QX ONE Droplet Digital PCR System and QX ONE Software

User Guide

Version 1.2
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## Revision History

<table>
<thead>
<tr>
<th>Document</th>
<th>Date</th>
<th>Description of Change</th>
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</table>
| QX ONE Droplet Digital PCR System and QX ONE Software Instrument Guide | December 2021 | Update with v1.2 enhancements, including the following:  
- Ability to cancel, recover, and reorder plates  
- New user privilege  
- Enhanced data recovery  
- Troubleshooting function |
| QX ONE Droplet Digital PCR System and QX ONE Software Instrument Guide | October 2020 | Add topics for the following:  
- Aspirating the waste reservoir  
- Loading the plate into the centrifuge  
- Recovering the plate from a failed run |
Table of Contents

Revision History ...................................................................................................................... iii

Safety and Regulatory Compliance .................................................................................... 9
  Safety Warning Labels ........................................................................................................ 9
  Safe Use Specifications ...................................................................................................... 10
  Regulatory Compliance ..................................................................................................... 11
  Hazards ............................................................................................................................. 13
    Biohazards ...................................................................................................................... 13
    General Precautions ....................................................................................................... 13
  Surface Decontamination ................................................................................................. 14
    Chemical Hazards ........................................................................................................... 14
    Explosive or Flammability Hazards ............................................................................... 14
    Electrical Hazards .......................................................................................................... 15
  Decommissioning and Disposal ....................................................................................... 15
  Transport ........................................................................................................................... 15
  Warranty ............................................................................................................................ 15

Chapter 1 Introduction to Droplet Digital PCR ................................................................... 17
  ddPCR Workflow ............................................................................................................... 18
  Droplet Generation .......................................................................................................... 19
  PCR Amplification ............................................................................................................. 19
  Droplet Reader .................................................................................................................. 19
  Finding Out More ............................................................................................................. 19

Chapter 2 QX ONE Droplet Digital PCR System ............................................................... 21
  General Operating Instructions and Routine ................................................................... 22
    Turning on the Instrument ............................................................................................... 22
    Replacing Oil and Waste Bottles .................................................................................... 23
    Turning off the Instrument ............................................................................................. 25
  Instrument Specifications ................................................................................................. 26
  Environmental Requirements ............................................................................................ 27
Table of Contents

QX ONE Droplet Digital PCR System Components .......................................................... 28

Chapter 3 Getting Started ............................................................................................. 29
  Signing In, Viewing Privileges, and Changing Preferences ........................................ 29
    Signing into the Software ......................................................................................... 30
    Viewing Your User Privileges .................................................................................. 32
    Managing Your Preferences ...................................................................................... 34
    Signing Out or Changing Users ................................................................................. 36
  About QX ONE Software ........................................................................................... 37
  Instrument Status Bar .................................................................................................. 39
    Tabs to Functional Windows ..................................................................................... 40
    Compatible File Types .............................................................................................. 41
    Touch Screen Differences ........................................................................................ 42

Chapter 4 Preparing a Sample Experiment ................................................................... 43
  Required Components ................................................................................................. 43
  Creating the Sample Mix ............................................................................................. 44
  Filling the GCR96 Cartridge ....................................................................................... 45
  Sealing the GCR96 Cartridge ...................................................................................... 46
  Loading the GCR96 Cartridge into a Centrifuge ......................................................... 47

Chapter 5 Adding Plates in the QX ONE ..................................................................... 49
  Adding the Plate .......................................................................................................... 52
  Plate Configuration Window ....................................................................................... 55
    Plate Information Tab ............................................................................................... 56
    Well Selection Tab ................................................................................................... 58
    Well Information Tab ............................................................................................... 60
  Experiment Types ....................................................................................................... 62
  Sample Descriptions .................................................................................................. 63
  Sample Types .............................................................................................................. 63
  Supermixes .................................................................................................................. 64
  Assay Types and Fluorophores ................................................................................... 65
  Creating or Modifying Plate Templates ...................................................................... 67
  Creating or Modifying Protocol Templates ................................................................. 68

Chapter 6 Running Experiments .................................................................................. 69
  Using the One Step RT ddPCR Advanced Kit for Probes ........................................ 69

vi  QX ONE Droplet Digital PCR System and QX ONE Software
<table>
<thead>
<tr>
<th>Chapter 7 Data Analysis Overview</th>
<th>81</th>
</tr>
</thead>
<tbody>
<tr>
<td>Data Analysis Module</td>
<td>82</td>
</tr>
<tr>
<td>Analysis Dashboard</td>
<td>83</td>
</tr>
<tr>
<td>Plate View Windows</td>
<td>83</td>
</tr>
<tr>
<td>Viewing Run and Lot Information</td>
<td>84</td>
</tr>
<tr>
<td>Analysis Reporting</td>
<td>85</td>
</tr>
</tbody>
</table>

<table>
<thead>
<tr>
<th>Chapter 8 System Utilities</th>
<th>87</th>
</tr>
</thead>
<tbody>
<tr>
<td>System Settings Tab</td>
<td>87</td>
</tr>
<tr>
<td>Preferred Location</td>
<td>88</td>
</tr>
<tr>
<td>Shared Settings</td>
<td>89</td>
</tr>
<tr>
<td>System Log Files</td>
<td>90</td>
</tr>
<tr>
<td>Event Log</td>
<td>90</td>
</tr>
<tr>
<td>Maintenance Log</td>
<td>91</td>
</tr>
<tr>
<td>Maintenance Reports</td>
<td>93</td>
</tr>
<tr>
<td>System Data</td>
<td>94</td>
</tr>
<tr>
<td>Archiving Data</td>
<td>94</td>
</tr>
<tr>
<td>Troubleshooting Data</td>
<td>95</td>
</tr>
<tr>
<td>Recovering Data</td>
<td>96</td>
</tr>
<tr>
<td>System Calibration</td>
<td>97</td>
</tr>
</tbody>
</table>

<table>
<thead>
<tr>
<th>Chapter 9 Managing Users</th>
<th>99</th>
</tr>
</thead>
<tbody>
<tr>
<td>Adding Users</td>
<td>101</td>
</tr>
<tr>
<td>Adding or Removing User Privileges</td>
<td>102</td>
</tr>
<tr>
<td>Modifying User Preferences</td>
<td>105</td>
</tr>
<tr>
<td>Removing Users</td>
<td>105</td>
</tr>
</tbody>
</table>

<table>
<thead>
<tr>
<th>Appendix A Instrument Maintenance</th>
<th>107</th>
</tr>
</thead>
<tbody>
<tr>
<td>General Maintenance</td>
<td>107</td>
</tr>
</tbody>
</table>
# Table of Contents

- Waste Removal ........................................................................................................... 108
- Vent Reservoir Maintenance .................................................................................... 109
  - Aspirating the Vent Reservoir .............................................................................. 110
  - Cleaning and Decontamination of the QX ONE Droplet Digital PCR System ........ 112

**Appendix B Additional Computers** ............................................................................. 115
- Computer Requirements ............................................................................................. 115
- Installing or Updating the Software ........................................................................ 116

**Appendix C Ordering Information** ............................................................................ 117
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.

Safety Warning Labels

Warning labels posted on the instrument and in this manual warn you about sources of injury or harm. Table 1 defines each safety warning label.

Table 1. Meaning of safety warning labels

<table>
<thead>
<tr>
<th>Icon</th>
<th>Meaning</th>
</tr>
</thead>
</table>
| !     | **Warning about risk of harm to body or equipment**
|      | Operating the QX ONE Droplet Digital PCR 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 burning**
|      | A thermal cycler generates enough heat to cause serious burns. Wear safety goggles or other eye protection at all times during operation, and do not try to open the front door. Always allow maximum clearance to avoid accidental skin burns. |
| ♂♀   | **Warning about pinch or crush risk**
|      | If the instrument is used in certain states, there may be pinch/crush hazards present. Ensure the instrument is installed on a level, flat top lab bench or sturdy table that is large enough for the entire instrument. |
Table 1. Meaning of safety warning labels, continued

<table>
<thead>
<tr>
<th>Icon</th>
<th>Meaning</th>
</tr>
</thead>
</table>
| ![Flash](image) | **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 QX ONE Droplet Digital PCR System, Bio-Rad Laboratories, Inc. strongly recommends that you comply with instructions listed in this section and in Instrument Maintenance on page 107.

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 27.
Regulatory Compliance

The QX ONE Droplet Digital PCR System has been tested and found to be in compliance with all applicable requirements of the following safety and electromagnetic standards:

- EC 61010-2-010:2014, Safety requirements for electrical equipment for measurement, control, and laboratory use - Part 2-010: Particular requirements for laboratory equipment for the heating of materials
- 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
- CAN/CSA C22.2 No 61010-1-04, Safety requirements for electrical, equipment for measurement, control, and laboratory use, Part 1: General requirements
- 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
Safety and Regulatory Compliance

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.

The CE symbol indicates that the manufacturer ensures the product conforms with the essential requirements of the applicable EN directives.

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.

Note: 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.

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.
Hazards

The QX ONE Droplet Digital PCR System 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 QX ONE Droplet Digital PCR System.

Biohazards

The QX ONE Droplet Digital PCR System 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.

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.
- 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.
- Change gloves frequently. 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).
Surface Decontamination

**WARNING!** To prevent electrical shock, always turn off and unplug the instrument prior to 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 generation, thermal cycling, and droplet reading surfaces
- Control panel and display

To prepare and apply the disinfectant, refer to the instructions provided by the product manufacturer. For more information on surface cleaning, see Appendix A, Instrument Maintenance. For questions regarding the use of other cleaning agents, contact Bio-Rad Technical Support.

**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.

**Disposal of Biohazardous Material**

Dispose of the following potentially contaminated materials in accordance with laboratory local, regional, and national regulations:

- Clinical samples
- Reagents
- Used reaction vessels or other consumables that may be contaminated

**Chemical Hazards**

The QX ONE Droplet Digital PCR System contains no potentially hazardous chemical materials.

**Explosive or Flammability Hazards**

The QX ONE Droplet Digital PCR System poses no uncommon hazard related to flammability or explosion when used in a proper manner as specified by Bio-Rad.
**Electrical Hazards**

The QX ONE Droplet Digital PCR System poses no uncommon electrical hazard to operators if installed and operated properly without physical modification and connected to a power source of proper specification.

**Decommissioning and Disposal**

The QX ONE Droplet Digital PCR System 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.

To decontaminate the QX ONE Droplet Digital PCR System before decommissioning and disposal, see Cleaning and Decontamination of the QX ONE Droplet Digital PCR System on page 112.

**Transport**

You must perform the specified decontamination procedures before moving or shipping the QX ONE Droplet Digital PCR 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**

The QX ONE Droplet Digital PCR System 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.

This 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.
Safety and Regulatory Compliance
Chapter 1 Introduction to Droplet Digital PCR

Droplet digital polymerase chain reaction (ddPCR) is a digital PCR 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 ±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 approximately 20,000 uniform nanoliter-sized droplets, with target and background DNA distributed randomly into the droplets during the partitioning process.

- Following droplet generation, the droplets are amplified 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. Positive droplets containing at least one copy of the target DNA molecule exhibit increased fluorescence compared to negative droplets.
Droplet Generation

Before droplet generation, ddPCR reactions are prepared in a similar manner as real-time PCR reactions that use TaqMan hydrolysis probes labeled with reporter fluorophores or an intercalating dye. ddPCR is performed with Bio-Rad’s proprietary reagents that were developed specifically for droplet generation.

The QX ONE Droplet Digital PCR System runs the plates containing your DNA samples through the Droplet Generator as the first component in the three-part process. The Droplet Generator uses specially developed reagents and microfluidics to partition each sample into approximately 20,000 nanoliter-sized droplets. Target and background DNA are distributed randomly into the droplets during the partitioning process. Droplet generation produces uniform droplets for the sample, enabling precise target quantification.

PCR Amplification

After droplet generation, the plates are moved through the internal thermal cycler, which performs repeated heating and cooling processes. This amplifies one or more copies of a particular DNA segment, thereby multiplying strands of DNA sequencing into the thousands or millions.

Droplet Reader

Following PCR amplification of the nucleic acid target in the droplets, the plate moves to the Droplet Reader, which analyzes each droplet individually using a -color detection system, depending on the number of channels specified in the experiment.

The autosampler in the Droplet Reader picks up the droplets from each well in the plate. The droplets are spaced out individually for fluorescence reading and fluorescence is then measured in each droplet. Positive droplets, which contain at least one copy of the target DNA molecule, exhibit increased fluorescence compared to negative droplets and are presented in color in the analysis displays. Negative droplets are presented in grayscale.

Finding Out More

Tap the Help tab (2) on the left, and then tap the Bio-Rad Website link 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 1 Introduction to Droplet Digital PCR

QX ONE Droplet Digital PCR System and QX ONE Software
Chapter 2 QX ONE Droplet Digital PCR System

The QX ONE Droplet Digital PCR System combines droplet generation, thermal cycling, and droplet reading technologies into a single instrument, as described below:

- The droplet generator partitions samples into approximately 20,000 uniform droplets.
- The internal thermal cycler performs repeated heating and cooling processes to amplify one or more copies of a particular DNA segment in each droplet.
- The droplet reader streams the droplets in single file, and then counts the fluorescent positive and negative droplets to calculate and quantify target DNA concentration.

The QX ONE Droplet Digital PCR System also features a touch screen computer, which is integrated onto the instrument and has QX ONE Software already installed.

When you open QX ONE Software from the touch screen computer, the software automatically recognizes the QX ONE Droplet Digital PCR System.

**Note:** You cannot directly connect standalone computers to the instrument. However, from a standalone computer you can access files stored on the touch screen computer’s internal drive, or from another shared location, as long as all units are connected through the same company network. You can also access the data with a USB drive connected to the touch screen.
General Operating Instructions and Routine

**Important:** The QX ONE Droplet Digital PCR System must be installed and calibrated by a Bio-Rad service engineer.

Note the following requirements:

- Your instrument should be placed on a solid surface and away from other instruments that can cause vibration.
- To ensure adequate ventilation, leave a minimum of 4 in (10 cm) of clear space behind, and a minimum of 14 in (35.5 cm) of clear space to the right and left of the instrument.
- Position the instrument where the power cord and power source can be easily accessed when it is time for equipment service and maintenance.
- Use only the cord provided with the instrument to connect to the power source.

**Turning on the Instrument**

**Note:** If you are restarting the instrument after turning it off, ensure at least 5 min have passed before turning the instrument on again.

**To turn on the instrument**

1. Press the switch on the back of the instrument to power on the QX ONE Droplet Digital PCR System.
2. Press the power button on the front of the instrument. The button flashes green to indicate power is on and the system is initializing.
3. Press the switch on the side of the touch screen to power on the computer.
4. Wait 5 min, and then launch QX ONE Software on the touch screen.
5. Log in with your user name and password. For information, see Signing into the Software on page 30.
6. Wait until the instrument status changes from Busy to Ready (between 2 and 2.5 min).

**Important:** Do not touch the instrument or touch screen while the instrument is busy initializing.

**Note:** If the instrument does not fully initialize after 10 min, close the software, turn off the computer, and then turn off the instrument. Wait 2 min, and then repeat steps 2, 3, and 4.
Replacing Oil and Waste Bottles

Ensure that you have enough QX ONE Droplet Digital PCR System Droplet Generation Oil and Droplet Reader Oil in the oil bottles, and enough space in the waste bottles, to complete your experiment.

**Important:** Use the Bio-Rad QX ONE Droplet Digital PCR System Droplet Generation Oil for Probes and Droplet Reader Oil. For catalog numbers, see Appendix C, Ordering Information.

The instrument status bar on the QX ONE Software main screen displays oil bottle levels. If current levels are not sufficient for the scheduled run, the instrument initiates an alert and QX ONE Software displays an advisory message to replace one or more bottles.

**Important:** If an error message appears (error #111), stating that there is not enough Droplet Reader Oil in the bottle to run the loaded plates, you must add a bottle or switch the bottle within 15 minutes. If the time elapsed exceeds 15 minutes before bottle replacement, the instrument might fail processing. For instances when the bottle has not been replaced within 15 minutes, Bio-Rad recommends restarting the QX ONE Droplet Digital PCR System.

Always use a full bottle as a replacement. Because the waste bottles fill up more quickly than the oil bottles are depleted, replace the existing waste bottle with an empty waste bottle whenever you change a droplet oil bottle.

As shown in the following graphics, the instrument features two slots for the droplet generation oil bottles (small spaces marked with a “P”) and two slots for the droplet reader oil bottles (larger spaces). Leave the two additional slots on the right (marked with “E”) empty.
The QX ONE Droplet Digital PCR System Droplet Generation Oil for Probes and Droplet Reader Oil bottles are prefilled and sealed with instrument-specific caps, as shown in the following graphics:

The notched top helps with bottle alignment and insertion. The RFID label allows the instrument to recognize each bottle as correct for its position, and monitor the oil levels in each bottle.

**To release an empty bottle from its position**

1. Firmly push the bottle down to the bottom of the slot and then release. The bottle pops up from the slot.
2. When the bottle pops up from the slot, lift it out of its position.

**To insert a bottle into its corresponding position**

1. Turn the bottle upside down so the notched cap is facing down.
2. Slowly twist the bottle until the notched cap clicks into place.
3. Firmly push down on the bottle to the bottom of the slot to snap it into place. When secured, the bottle should not be easily lifted out of position and should not twist in place.
Turning off the Instrument

Bio-Rad recommends turning off and restarting the instrument one day a week. When you turn off the instrument, wait at least 5 min before turning it back on.

To turn off the instrument

1. Close QX ONE Software on the touch screen, and then turn off the touch screen, as follows:
   a. Tap the Windows icon in the lower-left corner.
   b. Tap the Power icon and then select Shut down.
   c. Wait until the touch screen has powered off.
2. Turn off the power switch on the back of the instrument.

   Note: Wait at least 5 min before turning the instrument on again.
Chapter 2 QX ONE Droplet Digital PCR System

Instrument Specifications

Table 2. QX ONE Droplet Digital PCR System instrument specifications

<table>
<thead>
<tr>
<th>Element</th>
<th>Specifications</th>
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</thead>
<tbody>
<tr>
<td>Size (W x D x H)</td>
<td>122 cm (48 in) x 66 cm (26 in) x 38 cm (15 in)</td>
</tr>
<tr>
<td></td>
<td>76 cm (30 in) top of monitor</td>
</tr>
<tr>
<td>Weight</td>
<td>100 kg (220 lb) (includes the monitor but not reagents)</td>
</tr>
<tr>
<td>Capacity</td>
<td>5 plates</td>
</tr>
<tr>
<td>Samples</td>
<td>Up to 96 per plate</td>
</tr>
<tr>
<td>Partitions per sample</td>
<td>Approximately 20,000 droplets</td>
</tr>
<tr>
<td>Detection channels</td>
<td>4 (FAM, HEX, Cy5, and Cy5.5), plus scatter channel</td>
</tr>
<tr>
<td>Thermal gradient</td>
<td>Yes</td>
</tr>
<tr>
<td>Onboard computer</td>
<td>24 in, 1920 x 1080 resolution</td>
</tr>
<tr>
<td></td>
<td>Holds data for approximately 100 plates</td>
</tr>
<tr>
<td>Time to results</td>
<td>6.5 hours for the first plate, and approximately 4 hours for each plate</td>
</tr>
<tr>
<td></td>
<td>thereafter, based on a standard 2-step ddPCR cycling protocol</td>
</tr>
<tr>
<td>Benchtop requirements</td>
<td>The following requirements apply to the benchtop or table on which the</td>
</tr>
<tr>
<td></td>
<td>instrument will be installed.</td>
</tr>
<tr>
<td></td>
<td>The benchtop or table must be 76 in W x 30 in D x 50 in H (193 cm x 76 cm x</td>
</tr>
<tr>
<td></td>
<td>126 cm), single benchtop rated for more than 250 lbs (114 kg) load. Ensure</td>
</tr>
<tr>
<td></td>
<td>there is enough room above the instrument to ensure ergonomic safety.</td>
</tr>
<tr>
<td></td>
<td><strong>Important</strong>: Do not place instrument over multiple benchtop surfaces.</td>
</tr>
<tr>
<td></td>
<td>Level within approximately 1” about each axis.</td>
</tr>
</tbody>
</table>
## Environmental Requirements

Table 3 lists the environmental requirements for Bio-Rad’s QX ONE. The supplied shielded cables must be used with these instruments to ensure compliance with the Class A FCC limits.

### Table 3. Conditions for safe use

<table>
<thead>
<tr>
<th>Usage aspect</th>
<th>Requirements</th>
</tr>
</thead>
<tbody>
<tr>
<td>Rated input power</td>
<td>Input: 100–240 V, 50–60 Hz, 10A-4.6A plugs into standard AC receptacle</td>
</tr>
<tr>
<td>Fuses</td>
<td>10A 250V SLOW BLOW</td>
</tr>
<tr>
<td>Voltage fluctuations</td>
<td>± 10% for the included external power supply</td>
</tr>
<tr>
<td></td>
<td><strong>Note</strong>: Use only the power cord supplied with the equipment.</td>
</tr>
<tr>
<td>Pollution degree</td>
<td>2</td>
</tr>
<tr>
<td>Usage temperature</td>
<td>18–30°C</td>
</tr>
<tr>
<td>Relative humidity</td>
<td>20 to 80% non-condensing</td>
</tr>
<tr>
<td>Altitude</td>
<td>0 to 8,200 ft (0 to 2,500 meters) above sea level</td>
</tr>
<tr>
<td>Installation category</td>
<td>II (external power supply plugs into a standard AC receptacle)</td>
</tr>
<tr>
<td></td>
<td>Indoor use only</td>
</tr>
<tr>
<td>Ventilation requirements</td>
<td>The following distances should be unobstructed for proper ventilation:</td>
</tr>
<tr>
<td></td>
<td>- 14 in (35.5 cm) on the left and right sides of the instrument</td>
</tr>
<tr>
<td></td>
<td>- 4 in (10 cm) behind the instrument</td>
</tr>
</tbody>
</table>
QX ONE Droplet Digital PCR System Components

The QX ONE Droplet Digital PCR System ships with the following items:

- The instrument, which performs the sequential ddPCR processes
- Attached touch screen, which is a computer that displays the software interface and connects to the instrument
- QX ONE Software, which is installed on the instrument touch screen computer, and provides a user interface to
  - Control the instrument
  - Set up assays
  - Collect analysis data
  - Recover plates from run failures
  - Store analysis data
  - Display analysis data

- QX ONE Droplet Digital PCR System Accessory Kit, which includes the following:
  - PX1 GCR Sealer Support Block
  - QX ONE Droplet Digital PCR System Waste Bottles
  - Keyboard
  - Power cord (compatible within North America)
  - Warranty card

For information on additional ddPCR components and supplies, see Appendix C, Ordering Information.
Chapter 3 Getting Started

Use the information in this chapter to

- Sign into the software or change users
  
  **Important:** If the User Management module was not enabled during the QX ONE Software Standard Edition installation, a single generic user is added and functions associated with the User tab are not available.

- Become familiar with the main functional windows

- Understand the compatible file types for your software edition

- Learn the differences between the software installed on the instrument touch screen versus a standalone computer

**Tip:** If QX ONE Software goes into standby mode after being idle, tap 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 ONE Software, and communicates the information to you. Note the following:

- You cannot change your user privileges unless you are assigned the Add/Manage Users privilege.

- You can change your user preferences, which are set to defaults. For information, see Managing Your Preferences on page 34.

You can access partial preferences only, using the generic user link, as shown in the following graphic. See Partial Preferences on page 35 for more information.

**Note:** If you did not enable the User Management module during the software installation, QX ONE Software creates a generic user, which is already logged in, and is available to all users for setting up and running plates.
Signing into the Software

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

**Note:** In the Standard Edition, unless User Management is enabled during the software installation, the generic user is already logged in when you open the software.

To sign in

1. Tap the QX ONE Droplet Digital PCR System touch screen to open 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.
If the label is blank or shows a different domain then enter it as `<network or local computer name\user name>.

**Note:** If you are logging into the network and you don’t know the network domain name, contact your system administrator.

3. Enter the password, and then tap Sign in.

On first use, you must agree to the end user license agreement in order to use the software.

**Note:** The Do not show EULA on sign in checkbox is selected by default. 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.

4. Tap I Agree.

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

The Add Plate window opens on the instrument touch screen.

If you are opening the software on a standalone computer, the Data Analysis window opens.
Chapter 3 Getting Started

Viewing Your User Privileges

Important: If you did not enable the User Management module during the installation, the related functions are not available.

In the User Setup and Preferences window, your privileges are identified by the selected checkboxes.

To view your assigned privileges

> Select the User Setup and Preferences tab.

![User Management Screen](image)

Checkboxes in the display are not enabled unless you are assigned the Add/Manage Users privilege.

Table 4 lists the available privileges, which can be assigned in any combination.

Table 4. User privileges

<table>
<thead>
<tr>
<th>Privilege</th>
<th>Purpose</th>
</tr>
</thead>
<tbody>
<tr>
<td>Add and manage users</td>
<td>Add or remove users, set privileges, and change preferences. 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.</td>
</tr>
<tr>
<td>Create new templates</td>
<td><strong>Save a design for a plate, thermal cycling protocol, or analysis report as a template.</strong></td>
</tr>
</tbody>
</table>
### Table 4. User privileges, continued

<table>
<thead>
<tr>
<th>Privilege</th>
<th>Purpose</th>
</tr>
</thead>
<tbody>
<tr>
<td>View data files (created by other users)</td>
<td>View files created by other users.</td>
</tr>
<tr>
<td>Overwrite existing data file name</td>
<td>Use Save or Save As capability.</td>
</tr>
<tr>
<td></td>
<td>Selecting Save replaces the original file content with any changes made by the user, without changing the file name.</td>
</tr>
<tr>
<td></td>
<td>Selecting Save As allows the user to save either existing or new content with a different file name.</td>
</tr>
<tr>
<td></td>
<td><strong>Note</strong>: Users without this permission can open files and perform analysis but cannot save changes.</td>
</tr>
<tr>
<td>System settings</td>
<td>View logs, and view and modify the preferred or shared data file and template locations.</td>
</tr>
<tr>
<td></td>
<td><strong>Note</strong>: All users can view the file locations.</td>
</tr>
<tr>
<td></td>
<td><strong>Important</strong>: if your system administrator selected the Preferred Locations checkbox in System Settings, individual user location preferences are overridden.</td>
</tr>
<tr>
<td>Maintenance</td>
<td>View instrument maintenance information, such as the maintenance log and maintenance reports.</td>
</tr>
<tr>
<td>System Data</td>
<td>Move raw data from the touch screen computer to free up disk space for run files, access troubleshooting data, and recover data from a temporary file created for a failed run.</td>
</tr>
<tr>
<td>Access the Module Recovery Tool</td>
<td>For manual plate recovery, you can specify the processes to be rerun.</td>
</tr>
<tr>
<td>Manage Plate</td>
<td>You can cancel runs on the QX ONE Droplet Digital PCR System and you can reorder plates that are still in the instrument Inbox.</td>
</tr>
</tbody>
</table>
Managing Your Preferences

Use the User Setup and Preferences window to modify your personal user preferences.

Important: If you enabled User Management during the software installation, the functionality discussed here is available. If not, the User Management module is not included and a truncated set of preferences is available. See Partial Preferences on page 35 for information.

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.

Important: If your administrator has enabled Preferred Locations in System Settings, storage folder locations in User Preferences are overridden, and all files are saved to the preferred folder paths.

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.

2. Do one of the following:
   - If you are setting preferences for a new user, tap Add.
   - If you are updating your own preferences, tap Save.

3. When the confirmation message appears, tap Yes to save the changes, and then tap OK.
Partial Preferences

If User Management was not enabled during the software installation, you can still access partial functionality.

To open the menu

1. Click the user name link in the upper-right corner to display the pop-up.

2. Click User Preferences to display the dialog box.

3. You can change the following:
   - System time out in minutes (default is 5 minutes)
   - Number of plates in the completed section of the status screen (default is 7)

4. After you have made your changes, tap Save.
Chapter 3 Getting Started

Signing Out or Changing Users

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

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

**Important:** 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. Tap the user name link in the upper-right corner and select Sign Out.
2. Tap Yes to confirm.

   If there are unsaved changes, QX ONE Software displays a prompt.
   - To discard the changes and proceed, tap Yes.
   - To cancel the sign out, tap No. Save the changes, and then repeat Steps 1 and 2.

**To change users**

1. Tap 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.

   ![Sign in window](image)

   - 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 tap Sign in.
About QX ONE Software

QX ONE Software, as part of your QX ONE Droplet Digital PCR System, provides all necessary functionality to create, run, and analyze ddPCR experiments on your samples.

Note: In the QX ONE guides, you are instructed to tap options on the touchscreen. If you are using QX ONE Software with a keyboard and mouse, click the options instead. If QX ONE Software goes into standby mode after being idle, tap in the lockout screen to sign into the software again and display the last active window.

This instrument guide contains abridged information on QX ONE Software. For information on all software capabilities, refer to the QX ONE Software User Guide, Standard Edition or Regulatory Edition.

The following image highlights functional areas:

LEGEND

1. The status bar displays information about the instrument and user.

2. Tabs provide access to the main functional windows.

3. The main pane displays the details of the selected tab.

A Bio-Rad service engineer sets up your QX ONE Droplet Digital PCR System instrument and installs QX ONE Software onto the touch screen computer. Your installation includes activation of a primary superuser account, to be used by a system administrator in your organization. The superuser is responsible for setting up additional users and assigning user privileges as required for regulatory compliance. For information, see Managing Users on page 99.
Note: If you will be installing the software on additional computers to use for ddPCR file analysis, see Appendix B, Additional Computers.

Using the instrument and software, you can

- Set up customized ddPCR experiments for plate runs in the QX ONE Droplet Digital PCR System
- Create and store templates for plate layouts, thermal cycling protocols, and reports.
- Use the live analysis function during the droplet reading phase
- With the Manage Plate user privilege, cancel runs and reorder plates while they are in the instrument Inbox
- Analyze your data files in a variety of charts and tables in the Analysis module
- Generate reports on your data
- Produce system and experiment audit logs to ensure clear audit trails for activities and consumable/reagent use through the Event Log, Data Archive, and automatic Lot Management functions (Regulatory Edition only)
- In the Regulatory Edition, safeguard user access with secure logins and traceable user activity
- Restrict template access to ensure the use of approved templates only

Note: When you open QX ONE Software on the touch screen, the Add Plate window appears by default. If you open the software from a standalone computer, the Data Analysis window appears by default and the Add Plate and Run Status tabs are not available.
Instrument Status Bar

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

1. Software name and version
2. Instrument name and status information
3. Number of runs in progress and runs available
4. Oil and waste levels
5. Open or close the Inbox or Outbox
6. Used and free disk space
   - Storage availability is displayed in GB and by number of plates that you can run before running out of space on the touch screen computer.
   - **Note**: When QX ONE Software detects low disk space, the software displays an advisory prompt to archive data.
7. Name of the current user, along with the current date and time
8. Closes the QX ONE Software application

**LEGEND**
**Tabs to Functional Windows**

*Table 5* describes the functional areas and primary windows in QX ONE Software. For more information, refer to the QX ONE Software User Guide.

**Table 5. Window Tabs**

<table>
<thead>
<tr>
<th>Tab</th>
<th>Name</th>
<th>Purpose</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Add Plate</td>
<td>- Add and configure a plate and protocol for a ddPCR run.</td>
</tr>
<tr>
<td></td>
<td></td>
<td>- Recover a plate or run data from a failed run.</td>
</tr>
<tr>
<td></td>
<td>Run Status</td>
<td>- In the top pane, view the runs in progress.</td>
</tr>
<tr>
<td></td>
<td></td>
<td>- In the bottom pane, view up to 100 runs (finished, failed, recovered).</td>
</tr>
<tr>
<td></td>
<td>Data Analysis</td>
<td>- Access the Data Analysis and Gene Study modules.</td>
</tr>
<tr>
<td></td>
<td></td>
<td><strong>Note:</strong> In QX ONE Software, Gene Study is available on a standalone computer, but not from the touch screen.</td>
</tr>
<tr>
<td></td>
<td>Template Setup</td>
<td>- Set up templates for plates thermal cycling protocols, and reports, and search for existing template files.</td>
</tr>
<tr>
<td></td>
<td>System Utilities</td>
<td>- View storage locations, logs, and maintenance reports.</td>
</tr>
<tr>
<td></td>
<td></td>
<td>- Archive raw data, access troubleshooting information, and recover data from a temporary file created for a failed run.</td>
</tr>
<tr>
<td></td>
<td></td>
<td><strong>Important:</strong> Available functionality depends on your assigned user privileges. Instrument calibration is available only to the Bio-Rad service engineer.</td>
</tr>
<tr>
<td></td>
<td>Users and</td>
<td>- View the user privileges you have been assigned, and view and modify your personal preferences.</td>
</tr>
<tr>
<td></td>
<td>Preferences</td>
<td>- Create, edit, or remove QX ONE Software users (if you are assigned the Add/Manage Users privilege).</td>
</tr>
<tr>
<td></td>
<td></td>
<td><strong>Important:</strong> In Standard Edition, user functionality is available only if the User Management was enabled during software installation.</td>
</tr>
<tr>
<td></td>
<td>Help</td>
<td>- Access software version information, current calibration values, the End User License Agreement, the Bio-Rad website, open source software license information</td>
</tr>
</tbody>
</table>
Compatible File Types

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

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

- You can open files created using Regulatory Edition only in Regulatory Edition.
- If you open a .qlp or .ddpcr file in QX ONE Software, Standard Edition, the file opens and is saved as a .ddpcrone file.
- If you open a .qlps or .ddpcrs file in QX ONE Software, Regulatory Edition, the file opens and is saved as a .ddpcrsone file.

Table 6. Compatible file types

<table>
<thead>
<tr>
<th>File Type</th>
<th>Extension</th>
<th>Details</th>
</tr>
</thead>
<tbody>
<tr>
<td>Plate</td>
<td>.ddplt</td>
<td>Plate template file containing setup details to perform experiments; this file type opens in the Plate Editor.</td>
</tr>
<tr>
<td>Protocol</td>
<td>.ddthp</td>
<td>Protocol template file containing setup details to perform experiments; this file type opens in the Protocol Editor.</td>
</tr>
<tr>
<td>Data</td>
<td>.ddcrone</td>
<td>Contains the results of an experiment run performed on the QX ONE Instrument using QX ONE Software, Standard Edition.</td>
</tr>
<tr>
<td>Data</td>
<td>.ddcrsone</td>
<td>Contains the results of an experiment run performed on the QX ONE Instrument using QX ONE Software, Premium Edition.</td>
</tr>
</tbody>
</table>
# Touch Screen Differences

As shown in Table 7, QX ONE Software functionality varies between the QX ONE Droplet Digital PCR System touch screen and a standalone computer.

Table 7. Touch Screen Differences

<table>
<thead>
<tr>
<th>Feature</th>
<th>Instrument Touch Screen</th>
<th>Standalone Computer</th>
</tr>
</thead>
<tbody>
<tr>
<td>Gene Study</td>
<td>Not available</td>
<td>Available</td>
</tr>
<tr>
<td>Files open</td>
<td>Only one file at a time</td>
<td>Up to 5 files concurrently</td>
</tr>
<tr>
<td>Run Setup</td>
<td>Enabled</td>
<td>Disabled</td>
</tr>
<tr>
<td>Run Status</td>
<td>Enabled</td>
<td>Disabled</td>
</tr>
<tr>
<td>Maintenance Log</td>
<td>Enabled</td>
<td>Disabled</td>
</tr>
<tr>
<td>Archive Data</td>
<td>Enabled</td>
<td>Disabled</td>
</tr>
</tbody>
</table>
Chapter 4 Preparing a Sample Experiment

This section contains a standard protocol for sample preparation and plate setup for any assay performed on the QX ONE Droplet Digital PCR System. The following are recommended steps using the compatible GCR96 cartridges and reagents.

Important: Use only Rainin pipets and tips for all reagent and sample handling steps.

Required Components

This section describes the required equipment and materials for creating your samples. For a list of catalog numbers, see Appendix C, Ordering Information.

You need the following equipment:

- PX1 PCR Plate Sealer
- PX1 GCR Sealer Support Block
- Centrifuge, with plate rotor capable of 1,150 rcf minimum
- Vortex mixer

You need the following materials:

- PCR supermix
- Buffer control
- Pipets (Rainin only)
- Pipet tips (Rainin only)
- QX ONE Droplet Digital PCR System Droplet Generation Oil
- QX ONE Droplet Digital PCR System Droplet Reader Oil
- GCR96 cartridges
- Foil plate seal
- Reagent trough
- 5 mL mixing tube
Creating the Sample Mix

This section contains a standard protocol using the ddPCR Supermix (No dUTP). You can use this protocol to prepare your samples and set up plates for any assay you perform on the QX ONE Droplet Digital PCR System. The protocol below makes enough reagent for one plate (96 wells). Volumes can be adjusted for the number of plates you need. You can insert up to five plates into the instrument.

Important: You must combine the ddPCR Supermix (No dUTP) and the Reaction Mix in a 1:1 ratio. Final concentrations for components are included in the table below for a total of 20 µl per reaction. For Reaction Mix preparation, refer to each Supermix protocol.

To prepare the sample

1. Remove two ddPCR Supermix (No dUTP) vials of choice and two Reaction Mix vials from storage (-20°C) and let thaw at room temperature for approximately 30 min.
2. Vortex for 10 sec at medium to high speed and spin down in a small tube centrifuge.
   Important: Proper mixing is critical for both the ddPCR Supermix (No dUTP) and the Reaction Mix.
3. Prepare master mix as described in the following table (use 5 mL tubes to prepare mix). Be as accurate as possible in adding the correct volume of each reagent.

<table>
<thead>
<tr>
<th>Component</th>
<th>Volume, µl</th>
<th>Final Concentration</th>
</tr>
</thead>
<tbody>
<tr>
<td>2x ddPCR Supermix (no dUTP)</td>
<td>1,300</td>
<td>1x</td>
</tr>
<tr>
<td>2x Reaction Mix</td>
<td>1,300</td>
<td>1x</td>
</tr>
<tr>
<td><strong>TOTAL VOLUME</strong></td>
<td><strong>2,600</strong></td>
<td><strong>1x</strong></td>
</tr>
</tbody>
</table>

Note: The mixes can be highly viscous. Bio-Rad recommends slow pipeting for more accurate dispensing of all liquid.

a. Carefully pipet 650 µl from each of the two tubes of ddPCR Supermix (No dUTP) into the 5 mL mixing tube, changing tips with each addition.

b. Carefully pipet 650 µl from each of the two tubes of Reaction Mix into the 5 mL mixing tube, changing tips with each addition.

4. Cap the tube securely.
5. To mix, vortex for 10 sec at maximum speed.
Filling the GCR96 Cartridge

You must use GCR96 cartridges with the QX ONE Droplet Digital PCR System. The GCR96 cartridge plate contains 6 DG-16 individual cartridges representing 6 sets of 16 wells. Each set of 16 must be filled with sample or control buffer for the droplet generator to perform correctly.

Important: GCR cartridges are intended for single-use only.

The following graphic highlights the two columns of the first well set in the cartridge.

1. Set the PX1 PCR Plate Sealer to 180° C and 0.5 sec.  
   You will use the plate sealer when performing the steps in the next section.

2. Set the 8-channel pipet to 20 µl.

3. Pour the vortexed and centrifuged master mix from the 5 mL tube into a reagent reservoir.

4. Use a 20 µl 8-channel pipet to immediately transfer 20 µl of the master mix from the reagent reservoir into the wells of the first column of the cartridge.

5. Confirm the correct positioning of the cartridge, with the notched corner in the upper left.
   Note: Do not use the second depression on the pipet. While there will be a minute amount of sample left in the pipet tips, full depression can introduce air bubbles into the wells and disrupt proper droplet generation.

6. Repeat for columns 2–12 until all 96 wells are filled with 20 µl of master mix.
Sealing the GCR96 Cartridge

This procedure describes the plate sealing procedure for the PX1 PCR Plate Sealer. If you are using a different instrument, consult the applicable user manual.

Note: References to “plate” indicate a full GCR96 cartridge holder.

To seal the plate

1. Press the Eject icon to open the PX1 PCR Plate Sealer.
2. Ensure the following:
   - The temperature is set to 180° C.
   - The time is set to 0.5 sec.
3. Load the GCR96 cartridge on the appropriate support block, making sure that:
   - The notched corner (A1) is at the upper left.
   - The cartridge is securely set into the support block.
4. Place a foil seal on top of the GCR96 cartridge.

   Important: The foil seal must cover the frame of the plate with no overhang on any side. Overhanging foil can interfere with the movement of the robotic arm of the instrument.
5. Line up the top of the foil seal with the middle of the numbers on the top of the plate.
   - As the plate moves into the sealer, this alignment allows room for the foil to move into its final and correct position.
6. Press the green Seal button. When done, the plate sealer opens automatically.
7. Carefully rotate the plate 180° Con the support block.
8. Press the green Seal button again. When done, the plate sealer opens automatically.
9. Remove the plate from the plate sealer.
10. Remove the support block from the plate sealer.

   Caution: Do not leave the support block in the plate sealer, or the block will heat up. This creates a potential burn risk to users.

11. Continue to Loading the GCR96 Cartridge into a Centrifuge on page 47.
Loading the GCR96 Cartridge into a Centrifuge

Before you begin this procedure, ensure the following:

- You are wearing the proper personal protective equipment (PPE). For information, see General Precautions on page 13.
- The plate is sealed. For information on using the PX1 Plate Sealer, see Sealing the GCR96 Cartridge on page 46.

To load the plates

1. Push the individual DG16 cartridges in the GCR96 frame toward the numbered edge as far as they will go, to ensure there is no gap between the frame and cartridges.

2. Open the centrifuge instrument and insert a sealed plate into each side of the plate holder.

3. If processing only one cartridge, load a dummy cartridge on the other basket to balance the centrifuge.

4. Slide each plate toward the rotor axis.
5. Ensure the numbered sides touch the edges of the centrifuge baskets.

Important: Ensure the plates are oriented correctly, or the tabs on the cartridges can break. Broken tabs can cause separation between cartridges and the cartridge holder during the centrifuge process, and cause a failure of the QX ONE Droplet Digital PCR System.

6. Close and start the centrifuge.

7. Spin for 30 sec at 1150 rcf.

8. When centrifuging is complete, open the instrument and remove the plates.

9. Press the edges of the foil seal against the plate, in case centrifuging loosened the seal.

10. Continue to Adding Plates in the QX ONE on page 49.
Chapter 5 Adding Plates in the QX ONE

After you sign into QX ONE Software on the touch screen, the software opens to the Add Plate window.

The plate Inbox is located on the right side of the instrument, as shown in the following graphic. The instrument has slots where you can insert up to five plates.

Each GCR96 cartridge has an RFID label embedded on its side, which allows the instrument to scan and track each plate and the data generated from each plate.
You must use the scanner on the instrument to scan the RFID label on each plate before you can insert the plate into the Inbox. QX ONE Software prompts you to scan the plate RFID label, and then insert the plate.

- The Inbox door is opening
- The plate should be placed in the specified slot

**Note:** If plates from earlier runs occupy the QX ONE Droplet Digital PCR System Outbox, the instrument tells you to load your plate into the Inbox slot opposite the first empty Outbox slot. If there are not enough slots available for the plates you need to run, remove the completed plates from the Outbox.

1. Position the plate with the RFID label facing the empty slot with the amber lights, and then insert the plate.

Note the following:

- If the plate is inserted properly, the amber lights turn green.
If not, the lights turn red and you must remove and reinsert the plate.

The Inbox remains open as long as you are adding plates. When 30 sec have passed without a plate being added, the Inbox door closes automatically.

**Tip:** You can also tap the Open Inbox/Closed Inbox toggle button on the instrument status bar in QX ONE Software.

As each plate is added, a row containing plate information, including the plate barcode from the scan process, appears in the grid and QX ONE Software enables the Configure Plate button.

2. Tap Configure Plate.
Adding the Plate

To add your plates

1. Tap the Add Plate tab.

   - If the Runs Available number in the Instrument Status bar is 1 or higher, the Add Plate button is enabled.

2. Tap the + icon next to Add Plate.

   QX ONE Software prompts you to scan the RFID label on the GCR96 cartridge.

   Use the scanner on the front of the instrument, as shown below.

3. Scan the RFID label.

   **Tip:** The RFID label is on the opposite side of the bar code. The barcode is not scanned.
Adding the Plate

After the scan is complete, QX ONE Software displays advisory messages to indicate the following:

- The Inbox door is opening
- The plate should be placed in the specified slot

**Note:** If plates from earlier runs occupy the QX ONE Droplet Digital PCR System Outbox, the instrument tells you to load your plate into the Inbox slot opposite the first empty Outbox slot. If there are not enough slots available for the plates you need to run, remove the completed plates from the Outbox.

4. Position the plate with the RFID label facing the empty slot with the amber lights, and then insert the plate.

Note the following:

- If the plate is inserted properly, the amber lights turn green.

- If not, the lights turn red and you must remove and reinsert the plate.
Chapter 5 Adding Plates in the QX ONE

- The Inbox remains open as long as you are adding plates. When 30 sec have passed without a plate being added, the Inbox door closes automatically.

  **Tip:** You can also tap the Open Inbox/Close Inbox toggle button on the instrument status bar in QX ONE Software.

As each plate is added, a row containing plate information, including the plate barcode from the scan process, appears in the grid and QX ONE Software enables the Configure Plate button.

```
<table>
<thead>
<tr>
<th>Priority</th>
<th>Plate Name</th>
<th>Run Status</th>
<th>Barcode</th>
<th>Thermal Cycling Protocol</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td></td>
<td>Loaded</td>
<td>12-02-03-04-05</td>
<td></td>
</tr>
<tr>
<td>2</td>
<td></td>
<td>Loaded</td>
<td>01-12-03-04-05</td>
<td></td>
</tr>
<tr>
<td>3</td>
<td></td>
<td>Loaded</td>
<td>01-02-10-04-05</td>
<td></td>
</tr>
</tbody>
</table>
```

5. Tap Configure Plate.
Plate Configuration Window

From the Plate Configuration window, all users can start runs on selected plate templates using the initial configuration, an altered configuration, or just the plate name, data file name, supermix, and thermal cycling protocol.

Tip: Except for the supermix, which you must select before the run, you can configure the plate experiment parameters before, during, or after a run.

The window contains the following three tabs for identifying and configuring your plates and protocols for the run:

- **Plate Information** — Default display, where you can create or select existing plate and protocol templates, or start a run immediately with a minimum of information and configure experiment parameters later

- **Well Selection** — Optional task, where you can exclude wells from processing

  Note: The QX ONE Droplet Digital PCR System processes the entire plate by default. If you have filled fewer than 96 wells with sample, you should exclude those wells from processing.

- **Well Information** — Where you can create or modify the experiment parameters; optional if you are working with an existing plate layout

When configuring plates, note the following:

- Users with the Create New Template user privilege can save a new or modified plate or protocol design as a template before starting the run.

- All users can set up experiment parameters and run the plate without saving the layout.

- Users can identify experiment parameters in the Analysis module.

  **Important:** If you are configuring a plate in which the One-Step RT kit is being used, see Using the One Step RT ddPCR Advanced Kit for Probes on page 69.
Chapter 5 Adding Plates in the QX ONE

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
- Create and run a plate design using a manual configuration or a template
- 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 have the option to change it. 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 a thermal cycling protocol
- Select to acquire wells by columns or rows
Setting Plate Information

When you select Configure Plate, the following window appears.

**Important:** If you have been assigned the Access Module Recovery Tool user privilege, the Module Recovery Tool section appears in the bottom-left corner.

To set plate information

1. Under Plate Template, do one of the following to define your plate layout:
   - To use an existing template, tap the dropdown arrow and select a template. To modify the layout, select the Well Information tab.
   - To create a new template, tap Create New and see *Creating or Modifying Plate Templates on page 67* for more information on creating plate layouts.

2. Under Protocol template, do one of the following to define your protocol:
   - To use an existing template, tap the dropdown arrow and select a template.
   - To create a new template, tap Create New. For information on creating or editing templates, refer to the QX ONE Software User Guide.

3. Under Create New Plate, select a supermix.

   **Tip:** To run the plate without first setting experiment parameters, you can enter a plate name and file name, and then select a supermix to enable the Start Run button immediately.

4. (Optional) Select the alternate well acquisition method.

**Important:** The QX ONE Droplet Digital PCR System processes all 96 wells in the plate by default. If you have filled fewer than 96 wells with sample, you must exclude the remainder. For information, see *Well Selection Tab on page 58*.
Well Selection Tab

The QX ONE Droplet Digital PCR System processes the entire 96-well plate by default. If you have added your sample to fewer than 96 wells in the plate, you must exclude the remainder.

When you select the Well Selection tab, a blank plate grid appears.

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

Use this window to select wells to exclude from droplet generation and reading, or just droplet reading.

Excluding Wells in the Plate Layout

Use the Well Selection tab or the Exclude button to exclude wells from the droplet generation and droplet reading run phases, or droplet reading only.

The GCR96 cartridges that are inserted into the instrument contain six separate sections, each with two rows of eight wells each. The QX ONE Droplet Digital PCR System automatically generates droplets by section rather than by well or by individual column. In the QX ONE Software well exclusion design, the first two columns represent the first section, the second two columns the second section, and so forth.

Note the following:

- When you select Droplet Generation and Droplet Reading, the software allows you to exclude each section from both processes, but if you try to exclude an odd number of columns, QX ONE Software automatically selects the remaining column.
- When you select Droplet Reading, the software allows you to exclude any well or group of wells from the droplet reading phase.

Wells excluded from Droplet Generation and Droplet Reading are displayed with a criss-cross pattern.
Wells excluded from droplet reading are identified by a diagonal pattern.

To exclude wells

1. Select one or more column pairs to exclude, and then tap Droplet Generation and Droplet Reading or Droplet Reading.

   **Note:** If you selected an odd number of columns, the software automatically selects the remaining column to represent a section. You can exclude that column from droplet reading only.

2. Tap Exclude Selected Wells.

   **Note:** The Exclude Selected Wells button is a toggle button. If you select wells that have been excluded, the button name changes to Include Selected Wells. If you excluded one or more wells that should be read, you can select the wells and then tap the button to include them again.
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. However, you must select a supermix, and identify the plate and file name before the Start Run button is enabled. For information, see Defining or Editing Well Information on page 60.

Note: QX ONE 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 63.

Defining or Editing Well Information

You can assign different experiment parameters to each well or groups of wells. For information on each field, see Experiment Types on page 62, Sample Types on page 63, Supermixes on page 64, and Assay Types and Fluorophores on page 65.

Note: Each plate you configure appears as an icon on the left.

You can apply your entries at any time.

Note: If you tap the Save button before the plate layout is complete, the file closes and you must reopen it from the saved template location.
To define the configuration for the wells

**Important:** You must tap Apply for the software to recognize your new or edited information. If you enter information in a field and need to apply earlier changes, QX ONE Software displays an advisory message.

1. Select a well or group of wells.
2. Select an experiment type. For more information, see Experiment Types on page 62.
3. In the Sample Description fields, enter up to four words or phrases that describe the sample. For more information, see Sample Descriptions on page 63.
4. Select a sample type. For more information, see Sample Types on page 63.
5. Select a supermix. For more information, see Supermixes on page 64.
6. Select an Assay Type. Available assay types vary, depending on the selected Experiment Type. For information, see the fluorophore options in Assay Types and Fluorophores on page 65.
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. Tap the field dropdown arrow and select an alternative.
   
   b. To delete a row, tap the minus (-) icon next to the target.
   
   c. To add a row, tap the plus (+) icon.
8. (Optional) To clear one or more wells at any time, select the wells and tap Clear Selected Wells.
9. Tap Apply.

**Tip:** After you have applied your entries and selections, you can pause the cursor on a well to see the well information.

10. When you finish configuring the plate, tap Save.
Chapter 5 Adding Plates in the QX ONE

Experiment Types

QX ONE Software offers seven experiment types, each enabling different assay options and analysis tools. Table 8 briefly explains each experiment type.

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

Table 8. Experiment types

<table>
<thead>
<tr>
<th>Type</th>
<th>Description</th>
</tr>
</thead>
<tbody>
<tr>
<td>Direct quantification (DQ)</td>
<td>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. <strong>Note:</strong> 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.</td>
</tr>
<tr>
<td>Copy number variation (CNV)</td>
<td>CNV determines concentration and calculates the copy number of an unknown (CNV) target relative to a known reference or references within the same well.</td>
</tr>
<tr>
<td>Mutation detection (MUT)</td>
<td>MUT determines concentration and calculates the fractional abundance of an unknown mutant present at a low frequency in a wild-type background.</td>
</tr>
<tr>
<td>Rare event detection (RED)</td>
<td>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.</td>
</tr>
<tr>
<td>Drop-off (DOF)</td>
<td>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.</td>
</tr>
<tr>
<td>Gene expression (GEX)</td>
<td>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.</td>
</tr>
<tr>
<td>Residual DNA quantification (RDQ)</td>
<td>RDQ provides a precise method for residual host cell DNA quantification.</td>
</tr>
</tbody>
</table>

62 QX ONE Droplet Digital PCR System and QX ONE Software
Sample Descriptions

QX ONE 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 ONE 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**
Supermixes

QX ONE 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.

<table>
<thead>
<tr>
<th>ddPCR Supermix</th>
<th>Description</th>
</tr>
</thead>
<tbody>
<tr>
<td>Multiplex Supermix</td>
<td>For use in nucleic acid sample preparations to amplify and detect DNA targets in probe-based experiments involving multiple targets.</td>
</tr>
<tr>
<td>Supermix for Probes (No dUTP)</td>
<td>For use in nucleic acid sample preparations, with sensitivity for the amplification and detection of DNA targets using hydrolysis probe-based assays.</td>
</tr>
<tr>
<td>Supermix for Probes</td>
<td>For use in sample preparation with uracil N-glucosylase (UNG) decontamination protocols to prevent the reamplification of carryover PCR products between experiments.</td>
</tr>
<tr>
<td>Supermix for Residual DNA Quantification</td>
<td>For use in residual DNA detection.</td>
</tr>
</tbody>
</table>
| One-Step RT Advanced Kit for Probe | For use in absolute quantification of target RNA molecules.  
  **Important**: Load only one (1) one-step RT plate, using the first slot in the QX ONE Droplet Digital PCR System plate Inbox. See Using the One Step RT ddPCR Advanced Kit for Probes on page 69 for more information. |
Assay Types and Fluorophores

QX ONE Software offers the assay methods explained in Table 9.

Table 9. Assay types

<table>
<thead>
<tr>
<th>Assay type</th>
<th>Description</th>
</tr>
</thead>
</table>
| Single target per channel | Method assuming up to four probe colors (FAM, HEX/VIC, Cy5 and Cy5.5) and up to four targets per well, with a single target detected per channel.  
This is the default assay type, with the maximum four rows displayed. You can delete up to three rows. |
| Amplitude multiplex    | Method to increase multiplexing up to eight 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.                                  |
| Probe mix triplex      | Triplex mode that allows two groups of three targets each.  
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.  
You can delete a group, but not the individual targets within the group.                                           |
| Advanced classification | Method to increase multiplexing rate up to 10 targets per well in any combination of method targets detected per channel. This method requires manual classification of droplets.                                      |
| Basic drop-off         | This mode allows two groups of two targets each. Group 1 is shown by default, with one target in FAM and HEX/VIC and one target in FAM or HEX/VIC.  
This assay type is available only for the DOF experiment type.  
You can add one more group of two targets, with one target in Cy5 and Cy5.5, one target in Cy5 or Cy5.5. You can delete a group, but not the individual targets in the group.  
**Note:** The channels within each group are not interchangeable therefore FAM is always paired with HEX/VIC and Cy5 with Cy5.5. |
The experiment type you select determines the available assay methods. *Table 10* displays the sample type associated with each assay method.

**Table 10. Assay types by experiment type**

<table>
<thead>
<tr>
<th>Assay type</th>
<th>DQ</th>
<th>CNV</th>
<th>MUT</th>
<th>DOF</th>
<th>GEX</th>
<th>RDQ</th>
<th>RED</th>
</tr>
</thead>
<tbody>
<tr>
<td>Single target per channel</td>
<td>✓</td>
<td>✓</td>
<td>✓</td>
<td>✓</td>
<td>✓</td>
<td>✓</td>
<td>✓</td>
</tr>
<tr>
<td>Amplitude multiplex</td>
<td>✓</td>
<td>✓</td>
<td>✓</td>
<td>✓</td>
<td>✓</td>
<td>✓</td>
<td>✓</td>
</tr>
<tr>
<td>Probe mix triplex</td>
<td>✓</td>
<td>✓</td>
<td>✓</td>
<td>✓</td>
<td>✓</td>
<td>✓</td>
<td>✓</td>
</tr>
<tr>
<td>Advanced classification</td>
<td>✓</td>
<td></td>
<td></td>
<td></td>
<td>✓</td>
<td>✓</td>
<td>✓</td>
</tr>
<tr>
<td>Basic drop-off</td>
<td>✓</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

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

Use *Table 11* to select from the fluorophore options for each assay method listed.

**Table 11. Fluorophore options**

<table>
<thead>
<tr>
<th>Assay method</th>
<th>Signal Ch1</th>
<th>Signal Ch2</th>
<th>Signal Ch3</th>
<th>Signal Ch4</th>
</tr>
</thead>
<tbody>
<tr>
<td>Single target per channel, for 1 to 4 targets</td>
<td>FAM</td>
<td>HEX</td>
<td>Cy5</td>
<td>Cy5.5</td>
</tr>
<tr>
<td></td>
<td>None</td>
<td>VIC</td>
<td>None</td>
<td>None</td>
</tr>
<tr>
<td>Amplitude multiplex, for 1 to 8 targets</td>
<td>FAM Hi</td>
<td>HEX Hi</td>
<td>Cy5 Hi</td>
<td>Cy5.5 Hi</td>
</tr>
<tr>
<td></td>
<td>FAM Lo</td>
<td>HEX Lo</td>
<td>Cy 5 Lo</td>
<td>Cy 5.5 Lo</td>
</tr>
<tr>
<td></td>
<td>None</td>
<td>VIC Hi</td>
<td>None</td>
<td>None</td>
</tr>
<tr>
<td></td>
<td></td>
<td>VIC Lo</td>
<td></td>
<td></td>
</tr>
<tr>
<td></td>
<td></td>
<td>None</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Probe mix triplex, for 6 targets</td>
<td>FAM</td>
<td>HEX</td>
<td>Cy5</td>
<td>cy5.5</td>
</tr>
<tr>
<td></td>
<td>None</td>
<td>VIC</td>
<td>None</td>
<td>None</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Advanced classification</td>
<td>FAM 1-10</td>
<td>HEX 1-10</td>
<td>Cy5 1-10</td>
<td>Cy5.5 1-10</td>
</tr>
</tbody>
</table>
Creating or Modifying Plate Templates

When you choose to add or modify a plate template, the Plate Editor opens, featuring options to set up your samples in the plate layout and exclude certain wells from droplet generation and droplet reading or droplet reading only.

LEGEND

1. Buttons at the top of the window provide quick access to plate editing functions.
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 is displayed, the button name changes to Plate Layout. You cannot edit the Well Data table.

For detailed information on creating plate templates, refer to the QX ONE Software User Guide.
Creating or Modifying Protocol Templates

Thermal cycling protocols are specific to the thermal cycling ddPCR phase. When you add or modify a protocol template from the Template Setup window, the Protocol Editor opens. By default, the editor opens to a generic 4-step protocol, which includes standard protocol controls to quickly create protocols, as well as the ability to

- Quickly calculate a gradient for a specific temperature range and run time for the plate type
- Edit protocol steps
- Save protocols for reuse

The lower pane displays the following toolbar commands:

- The trashcan command deletes the selected step.
- The plus sign to the left of the trashcan adds a step before the selected step.
- The plus sign to the right of the trashcan adds a step after the selected step.

The upper pane displays the protocol controls that you can use to customize the protocol.

The main pane displays a graphical representation of the protocol.

The information bar provides quick access to the name of the protocol, view the estimated protocol run time, and to save or cancel changes to the protocol.

For detailed information on creating protocol templates, refer to the QX ONE Software User Guide.
Chapter 6 Running Experiments

When the Start Run button is enabled for a plate, you can start the run. You can also cancel one or more runs during the droplet generation or droplet reading phases, and you can recover a canceled or failed run during the droplet generation or droplet reading phases. For information, see Canceling a Run on page 76 and Recovering Plates From Failed or Canceled Runs on page 77.

Using the One Step RT ddPCR Advanced Kit for Probes

The One Step RT ddPCR Advance Kit for Probes supermix is time and temperature-sensitive. Therefore, if you are performing an experiment using this supermix, it is important that the plate does not sit idle in one of the Inbox slots.

To perform an experiment using this kit

1. Wait until all plates in the QX ONE Droplet Digital PCR System have processed.
2. Place the One Step RT plate into the first slot.
3. Quickly set up and start the run.

For information, see Plate Configuration Window on page 55 and Running Experiments on page 69.
Starting the Run

When the required elements to set up one or more plates are satisfied, QX ONE 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 window.

Important: Ensure that at least one full bottle of QX ONE Droplet Digital PCR System Droplet Generation Oil for Probes and one full bottle of QX ONE Droplet Digital PCR System Droplet Reader Oil are installed, and at least one waste bottle is completely empty.

To start the droplet reading run

When all plate setup requirements have been satisfied and the Start Run button is enabled, tap Start Run.

The QX ONE Droplet Digital PCR System begins processing each plate in the order you specified.

Note: The enabled button is available in the windows associated with the Add Plate function.

Important: The Regulatory Edition prompts you to log in again and reauthenticate your user credentials. Enter your user name and password, and then tap Sign in.
Changing the Plate Processing Order

If you have been assigned the Manage Plates user privilege, you can change the processing order of plates still in the instrument Inbox, even if other plates are running.

To change the plate processing order

1. Select a row representing a plate in the Inbox.

2. Drag the row to the new location.

3. Continue reordering, as applicable. You can reorder up to five plates in the Inbox.
Running One or More Experiments

The instrument allows you to do the following:

- During the droplet reading phase of the run, perform real-time analysis on the droplets in each well after the well has been read
- Reorder plates while they are still in the Inbox
- Cancel a run
- When the run concludes and the plate has been removed from the outbox, QX ONE 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 one of the following locations:

- Your personal file location specified in your user preferences
- The preferred location in System Settings

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

  **Important:** if your system administrator has designated preferred locations for all users, these file paths override the file paths indicated in your preferences.

Tracking the Run Status

You can track each run from the Run Status window.

The top pane shows the runs 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 12.

Table 12. Run status indicators

<table>
<thead>
<tr>
<th>Status Icon</th>
<th>Description</th>
</tr>
</thead>
<tbody>
<tr>
<td>));</td>
<td>Ready to begin first or next phase</td>
</tr>
<tr>
<td>⚪</td>
<td>Droplet generation phase</td>
</tr>
<tr>
<td>⚪</td>
<td>Droplets generated, plate waiting in Inbox</td>
</tr>
<tr>
<td>⚪</td>
<td>Thermal cycling protocol phase</td>
</tr>
<tr>
<td>⚪</td>
<td>Droplet reading phase</td>
</tr>
<tr>
<td>✔</td>
<td>Run completed, plate in Outbox</td>
</tr>
<tr>
<td>✔</td>
<td>Run completed, plate removed from Outbox</td>
</tr>
<tr>
<td>☑</td>
<td>Plate successfully recovered</td>
</tr>
<tr>
<td>⚠</td>
<td>Run stopped or canceled by the system</td>
</tr>
</tbody>
</table>

When the run is finished and the plate is removed from the Outbox, 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 34.

The content of each column is explained in Table 13.
# Table 13. Run Information Columns

<table>
<thead>
<tr>
<th>Column Name</th>
<th>Location</th>
<th>Description</th>
</tr>
</thead>
<tbody>
<tr>
<td>Plate name</td>
<td>Top and bottom panes</td>
<td>Name entered under Create New Plate</td>
</tr>
<tr>
<td></td>
<td></td>
<td><strong>Note:</strong> If you selected an existing template, the template name appears.</td>
</tr>
<tr>
<td>Bar code</td>
<td>Top and bottom panes</td>
<td>Bar code, as read by the instrument</td>
</tr>
<tr>
<td>Run status</td>
<td>Top and bottom panes</td>
<td>Icons for each process step, as explained in Table 12 on page 73</td>
</tr>
<tr>
<td>Location</td>
<td>Top pane only</td>
<td>Process step location (for example Droplet Reader)</td>
</tr>
<tr>
<td>Time remaining</td>
<td>Top pane only</td>
<td>Hours and minutes until the run will be finished</td>
</tr>
<tr>
<td>Time elapsed</td>
<td>Top pane only</td>
<td>Time, in hours and minutes the run has been in progress</td>
</tr>
<tr>
<td>Total time</td>
<td>Below the top pane</td>
<td>Total hours and minutes remaining for all in-progress runs</td>
</tr>
<tr>
<td>remaining</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Run completed</td>
<td>Bottom pane only</td>
<td>Date and time stamp showing when the instrument completed the run</td>
</tr>
<tr>
<td>File name</td>
<td>Bottom pane only</td>
<td>Plate name and system-generated identifier</td>
</tr>
</tbody>
</table>

After run completion, the software saves the analysis data file in either your personal file location or to a preferred location specified by your system administrator. If your system is connected to a network, and shared folders are identified in the global preferences, the data file is also copied to a shared folder.

**Important:** If your system administrator has specified preferred locations in System Settings for all users, files are saved there, rather than to the path specified in user preferences. For information, see Preferred Location on page 88.
Running One or More Experiments

Using Live Analysis

When the QX ONE Droplet Digital PCR System reaches the droplet reading phase, QX ONE 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 ONE 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 (跑了) for the run in progress.
   
   The Droplet Reading dialog box opens.

2. Tap Open Live Analysis.
3. Select an analysis window.
   
   For brief descriptions of the analysis windows, see Data Analysis Overview on page 81. For more detailed information, refer to the QX ONE Software User Guide.

4. In the well selector, select one or more wells that are enabled.
   
   Important: QX ONE 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.
Chapter 6 Running Experiments

Canceling a Run

You can cancel one or more runs at any time during the process.

1. In the Run Status window, tap the row for the run to be canceled and tap the X on the right.

2. In the pop-up that appears, tap Abort Plate Run.

3. When the Are you sure message appears, tap Yes.

   The instrument finishes reading the current well, stops the process and either proceeds to the next plate or reverts to its pre-run state.

   **Note:** If you tap 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 ONE Software creates the data file for the wells that were processed on the canceled plate, and the run end status in the Event Log is specified as “run aborted by user.”
Recovering Plates From Failed or Canceled Runs

If a user cancels a run, or if a component experiences a problem during a run, the run fails and QX ONE Software displays an error message on the touch screen.

If the failure occurred during the thermal cycling phase, the plate is not recoverable and must be discarded. However, if the run failed during the droplet generation or droplet reading phase, you can use the Recover Plate option in QX ONE Software.

When a cancellation or failure occurs, QX ONE Software moves the run to the lower pane in the Run Status screen and displays a red exclamation point under Run Status, as shown in the following graphic.
To recover or discard the plate in QX ONE Software

1. Before you begin the plate recovery process, do one of the following:
   - If the error did not stop the instrument and plates are still running through ddPCR processes, allow the remaining runs to finish and then restart the instrument.
   - If the error did stop the instrument, resolve the instrument problem and then restart the instrument.

2. When the instrument restarts, the recover plate screen appears automatically. Continue to Step 3.

   **Important:** If the window is inadvertently closed, select the Add Plate tab and then tap Recover Plate.

3. Do one of the following:
   - If the plate failed during thermal cycling, manually discard the plate using safety protocols, and then select the Discard check box and tap OK. A message appears, advising that you cannot recover the plate after it is discarded. To permanently discard the plate, tap OK again.
   - For droplet generation or droplet reading failures where you intend to recover the plate, select the Recover Plate check box and tap OK.

4. Tap Add Plate.
5. Follow the prompts to scan the RFID code and insert the plate into the specified slot.

If you are recovering multiple plates, the system prompts you to add and scan each plate you identified for recovery.

6. Tap Start Run.

The QX ONE Droplet Digital PCR System resumes the process where it was interrupted for each plate.

When the run for each recovered plate successfully concludes, QX ONE Software shows the plate with an amber check mark in the Run Status column, as shown in the following graphic.
Manually Rerunning ddPCR Processes

QX ONE Software also provides the capability to manually recover a plate by identifying the failed process, and then rerunning the plate from that point. Users must be assigned the Access Module Recovery Tool user privilege to omit the processes that will not be rerun.

**Important:** Because using the manual process requires the user to identify in the log file the precise point of failure, Bio-Rad instead recommends using the automatic plate recovery functionality described in *Recovering Plates From Failed or Canceled Runs on page 77*. If you are using manual recovery, contact Bio-Rad Technical Support for assistance before proceeding.

To manually rerun the plate

1. Wait for plates currently in process to complete.
2. In QX ONE Software, tap Add Plate.
3. When prompted, insert the plate into the Inbox in accordance with the displayed message.
4. Tap Configure Plate.
   
   The Module Recovery Tool pane appears below the plate and thermal cycling protocol panes.

   ![Module Recovery Tool](module_recovery_tool.png)

   By default, all checkboxes are selected.

5. If necessary, clear the checkbox for each process that will not be repeated.

   **Important:** You cannot clear the Plate Handler Present checkbox.

6. Make any necessary configuration changes to the plate.
7. When the Start Run button is enabled, click Start Run.
   
   The QX ONE Droplet Digital PCR System enables and runs each specified module.
Chapter 7 Data Analysis Overview

From the Data Analysis tab, you can browse for a particular data file, or open a data file that has been recently viewed.

This section summarizes the functionality available in the Analysis module, and also describes the general options governing your chart displays. See the QX ONE Software User Guide for detailed information on analyzing run files in the Analysis module and the Gene Study module. Gene Study functionality is not available from the touch screen.
Data Analysis Module

The Data Analysis module functionality allows you to

- View your droplet data in 1D, 2D, and 3D amplitude
  - View thresholds automatically assigned by the software based on the distance of the threshold from the means and standard deviations of the positive and negative clusters
  - Manually adjust thresholds or classify droplets using cluster modes
  - View data in amplitude plots, histograms, and heat maps
  - Apply tilt correction to automatic analysis
- View charts reflecting calculated data visualizations for concentration, copy number, ratio, and fractional abundance
- 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
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 increase or decrease the number of panes that appear in the Dashboard, and you can also change the selection of panes that appear.

![Dashboard Image](image)

**Note:** If you open a .qlp 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.

Plate View Windows

In the QX ONE 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
Chapter 7 Data Analysis Overview

Viewing Run and Lot 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 the Regulatory Edition, you can also view the audit logs.
Analysis Reporting

Use the QX ONE Software Reporting function to generate a variety of reports.

As shown in the following graphic, any user can open, modify, and run the default layout to generate a report from the Reports window in the Analysis module.

If specific reports are required on a regular basis, users who are assigned the Create New Templates user privilege can modify the default layout and save it as a new template report file for reuse.

For the checkboxes you select in the report configuration, QX ONE Software displays corresponding charts and data for the selected wells. In each report header, the software automatically displays the user name of the person creating the report, date and time stamps, and the name of the analysis file used to generate the report.

For more information on the Reporting function, refer to the QX ONE Software User Guide.
Chapter 7 Data Analysis Overview
Chapter 8 System Utilities

From the System Utilities tab, you can access the following tabs for managing tasks and data related to the instrument, software, data, and 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 raw data to clear up space in your primary storage areas and access data for troubleshooting.

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

**System Settings Tab**

When you select the System Utilities tab, the System Settings window opens by default, and contains the following data file and template storage options set by your 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 shared locations are defined, but users are not required to save their files to the specified folders. The administrator can also define a backup location for data file storage if the priority folder is full.

**Note**: You can choose one option only. For Shared Settings, users can choose to save files to those locations or to the locations specified in their user preferences.
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.

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. Tap Save.
4. When the confirmation message appears, tap OK.
Shared Settings

Under Shared Settings, you can specify two shared data file storage folders and one shared template storage folder. Because data files are often very large, the software allows you 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 can change storage locations in the System Settings window.

**To specify shared folders**

1. In each of the storage path fields, enter a file path or tap Browse to search for the folder.
2. Tap Save.
3. When the confirmation message appears, tap OK.
System Log Files

From the System Utilities 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.

To view the Event Log

1. Select the System Settings tab.

2. Select Event Log and scroll to and select an event.

3. Tap Open Log to display a sequence of subevents.

4. (Optional) To automatically generate and display a PDF document, tap 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. The Maintenance Log tab is available from the instrument touch screen only.

**Tip:** This guide also contains a form on which you can document completion of maintenance activities. For information, see *Instrument Maintenance Form on page 113.*

Users who are assigned the Maintenance user privilege can set up maintenance records, specify associated activities, and generate PDF reports.

To add or view an activity

1. Select the System Settings tab.

2. Select Maintenance Log. 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 activities, but you cannot modify them.
   - Add an activity. Continue to step 4.

4. Tap the plus sign (+) next to New Activity.
   A line item appears in the bottom section, with all fields except Activity completed automatically.

5. In the Activity field in the bottom section, type the activity into the Activity field.

6. Tap Save.

7. Repeat to add another activity.
   After saving, you can create a PDF report of the added activities.

8. (Optional) Tap PDF Report.
Maintenance Reports

Reports generated from the Bio-Rad service engineer when performing maintenance on your instrument are stored on the Maintenance Reports tab. The Maintenance Reports tab is available from the instrument touch screen only.

To open a report, tap the list item.

Following is an example of a Color Calibration report:
System Data

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

Archiving Data

When the storage space on the QX ONE instrument touch screen QX ONE Droplet Digital PCR System approaches the limit displayed under Disk Space, you can free up space by moving raw data to an archive storage folder in a different location.

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

To archive files

1. Select the System Settings tab.

2. Select System Data.

The Archive Raw Data tab is selected by default.

3. Under Archive Progress, tap the calendar icons and select a From date and a To date to define a date span.
4. Tap Query to search for files dated within the date span.
5. Tap Browse to search for a select the target storage location.
6. Tap Archive.
Troubleshooting Data

If issues occur with the QX ONE 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 access data for troubleshooting purposes

1. Select the System Settings tab.

2. Select System Data > Troubleshooting.

3. Tap the From and To calendar icons to select a date range.

4. Select any of the following checkboxes:
   - System Logs
   - Raw Plate Data
   - Version Data
   - Calibration Data

5. Tap Browse and navigate to the data folder in the target storage location.

6. Tap Collect Data.
Recovering Data

If a run fails on the QX ONE Droplet Digital PCR System, you can use the Data Recovery tab in System Data to recover data that has been saved to a temporary file.

Important: You must be assigned the System Data user privilege to access the Data Recovery tab.

To create a .ddpcrone file from the data in the temporary file

1. Select the System Settings tab.

2. Select System Data and then select the Data Recovery tab.

Temporary files that are available for conversion are listed in the top pane, which is scrollable.

3. Tap a file, and then tap Browse.

4. Navigate to the folder on your computer or network and then click Recover.

The following advisory message appears.

The file(s) selected for recovery are temporary file(s) and as such may contain incomplete plate data. Do you wish to recover the data contained in the temp file?

Yes  No
5. Click Yes.

A dialog box appears with a progress bar, and automatically closes when the conversion is complete.

The .ddpcrone file is stored in the folder you specified.

System Calibration

You cannot execute runs on the QX ONE Droplet Digital PCR System unless the instrument is properly calibrated.

Only a Bio-Rad service engineer can calibrate your instrument and use the Instrument Calibration options in QX ONE Software. The option is available from the touch screen only, and is disabled for all other users.

If your instrument requires service, contact Bio-Rad Technical Support.
Chapter 8 System Utilities
Chapter 9 Managing Users

**Important:** This chapter applies only if you enabled the User Management module during the installation of QX ONE Software Standard Edition. If you did not enable the module, QX ONE Software creates a generic user with the ability to set up, run, and analyze experiments.

Use the User Setup and Preferences window to add, change, or remove QX ONE Software users, and set or change user privileges and preferences.

The Bio-rad engineer sets up an initial user with all privileges, including Add/Manage users, on the touch screen. Your software administrator can use this account to set up additional users with appropriate privileges.

**Note:** 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 instrument touch screen computer connected to the QX ONE Droplet Digital PCR System:

- 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 ONE 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.

However, QX ONE Software must be able to validate the user in either the local or domain user group before you can add the user in the software. Note the following:

- For local IDs, you must set up each user as a local Windows user account on each computer where the software is installed.
- For Active Directory IDs, your IT department must connect the QX ONE Droplet Digital PCR System instrument touch screen, as well as any separate computers running the software, to your network.
Adding Users

This section applies only if the User Management module was enabled during the installation of QX ONE Software Standard Edition.

Before you can add users to QX ONE 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 ONE Software and sign into the application.
2. Tap 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*.
     **Important:** Active Directory users must log in every time with the domain name preceding the user name.
   - If the computer is not connected to a network, enter `<local computer name>/<user name>`. For example, *analysiscomputer/johnsmith*.
     **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. Tap 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, tap OK, and then tap Add at the bottom of the
   pane.
   An Are you sure you want to add this user? message appears.

6. Tap Yes.
   A confirmation message appears.

7. Tap OK.

8. Continue until all users are added.

Adding or Removing User Privileges

This section applies only if the User Management module was enabled during the installation of

Only a user assigned the Add/Manage users privilege can add or remove user privileges.

By default, individual users without the privileges described in Table 14 can set up and execute a run,
open files, view and analyze results.

Table 14 lists the available privileges, which can be assigned in any combination.
Adding or Removing User Privileges

Table 14. User privileges

<table>
<thead>
<tr>
<th>Privilege</th>
<th>Purpose</th>
</tr>
</thead>
<tbody>
<tr>
<td>Add and manage users</td>
<td>Add or remove users, set privileges, and change preferences.</td>
</tr>
<tr>
<td></td>
<td><strong>Note:</strong> Only the superuser can remove this privilege from other users.</td>
</tr>
<tr>
<td>Create new templates</td>
<td>Save a plate, thermal cycling protocol, or analysis report design as a template.</td>
</tr>
<tr>
<td>View data files (created by other users)</td>
<td>View files created by other users.</td>
</tr>
<tr>
<td>Overwrite existing data file name</td>
<td>Use Save or Save As capability.</td>
</tr>
<tr>
<td></td>
<td>■ Selecting Save replaces the original file content with any changes made by the user, without changing the file name.</td>
</tr>
<tr>
<td></td>
<td>■ Selecting Save As allows the user to save either existing or new content with a different file name.</td>
</tr>
<tr>
<td></td>
<td>Whether or not changes are made to the file after you open it, note the following:</td>
</tr>
<tr>
<td></td>
<td>.qlp and .ddpcr files are saved as .ddpcrone files</td>
</tr>
<tr>
<td></td>
<td>.qlps and .ddpcrs files are saved as .ddpcrsone files</td>
</tr>
<tr>
<td></td>
<td><strong>Note:</strong> Users without this permission can open files and perform analysis but cannot save their changes.</td>
</tr>
<tr>
<td>System settings</td>
<td>View logs, and view and modify the preferred or shared data file and template locations.</td>
</tr>
<tr>
<td></td>
<td><strong>Note:</strong> All users can view the file locations.</td>
</tr>
<tr>
<td></td>
<td><strong>Important:</strong> Your system administrator can set preferred locations in System Settings, which override individual user location preferences.</td>
</tr>
<tr>
<td>Maintenance</td>
<td>View the event log.</td>
</tr>
<tr>
<td></td>
<td><strong>Note:</strong> All users can view the maintenance log and maintenance reports.</td>
</tr>
<tr>
<td>Data archive</td>
<td>Move raw data from the touch screen computer to free up disk space needed for runs on the instrument.</td>
</tr>
<tr>
<td>Access the Module Recovery Tool</td>
<td>Specify the processes to be rerun for manual recovery.</td>
</tr>
<tr>
<td>Manage Plate</td>
<td>Cancel plates that are running and change the order that plates are run.</td>
</tr>
</tbody>
</table>

Table 15 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 15. Example User Roles

<table>
<thead>
<tr>
<th>Privilege</th>
<th>Superuser</th>
<th>Lab Manager</th>
<th>Group Lead</th>
<th>Technician/Student Intern</th>
</tr>
</thead>
<tbody>
<tr>
<td>Add/manage users</td>
<td>✓</td>
<td>✓</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Create new templates*</td>
<td>✓</td>
<td>✓</td>
<td>✓</td>
<td>✓</td>
</tr>
<tr>
<td>View data files created by other users</td>
<td>✓</td>
<td>✓</td>
<td>✓</td>
<td>✓</td>
</tr>
<tr>
<td>Override existing ddPCR file name</td>
<td>✓</td>
<td>✓</td>
<td>✓</td>
<td></td>
</tr>
<tr>
<td>System settings</td>
<td>✓</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Maintenance</td>
<td>✓</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Archive data files</td>
<td>✓</td>
<td>✓</td>
<td>✓</td>
<td>✓</td>
</tr>
<tr>
<td>Access the module recovery tool</td>
<td>✓</td>
<td>✓</td>
<td>✓</td>
<td>✓</td>
</tr>
<tr>
<td>Manage Plates</td>
<td>✓</td>
<td>✓</td>
<td>✓</td>
<td></td>
</tr>
</tbody>
</table>

*You can assign this 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 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. Tap 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, tap Yes to save the changes, and then tap OK to close the pop-up.
Modifying User Preferences

This section applies only if the User Management module was enabled during the installation of QX ONE Software Standard Edition.

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. Tap 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.
     Important: You can also specify Preferred Locations in System Settings, which override all file paths specified in individual preferences. For information, see System Settings Tab on page 87.
   - Select or clear the checkbox to change user data file and template privacy settings.
   - 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. Tap Save. QX ONE Software displays a confirmation message.
4. Tap Yes to save the changes, and then tap OK to close the dialog box.

Removing Users

This section applies only if the User Management module was enabled during the installation of QX ONE Software Standard Edition.

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. Tap the Add/Manage Users tab to open the User Management window.
2. In the Current Users pane, select the user and tap Remove. QX ONE Software displays a confirmation message.
3. Tap Yes to remove the user, and then tap OK to close the dialog box.
Chapter 9 Managing Users
Appendix A Instrument Maintenance

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

- Keeping the surface areas clean
- Regularly checking the fluid levels
- Inspecting the equipment for damaged parts or wiring

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

General Maintenance

Follow these guidelines to properly maintain your instrument:

- Surfaces of the instrument might require general cleaning.
  - Use a cloth, slightly dampened with deionized/distilled water, to wipe down the instrument.
  - For regular cleaning and decontamination, use 10% bleach solution. For information, see Cleaning and Decontamination of the QX ONE Droplet Digital PCR System on page 112.
  - **Important:** Do not use acetone or tap water.

- Inspect the equipment regularly for damaged external components or wiring. Do not use if damaged.

- Apply standard SDS (Safety Data Sheet) and OSHA practices when handling and disposing of generated waste.

- 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. These nonflammable fluids are inert and have low environmental impact and low toxicity.
Droplets made with Bio-Rad supermix have antimicrobial properties, but microbial growth is possible. The waste profile should contain the following:

- Fluorinated hydrocarbons
- Water
- Fluorescent dye (from probes)
- Protein
- Nucleic acids

Do not replace the included detachable power cord with an uncertified or an inadequately rated cord.

Since the QX ONE Droplet Digital PCR System contains moving parts, fluids, tubing, and pumps, Bio-Rad strongly recommends semi-annual preventive maintenance on the instrument by a Bio-Rad service engineer or an authorized agent.

**Waste Removal**

Empty droplet reader oil bottles can be moved to the waste positions on the left side of the instrument.
Waste bottle replacement is identical to oil bottle replacement procedures.

**Important:** Use each empty oil supply bottle as a new waste bottle only once, then discard. Reuse of a waste bottle might lead to microbial growth, instrument damage, or run failures.

**Important:** Waste bottles are filled more quickly than the QX ONE Droplet Digital PCR System Droplet Generation Oil or Droplet Reader Oil bottle contents are depleted. Bio-Rad recommends purchasing QX ONE Droplet Digital PCR System waste bottles each time a total of 20 plates have been run. For the catalog number, refer to the Consumables section in Appendix C, Ordering Information. Bio-Rad does not recommend reusing waste bottles.

**Vent Reservoir Maintenance**

The reservoir in the QX ONE instrument collects liquid that escapes from waste bottles as the plates are being processed in the QX ONE Droplet Digital PCR System. As the liquid accumulates, QX ONE Software prompts you to remove the liquid:

- **When the liquid level in the reservoir exceeds** 25 mL, QX ONE Software displays an advisory message, prompting you to aspirate the reservoir before adding new plates to the Inbox.
  
  Bio-Rad recommends aspirating at this time. You can perform the aspiration procedure while the instrument is processing ddPCR runs.

- **When the liquid in the reservoir approaches** 50 mL, QX ONE Software displays a more urgent message advising immediate action.

  If you do not aspirate the reservoir before the volume reaches 50 mL, the instrument stops and QX ONE Software displays an error message.
Appendix A Instrument Maintenance

Aspirating the Vent Reservoir

You can aspirate the liquid in the vent reservoir at any time, but you must aspirate the reservoir when the level reaches 50 mL, as instructed by QX ONE Software.

To aspirate the liquid

1. From the QX ONE Software touch screen, tap Open Outbox on the toolbar.

   The following message appears.

   **Important:** You must perform this procedure within the allotted 3 min, at which time the outbox door automatically closes.

2. Locate the red luer-lock port on the bottom-left side of the outbox.
3. Using a twisting motion, attach a 50 mL luer-lock syringe to the port.

**Tip:** Bio-Rad recommends the 50 mL BD Luer-Lok Tip Syringe. For information, see Ordering Information on page 117.

4. Draw liquid into the syringe, up to the 50 mL point.

5. Remove the syringe from the port.

6. Dispose of the syringe using required procedures for bio-hazardous materials. For information, see Disposal of Biohazardous Material on page 14.

**Important:** Use each syringe only once.
Cleaning and Decontamination of the QX ONE Droplet Digital PCR System

The following protocol can be used to clean and decontaminate the QX ONE Droplet Digital PCR System, Bio-Rad recommends performing this procedure weekly, at a minimum, as outlined on the Maintenance Log.

To clean and decontaminate the QX ONE Droplet Digital PCR System, you must have 10% bleach solution (either spray or pre-soaked wipes). If you have questions regarding the use of other cleaning agents, contact Bio-Rad Technical Support.

Important: You do not need to pass 10% bleach through the system itself.

To clean the instrument surface and touch screen using 10% bleach

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

2. Wipe the plate holders and surrounding areas in each Inbox and Outbox.

3. Wipe the touch screen.

4. Identify the equipment as "Decontaminated."
# Instrument Maintenance Form

## QX ONE Droplet Digital PCR System

### Maintenance Log

<table>
<thead>
<tr>
<th>Date</th>
<th>Time</th>
<th>Task Description</th>
<th>Frequency</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td></td>
<td>Instrument Serial Number</td>
<td>Last Annual Maintenance Date</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

- Weekly at start of week:
  - Check all expansion lines
  - Replace any necessary
  - Replace the touch screen controller

- Monthly:
  - Clean, sanitize, and recalibrate
  - Replace any necessary

- Quarterly:
  - Check the system for any maintenance instructions
  - Follow specific maintenance guide

- Annually:
  - Call Bi-Taq for service
Appendix A Instrument Maintenance
Appendix B Additional Computers

This section explains the recommended requirements for QX ONE Software, and provides instructions for installing or updating the software.

Computer Requirements

Standalone computers are for analysis purposes only. The computers should meet the requirements specified in Table 16.

Table 16. Standalone computer requirements

<table>
<thead>
<tr>
<th>System Component</th>
<th>Minimum</th>
<th>Recommended</th>
</tr>
</thead>
<tbody>
<tr>
<td>Operating system</td>
<td>Windows 10 64-bit</td>
<td>Windows 10 64-bit</td>
</tr>
<tr>
<td>CPU</td>
<td>6th generation Intel 2 core processor</td>
<td>8th generation Intel 4 core processor</td>
</tr>
<tr>
<td>Hard disk space</td>
<td>500 GB</td>
<td>1 TB</td>
</tr>
<tr>
<td>System memory</td>
<td>8 GB</td>
<td>16 GB</td>
</tr>
<tr>
<td>Display resolution</td>
<td>1920 x 1080</td>
<td>1920 x 1080</td>
</tr>
<tr>
<td>Ports</td>
<td>1 USB</td>
<td>1 USB</td>
</tr>
</tbody>
</table>

Notes:

(1) The software is compatible with Windows Defender and Trend Micro Office Scan anti-virus applications.

Important: Do not update the anti-virus or perform a scan while the instrument is acquiring wells or data may be lost.

(2) Enabling FIPS security on your Windows 10 computer does not interfere with instrument communication or application functionality.
Installing or Updating the Software

The QX ONE Droplet Digital PCR System must complete all runs and be in an inactive state when you upgrade the software on the installed touch screen computer, or the instrument firmware.

QX ONE Software can also be installed on additional computers to facilitate run analysis.

If you are already logged into QX ONE Software and you have open windows, save your data and close the windows before proceeding with the software update.

When the software is updated, the new version overwrites the existing version. If you are reverting to an older version, you must uninstall the newer version before installing the older version.

For more information on installing QX ONE Software, call Bio-Rad Technical Support.
Appendix C Ordering Information

This appendix contains descriptions and catalog numbers for new or replacement items.

ddPCR System

Table 17 contains information on the QX ONE Droplet Digital PCR System components.

Table 17. QX ONE Droplet Digital PCR System

<table>
<thead>
<tr>
<th>Product</th>
<th>Description</th>
<th>Catalog number</th>
</tr>
</thead>
<tbody>
<tr>
<td>QX ONE Droplet Digital PCR System</td>
<td>Instrument used for droplet generation, PCR, droplet reading, and data analysis</td>
<td>12006536</td>
</tr>
<tr>
<td></td>
<td>This purchase includes the QX ONE Accessory Kit (formerly catalog number 12011478), which contains the following:</td>
<td></td>
</tr>
<tr>
<td></td>
<td>- PX1 GCR Sealer Support Block</td>
<td></td>
</tr>
<tr>
<td></td>
<td>- QX ONE Droplet Digital PCR System ddPCR System Waste bottles</td>
<td></td>
</tr>
<tr>
<td></td>
<td>- Keyboard</td>
<td></td>
</tr>
<tr>
<td></td>
<td>- Power cord, compatible within North America</td>
<td></td>
</tr>
<tr>
<td></td>
<td>- Warranty card</td>
<td></td>
</tr>
<tr>
<td>QX ONE Software</td>
<td>Connects to the QX ONE Droplet Digital PCR System for instrument control, assay setup, and data collection and analysis</td>
<td>N/A</td>
</tr>
<tr>
<td></td>
<td>Standard Edition, N/A</td>
<td>12012078</td>
</tr>
<tr>
<td></td>
<td>Regulatory Edition</td>
<td></td>
</tr>
<tr>
<td>Plate Sealer</td>
<td>PX1 PCR Plate Sealer</td>
<td>1814000</td>
</tr>
</tbody>
</table>
**ddPCR Materials and Consumables**

Table 18 contains information on additional items for performing ddPCR.

**Table 18. Additional ddPCR materials and consumables**

<table>
<thead>
<tr>
<th>Product</th>
<th>Description</th>
<th>Catalog Number</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>Reagents for Probe Detection</strong></td>
<td></td>
<td></td>
</tr>
<tr>
<td>PCR Supermix</td>
<td>ddPCR Supermix for Probes (No dUTP)</td>
<td>1863023</td>
</tr>
<tr>
<td></td>
<td></td>
<td>1863024</td>
</tr>
<tr>
<td></td>
<td></td>
<td>1863025</td>
</tr>
<tr>
<td></td>
<td>ddPCR Supermix for Probes</td>
<td>1863026</td>
</tr>
<tr>
<td></td>
<td></td>
<td>1863010</td>
</tr>
<tr>
<td></td>
<td></td>
<td>1863027</td>
</tr>
<tr>
<td></td>
<td></td>
<td>1863028</td>
</tr>
<tr>
<td></td>
<td>ddPCR Multiplex Supermix</td>
<td>12005909</td>
</tr>
<tr>
<td></td>
<td></td>
<td>12005910</td>
</tr>
<tr>
<td></td>
<td></td>
<td>12005911</td>
</tr>
<tr>
<td></td>
<td>ddPCR Supermix for Residual DNA Quantification</td>
<td>1864037</td>
</tr>
<tr>
<td></td>
<td></td>
<td>1864038</td>
</tr>
<tr>
<td></td>
<td></td>
<td>1864039</td>
</tr>
<tr>
<td></td>
<td></td>
<td>1864040</td>
</tr>
<tr>
<td></td>
<td>One-Step RT-ddPCR Advanced Kit for Probes</td>
<td>1864021</td>
</tr>
<tr>
<td></td>
<td></td>
<td>1864022</td>
</tr>
<tr>
<td>DTT (dithiothreitol)</td>
<td>2 mL DTT</td>
<td>12012171</td>
</tr>
<tr>
<td>Control</td>
<td>ddPCR Buffer Control for Probes</td>
<td>1863052</td>
</tr>
<tr>
<td>QX ONE Droplet Digital PCR System</td>
<td>QX ONE Droplet Generation Oil for Probes, 15 plates</td>
<td>12006058</td>
</tr>
<tr>
<td>Droplet Generation Oil</td>
<td></td>
<td></td>
</tr>
<tr>
<td>QX ONE Droplet Digital PCR System</td>
<td>QX ONE Droplet Reader Oil, 10 plates</td>
<td>12006057</td>
</tr>
<tr>
<td>Droplet Reader Oil</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Product</td>
<td>Description</td>
<td>Catalog Number</td>
</tr>
<tr>
<td>------------------</td>
<td>-----------------------------------------------------------------------------</td>
<td>----------------</td>
</tr>
<tr>
<td><strong>Consumables</strong></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Cartridge</td>
<td>GCR96 Cartridge, pk of 1</td>
<td>12006858</td>
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<tr>
<td></td>
<td>GCR96 Cartridges, pk of 10</td>
<td>12006859</td>
</tr>
<tr>
<td>Foil plate seal</td>
<td>GCR96 Foil Heat Seal</td>
<td>12006843</td>
</tr>
<tr>
<td>Waste bottle</td>
<td>QX ONE Droplet Digital PCR System ddPCR System Waste bottle</td>
<td>12006060</td>
</tr>
<tr>
<td><strong>Materials from Other Suppliers</strong></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Pipets (Rainin)</td>
<td>8-channel multichannel pipet, 2–20 µl</td>
<td>L8-20 XLS</td>
</tr>
<tr>
<td></td>
<td>Single channel pipet, 2–20 µl</td>
<td>L-20 XLS</td>
</tr>
<tr>
<td></td>
<td>Single channel pipet, 100–1,000 µl</td>
<td>L-1000 XLS</td>
</tr>
<tr>
<td>Pipet tips (Rainin)</td>
<td>20 µl filter tips</td>
<td>GP-LTS-A-10µl/-F-960/10</td>
</tr>
<tr>
<td></td>
<td>1,000 µl filter tips</td>
<td>GP-LTS-A-1000µl/-F-/768/8</td>
</tr>
<tr>
<td>Reagent trough</td>
<td>Matrix Reagent Reservoirs (Thermo Fisher Scientific)</td>
<td>8093</td>
</tr>
<tr>
<td>Vortexer</td>
<td>Any</td>
<td>N/A</td>
</tr>
<tr>
<td>Centrifuge</td>
<td>Centrifuge with plate rotor capable of at least 1,150 rcf</td>
<td>N/A</td>
</tr>
<tr>
<td>50 mL BD Luer-Lok Tip Syringe</td>
<td>Syringe for removing liquid from the overflow reservoir</td>
<td>309653</td>
</tr>
<tr>
<td><strong>Services</strong></td>
<td></td>
<td></td>
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<tr>
<td>Bio-Rad service</td>
<td>QX ONE Droplet Digital PCR System IQ/OQ/PQ service</td>
<td>IQ 12012331</td>
</tr>
<tr>
<td></td>
<td></td>
<td>OQ 12012333</td>
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<tr>
<td></td>
<td></td>
<td>PQ 12012332</td>
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</table>