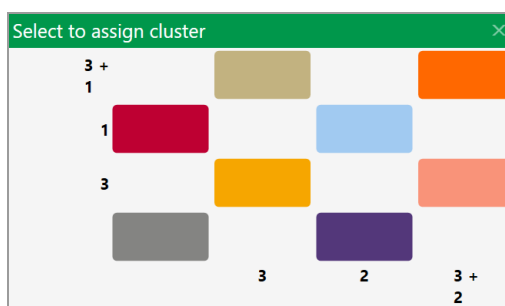
Viewing and Adjusting Results

Note: Linear thresholding is not available for Probe Mix Triplex experiments.

To see the results

1. After all wells are configured, select the 2D Amplitude tab to evaluate the data and manually set clusters.
2. Click any of the three cluster mode buttons (square, circle, or freehand).
3. Press the left mouse button and draw the chosen shape around the cluster of droplets to be classified first.

Tip: When you release the mouse button, a pop-up tool appears to help you select the correct target combination. Target names are shown along the two axis based on the signal values assigned during plate setup.



4. To classify droplets as positive or negative for each given target, select the colored box that best represents the location of the cluster of droplets.

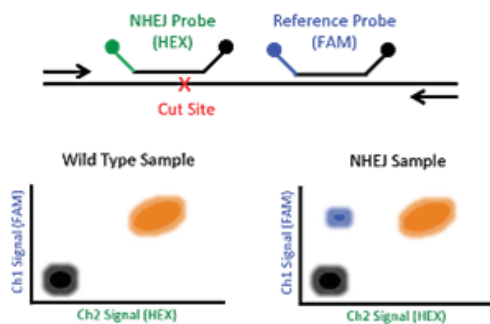
Drop-Off Assays for Genome Edit Detection

Optionally, to expedite plate setup you can copy and paste data from one well to another, and you can click Undo to undo an applied well setting.

Two configurations of drop-off assays are possible and are shown below. Assign a signal (expected cluster position) to wild-type and NHEJ indel alleles. The signal produced by a wild-type allele will always be positive for both FAM and HEX, while the signal produced by an NHEJ indel allele depends on the assay configuration used (as shown below left and right).

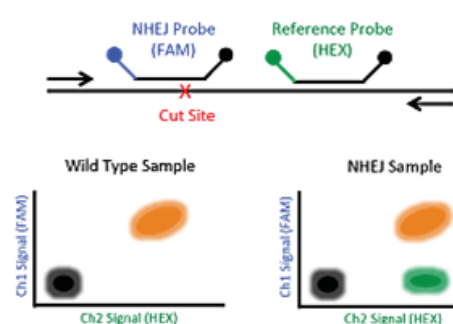
Drop-Off Probe in HEX

NHEJ Probe = HEX and Reference Probe = FAM



Drop-Off Probe in FAM

NHEJ Probe = FAM and Reference Probe = HEX



This type allows three groups of two targets per well; Group 1, shown by default, with 1 target in FAM and HEX (reference), and 1 target in FAM or HEX (unknown). You can add a second group with 1 target in Cy5 and Cy5.5 (reference) and 1 target in Cy5 or Cy5.5 (unknown) and a third group with 1 target in ROX and ATTO 590 (reference) and 1 target in ROX and ATTO 590 (unknown).

Note: The channels within each group are not interchangeable therefore FAM is always paired with HEX/VIC. Cy5 with Cy5.5. and ROX with ATTO 590.

Setting Up Experiment Parameters

To set experiment parameters

1. Open the analysis data file and select the Plate Editor tab.
2. Select one or more wells to be assigned well analysis attributes.
3. Set the experiment type to Drop-Off (DOF) and click Apply.

The screenshot shows the 'Experiment Type' dropdown set to 'Drop Off (DOF)' with an 'Apply' button. Below are four 'Sample Description' fields (1-4) with an 'Apply' button. The 'Sample Type' dropdown is set to 'Unknown'. The 'Supermix' dropdown is set to 'One-Step RT-ddPCR Advanced Kit for Probes'. The 'Assay Type' dropdown is set to 'Basic Drop-Off' with an 'Apply' button. The 'Target Info' table is as follows:

Target Name	Target Type	Signal Ch1	Signal Ch2	Signal Ch3	Signal Ch4	Signal Ch5	Signal Ch6	Ref Copies
Target-1	Ref	FAM	HEX	None	None	None	None	2
Target-2	Unkn	FAM	None	None	None	None	None	
3	Ref	None	None	Cy5	Cy5.5	None	None	2
4	Unkn	None	None	Cy5	None	None	None	
5	Ref	None	None	None	None	ROX	ATTO590	2
6	Unkn	None	None	None	None	ROX	None	

4. (Optional) Change the sample description identifiers.
5. (Optional) Change the sample type. The default type is Unknown.

Tip: For automatic thresholding of positive control wells, you must select Pos Ctrl.

Note: You cannot change the supermix that was used during data collection.

6. Click Apply.

QX Manager Software automatically assigns the assay method as Basic Drop-Off.

7. Enter the target names, and then designate one target as Reference and the other as Unknown.

The Reference target enables ratio calculation of results.

8. Click Apply.

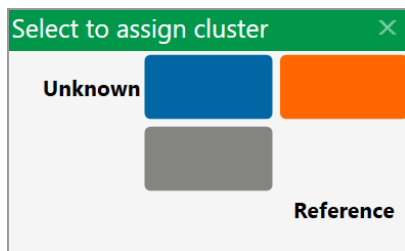
Viewing and Adjusting Results

Thresholding is not available for Drop Off experiments.

To apply and adjust clusters

1. After all wells are configured, select the 2D Amplitude tab to evaluate the data and manually set clusters.
2. Click any of the three cluster mode buttons (square, circle, or freehand).
3. Press the left mouse button and draw the chosen shape around the cluster of droplets to be classified first.

Tip: When you release the mouse button, a pop-up tool appears to help you select the available cluster assignments. Target names are shown along the two axis based on the signal values assigned during plate setup.



4. To classify droplets as positive or negative for each given target, select the colored box that best represents the location of the cluster of droplets.

Direct Quantification with the Advanced Classification Method

The Advanced Classification Method analysis mode allows the user complete control of droplet identification, and does not force or preassign any clusters. This allows a higher degree of multiplexing through radial dye mixing. Optionally, to expedite plate setup you can copy and paste data from one well to another, and you can click Undo to undo an applied well setting.

Important: This mode is intended for experienced users of Droplet Digital PCR.

Setting Up Experiment Parameters

To use the advanced classification method

1. Open the analysis file and select the Plate Editor tab.
2. Select one or more wells to be assigned well analysis attributes.
3. Set the experiment type to Direct Quantification (DQ) or RED and click Apply.
4. (Optional) Change the sample description identifiers.
5. (Optional) Change the sample type. The default type is Unknown.

Tip: For automatic thresholding of positive control wells, you must select Pos Ctrl.

Note: You cannot change the supermix that was used during data collection.

6. Click Apply.
7. Under Assay Type, select Advanced Classification Method and click Apply.
8. Use the + icon to add up to ten targets.
9. Enter the target name and type (unknown or reference) for each target in the well.
10. (Optional) If you select Reference, you can change the number of reference copies. The default is 2.

- If no reference is selected, the target with the highest dye concentration in the highest channel is used as the reference. For example, if there are targets in all six channels, the target in Channel 6 is used; however, if there are targets only in the first five channels, then Channel 5 is the reference target.

- If more than one reference is selected in the well, checkboxes appear to the right for using the reference in the plot.

Note: Each Reference target enables ratio calculations in the results.

11. Assign the first target an expected signal in a channel.

12. Assign your last target a signal in a channel that was not selected for your first target.

Each target in between will be assigned a signal depending on mixture ratio of the target.

13. Click Apply to confirm all well settings.

Viewing and Adjusting Results

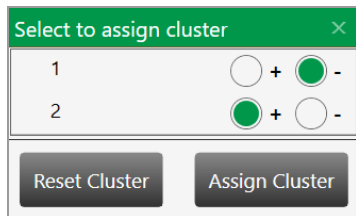
To view and adjust results

1. Click the 2D Amplitude tab.

All droplets are unclassified by default and are colored red. Only the Threshold Cluster mode is available for setting thresholds in Advanced Classification experiments in the 2D plot.

2. Use one of the Threshold Cluster mode tools (box, circle or freehand) to assign droplet clusters to targets.

When you select a droplet cluster, a cluster assignment box appears.



The image shows a dialog box titled "Select to assign cluster" with a close button (X) in the top right corner. It contains two rows, labeled 1 and 2. Each row has two radio buttons: a white one followed by a plus sign, and a green one followed by a minus sign. In row 1, the green minus button is selected. In row 2, the green plus button is selected. At the bottom of the dialog are two buttons: "Reset Cluster" and "Assign Cluster".

Cluster	+	-
1	<input type="radio"/>	<input checked="" type="radio"/>
2	<input checked="" type="radio"/>	<input type="radio"/>

Reset Cluster Assign Cluster

3. Select a radio button to identify the composition of the cluster.

For example, if the cluster is composed of positive droplets for Target-2, select the positive radio button for Target-2 and the negative radio buttons for the remainder.

Important: For calculations to occur, you must have at least one positive cluster for each target, as well as the assigned negative cluster.

Note: The software sets aside unassigned for calculation purposes. If you are unsure of droplet/cluster composition, leave it unassigned (red).

You can view the concentration results in the data table and in the Concentration tab.

If one of your samples was identified as a Reference under AssayType, you can view a ratio of concentration for unknown versus reference in the Ratio tab.

Appendix B Instrument Maintenance

Bio-Rad recommends regular maintenance of your equipment, which includes

- Keeping the surface areas clean, post-run or as necessary

Note: Wipe down external panels and the plate holder with 70% ethanol. See [Cleaning or Decontaminating the Instrument on page 214](#) for information.

- Regularly checking the fluid levels
- Inspecting the equipment for damaged parts or wiring

Bio-Rad also recommends that a Bio-Rad service engineer perform annual preventive maintenance on your instrument. Contact Bio-Rad if your instrument requires service or recalibration.

Fluid Maintenance and Waste Management

You must ensure there is enough oil for at least one full run, and you must empty the waste container when it approaches a full state.

You can verify your fluid levels by checking the following:

- Instrument status bar
- Instrument indicator lights
- The oil and waste bottles

[Table 41](#) explains fluid level maintenance for your instrument.

Table 41. Fluid Levels

Fluid Level	Status
Oil greater than 40% or waste less than 70%	Bottle levels OK. You can start your run.
Oil less than 40% or waste greater than 70%	<p data-bbox="613 506 1320 548">You may be able to start a run with fewer than 96 wells.</p> <p data-bbox="613 554 1320 695">If there is not enough oil in the dispensers to perform a run on the number of wells you set up in the Plate Editor, the software prevents the run until you fill the oil dispenser and empty the waste container.</p> <p data-bbox="613 701 1320 835">Note: Pay attention to the fluid level by checking instrument status bar in the QX Manager Software or by checking the oil bottles.</p>
Oil less than 10% or waste greater than 90%	The software prevents the run until you fill the oil dispenser and empty the waste container.

Changing the Droplet Reader Oil Bottle

To replace the oil bottle

1. Slide the container carriage out using the handle on the side of the instrument.
2. Remove the oil bottle and set it aside, to use when changing the waste bottle.
3. Insert a new bottle of oil and tighten the cap into place.
4. Prime and flush the instrument at least once after the new oil bottle is installed. See [Priming and Flushing the Instrument on page 213](#) for information.

Emptying the Waste Container

Important: Use each empty oil bottle as a new waste bottle once and then discard it. Before discarding, add 50 mL of 10% bleach solution to the bottle to prevent microbial growth, and then label the bottle as waste.

To replace the waste bottle

1. Slide the container carriage out using the handle on the side of the instrument and remove the waste bottle.
2. Place the new Waste label over the Oil label on the oil bottle.
3. Insert the last empty oil bottle into the waste container slot.
4. Flush the instrument after the new waste bottle is installed. See [Priming and Flushing the Instrument on page 213](#) for information.

Managing Waste

You must apply standard Material Safety Data Sheet (MSDS) and OSHA practices when handling and disposing of generated waste. A typical waste profile should contain the following:

Table 42. Managing Waste

Chemical name	Weight %
Fluorinated fluids	>50%
Water	<50%
Bleach	<10%
Other materials (proteins, nucleic acids, fluorescent dye)	<10%

Note: The chemical identities and exact percentages in the above table are withheld as trade secrets.

Bio-Rad droplet generation and reader fluids are based on fluorinated hydrocarbon chemistry, and should be disposed of in accordance with institutional, state and local regulations. Avoid release to the environment and prevent entry into sewer systems or bodies of water. As a disposal alternative, incinerate in a permitted high-temperature waste incineration facility.

These nonflammable fluids are inert and have low environmental impact and low toxicity.

Toxicological Information

Chemical name: (mixture)

Oral LD50: >5000 mg/kg (Rat)

Dermal LC50: >5000 mg/kg (Rabbit)

Inhalation LD50: >8838 ppm (Rat/8-hr)

Ecological Information

Acute aquatic hazard: LC50 > 500 mg/l (96-hr./static; Pimephales promelas/Fathead minnow)

Chronic aquatic hazard: No information available (GHS Chronic 4)

Priming and Flushing the Instrument

You should prime the instrument after you replace the oil bottle and you should prime and flush the instrument when you change the waste bottle.

Important: Before you initiate a prime or flush, *the lid must be closed*. Do NOT press the button to open the lid while a prime or flush is in progress.

To prime the instrument

1. Select the System Utilities tab.
2. Tap Prime.
A message appears, asking if you want to prime the instrument.
3. Tap Yes to begin.
4. A "Performing Prime" message appears.
When the priming process is finished, an operation complete message appears.
5. Tap OK.

To flush the instrument

1. Select the System Utilities tab.
2. Tap Flush.
A message appears, asking if you want to flush the instrument.
3. Tap Yes to begin.
4. A "Performing Flush" message appears.
When the flushing process is finished, an operation complete message appears.
5. Tap OK.

Recovery From Idle Mode

Complete the steps below if the instrument has been idle:

1. Check the expiration date on the oil bottle. If expired, replace the oil bottle.
2. Ensure the waste bottle is not full.
3. If needed, add 50 mL of 10% bleach to the waste bottle.
4. If you have changed the bottles, or if you have not used the system for a week or more, perform the prime and flush processes as specified in [Priming and Flushing the Instrument on page 213](#).

Cleaning or Decontaminating the Instrument

Use the protocols specified below for the following:

- Regular post-run instrument cleanup
- Instrument decontamination, as needed

If you have questions regarding the use of cleaning agents other than those specified, contact Bio-Rad Technical Support.

To clean the instrument

1. Remove the plate after a run is complete and ensure it is properly discarded.
2. Using 70% ethanol, wipe down the external panels, as well as the plate holder and its compartment.

To decontaminate the instrument

1. Remove the plate after a run is complete and ensure it is properly discarded.
2. Wipe down the plate holder and its compartment with 10% bleach.

Important: Do not use bleach inside the system.

3. Wipe down all external surfaces with 10% bleach.
4. Identify the equipment as *Decontaminated*.

Instrument Maintenance Log

[illegible]

Appendix C Troubleshooting

This section contains information on troubleshooting and correcting minor issues you may experience with your instrument or software.

Event Log

The Event Log contains records for all system and user activities, including system errors. If you do not understand, or cannot find an immediate remedy to, the error event, contact Bio-Rad Technical Support.

Instrument Errors

If an error occurs during a run, the software cancels the run and displays an X in the Status column, along with a Run Aborted by System message as the Run End status. [Table 43](#) contains error grouped by type.

1. Tap the X to display the corresponding error code and description.
2. Perform the steps in the solution column before starting a new run.
3. If the suggested solution does not resolve the problem, contact Bio-Rad Technical Support immediately.

Note: The error code and description are also displayed in the Run Information window .

Communication errors can be caused by instrument, connectivity, or power failures.

Table 43. Communication errors

Error Code	Description	Solution
102, 103, 104*, 110*, 111	Communication loss between the instrument and software, detector board error, loss during a run, or unable to reestablish a connection Note: For codes marked with an asterisk (*), some data may be usable.	Disconnect and reconnect the USB cable, and then power cycle the instrument.
302, 308	Communication error involving pump Note: Some data might be usable.	Power cycle the instrument.

Table 43. Communication errors, continued

Error Code	Description	Solution
502	Invalid checksum value	Retry.
900	Invalid command	

An electrical error is typically triggered by an invalid setting.

Table 44. Electrical error codes

Error Code	Description	Solution
406	Electrical Error; invalid power value setting. Note: Some data might be usable.	Power cycle the instrument.

Mechanical errors can be caused by fluid issues (low oil, high waste), instrument operation errors, or binary data collection errors.

Table 45. Mechanical error codes

Error Code	Description	Solution
105, 106	Fluidics or motion response was invalid. Note: Some data may be usable.	Power cycle the instrument.
201	Motors disabled Note: Some data may be usable.	<ol style="list-style-type: none"> 1. Remove and reinsert the plate and plate lid. 2. Check for incorrect or multiple heat seal foil layers. 3. Check for strain on all motor axes. 4. Power cycle the instrument.
205, 207, 210, 211, 212, 218	X, Y, or Z-axis, unable to reach target location or home in allotted time. Note: Some data might be usable.	Check for motor obstructions, and then power cycle the instrument.
301, 303, 304, 305, 307, 312, 316, 317	Pump errors Note: Some data might be usable.	Power cycle the instrument.

Appendix D Ordering Information

This appendix contains descriptions and catalog numbers for new or replacement instruments, accessories, and consumables for Bio-Rad ddPCR products, including your QX600 Droplet Reader.

ddPCR System and Instrument Packages

Table 46. ddPCR system and instrument packages

Product	Description	Catalog Number
QX200 Droplet Digital PCR System	Includes the QX200 Droplet Generator, QX200 Droplet Reader, computer, software, and consumables	1864001
QX600 Droplet Digital PCR System	Includes the QX200 Droplet Generator, QX600 Droplet Reader, computer, software, and consumables	17007769
QX200 AutoDG™ Droplet Digital PCR System	Includes the AutoDG, QX200 Droplet Reader, computer, and QX Manager Software Standard Edition	1864100
QX600 AutoDG Droplet Digital PCR System	Includes the AutoDG, QX600 Droplet Reader, computer, QX Manager Software Standard Edition, and consumables	17008371
Automated Droplet Generator	Includes the AutoDG (1), cooling block accessory (1), oil purge reservoir (1), and power cord (1)	1864101
QX200 Droplet Generator	Includes the droplet generator (1), cartridges and gaskets (24 each), cartridge holders (2), and power cord (1)	1864002
QX200 Droplet Reader	Includes the droplet reader, two plate holders, USB cable, and power cord	1864003
QX600 Droplet Reader	Includes the droplet reader, two plate holders, USB cable, and power cord	12013328

Thermal Cyclers and Plate Sealer

Table 47. Thermal cycler and plate sealer

Product	Description	Catalog Number
C1000™ Touch Thermal Cycler with 96–Deepwell Reaction Module	Includes the C1000 Touch Thermal Cycler chassis, 96–deepwell reaction module, USB flash drive	1851197
PTC Tempo Deepwell Thermal Cycler	Includes the thermal cycler, power cord, USB cable, and Ethernet cable	12015392
PX1 PCR Plate Sealer	PCR plate sealer, includes heat sealing instrument, plate support block that holds 96-well and 384-well plates, sealing frame, power cord	1814000
Piercable Foil Heat Seal	Heat seal for the PX1 PCR Plate Sealer	1814040

AutoDG Accessories

Table 48. Automated Droplet Generator accessories

Product	Description	Catalog Number
Cooling block accessory	Prevents evaporation during droplet generation	12002819
Oil purge reservoir	Collects oil waste from priming and flushing Note: Contact Bio-Rad Technical Support for replacement information.	N/A
Power cord	Connects the AutoDG to the power source Note: Contact Bio-Rad Technical Support for replacement information.	N/A

QX200 Droplet Generator Accessories

Table 49. QX200 Droplet Generator accessories

Product	Description	Catalog Number
DG8 droplet generator cartridges and gaskets	Microfluidic cartridges used to mix sample and oil for droplet generation, and gaskets to seal the cartridges Note: You must seal the cartridge to prevent evaporation and to apply required pressure for droplet generation.	1864007
Droplet generator cartridge holder	Positions and holds the droplet generator cartridge in the instrument compartment	1863051
Power cord	Connects the QX200 Droplet Generator to the power source Note: Contact Bio-Rad Technical Support for replacement information.	N/A

QX600 Droplet Reader Accessories

Table 50. QX600 Droplet Reader accessories

Product	Description	Catalog Number
Computer	Computer that connects to the QX600 Droplet Reader for data collection and analysis	12017458
USB cable and power cord	Cable connecting the computer to the instrument and power cord connecting the instrument to the power source Note: Contact Bio-Rad Technical Support for replacement information.	Included
Droplet reader plate holders (2)	Used to position the 96-well plate in the droplet reader plate compartment	12006834

QX200 Droplet Reader Accessories

Table 51. QX200 Droplet Reader accessories

Product	Description	Catalog Number
Computer	Computer that connects to the QX200 Droplet Reader for data collection and analysis	12017458
USB cable and power cord	Cable connecting the computer to the instrument and power cord connecting the instrument to the power source Note: Contact Bio-Rad Technical Support for replacement information.	Included
Droplet reader plate holders (2)	Used to position the 96-well plate in the droplet reader plate compartment	12006834

ddPCR Consumables and Other Materials

Table 52. Consumables and other materials

Product	Description	Catalog Number
ddPCR 96-Well Plates	Clear well/clear shell semi-skirted plates	12001925 (pkg of 25)
DG32 Droplet Generation Cartridges	Microfluidic cartridge with gasket for the AutoDG	1864108 (pkg of 30) 1864109 (pkg of 60)
Pipet Tips	Pipet tips for the AutoDG	1864120 (pkg of 20) 1864121 (pkg of 40)
Pipet Tip Bins	Pipet tip waste bins for the AutoDG	1864125
Automated Droplet Generation Oils	Automated droplet generation oils for the AutoDG: ■ Automated Droplet Generation Oil for Probes ■ Automated Droplet Generation Oil for EvaGreen®	1864110 1864112
Droplet Generation Oils	Droplet generation oil for the QX200 Droplet Generator: ■ Droplet Generation Oil for Probes, 10 x 7 ml ■ Droplet Generation Oil for EvaGreen®, 2 x 7 ml ■ Droplet Generation Oil for EvaGreen®, 10 x 7 ml	1863005 1864005 1864006
Rainin Pipets	■ 20 µl for sample loading ■ 50 µl for droplet transfer ■ 8-channel, 200 µl for oil	L-20, L8-20 L-50, L8-50 L8-200
Rainin Pipet Tips	Filtered	GP-L10F GP-L200F
Foil Plate Seals for PX1	Pierceable foil plate seals for PX1 Plate Sealer	1814040
ddPCR Droplet Reader Oil	Droplet reader oil for the QX600 Droplet Reader and QX200 Droplet Reader	1863004
Droplet reader waste bottle	You can use an empty droplet reader oil bottle to collect the waste from droplet reading.	N/A

ddPCR Supermixes

Table 53. ddPCR supermixes

Supermix	Description	Catalog number	
ddPCR EvaGreen® Supermix	2x supermix, for use in nucleic acid sample preparation with the QX600 and QX200 ddPCR systems	1864033	2ml (2 x 1ml)
		1864034	5ml (5 x 1ml)
		1864035	25ml (25 x 1ml)
		1864036	50ml (50 x 1ml)
ddPCR Supermix for Probes (no dUTP)	2x supermix, for use in nucleic acid sample preparation with the QX600 and QX200 ddPCR systems	1863023	2ml (2 x 1ml)
		1863024	5ml (5 x 1ml)
		1863025	25ml (25 x 1)
ddPCR Supermix for Probes	2x supermix, for use in sample preparation with the QX600 and QX200 ddPCR systems	1863026	2ml (2 x 1ml)
		1863010	5ml (5 x 1ml)
		1863027	25ml (25 x 1ml)
		1863028	50ml (50 x 1ml)
ddPCR Multiplex Supermix	4x supermix especially suited for probe-based detection of multiple targets in DNA samples using the QX600 and QX200 ddPCR systems	12005909	1.2ml (2 x 0.6ml)
		12005910	3 ml (5 x 0.6 ml)
		12005911	12.5 ml (5 x 2.5 ml),
ddPCR Supermix for Residual DNA Quantification	2x supermix, for use in residual DNA detection with the QX600 and QX200 ddPCR systems	1864037	2ml (2 x 1ml)
		1864038	5ml (5 x 1ml)
		1864039	25ml (25 x 1ml)
		1864040	50ml (50 x 1ml)
One-step RT- ddPCR Advanced Kit for Probes	200 or 500x 20 µl reaction kit, for absolute quantification of target RNA in a one-step format with the QX600 and QX200 ddPCR systems	1864021	2ml (2 x 1ml)
		1864022	5ml (5 x 1ml)

Buffer Controls

Table 54. Buffer controls

Buffer	Catalog Number
ddPCR Buffer Control Kit for Probes	1863052
QX200 Buffer Control Kit for EvaGreen®	1864052

ddPCR Kits

Table 55. ddPCR Kits

Product	Description	Catalog number
ddPCR Mutation Screening Kits		
ddPCR KRAS G12/G13 Screening Kit		1863506
ddPCR KRAS Q61 Screening Kit		12001626
ddPCR BRAF V600 Screening Kit		12001037
ddPCR NRAS Q61 Screening Kit		12001006
ddPCR NRAS G12 Screening Kit		12001094
ddPCR NRAS G12/G13 Screening Kit		12001627
ddPCR EGFR Exon 19 Deletions Screening Kit		12002392
All kits include 20x multiplex assay and 2x ddPCR supermix for probes (no dUTP).		
ddPCR Residual DNA Quantification Kits		
ddPCR CHO Residual DNA Quantification Kit		17000031
ddPCR E.coli Residual DNA Quantification Kit		17000032
200x 20 µl reactions, includes 20x CHO or E.coli RDQ assay and 2x ddPCR Supermix for Residual DNA Quantification.		
ddPCR Copy Number Determination Kits		
ddPCR SMN1 Copy Number Determination Kit		1863500
ddPCR SMN2 Copy Number Determination Kit		1863503
200x 20 µl reactions, includes assay at 20x concentration, 2x ddPCR Supermix for Probes (no dUTP) and positive controls		
ddPCR Library Quantification Kit		
ddPCR Library Quantification Kit for Illumina TruSeq		1863040
200x 20 µl reactions, includes 1 vial of primers and probes at 20x concentration, 2x ddPCR Supermix for Probes (no dUTP) and positive controls		

ddPCR Assays

Table 56. ddPCR Assays

Product	Catalog number	
ddPCR HDR Gene Edit Assay	12002312	100 rxns
ddPCR HDR Gene Edit Assay	12002313	500 rxns
ddPCR HDR Gene Edit Package	12003796	1,000 rxns
ddPCR HDR Ref Assay, Predesigned	12003805	100 rxns
ddPCR HDR Ref Assay, Predesigned	12003806	500 rxns
ddPCR HDR Ref Package, Predesigned	12003793	1,000 rxns
ddPCR NHEJ Gene Edit Assay	12002314	100 rxns
ddPCR NHEJ Gene Edit Assay	12002315	500 rxns
ddPCR NHEJ Gene Edit Package	12003794	1,000 rxns



Bio-Rad
Laboratories, Inc.

Life Science
Group

Website bio-rad.com **USA** 1 800 424 6723 **Australia** 61 2 9914 2800 **Austria** 00 800 00 24 67 23 **Belgium** 00 800 00 24 67 23 **Brazil** 4003 0399
Canada 1 905 364 3435 **China** 86 21 6169 8500 **Czech Republic** 00 800 00 24 67 23 **Denmark** 00 800 00 24 67 23 **Finland** 00 800 00 24 67 23
France 00 800 00 24 67 23 **Germany** 00 800 00 24 67 23 **Hong Kong** 852 2789 3300 **Hungary** 00 800 00 24 67 23 **India** 91 124 4029300 **Israel** 0 3 9636050
Italy 00 800 00 24 67 23 **Japan** 81 3 6361 7000 **Korea** 82 080 007 7373 **Luxembourg** 00 800 00 24 67 23 **Mexico** 52 555 488 7670
The Netherlands 00 800 00 24 67 23 **New Zealand** 64 9 415 2280 **Norway** 00 800 00 24 67 23 **Poland** 00 800 00 24 67 23 **Portugal** 00 800 00 24 67 23
Russian Federation 00 800 00 24 67 23 **Singapore** 65 6415 3188 **South Africa** 00 800 00 24 67 23 **Spain** 00 800 00 24 67 23 **Sweden** 00 800 00 24 67 23
Switzerland 00 800 00 24 67 23 **Taiwan** 886 2 2578 7189 **Thailand** 66 2 651 8311 **United Arab Emirates** 36 1 459 6150 **United Kingdom** 00 800 00 24 67 23