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QIAxcel Connect System User Manual

For use with QIAxcel ScreenGel Software v2.1



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1 Introduction

Thank you for choosing the QIAxcel Connect. We are confident it will become an integral part of your laboratory.

Thank you for choosing the QIAxcel ScreenGel Software. We are confident it will become an integral part of your laboratory.

The QIAxcel ScreenGel Software version 2.0 and higher is designed for use in combination with the QIAxcel Connect system.

Before using the QIAxcel Connect, it is essential that you read the QIAxcel Connect system user manual carefully and pay particular attention to the safety information. The instructions and safety information in the user manual must be followed to ensure safe operation of the instrument and to maintain the instrument in a safe condition.

1.1 About this user manual

This user manual provides information about the QIAxcel Connect in the following sections:

- 1. Introduction
- 2. <u>Safety Information</u>
- 3. General Description
- 4. Installation Procedures
- 5. Operating Procedures
- 6. <u>QIAxcel ScreenGel Software</u>
- 7. Maintenance Procedures
- 8. Troubleshooting
- 9. Glossary
- 10. Appendix A Technical Data
- 11. Appendix B QIAxcel Connect methods
- 12. Appendix C QIAxcel Accessories
- 13. Appendix D DNA Analysis Algorithm Description
- 14. Appendix E Liability clause
- 15. Appendix F Informations de sécurité
- 16. Appendix G Sicherheitshinweise
- 17. Appendix H License Terms
- 18. Appendix I Revision History

This user manual provides information about the QIAxcel ScreenGel in the following sections:

- 1. Introduction
- 2. General Description
- 3. Installation Procedures
- 4. <u>QIAxcel ScreenGel Software</u>
- 5. Troubleshooting
- 6. Glossary
- 7. Appendix

1.2 General information

1.2.1 Technical assistance

Technical assistance

At QIAGEN, we pride ourselves on the quality and availability of our technical support. Our Technical Services Departments are staffed by experienced scientists with extensive practical and theoretical expertise in sample and assay technologies and the use of QIAGEN products. If you have any questions or experience any difficulties regarding the QIAxcel Connect, QIAxcel ScreenGel Software or QIAGEN products in general, do not hesitate to contact us.

QIAGEN customers are a major source of information regarding advanced or specialized uses of our products. This information is helpful to other scientists as well as to the researchers at QIAGEN. We therefore encourage you to contact us if you have any suggestions about product performance or new applications and techniques.

For technical assistance and more information, please see our Technical Support Center at <u>https://</u> <u>www.qiagen.com/de/service-and-support/technical-support/</u> or call one of the QIAGEN Technical Service Departments or local distributors (see back cover or visit <u>www.qiagen.com</u>).

For up-to-date information about the QIAxcel Connect system, visit the product page on QIAGEN.com

1.2.2 Policy statement

Policy statement

It is the policy of QIAGEN to improve products as new techniques and components become available. QIAGEN reserves the right to change specifications at any time.

In an effort to produce useful and appropriate documentation, we would appreciate your comments on this user manual. Please contact QIAGEN Technical Services.

1.2.3 Intended use of the QIAxcel Connect

The QIAxcel Connect is a multicapillary electrophoresis instrument, designed to perform fast and fully automated DNA fragment analysis, or qualitative and quantitative RNA analysis.

The QIAxcel Connect instrument is intended to be used only in combination with QIAxcel Kits for applications described in the respective QIAxcel Kit handbooks and QIAxcel ScreenGel software version 2.0 or higher.

The QIAxcel Connect instrument is intended for use by professional users, such as technicians and physicians trained in molecular biological techniques and the operation of the QIAxcel Connect instrument.

1.2.3.1 Requirements for QIAxcel Connect users

This table covers the general level of competence and training necessary for transportation, installation, use, maintenance, and servicing of the QIAxcel Connect.

Task	Personnel	Training and experience
Transportation	No special requirements	No special requirements
Installation	Laboratory technicians or equivalent	Appropriately trained and experienced personnel familiar with use of computers and automation in general
Instrument administratior	Laboratory technicians or equivalent	Appropriately trained and experienced personnel familiar with use of computers and automation in general
Routine use (running profiles)	Laboratory technicians or equivalent	Appropriately trained and experienced personnel familiar with use of computers and automation in general
Analyzing data	Laboratory technicians or equivalent	Appropriately trained and experienced personnel familiar with use of computers and automation in general
Process Profile design and verification	Scientist or equivalent	Appropriately trained or experienced personnel familiar with molecular biological techniques
Instrument maintenance	Laboratory technicians or equivalent	Appropriately trained and experienced personnel familiar with use of computers and automation in general
Servicing	QIAGEN-authorized representatives only	Trained and authorized by QIAGEN

2 Safety information

Before using the QIAxcel Connect, it is essential that you read this user manual carefully and pay particular attention to the safety information. The instructions and safety information in the user manual must be followed to ensure safe operation of the instrument and to maintain the instrument in a safe condition.

The following types of safety information appear throughout this manual:

WARNING	The term WARNING is used to inform you about situations that could result in personal injury to you or other persons.
	Details about these circumstances are given in a box like this one.

CAUTION	The term CAUTION is used to inform you about situations that could result in
^	admage to the instrument or other equipment.
	Details about these circumstances are given in a box like this one.

The advice given in this manual is intended to supplement, not supersede, the normal safety requirements prevailing in the user's country.

2.1 Proper use

WARNING/ CAUTION	Risk of personal injury and material damage [W1] Improper use of the QIAxcel Connect may cause personal injuries or damage to the instrument.
	The QIAxcel Connect must only be operated by qualified personnel who have been appropriately trained.
	Servicing of the QIAxcel Connect must only be performed by QIAGEN Field Service Specialists.

Perform the maintenance as described in the <u>Maintenance procedures</u> section. QIAGEN charges for repairs that are required due to incorrect maintenance.

WARNING/ CAUTION	
\wedge	

Risk of personal injury and material damage [W2]

The QIAxcel Connect is too heavy to be lifted by one person. To avoid personal injury or damage to the instrument, do not lift the instrument alone.

WARNING	Risk of personal injury and material damage [W3]
\wedge	Do not attempt to move the QIAxcel Connect during operation.

In case of emergency, switch off the QIAxcel Connect at the power switch at the front of the instrument and unplug the power cord from the power outlet.

CAUTION	Damage to the instrument [C1]
	If the Pressure 1 status is LOW , increase system pressure before performing the unlatch command. Removing the cartridge and unlatching with reduced pressure may damage the instrument.

CAUTION	Damage to the instrument [C2]
\wedge	Do not use bleach, solvents, or reagents containing acids, alkalis, or abrasives to clean the QIAxcel Connect.

CAUTION	Damage to the instrument [C3]
\wedge	Avoid spilling water, chemicals, and liquids in general onto the QIAxcel Connect. Damage caused by water or chemical spillage will void your warranty.

CAUTION	Damage to the instrument [C4]
	Do not stack instruments and do not place items on top of the QIAxcel Connect.

2.2 Electrical safety

Disconnect the line power cord from the power outlet before servicing.

CAUTION

WARNING	Electrical hazard [W4]
	Use only the cable supplied with the QIAxcel Connect.

WARNING	Electrical hazard [W5]
	Any interruption of the protective conductor (earth/ground lead) inside or outside the instrument or disconnection of the protective conductor terminal is likely to make the instrument dangerous.
	Intentional interruption is prohibited.
	Lethal voltages inside the instrument
	When the instrument is connected to line power, terminals may be live, and opening covers or removing parts is likely to expose live parts.

To ensure satisfactory and safe operation of the QIAxcel Connect, follow the advice below:

- The line power cord must be connected to a line power outlet that has a protective conductor (earth/ ground).
- Do not adjust or replace internal parts of the instrument.
- Do not operate the instrument with any covers or parts removed.
- If liquid has spilled inside the instrument, switch off the instrument, disconnect it from the power outlet, and contact QIAGEN Technical Services.
- When replacing the main fuse, replace only with the type and current rating specified on the rating label.

If the instrument becomes electrically unsafe, prevent other personnel from operating it, and contact QIAGEN Technical Services. The instrument may be electrically unsafe when:

- The line power cord appears to be damaged.
- It has been stored under unfavorable conditions for a prolonged period.
- It has been subjected to severe transport stresses.

2.3 Environment

Operating conditions

WARNING	Explosive atmosphere [W6]
\wedge	The QIAxcel Connect is not designed for use in an explosive atmosphere.

WARNING	Risk of explosion [W7]
$\mathbf{\Lambda}$	The QIAxcel Connect is intended for use with reagents and substances supplied with the QIAGEN QIAxcel Kits. Use of other reagents and substances may lead to fire or explosion.

CAUTION	Damage to the instrument [C6]
	Direct sunlight may bleach parts of the instrument and cause damage to plastic parts.
	The QIAxcel Connect must be located out of direct sunlight.

CAUTION	Damage to the cartridge [C7]
	When in use, the gel cartridge should not be left out of the Wash Park position of the buffer tray for more than 15 minutes. Failure to do so will cause the capillary tips to dry out, affecting correct function of the cartridge. Dry tips will void the warranty.
	The capillary tips are made of glass and are very fragile. Take care not to knock the tips on any hard surfaces. Failure to do so will cause the capillary tips to break, affecting correct function of the cartridge. Broken tips will void the warranty.

CAUTION	
\wedge	

Damage to the cartridge [C8]

If less than 12 samples are processed, fill the empty sample wells with the QX DNA (HS) Dilution Buffer or QX RNA (HS) Dilution buffer. Failure to do so may cause damage to the capillary channels.

CAUTION	Damage to the cartridge [C9]
\wedge	Direct sunlight may bleach the cartridge and reagents inside and cause damage to plastic parts.
	The cartridge must be located out of direct sunlight.

2.4 Chemicals

WARNING	Hazardous chemicals [W8]
\wedge	Some chemicals used with this instrument may be hazardous or may become hazardous after completion of the protocol run.
	Always wear safety glasses, gloves, and a lab coat.
	The responsible body (e.g. laboratory manager) must take the necessary precautions to ensure that the surrounding workplace is safe and that the instrument operators are not exposed to hazardous levels of toxic substances (chemical or biological) as
	defined in the applicable Safety Data Sheets (SDSs) or OSHA * , ACGIH † or COSHH ‡ documents.
	Venting for fumes and disposal of wastes must be in accordance with all national, state, and local health and safety regulations and laws.

* OSHA: Occupational Safety and Health Administration (United States of America).

† ACGIH: American Conference of Government Industrial Hygienists (United States of America).

‡ COSHH: Control of Substances Hazardous to Health (United Kingdom).



Risk of fire [W9]

When cleaning the QIAxcel Connect with alcohol-based disinfectant, leave the QIAxcel Connect doors open to allow flammable vapors to disperse.

2.5 Waste disposal

Used labware and containers may contain hazardous chemicals. Such wastes must be collected and disposed of properly according to local safety regulations.

For information on how to dispose of the QIAxcel Connect, see Appendix A.

2.6 Mechanical hazards

The cartridge door and sample door of the QIAxcel Connect must remain closed during operation of the instrument.

WARNING	Moving parts [W10]
\wedge	To avoid contact with moving parts during operation of the QIAxcel Connect, the instrument must be operated with the cartridge door and sample door closed.
	If the door sensors are not functioning correctly, contact QIAGEN Technical Services.

2.7 Symbols on the QIAxcel Connect

Symbol	Location	Description
CE	Type plate on the back of the instrument	CE mark for European Conformity
	Type plate on the back of the instrument	CSA listing mark for Canada and the USA
UK CA	Type plate on the back of the instrument	UKCA mark for Great Britain (England, Wales, Scotland)
	Type plate on the back of the instrument	Legal manufacturer
X	Type plate on the back of the instrument	Waste Electrical and Electronic Equipment (WEEE)
25)	Type plate on the back of the instrument	RoHS mark for China (the restriction of the use of certain hazardous substances in electrical and electronic equipment)
\oslash	Type plate on the back of the instrument	RCM mark for Australia and New Zealand
i	Type plate on the back of the instrument	Consult instructions for use
	Type plate on the back of the instrument	Date of manufacture



Rear of the instrument at the power inlet

Risk of electrical shock caution



Front of the instrument power On/Off switch

Push-On/Push-Off

3 General description

The QIAxcel Connect system performs fully-automated separation of DNA and RNA fragments according to their molecular weight and can process up to 96 samples without user intervention. The QIAxcel Connect system consists of the QIAxcel Connect instrument, a QIAxcel Kit (containing a QIAxcel gel cartridge and reagents), a computer, and QIAxcel ScreenGel Software, which have been optimized to work together across a wide variety of applications and provide unmatched resolution, speed, and throughput for DNA and RNA analysis.

The QIAxcel ScreenGel provides both Electropherograms and Gel images of the nucleic acid separation and can be used to perform the following types of analysis:

- Determination of the size and concentration of DNA fragments
- Determination of the 28S/18S ratio, concentration, and quality of total RNA, and the quality of cRNA/ cDNA and fragmented RNA/DNA

Note: We do not recommend determining nucleic acid concentration with the QIAxcel DNA Fast Analysis Kit.

The QIAxcel ScreenGel offers extensive tools that simplify data analysis, allow fast data interpretation, and provide flexibility in viewing results in Electropherogram and Gel image formats.

The software provides a wizard-based run setup and convenient profiles for standardized sample processing, and effortless, customizable documentation of results.

The QIAxcel ScreenGel is based on an unique algorithm, that enables to analyze separation data more reproducibly and accurately than other chromatographic analysis packages. It evaluates distribution profiles and calculates a variety of peak properties, such as peak number, height, width, and peak area.

3.1 QIAxcel Connect principle

The QIAxcel Connect uses capillary electrophoresis to enable high-resolution and sensitive separation of DNA and RNA samples. This 12-channel capillary electrophoresis instrument uses disposable, multiple-use cartridges for cost-effective analysis of up to 96 samples in as little as 25 minutes.

- 1. The QIAxcel Connect is set up with a gel cartridge, running buffer and wash buffer, and calibrated using intensity markers.
- 2. The samples for analysis (in a 96-well plate or 12-tube strips) are placed onto the sample plate holder.
- 3. Experimental run parameter settings are selected and the samples pass through the capillaries of the QIAxcel gel cartridge.
- 4. Data is analyzed using the QIAxcel ScreenGel.

3.2 External features of the QIAxcel Connect





- 1 Sample door
- 2 Cartridge door
- 3 Service door
- 4 Power Switch
- 5 Power connection for AC connection
- 6 Tube fitting for external N_2 supply
- 7 USB connection
- 8 N₂ regulator and cylinder

- 9 Transport lock and buffer tray location
- 10 Sample plate holder
- 11 Cartridge bay
- 12 Slot for smart key
- 13 Digital pressure display
- 14 Purge filter
- 15 Transport lock storage

Note: If using an external N₂ source, the output pressure must not exceed 75 psi. The QIAxcel Connect equipped with an internal regulator that will regulate the pressure provided by the external N₂ source to approximately 40 psi (37 to 45 psi), which is the instrument's operating pressure.

3.3 QIAxcel Kits and cartridges

The QIAxcel Connect can be operated using any of the QIAxcel Kits listed in the table below. Each kit contains a gel cartridge that has been specifically developed for particular applications. Each gel cartridge consists of a gel matrix with a proprietary linear polymer and fluorescent dye.

Kit	Application
QIAxcel DNA High-Sensitivity Kit (1200)	Analysis of DNA 15 bp – 3 kb, down to 5 pg/µl. 96 samples can be analyzed in about 75 minutes.
QIAxcel RNA High-Sensitivity Kit (1200)	Analysis of RNA down to 50 pg/µl. 96 samples can be analyzed in about 100 minutes.
QIAxcel DNA High Resolution Kit (1200)	Analysis of DNA 15 bp – 20 kb in size. 96 samples can be analyzed in about 90 minutes.
QIAxcel DNA Screening Kit (2400)	Fast analysis of DNA 15 bp – 5 kb in size. 96 samples can be analyzed in just 40 minutes.
QIAxcel DNA Fast Analysis Kit (3000)	Routine evaluation of DNA fragments (15 bp – 3 kb) in qualitative single or multiplex PCR applications. 96 samples can be analyzed in approximately 25 minutes.
QIAxcel RNA QC Kit v2.0 (1200)	Analysis of RNA quality and quantity. 96 samples can be analyzed in about 90 minutes.

Depending on each QIAxcel Kit contains the following additional reagents that are required for use of the QIAxcel Connect:

- QX (HS) Intensity Calibration Marker calibrates the signal intensity for each new gel cartridge
- QX (HS) Separation Buffer enables separation of DNA or RNA molecules
- QX FA DNA Separation Buffer (QIAxcel DNA Fast Analysis Kit) enables separation of DNA molecules (only for use with the QIAxcel DNA Fast Analysis Cartridge)
- QX (HS) Wash Buffer for washing the capillary tips to prevent cross-contamination
- QX DNA or RNA (HS) Dilution Buffer for optimization of sample concentration
- QX Mineral Oil for covering solutions and/or samples to prevent evaporation

- QX Alignment Marker used in every run to equalize the migration time variation across all channels (included in the QIAxcel DNA High-Sensitivity and RNA High-Sensitivity Kit, QIAxcel RNA QC Kit v2.0 and QIAxcel DNA Fast Analysis Kit only; for all other kits, see ordering information, <u>Appendix C</u>)
- QX Size Marker enables the creation of a reference marker table allowing size and concentration determination of samples (included in the QIAxcel DNA High-Sensitivity and RNA High-Sensitivity and QIAxcel RNA QC Kit v2.0. For all other kits, see ordering information, <u>Appendix C</u>)

3.4 Computer and software

A computer with the correct specifications for operation of the QIAxcel Connect instrument and the QIAxcel ScreenGel software is supplied as part of the QIAxcel Connect system. However, if a different computer is used to operate the QIAxcel Connect instrument or run the QIAxcel ScreenGel, then the following requirements are necessary:

Computer specifications:

- At least 2.3 GHz CPU
- Minimum 80 GB free hard drive capacity, NTFS formatted
- At least 4 GB RAM
- 1920 x 1080 screen resolution or higher
- USB port
- Pointer device (mouse or similar)
- Windows 10 (64 bit) with minimum required version 1607
- Ethernet or Wi-Fi network adapter in the case of QIAsphere usage
- Important: The computer has to be compliant with IEC/CAN/UL 60950-1 or IEC/CAN/UL 62368-1, and must have a double isolation integrated.

To view reports in PDF format, a PDF reader must be installed on the computer. Adobe Reader can be downloaded at <u>www.adobe.com</u>.

Additional configuration steps:

- Turn off all automatic processes or services, such as indexing or similar, which could produce a high workload, especially when connecting to the QIAxcel Connect instrument.
- Ensure that the energy saving options and hibernation are deactivated during the operation of the QIAxcel Connect.

• For Windows 10 only – If UEFI Boot is used, the Secure Boot option must be disabled. Otherwise, the Device Driver installation of the QIAxcel Connect device driver will fail during the installation of the QIAxcel ScreenGel software.

Note: In case of any concern regarding the Windows configuration, please contact your local IT support.

If connecting a computer to a network, you must consider the following:

- Ensure that the update processes for downloading and installing are only scheduled outside the operating hours of the QIAxcel Connect.
- Turn off the Windows security path auto-restart feature to to prevent the computer from automatically restarting during QIAxcel Connect operating hours.

Note: To complete a security update installation, perform a manual restart later.

If virus protection software is required, ensure that the following tasks are scheduled to run outside the operating times of the QIAxcel Connect operating hours:

- Scans (make sure that the files created by the QIAxcel ScreenGel are not scanned when they are created or accessed)
- All kind of updates (download and installation)

Note: ScreenGel is Federal Information Processing Standards (FIPS) compliant. By default, FIPS is disabled on the laptop delivered with the instrument. To activate FIPS, you need admin rights. Follow these steps:

- 1. Press the Windows key on the keyboard.
- 2. To open the Local Security Policy, enter secpol.
- 3. Click the Local Policies folder to expand.
- 4. Open the **Security Options** folder and select the **System cryptography** entry. Use FIPS compliant algorithms for encryption, hashing and signing.
- 5. Select Enabled and click OK.

To migrate the experiment, open QIAxcel ScreenGel and confirm the experiment migration. Marker files or profiles are not migrated automatically.

Note: The QIAxcel ScreenGel is tested by QIAGEN using the original image supplied with your laptop without connecting the laptop to the Internet. Updates on your operating system might lead to unexpected behavior of the QIAxcel ScreenGel software. In such cases, please contact your local IT support.

Note: The minimal screen resolution can be used in combination with small fonts. However, medium size fonts can be used in combination with an improved resolution. Larger font sizes are not supported.

4 Installation procedures

4.1 Requirements

This section of the QIAxcel Connect User Manual describes the requirements for operating the QIAxcel Connect.

4.1.1 Site

The QIAxcel Connect must be protected from direct sunlight, heat sources, vibration sources, and electrical interference. The operating conditions (temperature and humidity) can be found in <u>Appendix A</u>. The installation site should be free of excessive drafts, excessive moisture, excessive dust, and should not be subject to large temperature fluctuations.

Use a level workbench that is large enough to accommodate the QIAxcel Connect and the computer. The weight and dimensions of the QIAxcel Connect can be found in <u>Appendix A</u>.

Ensure that the workbench is dry, clean, vibration-proof, and has additional space for accessories. A clearance of approximately 72 cm (28 in.) above the workbench is recommended to allow loading of cartridges.

The QIAxcel Connect must be placed within approximately 1.5 m (4.92 ft.) of a properly grounded (earthed) AC power outlet. The power line to the instrument should be voltage regulated and surge protected.

Ensure that the power switch and the main power input socket at the rear of the instrument are easily reachable. Maintain sufficient clearance at the rear to unplug the power line from the main power input socket.

Note: We recommend plugging the instrument directly into its own wall socket and not to share the socket with other lab equipment. To achieve proper capillary electrophoresis separation, do not place the QIAxcel Connect on a vibrating surface or near vibrating objects.

WARNING	Explosive atmosphere [W6]
\wedge	The QIAxcel Connect is not designed for use in an explosive atmosphere.

WARNING	Risk of overheating [W11]
\wedge	To ensure proper ventilation, maintain a minimum clearance of 10 cm (3.94 in) at the sides and rear of the QIAxcel Connect.
	Slits and openings that ensure the ventilation of the QIAxcel Connect must not be covered.

4.1.2 Power requirements

The QIAxcel Connect operates at 100–240 V AC, 50–60 Hz, 100 VA.

Ensure that the voltage rating of the QIAxcel Connect is compatible with the AC voltage available at the installation site. Main supply voltage fluctuations are not to exceed $\pm 10\%$ of nominal supply voltages.

CAUTION	Damage to electronics [C5]
	Before switching on the instrument, make sure that the correct supply voltage is used. Use of incorrect supply voltage may damage the electronics. To check the recommended supply voltage, refer to the specifications indicated on the type plate of the instrument.

4.1.3 Grounding requirements

To protect operating personnel, the instrument is equipped with a 3-conductor AC power cord. To preserve this protection feature, do not operate the instrument from an AC power outlet that has no ground (earth) connection.

For replacement of the 3-conductor AC power cord, contact QIAGEN Technical Services for an authorized spare part.

WARNING	Electrical hazard [W5]	
	Any interruption of the protective conductor (earth/ground lead) inside or outside the instrument or disconnection of the protective conductor terminal is likely to make the instrument dangerous.	
	Intentional interruption is prohibited.	
	Lethal voltages inside the instrument.	
	When the instrument is connected to line power, terminals may be live and opening covers or removing parts is likely to expose live parts.	

4.2 Unpacking the QIAxcel Connect

First, check if the package is damaged. In case of damage, contact the transporter of the package.

Before unpacking the QIAxcel Connect, move the package to the installation site and check that the arrows on the package point upward.



When lifting up the QIAxcel Connect, slide your fingers under both sides of the instrument and keep your back straight.

WARNING/ CAUTION

Risk of personal injury and material damage [W2]

The QIAxcel Connect is too heavy to be lifted by one person. To avoid personal injury or damage to the instrument, do not lift the instrument alone.

After unpacking the QIAxcel Connect, check that the following documents are supplied:

- Packing list
- Warranty registration form

Read the packing list to check that you have received all items. If anything is missing, contact QIAGEN Technical Services.



The QIAxcel Connect User Manual is available on the software USB in PDF format.

Check that the QIAxcel Connect is not damaged and that there are no loose parts. If anything is damaged, contact QIAGEN Technical Services.

Retain the package in case you need to transport or ship the QIAxcel Connect in the future. Instructions for packing the QIAxcel Connect are given in section <u>Packing the QIAxcel Connect</u>. Using the original package minimizes damage during transportation of the QIAxcel Connect.

Carefully place the QIAxcel Connect onto the workbench where it will be used. For site requirements, please refer to section <u>Requirements</u>.

4.3 Installing the QIAxcel Connect

This section describes important actions that must be performed before operating the QIAxcel Connect:

Note: Make sure that the QIAxcel Connect has equilibrated to ambient temperature before operating it.

- Removing of the transport lock
- Setting up the instrument
- Connecting the power cable
- Installing the N_2 cylinder or an external N_2 supply
- Switching ON the QIAxcel Connect

4.3.1 Removing of the transport lock

The QIAxcel Connect is delivered with a transport lock that secures the tray/transport mechanism during shipment. This transport lock must be removed before the QIAxcel Connect can be put into operation.

Note: Please reinstall the transport lock each time you ship the instrument.

Remove the transport lock as follows:

1. Open the sample door.

Note: Please close the cartridge door before opening the sample door. Do not open the sample door if the cartridge door is open. This may damage the instrument.

2. Release the transport lock securing the buffer tray/sample plate holder by turning it counter clockwise.



3. Keep the transport lock in case you need to ship the QIAxcel Connect in the future. A storage bay for the transport lock is located under the Service Door for storage use.



4.3.2 Setting up the instrument

Set up the QIAxcel Connect as follows:

- 1. Set up the computer as described in the computer installation instructions provided with the computer.
- 2. Connect the USB cable from the instrument to the computer.

4.3.3 Connecting the power cable

Connect the QIAxcel Connect to the power outlet as follows:

- 1. Make sure that the power switch of the QIAxcel Connect is in the "off" position.
- 2. Check that the voltage specification on the label on the back of the QIAxcel Connect corresponds to the voltage available at the installation site.
- 3. Plug the power cord into the power cord socket on the back of the instrument.
- 4. Plug the power cord into a grounded power outlet.



Damage to electronics [C10]

Use only the cable supplied with the QIAxcel Connect.

4.3.4 Installing the N2 cylinder or an external N2 supply

Important: Use only N₂ cylinders provided by QIAGEN (Catalog No. 929705).

- 1. Make sure that the power switch of the QIAxcel Connect is in the "Off" position.
- 2. Open the sample door and pull carefully and gently the N₂ cylinder port upwards.
- 3. Screw the N_2 cylinder clockwise into the port.
- Turn until the needle inside the port punctures the N₂ cylinder.
 Note: Do not overtightened. The cylinder should only be finger-tight.
- 5. Gently push down the N_2 cylinder until it is in its stowed (down) position.
- 6. Close the sample door.

Alternatively, you can use an external N_2 supply and attach it to the rear value of the instrument.

4.4 Installation of the QIAxcel ScreenGel Operating Software

The QIAxcel ScreenGel software version 2.0 or higher must be installed before connecting the QIAxcel Connect to the computer for the first time. The QIAxcel ScreenGel software is available for installation from the USB drive provided with the QIAxcel Connect instrument. The QIAxcel ScreenGel software version lower than 2.0 will not work with the QIAxcel Connect.

Things to do before starting:

- 1. If you are using a computer other than provided with the QIAxcel Connect, ensure that the computer meets the minimum requirements (see <u>Computer and software</u>).
- 2. Use a Windows account with administrator privileges.

Note: Windows administrator privileges are only required for software installation.

- 3. Close any programs running on the computer and make sure that the QIAxcel Connect is not connected to the computer (i.e. unplug the cable from the computer).
- 4. Set up the computer as described in the computer installation instructions provided with the computer.
- 5. Before installing the QIAxcel ScreenGel software, close any programs and make sure that the QIAxcel Connect is not connected to the computer (i.e., unplug the connection from the computer).
- If the QIAxcel ScreenGel software shall be installed for the first time, follow the instructions in the next section, <u>First-time installation from a USB drive</u>. First time installation of the QIAxcel ScreenGel software is possible from a USB drive only.
- 7. If a previous version of the QIAxcel ScreenGel software is already installed on the computer, refer to <u>Updating the QIAxcel ScreenGel Software</u>. A dedicated update package is available to update the software. In this case, it is not possible to install the QIAxcel ScreenGel from a USB drive. If you have previously installed any version of the QIAxcel ScreenGel software and need to install it again from a USB drive, follow the <u>re-installation</u> procedure.

If the QIAxcel ScreenGel shall be installed for the first time, follow the instructions in the next section <u>First-time installation from a USB</u>.

Note: Default settings are recommended where appropriate for all installation steps.

- **Note:** We recommend having no other software installed on the computer for operating the QIAxcel Connect other than QIAxcel ScreenGel.
- 4.4.1 Switching ON the QIAxcel Connect

Check that QIAxcel Connect operates properly:

- 1. Make sure that the hood of the QIAxcel Connect is closed.
- 2. Power ON the QIAxcel Connect using the power switch on the front. The power switch lights up blue.

4.4.1.1 Getting started with the QIAxcel ScreenGel Software

Before using the software, read the <u>Concepts</u> of QIAxcel ScreenGel. Afterwards, configure the software. This can only be carried out by a user with the **Administrator** role.

- 1. Launch the QIAxcel ScreenGel software from the Windows start menu in the **QIAGEN/QIAxcel** folder or from the desktop icon.
- 2. Log in as Administrator and click OK (for this first login, no password is required).
- 3. Provide a valid password for the **Administrator** account. Leave the field for the old password empty, then provide a new valid password and confirm it.

Note: The password must contain one upper-case character, one lower-case character, and one digit. The minimum length of the password is eight characters.

- 4. The **Configuration** environment is displayed by default. Select the **User Manager** tab.
- 5. Create user accounts for all relevant users. For more information, refer to <u>User management</u> and <u>User</u> <u>roles</u>.
- 6. Configure which **COM** port is to be used for the connection to the QIAxcel Connect system. The default adjustment is **COM1**. For more detailed information, refer to <u>Settings</u>.
- 7. Configure the global settings for the QIAxcel ScreenGel software. For further details, refer to Settings.

4.4.2 First-time installation from USB

To install the QIAxcel ScreenGel software from a USB drive for the first time, follow the steps below:

- 1. Insert the USB drive in the computer's USB port.
- Double-click My Computer and double-click the USB drive. Launch the setup.exe program. This will start the QIAxcel ScreenGel software installation. The setup wizard will guide you through the setup process.

Note: Default settings are recommended where appropriate for all installation steps.

Note: It is possible to switch the language of the installed software later using the File menu.

3. To start the actual installation procedure, select the Install QIAxcel ScreenGel Software option.

Note: To read the user manual in the selected language, select the User Manual option.

- 4. The Microsoft Visual C++ 2010 x 86 Redistributable will be installed if it is not yet installed. In this case, a dialog opens. Accept the license agreement and click Install. Click Finish to complete the installation of this package. If you cancel or close the dialog before completing the installation, the QIAxcel ScreenGel software will not be able to operate.
- 5. The appropriate version of the Microsoft .NET Framework will be installed or updated at the beginning of the QIAxcel ScreenGel setup, if it is not yet installed. In this case, another dialog opens. Follow the steps to complete the installation of this package. If you cancel or close the dialog, the QIAxcel ScreenGel will not be able to operate.
- 6. Depending on the installed software packages from the step above, a computer restart might be required before proceeding with the setup. After the restart, the installation procedure resumes automatically.
- 7. The QIAxcel ScreenGel setup wizard opens. Click Next.
- 8. Accept the license agreement and click Next.
- Select the program installation path. The default path is C:\Program Files (x86)\QIAGEN\QIAxcel ScreenGel 2\. Click Next.

Note: If C:\Program Files (x86)\QIAGEN\QIAxcel ScreenGel 2\ is selected on a Windows 64 bit system, the installation will appear in C:\Program Files\x86\QIAGEN\QIAxcel\ScreenGel.

10. Select the data path for acquired data and all other application data. The default path is C: \ProgramData\QIAGEN\QIAxcel\ScreenGel 2\. Click **Next**.

Note: It is not possible to choose C:\Program Files (x86)\QIAGEN\QIAxcel ScreenGel 2\ also as the data path. It is also not possible to choose the system root path (e.g. **C:\Windows**) as the data path.

Note: All paths in this manual refer to the data path as %DATA_DIR%.

Note: The %DATA_DIR% directory and its sub-directories can be opened from QIAxcel ScreenGel using the **File/Open Data Directory** menu item.

Note: Read and write permissions are required for the data path. The permissions can be checked by rightclicking the folder, selecting **Properties**, and opening the **Security** tab.

11. To begin the installation, click Install. This may take some time.
- 12. The **QIAGEN QIAxcel Connect Driver Installer** dialog opens, if the driver is not yet installed on the computer. To install the driver, click **Next**. To complete the driver installation, click **Finish**.
- 13. To complete the QIAxcel ScreenGel setup wizard, click Finish.
- 14. To close the installation window, click Exit.
- 15. Connect the QIAxcel Connect to the computer.
- 16. After the installation, the QIAxcel ScreenGel can be launched either from the Windows Start menu under QIAGEN/QIAxcel or from the desktop icon. The user manual can be accessed in the available languages from the Windows Start menu.

4.4.3 Updating the QIAxcel ScreenGel Software

To update the QIAxcel ScreenGel software, a previous version of the software must already be installed on the computer and an upgrade package must be used. The upgrade package is available under the **Resources** tab on the product page at <u>www.qiagen.com/qiaxcel-connect</u>. Before updating, read the information provided on the web page. Experiments and user settings will be retained.

Important: It is not possible to update QIAxcel ScreenGel 1.6 to version 2.0.

To update the software from web download:

- 1. Use a Windows account with administrator privileges.
- 2. Close all programs running on the computer and disconnect the QIAxcel connect from the computer.
- 3. In your computer, download the latest version of the update package from the QIAGEN website. It is available under the **Resources** tab <u>www.qiagen.com/qiaxcel-connect</u>.
- 4. Unzip the update package.
- 5. Launch the setup.exe program. This will start the QIAxcel ScreenGel software installation.
- 6. From the drop-down menu, select the language in which QIAxcel ScreenGel shall be installed.

Note: It is possible to switch the language of the installed software later using the File menu.

- 7. To start the installation procedure, select the Install QIAxcel ScreenGel Software option.
- 8. The Microsoft Visual C++ 2010 x 86 Redistributable will be installed if it is not yet installed. In this case, a dialog opens. Accept the license agreement and click Install. Click Finish to complete the installation of this package. If you cancel or close the dialog before completing the installation, the QIAxcel ScreenGel will not be able to operate.
- 9. The appropriate version of the Microsoft .NET Framework will be installed or updated at the beginning of the QIAxcel ScreenGel software setup, if it is not yet installed. In this case, another dialog opens. Follow the steps to complete the installation of this package. If you cancel or close the dialog, QIAxcel ScreenGel will not be able to operate
- 10. Depending on the installed software packages from steps 8 to 10, a computer restart might be required before proceeding with the setup. After the reboot, launch the **setup.exe** program again and select **Install QIAxcel ScreenGel Software**.

- 11. The QIAxcel ScreenGel setup wizard opens. Click Next.
- 12. Accept the license agreement and click Next.
- Select the program installation path. The default path is C:\Program Files (x86)\QIAGEN\QIAxcel ScreenGel 2\. Click Next.

Note: If C:\Program Files (x86)\QIAGEN\QIAxcel ScreenGel 2\ is selected on a Windows 64 bit system, the installation will appear in C:\Program Files\x86\QIAGEN\QIAxcel\ScreenGel.

 Select the data path for acquired data and all other application data. Use the same data path as the installed software version. The default path is C:\ProgramData\QIAGEN\QIAxcel\ScreenGel 2\. Click Next.

Note: It is not possible to choose C:\Program Files (x86)\QIAGEN\QIAxcel ScreenGel 2\ also as the data path. It is also not possible to choose the system root path (e.g. **C:\Windows**) as the data path.

Note: All paths in this manual refer to the data path as %DATA_DIR%.

Note: The %DATA_DIR% directory and its sub-directories can be opened from the Windows Explorer directly from QIAxcel ScreenGel using the **File/Open Data Directory** menu item.

- 15. To begin the installation, click **Install**. This may take some time.
- 16. The **QIAGEN QIAxcel Connect Driver Installer** dialog opens, if the driver is not yet installed on the computer. To install the driver, click **Next**. To complete the driver installation, click **Finish**.
- 17. To complete the QIAxcel ScreenGel setup wizard, click **Finish**. To close the installation window, click **Exit**.
- 18. Connect the QIAxcel Connect to the computer.
- After the installation, QIAxcel ScreenGel can be launched either from the Windows Start menu under QIAGEN/QIAxcel or from the desktop icon. The user manual can be accessed in the available languages from the Windows Start menu.
- 20. Use one of the previously created user IDs for login.

Important: It is not possible to update from QIAxcel ScreenGel software version 1.6 to 2.0. However, run files can be loaded and analyzed.

4.4.4 Re-installation from a USB

Note: Downgrading (i.e., replacing a newer version with an older version of the software) the QIAxcel ScreenGel is not recommended. Experiments and profiles created with the newer version cannot be used with the older version. However, it may be necessary to re-install an older version of the software from USB to perform an upgrade. In this case, do not launch the software after re-installing it from a USB. Continue instead directly with the update procedure described above. It is not possible to launch the intermediate installation if the software version is below that which was initially installed on the computer.

If you have installed any version of the QIAxcel ScreenGel software before and need to install the QIAxcel ScreenGel software again from a USB, follow the steps below.

- 1. Ensure that the computer meets the minimum requirements (see Computer and software).
- 2. Use a Windows account with administrator privileges.

Note: Windows administrator privileges are only required for software installation.

3. Close any programs running on the computer and make sure that the QIAxcel Connect is not connected to the computer (i.e., unplug the connection from the computer).

To re-install from USB:

- Ensure that you know the data path of the installed QIAxcel ScreenGel software. Alternatively, launch QIAxcel ScreenGel software and login. Select **Open Data Directory** in the **File** menu. Keep open the launched Windows Explorer and close the software.
- 2. Uninstall the existing installation of the QIAxcel ScreenGel software through the Windows Control Panel.
- 3. If downgrading the QIAxcel ScreenGel software, rename or move the data directory of the existing version.
- 4. Place the USB drive in the computer's USB port.
- 5. Double-click **My Computer** and double-click the **USB drive**. Launch the **setup.exe** program. This will start the QIAxcel ScreenGel software installation. The setup wizard will guide you through the setup process.

Note: It is possible to switch the language of the installed software later using the File menu.

- 6. QIAxcel ScreenGel uses several software packages provided by Microsoft. If they are not installed on the system, these software packages are automatically installed at the beginning of the QIAxcel ScreenGel setup. When the respective dialog is displayed, click the **Install** button to install these required software packages. Depending on the installed software packages, a reboot of the system may be required before proceeding with the setup. After the reboot, the installation procedure is resumed automatically.
- 7. To start the installation procedure, select the Install QIAxcel ScreenGel Software option.
- 8. Accept the license agreement.
- 9. Select the program installation path. The default path is C:\Program Files (x86)\QIAGEN\QIAxcel ScreenGel 2\.
- 10. Select the data path for acquired data and all other application data. The default path is C: \ProgramData\QIAGEN\QIAxcel\ScreenGel 2\.
- 11. If you re-install an older version of the software from a USB drive, as a prerequisite for an update to the current version, select the path, mentioned in step 4.
- 12. If downgrading, it is important that the selected data path does not contain any files of the previously installed version. In particular, do not select the renamed directory from step 6.

Note: It is not possible to choose C:\Program Files (x86)\QIAGEN\QIAxcel ScreenGel 2\ also as the data path. It is also not possible to choose the system root path (e.g. **C:\Windows**) as the data path.

Note: All paths in this manual refer to the data path as %DATA_DIR%.

Note: The %DATA_DIR% directory and its subdirectories can be opened in the Windows Explorer directly from QIAxcel ScreenGel software, through the **File/Open Data Directory** menu item.

- 13. After the installation finished, click **Exit** to close the installation window.
- 14. If you installed an older version of the software from a USB drive, as a prerequisite for an update to the current version, do not launch the software. Continue directly with the <u>update procedure</u>.
- 15. After the installation, the QIAxcel ScreenGel software can be started either from the Windows Start menu under QIAGEN/QIAxcel or from the desktop icon. The user manual can be accessed in the available languages from the Windows Start menu.

Note: When downgrading, an error messages may appear indicating that one of the packages from the Operating System cannot be installed or that QIAxcel ScreenGel cannot be launched after installation. If the computer will only be used for the QIAxcel ScreenGel, open the Windows Control Panel and uninstall QIAxcel ScreenGel, all items starting with "Microsoft Visual C++ 20xx Redistributable - ...," and the "Microsoft.NET Framework" items. Then try to install the QIAxcel ScreenGel software again. If the problem persists, contact QIAGEN Technical Services. If the computer is not intended to be used only for the QIAxcel ScreenGel software, exercise care when uninstalling the listed items. These may be used by other software running on the computer. Have the installation package(s) of the other software at hand in case repair is necessary.

4.4.5 Uninstall software

Important: For updates, uninstallation of QIAxcel ScreenGel is not necessary. For this purpose, follow the instructions in section <u>Updating the QIAxcel ScreenGel Software</u>.

When uninstalling QIAxcel ScreenGel, experiment and report data within the predefined data directory is retained. This directory can be opened via **Open Data Directory** from the **File** menu. A **File Explorer** window will open.

To uninstall the QIAxcel ScreenGel, please follow these steps:

- 1. Close the QIAxcel ScreenGel software.
- 2. Open the Control Panel by clicking on the Windows Start menu and selecting Control Panel.
- 3. Depending on your layout settings, either click Programs and Features or Add/Uninstall Programs.
- 4. Select QIAxcel ScreenGel to be uninstalled and click Uninstall.

Note: Depending on the Windows configuration, there may be a pop-up message asking if you want to allow the program to install. Allow the program to install.

- 5. The installer window opens. Click Uninstall.
- 6. Follow the instructions of the installer to uninstall.
- 7. Finally, click Exit to close the installer window.

If you want to uninstall QIAxcel ScreenGel and delete all experiment and report data, open the %DATA_DIR % directory as described above before uninstalling QIAxcel ScreenGel. Then, delete all data using the File Explorer.

4.5 Packing the QIAxcel Connect

WARNING/ CAUTION

Risk of personal injury and material damage [W2]

The QIAxcel Connect is too heavy to be lifted by one person. To avoid personal injury or damage to the instrument, do not lift the instrument alone.

If you need to transport the QIAxcel Connect, pack the instrument as follows:

- 1. Switch on the QIAxcel Connect.
- 2. Switch on the computer and launch the QIAxcel ScreenGel.
- 3. Click in the <u>Status Information panel</u> to move the buffer tray to the front of the instrument.
- 4. Open the sample door and remove the buffer tray.

Note: Do not open the sample door if the cartridge door is open. This may damage the instrument. Please close the cartridge door before opening the sample door.

- 5. Remove the QIAxcel gel cartridge from the QIAxcel Connect and store in the QX Cartridge Stand as described in section <u>Unpacking a QIAxcel Kit</u>.
- 6. Remove the N₂ bottle as described in section <u>N2 cylinder replacement</u>.
- 7. Close the QIAxcel ScreenGel and switch off the QIAxcel Connect.
- 8. Disconnect the USB cable from the instrument to the computer and remove the AC power cord.
- 9. Reinstall the transport lock holding down the sample plate holder (see section <u>Installing the QIAxcel</u> <u>Connect</u>).
- 10. Place the QIAxcel Connect into the original packaging it was shipped in.

5 Operating procedures

This section describes how to operate the QIAxcel Connect.

Before proceeding, we recommend that you familiarize yourself with the features of the QIAxcel Connect by referring to the section <u>General Description</u>.

The QIAxcel Connect cartridge door, sample door, and service door must remain closed during operation of the instrument. Only open the doors when the instrument is not operating or when prompted by the software.

Note: Opening the cartridge door or sample door during operation of the QIAxcel Connect will cause the system to stop the action it is currently performing. Any sample run in progress cannot be recovered. However, due to the small volume of sample used, a new run can be performed.

Note: Do not open the sample door if the cartridge door is open. This may damage the instrument. Close the cartridge door before opening the sample door.

	Risk of personal injury and material damage [W3] Do not attempt to move the QIAxcel Connect during operation.
--	---

WARNING	Moving parts [W10]
\wedge	To avoid contact with moving parts during operation of the QIAxcel Connect, the instrument must be operated with the cartridge door and sample door closed.
	If the door sensors are not functioning correctly, contact QIAGEN Technical Services.

5.1 Setting up the QIAxcel Connect

The steps required for correct setup of the QIAxcel Connect instrument before each use are described in this section.

5.1.1 Unpacking a QIAxcel Kit

- 1. Carefully read the handbook supplied with the QIAxcel Kit before starting.
- Remove all buffer bottles from the kit. Refer to the handbook for a detailed description of the kit contents.

- 3. Add 10 ml QX (HS) Wash Buffer to both reservoirs of the QX Cartridge Stand (provided with the QIAxcel instrument) reservoir and cover it with 2 ml mineral oil.
- 4. Remove the QIAxcel gel cartridge from its packaging and carefully wipe off any soft gel debris from the capillary tips using a soft tissue.

Note: The QIAxcel gel cartridge is provided together with a smart key, which is used for fully automatic cartridge detection. Do not remove the smart key from the cartridge.

5. Remove the purge cap seal from the back of the QIAxcel gel cartridge and place it in the QX Cartridge Stand.



Note: A soft tissue should be used to wipe off any gel that may have leaked from the port.

Note: Ensure that the capillary tips are submerged in QX (HS) Wash Buffer.

Important: New cartridges must be allowed to equilibrate at room temperature for at least 20 minutes prior to use. Place the gel cartridge in the QX Cartridge Stand, protected from light with the QX Cartridge Stand Cover, or store the cartridge latched in the instrument in the Park position with QX (HS) Wash Buffer in the wash park (WP) position, and allow to stand at room temperature (15–25°C) for at least 20 minutes. Do not equilibrate the gel cartridges inside the kit box or in the black cartridge blister.

Note: Incorrect cartridge handling or shorter equilibration times may cause damage to your QIAxcel Connect instrument and could result in a loss of warranty.



5.1.2 Loading a QIAxcel gel cartridge and smart key

Note: Read section **Unpacking a QIAxcel Kit** in the QIAxcel Connect User Manual prior to performing this procedure.

- 1. Switch on the QIAxcel Connect at the power switch.
- 2. Switch on the computer and launch the QIAxcel ScreenGel software from the Windows start menu under **QIAGEN/QIAxcel ScreenGel** or from the desktop icon.
- In QIAxcel ScreenGel, select a mode (DNA or RNA) and log in (for detailed information, refer to the <u>User authentication</u> section). The **Process** environment opens and displays the first screen of the Process Setup wizard.

Note: The icon on the top left indicates that a connection is being established and the icon shows that the QIAxcel Connect is already connected. In the case that the instrument could not be connected, you will be notified that the instrument is not available. If you intentionally did not switch on the instrument, click Instrument not needed. Otherwise click Troubleshoot.

Follow the instructions in the following message. For more detailed instructions, refer to the troubleshooting

section <u>System setup</u>. Close the message. To retry to connect to the instrument, click the *I* icon.

Note: Integral part of QIAxcel Connect system, besides ScreenGel is software for the instrument itself. QIAxcel ScreenGel comes with an up-to-date instrument software for QIAxcel Connect instruments with serial numbers 50000 and higher. If ScreenGel connects to the instrument for the first time, it checks if the instrument software is up-to-date. If not, it updates the instrument software. This process takes a few minutes. Do not turn off the instrument or disconnect the cable between the computer and the instrument, and do not close the QIAxcel ScreenGel software until the update is completed. After a successful update, QIAxcel ScreenGel automatically connects to the instrument.

- 4. Remove the QIAxcel gel cartridge from the QX Cartridge Stand.
- Open the cartridge door and insert the QIAxcel gel cartridge into the QIAxcel Connect. The cartridge description label should be facing toward the front and the purge hole should be pointed toward the rear of the instrument.

Note: Ensure the purge cap seal has been removed as described in the section **Unpacking a QIAxcel Kit** in the QIAxcel Connect User Manual.

6. Insert the smart key into the smart key socket near the cartridge. The smart key can be inserted in either direction.



Note: The system will not recognize the cartridge and will not operate if the smart key is not inserted.

Note: If the option **Latch cartridge automatically** is selected, the cartridge will be automatically latched after the smart key is detected. Therefore, it is important to insert the smart key as soon as the cartridge has been inserted and the cartridge door has been closed. Refer to section <u>Settings</u> for information about settings.

7. Close the cartridge door. The cartridge ID and cartridge type will be displayed automatically in **Cartridge Status** in the <u>Status Information panel</u>.

► Instrument Status				
▼ Cartridge Status				
Cart	Cartridge ID			
C21	0507A99			
Cart	ridge Type	e		
DNA	A Screeni	ng		
Rem	aining Ru	ns	Calibration Runs Left	
83	5		2	
Expi	ration Sta	tus	Calibration Status	
Vali	d	ius.	OK	
•	Calibratio	on Details		
Calil	bration Da	ite	Accepted By	
07.0	05.2021 1	1:40:02	Service	
Ch.	Area	Cal. Factor	Result	
1	0,0051	1,4144	Passed	
2	0,0051	0,9689	Passed	
3	0,0050	0,8727	Passed	
4	0,0045	1,0018	Passed	
5	0,0047	0,8044	Passed	
6	0,0045	0,7396	Passed	
7	0,0051	1,0330	Passed	
8	0,0054	1,1428	Passed	
9	0,0051	2,0957	Passed	
10	0,0049	0,7969	Passed	
11	0,0051	1,1086	Passed	
12	0,0051	0,7276	Passed	

5.1.3 Removing a QIAxcel gel cartridge

To remove a QIAxcel gel cartridge, proceed as follows:

- 1. Prepare the QX Cartridge Stand as described in section Unpacking a QIAxcel Kit.
- 2. Open the cartridge door.
- 3. Remove the smart key.

Note: If the **Latch cartridge automatically** option is selected, the cartridge will be unlatched automatically after the smart key is removed. Therefore, it is important to remove the smart key first before trying to remove the cartridge. Refer to section <u>Settings</u> for information about settings.

Note: If the QIAxcel Connect is switched on and **Latch cartridge automatically** is unchecked, ensure the QIAxcel gel cartridge is unlatched before removing. If latched, first unlatch it. See section <u>Status Information</u> panel for information on how to do this.

- 4. Remove the QIAxcel gel cartridge.
- 5. Install the purge cap seal to close the purge port.
- 6. Place the QIAxcel gel cartridge in the QX Cartridge Stand.

Note: Ensure that the capillary tips are submerged in QX (HS) Wash Buffer.

5.1.4 Storing a QIAxcel gel cartridge

Storage of QIAxcel DNA gel cartridges after delivery

Please see the respective QIAGEN kit handbooks for information about the storage conditions of the QIAxcel cartridges and other kit components.

Daily use

If the QIAxcel gel cartridge is used on a daily basis, the cartridge must be latched and stored in the QIAxcel Connect in "Park Position."

Note: The QIAxcel Connect must be left ON if the cartridge is stored in the "Park Position" and the cartridge must be latched. Do not switch the QIAxcel Connect OFF.

If more than one QIAxcel gel cartridge is being used on a daily basis, store the second cartridge in the QX Cartridge Stand in the dark or protected with the cover. Make sure that the cartridge stand reservoir is filled with wash buffer and covered with mineral oil (refer to <u>Unpacking a QIAxcel Kit</u>). Alternatively, close the purge port with the purge port seal, return the QIAxcel gel cartridge to the blister package, inserting the capillary tips into the soft gel, and store the QIAxcel gel cartridge at room temperature in an upright position (see orientation label on blister package).

The QIAxcel gel cartridge can be stored in this manner until expiration date as indicated on the kit label.

5.1.5 Preparing the buffer tray

Please see the respective QIAGEN kit handbooks for information about the preparation of the buffer tray.

5.1.6 Loading the buffer tray

To load the buffer tray proceed as follows:

1. Close the cartridge door and the sample door.

Note: The QIAxcel Connect cartridge and sample doors must remain closed during operation of the instrument. Opening either door during operation will cause the system to stop any action it is currently performing.

- 2. Click in the <u>Status Information panel</u> at the left to move the buffer tray holder to the front of the instrument. Wait until the buffer tray holder has reached its stop position.
- 3. Open the sample door.

Note: Do not open the sample door, if the cartridge door is open. This may damage the instrument. Close the cartridge door before opening the sample door.

4. Carefully place the filled buffer tray into the buffer tray holder. Be careful not to spill any solutions in the instrument or cause any cross-contamination between buffers loaded on the buffer tray.

Note: The 12-tube strips should be positioned towards the front of the instrument with the buffers towards the back.



5. Close the sample door and click in the <u>Status Information panel</u> to move the buffer tray holder to the **Wash Park** position.

Note: Leave the sample door open if you want to load your samples immediately afterward.

Note: If you close the sample door, the buffer tray automatically moves to the **Wash Park** position after a period of 5 minutes.

5.2 Operating the QIAxcel Connect

Samples are processed using the **Process** environment of the QIAxcel ScreenGel Software.

5.2.1 Things to do before starting

- Carefully read the handbook supplied with the QIAxcel Kit.
- Prepare the reagents and QX Alignment Marker to be used. For instructions about how to do this, see section <u>Setting up the QIAxcel</u>, or refer to the QIAxcel Kit handbook.
- Prepare your samples. Refer to the handbook supplied with the QIAxcel Kit for instructions about how to do this. For optimal results, the sample solution should be pH 6–9 and should not have an ionic content greater than that of a typical PCR buffer.

Note: If you need to run a size marker side by side with the samples, prepare it in the required position on the sample plate. Refer to the handbook supplied with the QIAxcel Kit for instructions about how to do this.

CAUTION Damage to the cartridge [C8] If less than 12 samples are processed, fill the empty sample wells with the QX DNA (HS) Dilution Buffer or QX RNA (HS) Dilution buffer. Failure to do so may cause damage to the capillary channels.

5.2.2 Starting a run

To start a run, follow the steps in <u>Running a process</u>. User role Operator can only start predefined Process.

6 QIAxcel ScreenGel Software

The QIAxcel ScreenGel software has been specifically developed for use with the QIAxcel Connect system. The software offers extensive tools that simplify data analysis, allow fast data interpretation, and provide flexibility in viewing results in Electropherogram and Gel image formats.

The software provides a wizard-based run setup and convenient profiles for standardized sample processing, and effortless, customizable documentation of results.

The QIAxcel ScreenGel software is based on an unique algorithm, that enables to analyze separation data more reproducibly and accurately than other chromatographic analysis packages. It evaluates distribution profiles and calculates a variety of peak properties, such as peak number, height, width, and peak area.

6.1 Concepts

This section explains the following fundamental concepts that each user working with QIAxcel ScreenGel should become familiar with:

- Modes
- User management
- Environments
- Profiles

6.1.1 Modes

The QIAxcel ScreenGel has two modes – one for DNA and one for RNA analysis. The main purpose of the modes is to adapt the data analysis algorithms to the properties of the analytes. In addition, the respective modes ensure that correct cartridge types are used and that only data of the right type can be loaded from files.

	QIAxcel Screer	nGel	
QIAGEN -			
Mode			
DNA			
User ID			
JohnSm	ith	\otimes	Cancel
Password	1		
	•••	\rightarrow	Login

Note: QIAxcel ScreenGel offers the possibility to switch between DNA and RNA mode during analysis.



6.1.2 User roles

The QIAxcel ScreenGel software distinguishes between three different user roles, which differ in their permissions. Each user is assigned one of the three user roles. This section provides a short overview of the use and permissions of each user role:

Role	Used for	Tasks	Access
Administrator	Sample processing	Creating/modifying profiles	All environments (full
	Analysis of data with full	Sample processing	access)
	access to analysis parameters	Analysis of samples	
	Creation and modification	Report/export	
	of profiles	Installation of software	
	User management	Configuration of global settings	
	Software configuration	Administration of user accounts	
	QIAsphere	Administration of QIAsphere	
Developer	Sample processing	Creating/modifying profiles	All environments
	Analysis of data with full	Sample processing	(restricted access)
	access to analysis parameters	Analysis of samples	
	Creation and modification of profiles	Report/export	
		Configuration of global settings	
Operator	Sample processing and	Sample processing	All environments
	analysis of data without the option of modifying	Analysis of samples	(restricted access)
	profiles	Report/export	

Note: A "Routine User" role can be activated additionally. A user with this role is restricted to predefined Process Profiles. Routine User roles are limited to view experiments only. Contact QIAGEN's technical service for further information.

• Administrator

This role is intended for management of settings and users. Additionally they have full access to the functionality of the software.

Users assigned this role can change the global settings and perform user management, i.e., management of user accounts (rights, user role, etc.). Users have full access to profiles, the **Process** environment, the **Analysis** environment, the **Service** environment, or the **Configuration** environment.

• Developer

This role is intended for experienced users.

Users assigned this role have access to the full functionality of the software, except for user management and **QIAsphere**. Users can create analysis, process, and report profiles and therefore prepare the working base for other users. They can perform advanced analysis, such as comparison of results from different processes (see <u>Customizing experiments</u>) and modify profile parameters, if required.

• Operator

This role is intended for users operating the QIAxcel Connect/QIAxcel ScreenGel in combination with predefined profiles, particularly analysis profiles.

Users assigned this role have access to all environments of the software; however, they cannot create new or modify predefined profiles, such as analysis and Process Profiles.

While setting up a new process using predefined Process Profiles, they can modify marker settings and rows that need to be processed. In the analysis environment, they can open existing experiments and perform routine analysis using predefined analysis profiles. They can perform report and export actions using predefined report/export profiles with the option to modify the report/export parameters, but without the option to save these changes.

Note: One person can be assigned several user roles by creating several user accounts with different roles.

Refer to the <u>User management</u> section for information on how to create user accounts.

6.1.3 Environments

The QIAxcel ScreenGel software is composed of four environments providing different functionality. Only one environment can be used at a time.



To switch to an environment, click the environment icon. The functionality of the environments is briefly described in the following table:



Contains all functions for maintenance and troubleshooting of instrument and cartridges. For detailed information, refer to the <u>Service</u> section.



Configuration environment for administration of users and system configuration.

Provides access to software preferences (settings), profile management, user management and configuration. For detailed information, refer to the <u>Configuration</u> section.

The Main toolbar can be minimized or maximized by clicking on the icon at the right top corner of the screen:



Switch between the four environments by clicking on the **View** menu:



6.1.4 Profiles

Data acquisition and analysis in the QIAxcel ScreenGel software are controlled by **Profiles**. Profiles combine settings and parameters and can be conveniently re-used for different purposes (see table below).

The standard procedure is to use predefined profiles. QIAxcel ScreenGel will be installed with a set of predefined profiles that cannot be modified. However, if modifications are necessary, experienced users can create new profiles or change profile settings as required. See the following table:

Profile	Description
<u>Process Profile</u>	To define settings for data acquisition. Defines all parameters for the electrophoresis run.
<u>Analysis profile</u>	To define the settings for sample data analysis (analysis parameters).
Peak calling instruction	To define search criteria for peaks of interest.
	Note: Peak calling instructions are available only if <u>peak calling features are</u> <u>active</u> .
Distribution profile	To define criteria for quality assessment of a nucleic acid, such as the distribution of libraries, including quality checks of the molar ratio or height, as well as molarity or concentration.
	Note: Distribution profiles are available only if <u>distribution analysis features</u> <u>are active</u> .
<u>Report/export profile</u>	To define report and export of sample data and analysis results in various formats.

Note: Process Profiles can include an Analysis profile, a Peak Calling Instruction or a Distribution profile, and a Report/Export profile, which allows fully automated processing including analysis and report.

Note: Analysis, Distribution and Report/Export profiles and **Peak Calling Instructions** are included in a Process Profile by copying the profile name and all profile parameters into the Process Profile. That means that modifications to the original profiles are not automatically copied into the Process Profile. This prevents Process Profiles from being changed by mistake and ensures process stability. If modifications to the underlying profiles should also be present in the Process Profile, include the modified profile again.

Available profiles and methods are loaded once, upon starting the software.

Profiles created with a previous version of the QIAxcel ScreenGel software are automatically converted (migrated) to the newest version.

Note: A profile created or saved with a newer version of the QIAxcel ScreenGel software cannot be opened in previous software versions.

Note: The profile names of the predefined profiles which are installed together with the software are displayed in the selected software language. The profile names of user created profiles are displayed in the language in which the profiles were created. For information how to change the software language, refer to the end of section <u>Settings</u>.

6.1.5 General software usage

QIAxcel ScreenGel is a modern software with a state-of-the-art graphical user interface. The controls of the user interface behave in the same way as standard Windows controls. This section explains frequently used terms and several concepts which are specific for the QIAxcel ScreenGel user interface.

In the following table, user interface-related terms that are used in this manual are explained.

Left-click Press the left button of a computer mouse or touchpad.

Right-click	Press the right button of a computer mouse or touchpad.
Double-click	Left-click twice, very quickly.
Drag-and-drop	Move an object (sample, lane) to another place using a computer mouse or touchpad. Left-click to pick up the object. Hold and press the left mouse button while dragging the object to the new place. Release the mouse button to release the object in its new position.
	If more than one object was selected, the left-click picks up all selected objects.
Ctrl-click	Press and hold the Ctrl key while pressing the left mouse button.
Ctrl+A	Press and hold the Ctrl key while pressing the A key.
Context menu	When right-clicking certain controls, a context-sensitive menu is shown. Not all controls provide a context menu.

Experiments		Gel Image	Electropherog
> 🖥 🎘 🗗			م 🗘 ۲۷
▼ *FA_100504_NL	▲ ×		
▼ FA_100504_NL	R1 E1		
1 2 3 4	5 6 7 8 9 10 11 12		
	Visualize Selected Samples		0 th /
FA_100504_N	Superimpose		~
► FA_100504_N	Size Marker		4
	Remove Analysis Data		
	Transfer Analysis Instructions		
	Revise Sample Information or	Sample Comme	ent
	Remove Peak Calling Data		
	Transfer Peak Calling Instruct	lion	
	Pomovo Pow		

Tool tip When the mouse pointer hovers over a control without clicking it, a small text window appears. Not all controls provide a tool tip. Usually a tool tip gives useful information about the functionality of a control, or useful information about possible problems (errors or warnings).

Drop-down box Drop-down lists allow the selection of one item from a predefined list of items. First, left-click on the dropdown arrow to open the list. Then move the mouse down to the correct item. Left-click to select the item.

	🖲 DNA 🔿 RNA	
	Cartridge Type	
	DNA High Res.	▼
	DNA Fast Analysis	
-	DNA High Res.	
_	DNA High-Sensitivity)
	DNA Screening	

Toggle buttonLeft-click the button to toggle. When the button is pressed, the functionality is on. Left-
click the button again to turn the functionality off.

Mode

Rubber band Span a "rubber band" to define a rectangular region (for zooming in). Left-click to define the starting point, hold and press the mouse button while dragging. While dragging, a "rubber band" is shown, outlining the rectangular region. Release the mouse button to define the region. The "rubber band" disappears. The system performs the (zooming) functionality based on the defined region.

The status bar (the information line at the bottom of the application window) shows the current user logged in, the date and time, user login name, and application mode (DNA or RNA).

Login: 23.06.2021 08:53:10 Name: Developer Mode: DNA

Some areas in the software can be expanded or collapsed; e.g. collapse the **Experiment Explorer** and **Parameters Panel** in the **Analysis** environment by clicking ▼ and expand them again by clicking ►.

6.1.5.1 Editing data in tables

The graphical user interface of the QIAxcel ScreenGel software contains many tables. If the table data is editable, data or values can be changed in a special edit area at the bottom of the table. The table cells themselves are not editable.

Size [bp]	Conc.[ng/µl]
100	1,38 🔺
200	1,38
300	1,38
400	1,38
500	1,38
600	1,38
700	1,38
800	1,38
1000	3,44
1200	1,38
1500	1,38
2000	1,38
2500	1,38
	Add Delete
Size Co 610 bp	ncentration 1,38 ng/µl
	OK Cancel

An example of an editable table is a Size Marker table in the Process environment. See image below:

For example, to change the base pair size from 600 to 610:

- 1. Highlight the required row by left-clicking it.
- 2. Click on the **Size** edit field below the table.
- 3. Enter 610. While editing, only the fields and buttons of the edit area are enabled. In this case, the **Size** and **Concentration** edit fields as well as the **OK and Cancel** buttons are all enabled.
- 4. To accept the values into the Size and Concentration fields, click OK. To cancel editing and discard the changes, click Cancel.

Note: The **OK** and **Cancel** buttons are only enabled if the values in the editable fields have been changed and are valid.

6.1.5.2 Displaying errors and warnings

In QIAxcel ScreenGel, a yellow backgrounds indicate an error, a warning, an inconsistency or missing data.

Validation during editing

If an incorrect value is entered into a field, the background turns yellow. In addition, a tool tip will be displayed which explains the problem. The following screenshots show examples of errors and the corresponding tool tips.

Size Concentration	Size Marker
45 bp 20.0 ng/µl	Total conc.
	Size [bp] Conc.[ng/µl]
l.	▲ 15 ▲
	100 1,38
	200 1,38
	300 1,38
	400 1,38
	<u> </u>
Marker Selection	700 1.38
	800 1.38
O No Marker	1000 3.44
 Reference Marker Table 	
Run size marker side by side with sample	Duplicates are not allowed. AlignmentMarker Entry Limit exceeding is not allowed.
Alignment Marker	2500 1.38
	<u> </u>
Size Marker	Add Delete Size range start Size range end
No Size Marker selected	100 bp 2500 bp
	Size Concentration bp ng/µl
	OK Cancel

Process screens

If the **Process Setup** wizard screen contains an error or a warning, a yellow triangle appears besides the subsection title. If the instrument is not connected to the software, a plug icon with a yellow background is displayed in the **Status Information** panel.



Note: The screen marker **Run Check** remains blue (normal state) even if errors/warnings have been detected. To start a process, operate as described in the <u>Running a process</u> section.

Message boxes

Message boxes are used in QIAxcel ScreenGel to report problems.



6.1.5.3 Displaying modifications

The QIAxcel ScreenGel displays modifications in a Windows compliant way, using an asterisk at the left of the name of the modified object. This is shown by a few examples below.

Analysis environment/modification of an experiment (apply or remove analysis, change the lane order, etc.).



The name of the experiment will be displayed with an asterisk.

Note: A changed profile will be shown in the same way.

Process environment/change of any parameter in the Process wizard

The selected Process Profile will be displayed with an asterisk.

Set the process profile!	
PROFILE DEFINITION	Mode
1. Process Profile	● DNA ○ RNA
2. Run Parameters	Cartridge Type
3. Analysis	DNA High-Sensitivity
4. Marker	Process Profile
5. Peak Calling	Peak Calling Distribution Analysis
6. Report/Export	*Default DNA High-Sensitivity ▼
START A PROCESS	Profile
1. Sample Selection	Included Steps
2. Sample Information	Run Analysis Peak Calling Report
3. Run Check	Experiment Directory
	C:\ProgramData\QIAGEN\QIAxcel\ScreenGel 2\Data\Experiment\DNA
	Allow Directory Selection
	Notes
	Save process profile as Run

Change of parameters in the Peak Calling screen

The selected peak calling instruction in the **Peak Calling Instruction** drop-down list will also be displayed with an asterisk.

PROFILE DEFINITION		Peak Calling Instruction		
1. Process Profile	L	*Lock for 46 bp and 500 bp	•	Save as .
2. Run Parameters		Peaks of Interest		1
3. Analysis		Include size marker samples		
4. Marker		Find centered peak in interval		
5. Deck Collins		 Find highest peak in interval 		
5. Peak Calling	~	Table Definition		
6. Report/Export		Name	Position	Tol. [%]
START A PROCESS		46 bp	46 bp	5,00
1. Sample Selection		500 bp	500 00	3,00
2. Sample Information			Add	Delete
3. Run Check				
		Name		
		500 bp		
		Position	Tolerance	
		500 Size ▼	3 %	

Note: You can start a process or leave the **Process** environment without saving your changes. QIAxcel ScreenGel will not remind you to save a Process Profile or the corresponding analysis profile, report/export profile, or peak calling instruction. If you leave the **Process** environment, log out, or exit the application, please save your changes if they are to be re-used.

6.1.5.4 Getting help

The QIAxcel ScreenGel has a context-sensitive help system. After pressing the **F1** key, a context-specific help page is shown. The following image shows the help page corresponding to the login screen:



Note: The **Start** menu folder under **QIAGEN/QIAxcel** also contains links to the user manual in **PDF** and **CHM** format.

Alternatively, select Help from the Help menu.



Search for a description of the software features in the section <u>QIAxcel ScreenGel Software</u>, organized in sections corresponding to the environments of the software. Use the content tree on the left-hand side to navigate through the help system.



The help system provides several links to other sections. Use the navigation links or buttons of the help system to return to the previous topic.



6.2 User authentication

To use the QIAxcel ScreenGel, the user must first log in. User authentication is required before working with the instrument. For details about how to create/add users and modify user settings, refer to the <u>User management</u> section.

Log in

After launching the QIAxcel ScreenGel, the Log in dialog box appears:

🛞 Car	ncel
_	
-⊴ Log	jin
	⊗ Car ⊸] Log

To log in to the software:

- 1. Select the application mode (DNA or RNA mode).
- 2. Enter the User ID.
- 3. Enter the password.
- 4. Click Login.

Note: The DNA mode is preselected by default in the Mode drop-down list.

Note: The mode could also be changed in the application after login.

Note: If 21 CFR Part 11 support is activated in the <u>Settings</u>, the user account will be locked if the maximum number of failed login is exceeded. The maximum number of failed log in is set by the administrator. The administrator can also unlock the user account in the <u>user manager</u>. If the administrator account is locked, please contact QIAGEN Technical Services.

Note: Refer to the User management section for information on how to create user accounts.

Note: Refer to the <u>Getting started with the QIAxcel ScreenGel software</u> section for information about the first login.

Log out

To log out:

- 1. Select File/Logout from the application menu bar.
- 2. A dialog appears. Click Yes to confirm the log out.
- 3. After logging out, the log in dialog appears.

Note: Before exiting the QIAxcel ScreenGel, cancel the log in dialog first.

Changing the user password

To change your password:

- 1. Select File/Change Password from the application menu bar.
- 2. The Change password dialog is displayed.
- 3. Enter the old password.
- 4. Enter the new password.

Note: The password must contain one upper-case character, one lower-case character, and one digit. The minimum length of the password is eight characters.

- 5. Confirm the new password.
- 6. Click **OK**.

QIAGEN	Change password for user Developer
Re-enter yo	our old password
Enter a nev	v password
Confirm the	e new password

Locking the software

The software can be locked to block user access if the computer is left unattended.

To lock the software:

- 1. Select File/Lock Application from the application menu bar.
- 2. When the application is locked, the following dialog is shown:

Application locked				
QIAxcel ScreenGel Software is in use and has been locked. Only Administrator can unlock this application.				
Unlock Shutdown				

Automatic software lock

In order to support 21 CFR Part 11 requirements, the software can be automatically locked after a defined time without user interaction. You can activate 21 CFR Part 11 support and specify the auto-lock time period in the **Settings**. For further details, refer to the <u>Settings</u> section.

Unlocking the software

To unlock the software:

- 1. Click the **Unlock** button.
- 2. The Log in dialog appears.
- 3. Enter User ID and password, click Login.

Note: Only a user with an equal or higher <u>user role</u> can unlock the application.

6.3 Process

The Process environment provides all functionality required for data acquisition here:

- Defines process parameters
- Starts the sample process
- Monitors the data acquisition process

Reference ScreenGel			- 🗆 X
File View Help	Service Configuration		Notifications
Status Information	Process Setup		
	Set the process profile!		
► Instrument Status	PROFILE DEFINITION	Mode	
▼ Cartridge Status	1. Process Profile	DNA O RNA	
Cartridge ID C220609000	2. Run Parameters 3. Analysis	Cartridge Type DNA High Res.	
DNA High Res.	4. Marker	Process Profile	(i) IMPORTANT:
Remaining Runs Calibration Runs Left	5. Peak Calling	Peak Calling Distribution Analysis	Select the Distribution Analysis for NGS library QC or RNA degradation
Expiration Status Calibration Status	6. Report/Export	Default DNA High Resolution	assessment or other analysis based on smear. The list of available process profiles and the analysis features are
Valid Not OK	START A PROCESS	Destin	adjusted accordingly to the selection.
Calibration Details	1. Sample Selection 2. Sample Information 3. Run Check	Included Steps Plan Analysis Peak Calling Report Experiment Directory C:\ProgramData\GLAGEN\GLAXcel\ScreenGel 2\Data\Experiment\DNA V Allow Directory Selection Notes	
		Deteutu process prole for Unive High Hesolution Carthole, it applies; Method: OM500 Alignment Matker: OX 15 kp-5 kb Galage Stream (Stream) Analysis: Default DNA Analysis Report: DNA Fragment Analysis Save process profile as	

Parameters for a process can be stored for later re-use in a Process Profile.

If using a predefined Process Profile, parameters for data acquisition, for analysis, and report and export options are already set. Then, only the specific sample information can be adjusted and the data acquisition process can be started. A predefined Process Profile can be used by the **Operator**, **Developer** or **Administrator** users.

Refer to the <u>Running a process</u> section for detailed information about how to define process parameters and to start a process. There are some limitations for users with the user role **Operator** with respect to analysis parameters and methods.

To fully automate a process, define a <u>Process Profile</u> that optionally includes an **Analysis** profile for analysis parameters and marker information, as well as a peak calling instruction, and a **Report** profile for Report/ Export options. Refer to the <u>Creating a new Process Profile</u> and <u>Modifying a Process Profile</u> sections for information on how to prepare and save Process Profiles for re-use. To create and change Process, Analysis and/or Report profiles, you have to be logged in as **Developer** or **Administrator**. Data acquired during processing will be stored in an experiment file. Refer to the <u>Run parameters and result</u> <u>structure</u> and <u>Handling samples and experiments</u> sections for detailed information.

6.3.1 Running a process

Users with the **Administrator**, **Developer** or **Operator** user role can modify Process Profiles as required. Restrictions apply to users with the Operator role as they can, for example, start a modified Process Profile, but not save it. Details on how to create a new process profile are described in <u>Creating a new process</u> profile.

Follow these steps to perform runs with advanced options:

- 1. Switch on the QIAxcel Connect at the power switch.
- Switch on the computer, launch the QIAxcel ScreenGel from the Windows start menu under QIAGEN/ QIAxcel ScreenGel or from the desktop icon.
- In the QIAxcel ScreenGel software, select a mode (DNA or RNA) and log in (for detailed information, refer to the <u>User authentication</u> section). The **Process** environment opens and displays the first screen of the Process Setup wizard.

Note: The icon on the top left indicates that a connection is being established and the icon shows that the QIAxcel Connect is already connected. In the case that the instrument could not be connected, you will be notified that the instrument is not available. If you intentionally did not switch on the instrument, click Instrument not needed. Otherwise click Troubleshoot.

Follow the instructions in the following message. For more detailed instructions, refer to the troubleshooting

section <u>System setup</u>. Close the message. To retry to connect to the instrument, click the *I* icon.

Note: Integral part of QIAxcel Connect system, besides ScreenGel is software for the instrument itself. QIAxcel ScreenGel comes with an up-to-date instrument software for QIAxcel Connect instruments with serial numbers 50000 and higher. If ScreenGel connects to the instrument for the first time, it checks if the instrument software is up-to-date. If not, it updates the instrument software. This process takes a few minutes. Do not turn off the instrument or disconnect the cable between the computer and the instrument, and do not close the QIAxcel ScreenGel software until the update is completed. After a successful update, QIAxcel ScreenGel automatically connects to the instrument.

4. Install the QIAxcel gel cartridge to be used as described in <u>Installing a QIAxcel gel cartridge and smart</u> key.

Note: You can inspect the cartridge status in the **Status Information** panel to the left of the **Process** environment. Refer to the <u>Status Information panel</u> section for detailed information.

5. Place the buffer tray containing the QX Alignment Marker onto the buffer tray holder as described in Loading the buffer tray.

6. Place the sample strips (in position A) or a 96-well plate onto the sample plate holder.

7. Close the sample door.

Note: If the instrument doors are closed, the buffer tray automatically moves to the **Wash Park** position after about 5 minutes.

In the Process environment of QIAxcel ScreenGel, select a Process Profile in the Process Profile screen

 The Process Profile drop-down list shows Process Profiles for the current mode and cartridge type.
 The QIAxcel ScreenGel software comes with a set of predefined Process Profiles. As a starting point,
 select the Process Profile that fits best to your sample type.

Set the process profile!		
PROFILE DEFINITION	Mode	
1. Process Profile 1 >	DNA O RNA	
2. Run Parameters	Cartridge Type	
3. Analysis	DNA High-Sensitivity	
4. Marker	Process Profile	IMPORTANT: Select the Distribution Analysis for NGS
5. Peak Calling	Peak Calling Distribution Analysis	library QC or RNA degradation assessment or other analysis based on
6. Report/Export	Default DNA High-Sensitivity	smear. The list of available process profiles and the analysis features are adjusted accordingly to the selection
START A PROCESS	Profile	adjusted accordingly to the serveron.
1. Sample Selection 2. Sample Information	Included Steps	
3. Run Check	Experiment Directory	
	C.\ProgramData\QIAGEN\QIAxcef\ScreenGel 2\Data\Experiment\DNA Allow Directory Selection	
	Notes	
	Default process profile for DNA High-Sensitivity Cartridge, it applies; Method: DNA High-Sensitivity Alignment Marker: QX DNA High-Sensitivity Size Marker: QX DNA H S100 bp - 1 kb Analysis: DNA High-Sensitivity Analysis Report: DNA Fragment Analysis HS	
	Save process profile as Run	

Note: The **Process Profile** drop-down list does not simultaneously display profiles containing Peak Calling and Distribution Analysis options. If you miss a profile in the list, select either the **Peak Calling** or **Distribution Analysis** button from above the process profile drop-down list.

This setting is user specific. It may change automatically when you open an experiment for analysis in the **Analysis** environment that contained peak calling or distribution analysis options.

Note: Select NewProcessProfile to create a Process Profile from scratch. The system will show NewProcessProfile after selection. This is only possible for users with the **Developer or Administrator** role. However, it may be more convenient to select a predefined Process Profile close to your needs and modify it only slightly.

9. **Optional:** Modify the Process Profile options if required, as described in the <u>Process Profile options</u> section; restrictions for **Operator** user role are described here as well.

Note: Experiment directory, markers, and sample rows can be adjusted in the **Sample Selection** screen, if the Process Profile allows it. These adjustments override the options defined in the Process Profile, so modify these options in the Process Profile only if you want to save them for re-use.
ROFILE DEFINITION	()		Samula Daw Salastian
1 Process Profile	Experiment Name / Plate ID 3		Sample Row Selection 3
1. Trocess Trome		Provide Sample Information (8	1 2 3 4 5 6 7 8 9 10 11
2. Run Parameters	C:\ProgramData\OIAGEN\OIAycel\ScreenGe		
. Analysis	Reference Marker		B •••••••
Markor			C • • • • • • • • • • •
. Mai Kei	O No Marker		
Peak Calling	O Use selected marker table		
Depert/Evrent	Run size marker side by side with sample	e	E • • • • • • • • • • • •
Report/Export	Alignment Marker		F •••••••••
ART A PROCESS	QX DNA High-Sensitivity		G
. Sample Selection 2	> Size Marker		
Cample Information	QX DNA HS 100 bp - 1 kb 🛛 🔻		н
. sample information			Total Runs: 8
. Run Check	Show Lot Information (7)		Estimated Lime: About 91 minutes
	Alignment Marker	Size Marker	
	▼	▼	
	Wash Buffer	Dilution Buffer	
	▼	▼	
	Separation Buffer		
	▼	Last used	
			-
			 Loft click to coloct/docoloct complex

10. Optional: Switch to the Sample Selection screen (2).

11. Optional: Change the plate ID (3).

Note: The Experiment name/ Plate ID will be used to name all the results from this process, for the experiment's storing the sample data, as well as for generated report/export files. Therefore, a unique Experiment name/Plate ID is required. See <u>Run parameters and result structure</u> for further information.

Note: As input for the **Experiment name/Plate ID**, the software allows only alpha numeric characters. Special characters such as /&!% i'' @*+~#=<>|;, etc cannot be used. The software removes automatically blanks at the start and at the end.

Note: After the process, the Experiment name / Plate ID cannot be modified.

Note: If the settings option **Generate Experiment name/ Plate ID** is selected, the system automatically generates an Experiment Name / Plate ID following the rules specified in the settings. Refer to section <u>Settings</u> for more information. However, the automatically generated Experiment name / Plate ID may be modified before run is started.

12. Optional: Adjust the directory where the experiment will be saved (4).

Note: This can be a directory on the network. During the process, however, the experiment is always first saved to a local directory (refer to <u>Settings</u>), and then copied to the experiment directory defined here when the process is completed.

Note: Changing the experiment directory here is only possible if the Process Profile allows it (see <u>Selecting</u> <u>general process options</u>); however, you can switch to the **Process Profile** screen to modify this.

13. Optional: Change the selection of sample rows to be processed (5). Use this option if you want to process less rows than specified in the Run Parameters screen. Left-click the row to deselect/select.

Optional: Change the position of the size marker on the plate, by right-clicking the new marker position. Then, select the option **Toggle Analysis Marker** from the context menu. This size marker will be used for automatic analysis during processing, if Sample row H was deselected.



Sample row H was deselected.



Note: Changing sample rows or size marker position is only possible in this step if the Process Profile allows it (see <u>Selecting run parameters</u>); however, you can switch to the **Run Parameters** screen to modify this.

Note: It is not possible to deselect a row containing the size marker.

Optional: If you have an additional size marker in the sample plate, right-click the position and select the **Set As Additional Marker** option from the appearing context menu. A rotated ladder symbol appears. Right-click it again and select the **Size Marker Name** from the context menu. This information can be used for manual analysis, but will not be used for automated analysis during processing.

14. **Optional:** Select the size and alignment marker in the **Size Marker** and **Alignment Marker** drop-down lists as required (6). This information will be used for automated analysis during the process.

Note: If the **No Marker** option is selected, select the prepared alignment marker. If you do not select an alignment marker in the **Alignment Marker** drop-down list, you will have to acknowledge a warning later in the **Run Check** screen.

15. Optional: Provide lot information of buffers and markers by checking the Show Lot Information box (7).

Up to five last-used lot information are stored separately for each lot type. These lastly used values can be selected from drop-down list to be used again. Alternatively, the lot numbers can be typed into the edit field of the combo box. Restore the last used combination of the lot information by clicking on the **Last** used button.

Note: This option is only available if either the option **Enable** input fields for **Buffer lot IDs** or **Marker lot IDs** is selected in the settings (see <u>Settings</u>).

 Optional: Check the Provide Sample Information box if you want to provide sample information (8). If not, proceed to step 18.

Experiment Name / Plate ID	
experiment 1	Provide Sample Information

17. **Optional:** Provide sample information in the **Sample Information** screen, entered manually or imported from a file. For detailed information, refer to section <u>Provide Sample Information</u>, or click on one of the positions in the grid and press the **F1** key.

Note: The Provide Sample Information box must be checked in the Sample Selection screen to access this screen.

18. Switch to the **Run Check** screen (9). Confirm all check boxes. Resolve the problems indicated by warnings and errors.

Process Setup		
Confirm checks, fix warning	s and errors!	
PROFILE DEFINITION 1. Process Profile 2. Run Parameters 3. Analysis 4. Marker	Please Confirm All selected sample rows contain samples Alignment marker is loaded Size marker is loaded	Sample Row Selection 1 2 3 4 5 6 7 8 9 10 11 12 A B
 5. Peak Calling 6. Report/Export START A PROCESS 1. Sample Selection 2. Sample Information 3. Run Check S 	① Check all boxes so that processing can start. Errors and Warnings A The cartridge door is open. Image: Control of the process can not be started until the errors have been corrected and the warnings acknowledged.	E F G Estimated Time: About 91 minutes Method(s): DNA High-Sensitivity Size Marker: QX DNA HS 100 bp - 1 kb Alignment Marker: QX DNA High-Sensitivity Reference Marker: — Experiment: experiment 1
	Save process profile as Run	(1)

Note: The process can be started only when all run checks are confirmed and no errors are shown. Warnings must be acknowledged or removed by eliminating the cause of the warnings.

Note: Developer or Administrator users can acknowledge an uncalibrated cartridge and a red marked reference marker (**Operator** can acknowledge a yellow one).

Note: If the run button is still deactivated, check the <u>Status Information panel</u> for cartridge and instrument status.

19. Start the process by clicking the Run button (10).

The process wizard will be closed and a preview will be shown for each row currently processed. The actions performed by the instrument are shown in the **Instrument Status** panel on the left. A progress bar below the preview shows the progress of the whole process and the estimated remaining process time.

Depending on the report settings the generated report may be displayed.

Note: Opening the cartridge door or sample door during operation of the QIAxcel Connect will cause the system to stop. The process is stopped and cannot be recovered. No sample data is stored for the row currently processed. Sample data is only stored for completed rows. No analysis, peak calling or report/ export is performed. Thus, the stored sample data contains only raw data.

Actions during process run:

 You can switch between the Gel Image and Electropherogram preview by clicking on the respective tabs.



Optionally, adjust the separation time. To do so, move the Adjust Separation Time slider below the
preview. Moving it to the far left will shorten the separation time by 120 seconds, moving it to the far
right will extend the separation time by 120 seconds. Moving the slider takes effect for an ongoing
separation as well as for all following separations during the process.



Note: When starting the next process by clicking the **Run** button, the slider is automatically reset to the middle position and the separation time will be applied as defined in the method.

- Optionally, switch the check box Individual Scaling on/off to adapted colors to the overall maximum signal height of all samples. See <u>Gel view</u> for more information about individual scaling. The check box Individual Scaling is initially set to off.
- The process can be stopped by clicking the STOP button below the preview.

Note: If the instrument motors are currently moving, the process will stop after this movement.

Note: No sample data is stored for the currently processed row. Sample data is only stored for completed rows. No analysis, peak calling, or report/export instructions are performed. Thus, the stored sample data contains only raw data.

Actions after the process has finished:

- QIAGEN	Service Config	guration										
Status Information	Gel Image	Electropherogran	n									
				Experiment	experimer	nt 1						
Instrument Status		A1 A2	A3 A4	A5	A6 /	A7 A8	A9	A10	A11	A12		
▼ Cartridge Status												
Cartridge ID C210728D99												
DNA High-Sensitivity												
Remaining Runs Calibration Runs Left 83 3												
Expiration Status Calibration Status												
Calibration Details												
		_										
					_	_						
		Separation Time	Adjustment: 0									
		-120	120		In	ndividual Scalii	ng					
	·	Process finished.							Stop	Process	Back to	Wizard

- The next process can be started by clicking the **Back to Wizard** button below the preview.
- Switch to the **Analysis** environment to inspect the sample data. For more information, refer to sections <u>Environments</u>, <u>Handling samples and experiments</u> and <u>Viewing sample data</u>, respectively. Click the **Back to Wizard** button to return to the **Process** environment and to start the next process.

6.3.1.1 Provide sample information

To access the **Sample Information** screen, check the **Provide Sample Information** box in the **Sample Selection** screen.

Use the Sample Information screen to:

- Enter sample information, typically sample IDs
- Enter comments
- Import sample information and comments for all plate positions from a file
- Save sample information (without comments) for later re-use

The sample information grid becomes automatically active upon entering the **Sample Information** step. To switch to sample comments, click the **Sample Comments** tab. To switch back to the sample information grid, click the **Sample Information** tab.

	Sample In	formation		Sample Co	mments							
	1	2	3	4	5	6	7	8	9	10	11	12
A	100bp - 2.5 kb (20ng/µl)	sample info A2										
в												
с												
D												
E												
F												
G												
н												
							Im	port	Save as	Res	et Sample I	nformation

Note: The size marker positions are marked by a "ladder" symbol as shown in position A1.

Editing sample information

To provide sample information manually, enter the sample information into the grid. **Position A1** is preselected by default. Confirm the information for each position by pressing the **Enter** or the **Tab** key; the next position will be selected automatically for the next input. To provide sample information for a specific position, click the position and enter the sample information.

The sample information grid supports an auto-increment functionality. To number sample information texts consecutively:

- 1. Write the main sample information in the left uppermost cell.
- 2. Drag the small black square at the lower right corner of the cell border to extend the selection to all wanted cells.
- 3. Release the selection square. All selected cells will be filled with the main sample information entered in step 1. The cells will be numbered consecutively, starting with number 1.

To navigate through the sample information grid, use the arrow and **Tab/Shift+Tab** keys (just as in a Microsoft Excel[®] table).

To select multiple cells:

- Starting with one or more selected cells, hold **Ctrl** and left-click cells to add or remove them from the selection.
- Left-click in a cell and, starting in the cell with the cursor, drag to select a rectangular area of cells.

To copy the text of one selected cell into multiple cells:

- 1. Select the (source) cell.
- 2. Use the context menu item Copy or press the Ctrl-C key combination to copy the text into the clipboard.
- 3. Select the target cells.
- 4. Right-click one of the selected cells, but not the one with the cursor, and use the context menu item Paste. The text will be pasted from the clipboard into all selected cells. In addition, the first selected cell will remain highlighted after the text has been pasted.

To copy a column:

- 1. Select all cells of the column you want to copy.
- 2. Use the context menu item Copy to copy the text of all selected cells into clipboard.

Note: The Ctrl-C key combination captures only the text of the last selected cell.

- 3. Left-click on the first cell of the column from which you want the text to be pasted.
- 4. Use the context menu item **Paste** or press the **Ctrl-V** key combination. The cells from the clipboard will be pasted to the column starting with the cell selected in the last step.

Note: In the same way, it is possible to copy rows or other selected areas. To start pasting, click the uppermost cell to the left.

To copy multiple cells from a Microsoft Excel table:

- 1. Copy the cells from the **Microsoft Excel** table into the clipboard.
- 2. In the Sample Information screen, click on the most left cell to paste.
- 3. Use the context menu item **Paste** or press the **Ctrl-V** key combination. The cells from the clipboard will be pasted to the cells starting with the cell selected in the last step.

2 sample nformation for sample sample sample sample sample sample nformation nformation nformation nformation sample	3	4	5	6	7	8	9	10	11	12
sample nformation for sample sample nformation n for sample sample sample sample nformation n for sample nformation n for sample										
sample nformation for sample sample nformation for sample nformation for sample sample										
sample nformatio n for sample sample nformatio n for sample										
sample nformatio n for sample										
amplo										
nformatio n for sample										
sample nformatio n for sample										
sample nformatio n for sample										
sample nformatio n for sample										
nf sa sa nf sa nf sa	ormatio or mple ormatio or mple ormatio or mple ormatio or mple	ormatio for mple mple ormatio for mple ormatio for mple ormatio	ormatio for mple mple formatio for mple mple formatio for mple	ormatio for mple mple ormatio mple mple ormatio or mple	ormatio for mple mple formatio mple mple ormatio for mple	iormatio ior mple mple ormatio ior mple mple ormatio ior mple mple	iormatio ior mple mple ormatio ior mple mple ormatio ormatio ior mple	ormatio for mple mple ormatio mple mple ormatio or mple	iormatio ior mple ormatio ior mple ormatio or mple ormatio or mple	iormatio ior mple ormatio ior mple mple ormatio ior mple

Note: If the length of the pasted text exceeds the maximum value (30), the cell background will change as shown in sample column B.

To clear sample information, click the Reset Sample Information button.

Note: Users who are allowed to revise sample information (see <u>User management</u>) may edit the sample information after the run using the context menu of the **Experiment Explorer**. In the analysis environment, select the sample in the **Experiment Explorer**, right-click the sample, and select the context menu option **Revise Sample Information**, then enter the new sample information.

Editing sample comments

To enter sample comments, click **Sample Comments**. To enter a comment for a specific position, click the relevant cell in the **Sample Comments** tab and enter the text.

To sort the plate positions by row or by column, click the column headers Row and Col, respectively.

Sample	e Inforn	nation									
Row	Col			Sample Comment							
А	1	sample 1 fr	om experiment 101								
А	2	replicate fro	om sample A1								
А	3	sample 2 fr	om experiment 101								
А	4	replicate fro	plicate from sample A3								
Α	5	I									
А	6										
Α	7										
А	8										
А	9										
А	10										
А	11										
А	12										
В	1										
В	2										
В	3										
В	4										
В	5										
В	6										
В	7										
		1		T.	*						
				Import Save as Reset Sample Comme	ent						

To copy a comment from one cell into another cell:

- 1. Click the (source) cell.
- 2. Select the text to be copied using the mouse or by holding down the **Shift** key while using the end, leftarrow or right-arrow key.
- 3. Select the context menu item **Copy** or press the **Ctrl-C** key combination to copy the text into the clipboard.
- 4. Click the target cell.
- 5. Select the context menu item **Paste** or press the **Ctrl-V** key combination. The text will be pasted from the clipboard into the cell.

To clear all sample comments, click the **Reset Sample Comment** button.

Sample comments can also be imported from a file together with the sample information.

Note: Sample comments are restricted to 150 characters.

Importing sample information

The QIAxcel ScreenGel software provides the possibility to import sample information, with or without sample comments, from files. Three file types are supported: Microsoft Excel files (Excel 97–2003 Workbook (*.xls), Excel Workbook 2007–2010 (*.xlsx)), QIAGEN rack files, and QIAGEN qdef files, the QIAGEN data exchange format used in both QIAsymphony[®] and QIAgility[®] applications.

Note: The QIAGEN rack file format does not support sample comments.

To import sample information and/or sample comments from a file, click the **Import** button. In the lower part of the dialog that appears, select the type of file to be imported. The dialog lists all available files of this type. Alternatively, navigate to the directory where the sample information was stored, select the file and click **OK**. The imported data overwrites all sample information and comments previously provided.

Note: Examples of all file types can be found under the File menu by selecting Open Data Directory and then Sample Information.

Note: Imported sample information is read-only if the settings option **Prevent modification of imported sample** information is selected (see <u>Settings</u>).

For the import of sample information from Microsoft Excel files, the following options are available:

Row-wise	Select this option if the sample list in the Microsoft Excel file follows this order: A1, A2,, A12, B1, B2,, B12,, H1,, H12. All samples names must be listed in the first column, starting in the first line of the table. Sample names for all plate positions must be listed in the file. Optional sample comments should be listed in the second column.
Column-wise	Select this option if the sample list in the Microsoft Excel file follows this order: A1, B1,, H1, A2, B2,, H2,, A12, H12. All sample names must be listed in the first column, starting in the first line of the table. Sample names for all plate positions must be listed in the file. Optional sample comments should be listed in the second column.
Matrix	Select this option if the sample information in the Microsoft Excel file is written in matrix format spanning cells A1 to L8 in the table. That is: A1, A2,, A8 in the first row, and A1, B1,, H1 in the first column.
	This option does not support sample comments.

Note: The QIAxcel ScreenGel imports sample information from the first worksheet in an alphabetical listing of all worksheets of the Microsoft Excel file. That is, if you rename the worksheets of the Microsoft Excel file, ensure that the relevant sample information is entered in the worksheet that would appear first, when arranged alphabetically by worksheet name.

Saving sample information

To save the provided sample information in rack file format for later reuse, click the **Save as** button. In the resulting dialog, navigate to the directory where the sample information should be stored, enter a unique file name, and click **OK**.

Note: The Save as button will be disabled if one or more cells contain more than 30 characters.

Note: The QIAGEN rack file format does not support sample comments.

6.3.1.2 Sample information

The sample information grid becomes automatically active upon entering the **Sample Information** step. To switch to sample comments, click the **Sample Comments** tab. To switch back to the sample information grid, click the **Sample Information** tab.

	Sample In	formation		Sample Co	mments							
	1	2	3	4	5	6	7	8	9	10	11	12
A	100bp - 2.5 kb (20ng/µl)	sample info A2										
в												
c												
D												
E												
F												
G												
н												
							Im	port	Save as	Res	et Sample I	nformation

Note: The size marker positions are marked by a "ladder" symbol as shown in position A1.

Select one of the following topics:

- Editing sample information
- <u>Copy/paste sample information</u>
- Importing sample information from file
- Saving sample information for re-use
- Editing sample comments

6.3.1.2.1 Edit sample information

Editing sample information

To provide sample information manually, enter the sample information into the grid. **Position A1** is preselected by default. Confirm the information for each position by pressing the **Enter** or the **Tab** key; the next position will be selected automatically for the next input. To provide sample information for a specific position, click the position and enter the sample information.

The sample information grid supports an auto-increment functionality. To number sample information texts consecutively:

- 1. Write the main sample information in the left uppermost cell.
- 2. Drag the small black square at the lower right corner of the cell border to extend the selection to all wanted cells.
- 3. Release the selection square. All selected cells will be filled with the main sample information entered in step 1. The cells will be numbered consecutively, starting with number 1.

To navigate through the sample information grid, use the arrow and **Tab/Shift+Tab** keys (just as in a Microsoft Excel[®] table).

To select multiple cells:

- Starting with one or more selected cells, hold **Ctrl** and left-click cells to add or remove them from the selection.
- Left-click in a cell and, starting in the cell with the cursor, drag to select a rectangular area of cells.

To copy the text of one selected cell into multiple cells:

- 1. Select the (source) cell.
- 2. Use the context menu item **Copy** or press the **Ctrl-C** key combination to copy the text into the clipboard.
- 3. Select the target cells.
- 4. Right-click one of the selected cells, but not the one with the cursor, and use the context menu item Paste. The text will be pasted from the clipboard into all selected cells. In addition, the first selected cell will remain highlighted after the text has been pasted.

To copy a column:

- 1. Select all cells of the column you want to copy.
- 2. Use the context menu item Copy to copy the text of all selected cells into clipboard.

Note: The Ctrl-C key combination captures only the text of the last selected cell.

- 3. Left-click on the first cell of the column from which you want the text to be pasted.
- 4. Use the context menu item **Paste** or press the **Ctrl-V** key combination. The cells from the clipboard will be pasted to the column starting with the cell selected in the last step.

Note: In the same way, it is possible to copy rows or other selected areas. To start pasting, click the uppermost cell to the left.

To copy multiple cells from a Microsoft Excel table:

- 1. Copy the cells from the Microsoft Excel table into the clipboard.
- 2. In the Sample Information screen, click on the most left cell to paste.
- 3. Use the context menu item **Paste** or press the **Ctrl-V** key combination. The cells from the clipboard will be pasted to the cells starting with the cell selected in the last step.

Sample	Sample Information		Sample Comments								
1	2	3	4	5	6	7	8	9	10	11	12
sample A informa n for A1	tio n for sample										
B n for A2	tio informatio n for sample										
c informa c n for A3	tio informatio n for sample										
D informa D n for A4	tio informatio n for sample										
E informa	tio informatio n for sample										
F informa	tio informatio n for sample										
sample G informa n for A7	tio informatio n for sample										
sample informa n for A8	sample tio informatio n for sample										
L						Im	port	Save as	Rese	et Sample I	oformation
										- Sample ii	

Note: If the length of the pasted text exceeds the maximum value (30), the cell background will change as shown in sample column B.

To clear sample information, click the Reset Sample Information button.

Note: Users who are allowed to revise sample information (see <u>User management</u>) may edit the sample information after the run using the context menu of the **Experiment Explorer**. In the analysis environment, select the sample in the **Experiment Explorer**, right-click the sample, and select the context menu option **Revise Sample Information**, then enter the new sample information.

6.3.1.2.2 Edit sample comments

Editing sample comments

To enter sample comments, click **Sample Comments**. To enter a comment for a specific position, click the relevant cell in the **Sample Comments** tab and enter the text.

To sort the plate positions by row or by column, click the column headers **Row** and **Col**, respectively.

Sample	Inform	nation	Sample C	omments								
Row	Col				Sample	Comment						
А	1	sample 1 fr	om experiment 1	01								
А	2	replicate fro	om sample A1						Ι			
А	3	sample 2 fr	Imple 2 from experiment 101									
А	4	replicate fro	plicate from sample A3									
Α	5	I										
А	6											
А	7											
А	8											
А	9											
А	10											
А	11											
А	12											
В	1											
В	2											
В	3											
В	4											
В	5											
В	6											
В	7											
									1			
						Import	Savo ac	Docot Sample Com	mor			

To copy a comment from one cell into another cell:

- 1. Click the (source) cell.
- 2. Select the text to be copied using the mouse or by holding down the **Shift** key while using the end, leftarrow or right-arrow key.
- 3. Select the context menu item **Copy** or press the **Ctrl-C** key combination to copy the text into the clipboard.
- 4. Click the target cell.
- 5. Select the context menu item **Paste** or press the **Ctrl-V** key combination. The text will be pasted from the clipboard into the cell.

To clear all sample comments, click the Reset Sample Comment button.

Sample comments can also be imported from a file together with the sample information.

Note: Sample comments are restricted to 150 characters.

6.3.1.2.3 Import sample information

Importing sample information

The QIAxcel ScreenGel software provides the possibility to import sample information, with or without sample comments, from files. Three file types are supported: Microsoft Excel files (Excel 97–2003 Workbook (*.xls), Excel Workbook 2007–2010 (*.xlsx)), QIAGEN rack files, and QIAGEN qdef files, the QIAGEN data exchange format used in both QIAsymphony[®] and QIAgility[®] applications.

Note: The QIAGEN rack file format does not support sample comments.

To import sample information and/or sample comments from a file, click the **Import** button. In the lower part of the dialog that appears, select the type of file to be imported. The dialog lists all available files of this type. Alternatively, navigate to the directory where the sample information was stored, select the file and click **OK**. The imported data overwrites all sample information and comments previously provided.

Note: Examples of all file types can be found under the File menu by selecting Open Data Directory and then Sample Information.

Note: Imported sample information is read-only if the settings option **Prevent modification of imported sample** information is selected (see <u>Settings</u>).

For the import of sample information from Microsoft Excel files, the following options are available:

Row-wise	Select this option if the sample list in the Microsoft Excel file follows this order: A1, A2,, A12, B1, B2,, B12,, H1,, H12. All samples names must be listed in the first column, starting in the first line of the table. Sample names for all plate positions must be listed in the file. Optional sample comments should be listed in the second column.
Column-wise	Select this option if the sample list in the Microsoft Excel file follows this order: A1, B1,, H1, A2, B2,, H2,, A12, H12. All sample names must be listed in the first column, starting in the first line of the table. Sample names for all plate positions must be listed in the file. Optional sample comments should be listed in the second column.
Matrix	Select this option if the sample information in the Microsoft Excel file is written in matrix format spanning cells A1 to L8 in the table. That is: A1, A2,, A8 in the first row, and A1, B1, ,H1 in the first column.
	This option does not support sample comments

This option does not support sample comments.

Note: The QIAxcel ScreenGel imports sample information from the first worksheet in an alphabetical listing of all worksheets of the Microsoft Excel file. That is, if you rename the worksheets of the Microsoft Excel file, ensure that the relevant sample information is entered in the worksheet that would appear first, when arranged alphabetically by worksheet name.

6.3.1.2.4 Save sample information

Saving sample information

To save the provided sample information in rack file format for later reuse, click the **Save as** button. In the resulting dialog, navigate to the directory where the sample information should be stored, enter a unique file name, and click **OK**.

Note: The Save as button will be disabled if one or more cells contain more than 30 characters.

Note: The QIAGEN rack file format does not support sample comments.

6.3.2 Modifying a process profile

Note: Only users with the assigned role **Administrator**, **Developer** and **Operator** can modify the Process Profiles. Restrictions apply to users with the role **Operator** as they, for example, cannot save the modified Process Profile. Restrictions are annotated in this manual, where they apply.

To modify a Process Profile, proceed as follows:

1. Launch the Process environment.

If not already open, switch to the **Process** environment by clicking the **Process** environment icon. Select the **Process Profile** (1) screen.

Process Setup			
Set the process profile	e!		
PROFILE DEFINITION		Mode	
1. Process Profile 1	>	● DNA ○ RNA	
2. Run Parameters		Cartridge Type	
3. Analysis		DNA High-Sensitivity	
4. Marker	3	Process Profile	
5. Peak Calling		Peak Calling Distribution Analysis	
6. Report/Export		Default DNA High-Sensitivity 🔹 2	
START A PROCESS	•	Profile	
1. Sample Selection		Included Steps	
2. Sample Information		Run Analysis Peak Calling Report	
3. Run Check		Experiment Directory	
		C:\ProgramData\QIAGEN\QIAxcel\ScreenGel 2\Data\Experiment\DNA	
		Allow Directory Selection	
		Notes	
		Default process profile for DNA High-Sensitivity Cartridge, it applies;	
		Method: DNA High-Sensitivity Alignment Marker: QX DNA High-Sensitivity	
		Size Marker: QX DNA HS 100 bp - 1 kb	
		Report: DNA Fragment Analysis HS	
		Save process profile as 4	Run

Note: If the last process only just finished, click the Back to Wizard button on the right bottom.

2. Select the Process Profile to be modified (2).

Use the Process Profile drop-down list to select a profile.

Note: Each Process Profile is related to a certain cartridge type. Therefore, the system can ensure that a process (i.e., data acquisition) can be started only if the correct cartridge type is inserted. If the Process Profile you want to select is not listed, make sure that the correct cartridge type is selected. If the instrument is connected, the system automatically detects the inserted cartridge type. If you want to modify a Process Profile to use with another cartridge type, remove the cartridge, or at least the cartridge key. Select the correct cartridge type. The Process Profile you want to modify should now be listed in the **Process Profile** drop-down list. Select the Process Profile and proceed.

3. Change the Process Profile options as required (3).

PROFILE DEFINITION					
1. Process Profile	>				
2. Run Parameters					
3. Analysis					
4. Marker					
5. Peak Calling					
6. Report/Export					

See the Process Profile options section for detailed information.

4. Save the modified Process Profile under a new name (4).

Click the **Save Process Profile as** button below the process setup and enter a new unique profile name in the dialog that appears.

Note: This is only possible for users only with the user role Developer.

Note: If there are Process Profile screens containing incomplete or inconsistent data highlighted in yellow, the **Save Process Profile as** button is disabled. Select the marked Process Profile screens and correct the data. If all data is correct, the **Save Process Profile as** button becomes enabled and the Process Profile can be saved.

Note: You can start a process or leave the **Process** environment without saving your changes. QIAxcel ScreenGel will not remind you to save a Process Profile or the embedded analysis profile, report/export profile, or peak calling instruction. If you leave the **Process** environment, log out, or exit the application, you should save your changes if they are to be re-used.

6.3.3 Process profile options

The Process Profile options are grouped into several screens. To specify the complete Process Profile, follow the screen order as described below. To switch to the **Run Parameters** screen, click the screen name:

Process Setup		
Set the process profi	le!	
PROFILE DEFINITION	_	Mode
1. Process Profile	<u> </u>	● DNA ○ RNA
2. Run Parameters	⚠	Cartridge Type
3. Analysis		Drechightes.
4. Marker		Process Profile
5. Peak Calling		Peak Calling Distribution Analysis
6. Report/Export		*NewProcessProfile
START A PROCESS		Profile
1. Sample Selection		lashudad Stars
2. Sample Information		Run Analysis Peak Calling Report
3. Run Check		Experiment Directory
		C:\ProgramData\QIAGEN\QIAxcel\ScreenGel 2.0\Data\Experiment\DNA
		Allow Directory Selection
		Notes
		Save process profile as Run

Note: You can return to each screen by clicking the screen name. You can skip screens if modifications are not necessary.

Note: Screens with incomplete or incorrect options are marked with a yellow triangle besides the subsection title. There is no need to correct them straight away, but you cannot save or start an incomplete or incorrect Process Profile.

The Process Profile screen options are described below.

Process Profile	Specifies the process scope. In addition to data acquisition, it is possible to include additional steps such as analysis and report. For detailed information, refer to the <u>Selecting general process options</u> section.			
Run Parameters	Specifies the data acquisition details such as method used, rows to be processed, and size marker position. For detailed information, refer to the <u>Selecting run parameters</u> section.			
Analysis Parameters	pecifies the analysis parameters for fully automated analysis after data acquisition. For detailed information, refer to the <u>Selecting analysis</u> parameters section.			
	Note: This screen is disabled if analysis is not in the process scope.			
Marker	Specifies the markers, alignment marker, and size marker used for analysis. For detailed information, refer to the <u>Selecting marker</u> section.			
	Note: This screen is disabled if analysis is not within the process scope.			
Peak Calling	Specifies the parameters for peak calling based on the peak result table. For detailed information, refer to the <u>Selecting peak calling instructions</u> section.			
	Note: This screen is disabled if peak calling is not within the process scope.			
Distribution Analysis	Specifies the parameters for distribution analysis based on the smear result table. For detailed information, refer to the <u>Selecting distribution profiles</u> section.			
	Note: This screen is disabled if distribution analysis is not within the process scope.			
Report/Export	Specifies the report and/or export parameters for fully automatic report and export as last step during process. For detailed information, refer to the <u>Selecting report/export parameters</u> section.			
	Note: This screen is disabled if report/export is not in the process scope.			

Note: The peak calling screen or the distribution analysis screen are available only if the corresponding features (peak calling or the distribution analysis) are active. To activate, select the corresponding option right above the process profile drop down list.

Process Profile	
Peak Calling Distribution An	alysis
*NewProcessProfile	•

Note that only one set of features can be active at a given time.

6.3.3.1 Selecting general process options

1. Select the Process Profile screen (1).

Process Setup	
Set the process profile!	
PROFILE DEFINITION	Mode
1. Process Profile (1)	
2. Run Parameters	Cartridge Type
3. Analysis	DNA High-Sensitivity
4. Marker	Process Profile
5. Peak Calling	Peak Calling O Distribution Analysis
6. Report/Export	Default DNA High-Sensitivity
START A PROCESS	Profile
1. Sample Selection	Included Steps (2)
2. Sample Information	Run Analysis Peak Calling Report
3. Run Check	Experiment Directory
	C:\ProgramData\QIAGEN\QIAxcel\ScreenGel 2\Data\Experiment\DNA
	Allow Directory Selection 4
	Notes (5)
	Default process profile for DNA High-Sensitivity Cartridge, it applies;
	Method: DNA High-Sensitivity Alignment Marker: QX DNA High-Sensitivity
	Size Marker: QX DNA HS 100 bp - 1 kb
	Report: DNA Fragment Analysis HS
	Save process profile as Run

Refer to the Process Profile options screen for information on how to select the screen.

2. Specify the process scope (2).

Select the additional steps:

Analysis

Includes analysis in the process. This means after data acquisition is completed, the software analyzes the sample data automatically in the background, based on the process options set in the **Analysis** and **Marker** screens (described in the <u>Selecting analysis parameters</u> and <u>Selecting marker</u> sections).

	The Analysis Parameters and Marker screens become enabled.			
Peak Calling	Includes peak calling in the process, based on the analysis peak result table. This means that after analysis, peak calling will be automatically performed, based on the process options set in the Peak Calling screen (described in the <u>Selecting peak calling instructions</u> section).			
	Note: This step is disabled if Analysis is not within the process scope.			
	Note: This option is only available if peak calling features are active. To activate peak calling features, select Peak Calling from the Process Profile section above.			
Distribution Analysis	Includes distribution analysis in the process, based on the analysis smear result table. This means that after analysis, distribution analysis will be automatically performed based on the process options set in the Distribution Analysis screen (described in the <u>Selecting distribution profiles</u> section).			
	Note: This step is disabled if Analysis is not within the process scope. Additionally, the option Smear Analysis Profile option must be selected in the analysis profile selected.			
	Note: In DNA mode, This option is only available if distribution analysis features are active. To activate distribution analysis features, select Distribution Analysis from Process Profile section above.			
Report	Includes report and/or export as the last step in the process. This means that the software automatically generates a report and exports the data.			

3. Optional: Change the directory where the experiment will be saved (3).

Note: This can be a directory on the network. However, during the process, the experiment will always be saved to a local directory first (refer to <u>Settings</u>), and then copied to the experiment directory defined here, when the process finished.

4. Optional: Allow directory selection (4).

Select the option to allow directory selection in the **Sample Selection** screen while preparing a process. Using this option, also the user role **Operator** will be able to choose the directory where the run is saved.

5. Optional: Add or Edit a note in the Process Profile (5).

Use this field to add a short description or information about the profile. This note is displayed each time the Process Profile is selected. The note field is limited to maximal 2.000 characters and 25 lines.

6.3.3.2 Selecting run parameters

Define the methods and the rows that have to be processed under **Run Paramters**. Select at least one method. In addition, the position of the size marker can also be specified.

Note: During processing, the list will be processed consecutively. For information about the correlation between run parameters and result structure, refer to the <u>Run parameters and result structure</u> section.

Proceed as follows to define the Run Paramter (1):

1a. If the **Method Details** list is empty, click the **Add** button to start the first definition. A new empty entry appears in the **Method Details** list. Proceed with step 2 to specify the entry.

Note: This option is only available for users with the user role Developer and Administrator.

ROFILE DEFINITION	Plate Definition	Method Details		
1. Process Profile	Method Details			Sample Row Selection (2)
2. Run Parameters 🛈 🛕 >	Method Ran	ge M Inj. Time	RpR Runs Preview	1 2 3 4 5 6 7 8 9 10 11
3. Analysis		0	1 0	A
Marker				в ••••••••
				c
Distribution Analysis				D
Report/Export				E
RT A PROCESS				F
Sample Selection				G
Sample Information				
Run Check		on Allow marker	Insert Add	Delete
	Method		Range	Marker
	Injustion Time	V (3)	Duna nar Daw	
	0 sec			Left-click to add or remove sample rows
				Right-click to define

Add	Click the Add button to add a new entry at the bottom of the list. Proceed with step 2 to specify the entry.
	Note: This option is only available for users with the user role Developer and Administrator .
Insert	Select an entry by left-clicking and then selecting the Insert button to insert a new entry before the selected one. Proceed to step 2 to specify the entry.
	Note: This option is only available for users with the user role Developer and Administrator .
Modify	Select the entry you want to modify. Proceed to step 2 to modify the entry.
Delete	Select the entry you want to delete, then click the Delete button. The entry will be deleted from the list. Proceed to step 6.
	Note: This option is only available for users with user role Developer.

1b. If the Method Details list is not empty you have the following options:

2. Select the rows to be processed (2).

In the **Sample Row Selection** view to the right, left-click this view to add or remove a sample row to the plate. The **Range** field shows the row selection you have made.

Note: If you later refine a Process Profile that uses Distribution analysis, note the following:

- If you reduce the rows to process in the sample row selection in this screen, these rows will be removed automatically also in the **Distribution** analysis screen to ensure a consistent Process Profile.
- If you add rows later, there is no automatic adaptation in the **Distribution** Analysis screen.

Optional: Define the number of runs per row in the editable field **Runs** per row. The method will be applied as often as you specify here.

3. Optional: Define the size marker position.

Note: This position will be used for automatic analysis if the size marker is to be run side by side with the samples.

Right-click to define the position that will contain the size marker. The **Marker** field shows the position of the marker you set.

Note: Only one size marker position can be defined for an entry in the **Method Details** list. If you want to specify the Process Profile for a plate run containing a size marker at each row, create an entry for each row separately in the **Method Details** list.

4. Select the appropriate method from the Method drop-down list (3).

Note: This option is only available for users with the user role Developer and Administrator.

Note: The **Method** drop-down list only contains methods that are allowed for the cartridge type that the Process Profile is defined for.

Note: A number of default methods are available for each QIAxcel Kit. For a complete list, please refer to <u>Appendix B</u>. Refer to the <u>Viewing method details</u> section if you want to look at the details of the method you have selected.

If required, change the sample injection time in the **Injection Time** editable field. This time will be applied instead of the sample injection time defined in the method.

ROFILE DEFINITION		Plate Definition		Met	hod Details				
1. Process Profile		Method Details							Sample Row Selection
2. Run Parameters	>	Method	Range	М	Inj. Time	RpR	Runs	Preview	1 2 3 4 5 6 7 8 9 10 1
3. Analysis		0M1200	A - C,E,	A1	10	1	6		A = • • • • • • • • • •
4. Marker		0M800		D12	10	1	1		B • • • • • • • • • •
5. Distribution Analysis		0M500	F	F2	5	3	3		D 0 0 0 0 0 0 0 0 0 0 0 0 0 0 0 0 0 0 0
6. Report/Export									E
TART A PROCESS									F ••••••
1. Sample Selection	<u>^</u>								G • • • • • • • • • • •
2. Sample Information									н •••••••••
3. Run Check		Allow row desele	ection	AI	low marker d	efinition	Insert	Add Delete	
		Method 0M1200		V			Range A - C,E,	G,H A1	
		Injection Time 15 sec					Runs pe	r Row	
		* This method costs	1 regular	r run(s)	decrement in	the car	tridge key		 Left-click to add or remove sample rows
		This method costs	-						

5. Click **OK** to confirm.

Note: The OK button becomes enabled as soon as the rows are defined and a method is selected.

The specified parameters appear in the entry created entry in the Method Details list.

Note: Click **Cancel** to discard all specifications made during steps 2 to 5. The entry will not be created/the selected entry will not be modified.

Note: The column **Runs** in the **Method Details** list contains the total number of runs specified by this entry: if **Range** is A-C and **RpR** ("Runs per Row") is 2, **Runs** will therefore be 3*2=6.

6. Define all entries in the Method Details list repeating steps 1 to 5.

7. Optional: Allow row deselection.

Select the option to allow row deselection in the **Sample Selection** screen while preparing a process. This option is useful to, for example, prepare the Process Profile for a complete plate but only run the rows needed.

Note: This option is only available for users with the user role Developer and Administrator.

8. Optional: Allow marker definition.

Select the option, to allow the size marker position to be redefined in the **Sample Selection** screen while preparing a process.

Note: This option is only available for users with the user role Developer and Administrator.

6.3.3.3 Selecting analysis parameters

Note: The **Analysis** screen is only enabled if analysis is selected in the Process Profile. For details, refer to the <u>Selecting general process options</u> section.

Specify the analysis parameters as follows:

1. Select an analysis profile.

Use the **Profile** drop-down list to select the analysis profile. The analysis parameters of the selected profile are shown below the drop-down list.

Process Setup					
Set analysis profile a	nd prop	erties!			
PROFILE DEFINITION		Profile			
1. Process Profile		*NewAnalysis	Profile	Save as	
2. Run Parameters		Analysis Pro	operties		
3. Analysis	>	Peak	⊖ Smear ⊃ g	JDNA	
4. Marker	Δ	Smoothing	Start	Window	
5 Distribution Analysis		Filter	0,00 min		15 pts
o. Distribution Printy sis		Baseline	Start	Window	
6. Report/Export		Filter	0,00 min		40 sec
START A PROCESS		Minimum	Start	Value	
1. Sample Selection	Δ	Distance	0,00 min		0,25 sec
2. Sample Information		Suspend Integration	Start	End	
3. Run Check		Threshold	Start	Value	
			0,00 min		2,50 S/N
		Alignment Marker	10 S/N		•
				Add	Delete
		Define Param Select Para	neter 🛛 🔻		
				01	Cancel

Note: The analysis profile will be used for fully automatic analysis of all samples.

Note: Select **NewAnalysisProfile** to define the <u>analysis parameters</u> from scratch. This is only possible for users with the user role **Developer** and **Administrator**.

Note: All analysis parameters currently set in the selected analysis profile will be copied to the Process Profile. Further modifications of the underlying analysis profile do not affect the analysis parameters in this Process Profile. This prevents the Process Profile from unwanted changes and ensures process stability. If modifications in the underlying analysis profile are to be included in this Process Profile as well, include the modified analysis profile again by selecting it from the **Profile** drop-down list.

2. **Optional:** Modify the analysis parameters. You can modify the analysis parameters according to your needs in this screen as described in the <u>Modifying an analysis profile</u> section.

Note: This is only possible for users with the user role Developer or Administrator.

Note: If you define the **Suspend Integration** intervals using relative migration time, make sure that the relative migration times match the marker mode. See <u>Modifying an analysis profile</u> for details.

Note: The modifications will only be included in this Process Profile. If the modified parameters are also to be included in the selected analysis profile, save the analysis profile as described below.

3. Optional: Save the modified analysis parameters.

Note: This is only possible for users with the user role Developer or Administrator.

The modified analysis parameters can be saved by clicking the **Save as** button. A dialog appears, which includes the name of the selected analysis profile. You have two options:

Click **OK** if you want the modifications to be saved in the selected analysis profile; enter a new unique analysis profile name to save the modified analysis parameters as a new analysis profile, then click **OK**. The name of this new analysis profile and all of its analysis parameters will be included in the Process Profile.

6.3.3.4 Selecting marker

Note: The **Marker** screen is only enabled if analysis is within the scope of the Process Profile. For details, refer to the <u>Selecting general process options</u> section.

Process Setup		
Set the size/reference and a	lignment markers!	
PROFILE DEFINITION	Marker Selection	Size Marker
1. Process Profile 2. Run Parameters	O No Marker	Total conc.
3. Analysis	Run size marker side by side with sample	Size [bp] Conc.[ng/µl]
4. Marker 1	Alignment Marker	<u> </u>
	QX 15 bp-5 kb ▼ (3)	72 0,27
5. Distribution Analysis		118 0,44
6. Report/Export	Size Marker	234 0.87
	phiX174 HaeIII (20 ng per ul)	271 1.01
START & PROCESS	Save as	281 1,04
1. Sample Selection	Ouve as	310 1,15
		603 2,24
2. Sample Information		872 3,24
3. Run Check		1078 4
		1353 5,02
		Add Delete
		Size Concentration
	Save process profile as Run	

Define the markers to be used when running this Process Profile in the **Marker** step (1).

- 1. Select one of the size marker options (2)
- 2. Choose Alignment Marker (3) and Size Marker (4)

For information about marker options, see the descriptions below:

No Marker	Select this option if the automatic analysis is only for peak detection.		
	Note: No size marker will be used during analysis, therefore the determination of size and concentration of the samples are not possible.		
Reference Marker Table			
	If determination of size and/or concentration is to be performed, select this option. The determination of size and/or concentration is based on a previously saved reference marker table.		
	Select the reference marker table to be used from the Reference Marker Table drop- down list.		

Note: To ensure compatibility, the drop-down list contains only reference marker tables whose size markers were processed with the same method as defined in this Process Profile. The drop-down list is marked as invalid and empty if there is no compatible reference marker table. In this case, run a process with size marker side by side with the samples or create a reference marker table from a previous process (refer to the <u>Creating a reference marker</u> section).

Note: If no cartridge is currently inserted, the system cannot check the compatibility of the selected reference marker table. The drop-down list is marked as invalid and empty, or it contains a previously selected reference marker table only. Insert the cartridge to be processed by the profile and select a reference marker table compatible for this cartridge and method defined in this Process Profile.

Note: See the table below for explanation of the symbol at the right of the **Reference Marker Table** drop-down box:

•	The selected reference marker table is fully compatible.
	This means that the size marker used for the reference marker table was processed no more than 90 days (DNA)/60 days (RNA) ago with the cartridge that is currently inserted and with the same method as defined in this Process Profile.
•	The size marker used for the reference marker table was processed more than 90 days (DNA)/60 days (RNA) ago with the cartridge that is currently inserted and with the same method as defined in this Process Profile.
•	The size marker used for the reference marker table was processed with the same method as defined in this Process Profile, but with a cartridge other than that currently inserted.
•	The reference marker table is not compatible with the method defined in this Process Profile. This can happen if the method was changed in the Run Parameters screen and the reference marker table was not yet changed correspondingly. Select another reference marker table from the drop-down list that is compatible with the newly selected method.

Run size marker Select this option if the plate requires a size marker. side by side Select the size marker to be used from the Size Marker drop-down list. with sample Optional: If needed, change the total concentration of the size marker above the Size Marker table on the right. The individual concentrations of the fragments are then recalculated automatically. Note: This is only possible for user role **Developer** or **Administrator**. In the Alignment Marker drop-down list, select the alignment marker to be used in the MARKER1 (M1) position in the buffer tray. Note: If the Run size marker is selected side by side with sample option, but size marker and/or alignment marker is not specified, the Marker screen will be considered as invalid. Note: Using the correct size marker will increase the accuracy of size and concentration determination. Select the marker containing nucleic acid fragments close to the size of your targeted fragments. The nucleic acid fragments to be analyzed must fall within the smallest and largest fragment size of the size marker. Note: In addition, the range of the alignment marker must cover the range of the size marker. If not, the Size Marker table on the right shows this discrepancy with rows highlighted in yellow and the Marker screen will be considered as invalid. Note: Refer to the Selecting run parameters section for information on how to define the position of the size marker.

6.3.3.5 Selecting peak calling instructions

Note: The **Peak Calling** screen is only enabled if **Peak Calling** and **Analysis** are selected in the Process Profile and Profile Steps, respectively. For details, refer to the <u>Selecting general process options</u> section.

Refer to the Peak Calling section for an explanation of the peak calling concept.

Define the instruction for peak calling:

1. Select a peak calling instruction.

Use the **Peak Calling Instruction** drop-down list to select a predefined peak calling instruction. The instruction definition is shown below the drop-down list.

Note: All parameters currently set in the selected peak calling instruction will be copied to the Process Profile. Further modifications of the underlying peak calling instruction that were made afterwards do not affect the peak calling parameters in the current Process Profile. This protects the Process Profile from unwanted changes and ensures process stability. If modifications in the underlying peak calling instruction shall also be included in this Process Profile, include the modified peak calling instruction again by selecting it from the **Peak Calling Instruction** drop-down list.

Note: Creating and editing peak calling instruction can only be performed by user role **Developer** or **Administrator**.

Select **NewPeakCallingInstruction** to create a new instruction (for user **Developer** and **Administrator** only). The **Peak Calling** screen is marked with a warning symbol in yellow because the new peak calling instruction is still empty. Proceed to step 2 to define the instruction.

2. Modify the peak calling instruction. The peak calling instruction can be modified as required in this screen as described in the <u>Modifying a peak calling instruction</u> section.

Note: This can only be carried out by user role Administrator or Developer.

Note: If the position of a peak is defined by the relative migration time, make sure that the relative migration times match the marker mode. See <u>Modifying a peak calling instruction</u> for details.

Note: The modifications will only be included in the current Process Profile. If the modifications shall also to be included in the selected peak calling instruction, save the peak calling instruction as described below.

3. Save the modified peak calling instruction.

The modified peak calling instruction can be stored by clicking the **Save as** button. A dialog appears containing the name of the selected peak calling instruction. You have two options: **1**. Click **OK** if the modifications are to be stored in the selected peak calling instruction; **2**. Enter a new unique peak calling instruction name to save the modifications to a new peak calling instruction, then click **OK**. The name of this new peak calling instruction and all of its parameters will be included to the Process Profile.

6.3.3.6 Selecting distribution profiles

Distribution analysis can be performed automatically in the process after smear analysis. Therefore, for example, the **gDNA Analysis** profile can be selected in the **Analysis** screen. In addition, a size or reference marker has to be selected in the **Marker** screen. For more information, refer to <u>Distribution analysis</u>. Thereby, a pre-defined distribution profile is included as Distribution Analysis in a process. Alternatively, Distribution Analysis can be included as a step in a Process Profile. Distribution Profiles can be selected and assigned per sample row. All selections are listed in the **Assignment of Distribution Profiles** list, where each row corresponds to one distribution profile.

Note: This screen in the **Process Setup** is enabled only if analysis and distribution analysis are selected for the Process Profile. For details, refer to <u>Selecting general process options</u>.

To select distribution profiles:

- 1. Open the Distribution Analysis part of the profile definition (1).
- 1a. If the **Assignment of Distribution Profiles** list is empty, click the **Add** button to make the first assignment. An empty entry appears in the list. Proceed with step 2 to specify the entry.

Note: This option is only available for users with the user role Developer or Administrator.

Process Setup					
Set the distribution profiles!					
PROFILE DEFINITION	Assignment of Distribution profiles			Sample Row Selection ③	
1. Process Profile	Distribution	n Profile	Preview	1 2 3 4 5 6 7 8 9 10 11 12	
2. Run Parameters				A ••••••••••	
3. Analysis				В ••••••••••	
4. Marker				C • • • • • • • • • • • •	
1) 5. Distribution Analysis 🛕 〉				D ••••••••••	
6 Report/Export				E •••••	
START & PROCESS				F ••••••	
1 Sample Selection				G • • • • • • • • • • • • •	
				н •••••••••	
2. Sample Information					
3. Run Check					
			-		
			Add Delete		
	Distribution Profile	2 Range		Left-click to add or remove sample rows	
	Save as			A sample row can be assigned to one	
				distribution profile only.	
			OK		
	Save process profile as		Run		

1b. If the Assignment of Distribution Profiles list is not empty, the following options are available:

Add Click the **Add** button to add a new assignment at the bottom of the list. Proceed with step 2 to specify the assignment.

Note: This option is only available for Developer or Administrator users.

- Modify Select the assignment to be modified. Proceed to step 2 to modify the assignment.
- Delete Select the assignment and click the **Delete** button to remove it from the list. Proceed to step 4. **Note:** This option is only available for users with user role **Developer** or **Administrator**.
- 2. Select the desired distribution profile from the **Distribution** profile drop-down list (2).

Note: This option is only available for users with the user role Developer or Administrator.

Note: All parameters from the selected **Distribution Profile** will be preselected in the current Process Profile. Further modifications of the **Distribution Profile** (e.g. in the **Analysis** environment) will not affect the Process Profile. This protects the Process Profile from unwanted changes and ensures process stability. If modifications in the underlying **Distribution Profile** are to be included in this Process Profile, select the modified **Distribution Profile** again by selecting it from the **Distribution Profile** drop-down list.

3. Select the sample rows to be analyzed with the distribution profile (3).

In the **Sample Row Selection** panel, left-click on the sample rows to select or deselect them. The **Range** field shows the sample row selection made. It is possible to select only the rows that are to be analyzed in the process, that is, that have already been selected in the **Run Parameters** screen.

Note: A sample row that has already been assigned to a distribution profile in the assignment list, can no longer be selected anymore.

Note: If the rows to be processed in the **Run Parameters** were deselected during refinement of a Process Profile, these sample rows are now automatically removed automatically on this screen. If rows have been added in the **Run Parameters** screen, one must adjust the row selection in this screen if you would want to use **Distribution** Analysis for the added rows as well.

4. Click OK to confirm.

Note: The OK button becomes enabled as soon as rows are defined and a distribution profile is selected.

The specified parameters appear in the newly created entry in the Assignment of Distribution Profiles list.

Note: Click **Cancel** to discard all entries made during steps 2 and 3. The entry will not be created or the selected entry will not be modified.

5. Repeat steps 1 to 4 to define all assignments in the Assignment of Distribution Profiles list.

The parameters of distribution profiles cannot be viewed or edited in this screen. To view or edit the <u>distribution profile parameters</u>, switch to the **Analysis** environment.

If the underlying distribution profile is modified after being selected for the Process Profile, the name of the distribution profile is preceded with an "*". This indicates that the parameters that will be used by this Process Profile are not the same those visible in the **Analysis** environment. To view the distribution profile parameters that are defined for the current Process Profile:

- 1. Select the row with the distribution profile that is modified.
- 2. Click the **Save as** button below the **Distribution Profile** drop-down box and save the distribution parameters using a new distribution profile name.

Note: This option is only available for users with the user role Developer or Administrator.

- 3. Switch to the Analysis environment to inspect the parameters of the saved distribution profile.
- 4. You can delete obsolete distribution profiles using the Profile Manager in the Configuration environment.

6.3.3.7 Selecting report/export parameters

Note: The **Report/Export** screen is only enabled if **Report option** is selected as Included Step in the Process Profile. For details, refer to the <u>Selecting general process options</u> section.

Specify the report and export parameters as follows:

1. Select a report/export profile.

Use the **Report/Export Profile** drop-down list to select the report/export profile. The parameters of the selected profile are shown below the drop-down list.

Process Setup								
Set the report/export profile!								
Set the report/export profile: PROFILE DEFINITION 1. Process Profile 2. Run Parameters 3. Analysis 4. Marker 5. Peak Calling 6. Report/Export > START A PROCESS 1. Sample Selection 2. Sample Information 3. Run Check	Report/Export Profile *Default DNA v2.0 ▼ Poperault DNA v2.0 Report Options Directory imData\QIAGEN\QIAxcel\ScreenGel 2.0\Data\Report Save to origin experiment directory Format ✓ PDF RTF Options ✓ Display Report Print Report ✓ Overview Experiment Plate Comment Reported by Experiment Path Sample List L ot Information Peak Calling Result Table Overall Smear Result Table							
	✓ Get Image Overview Image options	• · · · · · · · · · · · · · · · · · ·						

Note: All report/export parameters from the report/export profile will be preselected for the Process Profile. Further modifications of the underlying report/export profile do not affect the report/export parameters of the current Process Profile. This prevents the Process Profile from unwanted changes and ensures process stability. If modifications in the underlying report/export profile are also to be included in this Process Profile, include the modified report/export profile by selecting it again from the **Report/Export Profile** drop-down list.

Note: Report/export profiles that have the **Use Images as Displayed** option enabled are not listed in the **Report/Export Profile** drop-down list. This option is only available in the analysis environment. Refer to the Report/Export options section for more information.
Note: In **DNA** mode, report/export profiles that have the Distribution Analysis option selected are not listed in the **Report/Export Profile** drop-down list if Peak Calling features are active. Similarly, report/export profiles with selected Peak Calling option are not listed if Distribution Analysis features are active.

2. Modify the report/export parameters.

The report/export parameters can be modified as required in this screen. Refer to the <u>Report/Export options</u> section for more information.

Note: The modifications will only be included in this Process Profile. If the modified parameters are also to be included in the selected report/export profile, save the report/export profile as described below.

Note: A report/export profile is only applicable if at least one format check box (**PDF** or **RTF**) and one of the content options (**Overview** or **Sample Details**) is selected for report, or if one export option is selected. The possible options are highlighted in yellow in the **Report/Export** screen if no selection is made.

3. Save the modified report/export parameters.

Note: This can only be completed by users with the user role Developer.

The modified report/export parameters can be saved by clicking the **Save as** button. A dialog box appears, which includes the name of the selected report/export profile. You have two options: Click **OK** if the modifications are to be stored in the selected report/export profile; or enter a new unique report/export profile name to save the modified report/export parameters to a new report/export profile, then click **OK**. The name of this new report/export profile and all of its parameters will be included in the Process Profile.

6.3.3.8 Run parameters and result structure

This section provides an example so that the relation between process definition and the structure of the result can be described:

With a QIAxcel DNA High Resolution Cartridge:

- Rows A and B will be run twice with Method OM1200, with size marker at position A1
- Rows C and D will be run 3 times with Method OM800, with the same size marker at position C1

The same alignment marker will be used for both methods.

See the **Run Parameters** and **Sample Selection** screens of the process definition (refer to the <u>Selecting run</u> <u>parameters</u> section):

Plate Definition Method Details								
Method Details								
Method	Range	М	Inj. Time	RpR	Runs	Preview		
0M1200	A,B	A1	10	2	4			
0M800	1800 C,D C1 10				6			
Allow row de	selection	✓ A	llow marker o	lefinition	Insert	Add Delete	<u>;</u>	
Method					Range	Marke	r	
0M800		▼			C,D	C1		
Injection TimeRuns per Row10 sec3								
* This method costs 1 regular run(s) decrement in the cartridge key.								
						OK Cano	el	

Two entries were defined because of the different methods. The rows and size marker positions were defined for each method, as well as for the runs per row (column **RpR** in the **Method Details** list). In the **Runs** column the total number of runs for each method is listed.

During the process, the software first runs Method OM1200 in the order row A and B (first run), and row A and B (second run). It then runs Method OM800 in the order row C and D (first run), row C and D (second run), and row C and D (third run).



The Sample row selection shows a summary of the run definition in the Run Parameters screen.

The total number of runs (Total Runs) in this example is 10:

2 times A and B = 4 runs for Method OM1200

3 times C and D = 6 runs for Method OM800

When the process is finished, the sample data is presented in the **Analysis** environment in the **Experiment Explorer**:



All sample data generated by the process are combined into one experiment and named according to the **Experiment Name / Plate ID**, which was specified in the **Sample Selection** screen.

All sample data is grouped by so called "plates" as they were generated by the process.

The name of the plate identifies the run. It is combined with:

Experiment name	The experiment name is created from the Experiment Name / Plate ID specified in the Sample Selection screen.
R#	Run number, where "#" represents a number, starting from 1. In this example for method OM1200, the run numbers R1 and R2 are generated.
E#	Entry number (entry in the Method Details list of the Run Parameters screen), where "#" represents a number starting from 1. In this example, the entry numbers E1 and E2 are generated, because the Method Details list contained 2 entries.

Therefore, the sample data for Method OM1200 for rows A and B are listed first and also separately for each run.

6.3.4 Creating a new process profile

Note: Only users with the assigned role Administrator and Developer can create a new Process Profile.

To create a new Process Profile, proceed as follows:

1. Open the Process environment (1).

If not open, switch to the **Process** environment by clicking the **Process** environment icon. Select the **Process Profile** screen.

Process Setup

Set the process profi	le!	
PROFILE DEFINITION		Mode
1. Process Profile (1)	▲ >	● DNA ○ RNA
2. Run Parameters	Δ	Cartridge Type
3. Analysis		DNA High Res. (2)
4. Marker		Process Profile
5. Peak Calling		Peak Calling Distribution Analysis
6. Report/Export		*NewProcessProfile
START A PROCESS		Profile
1. Sample Selection	Δ	
2. Sample Information		Included Steps Image: Report Image: Report Image: Report
3. Run Check		Experiment Directory
		C:\ProgramData\QIAGEN\QIAxcel\ScreenGel 2.0\Data\Experiment\DNA
		Allow Directory Selection
		Notes
		Save process profile as (4) Run

Note: If the last process was just completed, click the Back to Wizard button at the bottom right.

2. Select cartridge type (2).

Select the cartridge type that is to be used for the new Process Profile.

Note: Each Process Profile is related to a certain cartridge type. Therefore, the system can ensure that a process (i.e., data acquisition) can be started only if a cartridge with correct cartridge type is inserted.

Note: If the instrument is connected, the system automatically detects the inserted cartridge and the cartridge type cannot be changed. If you want to create the Process Profile for another cartridge type, remove the cartridge, or at least the cartridge key, or disconnect from the instrument.

3. Select a Process Profile (3).

Use the **Process Profile** drop-down list to select a Process Profile. The selected profile serves as a template for the creation of the new profile.

Note: Select **NewProcessProfile** to create a Process Profile from scratch. The system will show ***NewProcessProfile** after selection. This can only be carried out by users with the assigned role **Administrator** or **Developer**.

3. Set the Process Profile options according to your needs.

See section <u>Process Profile options</u> for detailed information.

4. Save the modified Process Profile under a new name (4).

Click the **Save Process Profile as** button below process setup and enter a new unique profile name in the dialog that appears.

Note: This can only be done by users with the user role Administrator or Developer.

Note: If there are Process Profile screens containing incomplete or inconsistent data highlighted in yellow, the **Save process profile as** button is disabled. Select the marked Process Profile screens and correct the data. If all data is correct, the **Save process profile as** button becomes enabled and the Process Profile can be saved.

6.3.5 Viewing method details

During definition of a Process Profile, it is possible to view the details of a method.

Proceed as follows:

- 1. Select a Process Profile in the Process Profile screen.
- 2. Switch to the **Run Parameters (1)** screen. Select the entry containing the method you want to view in the **Plate Definition** tab .
- 3. Switch to the Method Details (2) tab.
- 4. Select another method to view, if required.

Note: Selecting another method modifies the entry in the **Plate Definition** tab you came from. Ensure you have selected the correct method you want to process.

PROFILE DEFINITION	Plate Definition	Meth	od Details 🤇	2	
1. Process Profile	Method		Car	tridge Type	
2. Run Parameters 1	> OM500 V	0M500 V			
3. Analysis					
4. Marker	Method Definition				
5 Peak Calling	Action	Voltage	Duration	Position	
5. Feak Gailing	Purge	0,00 kV	30 sec	WP	
6. Report/Export	Inject	0,00 kV	0 sec	WI	
	Inject	4,00 kV	20 sec	M1	
TART A PROCESS	Inject	0,00 kV	0 sec	WI	
1. Sample Selection	SampleInject	5,00 kV	10 sec	None	
	Inject	0,00 kV	0 sec	WI	
2. Sample Information	Inject	0,00 kV	0 sec	WI	
	Inject	0,00 kV	0 sec	WI	
3. Run Check	Inject	0,00 kV	0 sec	WI	
	Inject	0,00 kV	0 sec	WI	
	Separate	5,00 kV	500 sec	Buf	

All actions defined in the method are listed. During the run, the actions will be performed sequentially.

For more information about what the position abbreviations mean, see the **Instrument Status** section of the **Status Information** panel to the left of the **Process** environment.

6.3.6 Status Information panel

The left panel of the screen in the **Process** environment is called the **Status Information** panel. It is divided into:

- A toolbar at the top
- Instrument Status
- Cartridge Status

🖳 QI	Axcel Scre	enGel			
File	View	Tools	Help		
			>>>>>>>>>>>>>>>>>>>>>>>>>>>>>>>>>>>>>>	L.A.L	
			Process	Analysis	
	AGEIN				
Status	Informatio	on			
Instrument Status					
Cartridge Status					

Instrument Status and **Cartridge Status** can be collapsed and expanded. The **toolbar** buttons and status icons are explained in the following table:

	Load Position button. Click the button to move the buffer tray to the front of the instrument to load samples and buffer.
	Note: The motors will not move if either the sample door or cartridge door is open.
	Wash Park Position button. Click the button to move the cartridge to the wash park position of the buffer tray.
	Note: The motors will not move if either the sample door or cartridge door is open.
a	Indicates that the currently inserted QIAxcel gel cartridge is latched . Click the button to unlatch the QIAxcel gel cartridge.
	Important: If the Pressure 1 status is LOW , increase system pressure before unlatching. Releasing the cartridge latching with reduced pressure may damage the instrument.
£	Indicates that the currently inserted QIAxcel gel cartridge is unlatched . Click the button to latch the QIAxcel gel cartridge.



The **Instrument Status** panel displays the buffer tray and sample plate. The current position of the capillaries of the QIAxcel gel cartridge is highlighted when the process is running. In the example below, the sample rows A–D are processed with the same method.

▼ Ins	trum	er	nt S	tat	us								Position description
WP WI BUF M1 M2	Ì												Wash Park Wash Inject (Seperation) Buffer Marker 1 Marker 2
А	0	0	0	0	0	0	0	0	0	0	0	0	Sample row A - currently beeing processed
В	0	0	0	0	0	0	0	0	0	0	0	0	Sample row B
С	0	0	0	0	0	0	0	0	0	0	0	0	Sample row C
D	0	0	0	0	0	0	0	0	0	0	0	0	Sample row D
Е	•	•	•	•	•	•	•	•	•	•	•		Sample row E
F	•		•	•	•	•	•	•	•	•	•	•	Sample row F
G	•	•	•	•	•	•	•	•	•	•	•	•	Sample row G
Н	•	•	•	•	•	•	•	•	•	•	•		Sample row H

The **Cartridge Status** panel displays information about the QlAxcel gel cartridge currently inserted, such as cartridge ID, cartridge type, remaining number of runs, remaining number of calibration runs, expiration status, and calibration status. When opening the **Calibration Details** expander, additional calibration details are shown.

	Instrume	nt Status	
•	Cartridge	Status	
Cart	ridge ID		
C21	0507A99		
Cart	ridge Type	è	
DNA	A Screeni	ng	
Rom	aining Ru	ne	Calibration Runs Loft
83	uning itu	110	2
E		4	Calibratian Status
Expi Vali	ration Sta	tus	
van	u		UK
•	Calibratio	on Details	5
Calil	pration Da	ite	Accepted By
07.0	05.2021 1	1:40:02	Service
Ch.	Area	Cal. Fact	or Result
1	0,0051	1,4144	Passed
2	0,0051	0,9689	Passed
3	0,0050	0,8727	Passed
4	0,0045	1,0018	Passed
5	0,0047	0,8044	Passed
6	0,0045	0,7396	Passed
7	0,0051	1,0330	Passed
8	0,0054	1,1428	Passed
9	0,0051	2,0957	Passed
10	0,0049	0,7969	Passed
11	0,0051	1,1086	Passed
12	0,0051	0,7276	Passed

Note: Calibration status **NOT OK** means that the cartridge is not calibrated and needs calibration. For information about calibration, refer to the <u>Calibrating a cartridge</u> section.

Note: The Status Information panel is shown in the Process environment and the Service environment.

6.4 Analysis

The **Analysis** environment provides sophisticated visualizations and analysis algorithms for electropherograms acquired by the QIAxcel Connect instrument.

The software provides several customizable views for sample raw data, for example, a Gel view, a Single Electropherogram view, and Electropherogram Superposition view. Electropherogram views also visualize analysis results, enabling fast assessment of the results.

The analysis algorithms provided by the QIAxcel ScreenGel software automatically determine a variety of peak properties. **Analysis** algorithms can be applied to single samples or collections of samples (called experiments), and can be customized by the user. The size and concentration of analytes can be determined. The **DNA** mode includes functions that enable quality assessment of samples, for example, after a target enrichment step in a workflow. These functions include checking for the presence of certain fragment sizes (peak calling), and determining and assessing the molarity and size distribution of DNA fragments (distribution analysis). The **RNA** mode provides the RNA Integrity Score (RIS) as a tool to objectively asses the quality of eukaryotic RNA samples. In addition, parameters such as 28S/18S ratio and concentration of total RNA can be calculated.

2 QIAxcel ScreenGel			- U X
File Edit View Help			
>>> 🖳			Ļ
Process Analysis	Service Configuration		Notifications
- GIAGEN			
Experiments	Gel Image Electropherogram Peak Calling Marker	Analysis Report Pe	eak Calling Properties >
🕞 🗔 🗾 📑	🔲 🏥 🖳 🔍 🖏 👗 🕅 🕂 Image Options Select All	▼ Mode	
- C400007400 2040 07 40 42 24 27 . Q. X		● DNA ○ RNA	
C100607A99_2010-07-16_13-34-37	0,900		
1 2 3 4 5 6 7 8 9 10 11 12	0.800	 Analysis Properties 	3
		Default DNA Analysis	V Save as
B		Deremetere	
	2 0.000 		
	5 0.500	Peak O Smear	⊖ gDNA
F	5 0,400-	Smoothing Start	Window
	5 0,300- 	Filter 0,00 min	15 pts
	8 ⁴	Baseline Start	Window
6 000000000	Ž 0,100	Filter 0,00 min	40 sec
	0,000	Minimum Start	Value
	15 72 194 271 803 1078 5000 [bp]	0,00 min	2,00 sec
	Peak Size	Suspend Start	End
	<u>↑</u> -0,093172 0,944492 RFU		
	I→ -0,651495 ←I 7,166452 min	Threshold Start	Value
	Sample Info: Position: A8 Experiment Name / Plate ID: C100807A99_2010-	0,00mm	5,00 3/14
	# Size [bp] Conc. [ng/µ] Saturated	Marker 20	S/N
	2 564 0,31 No		
Add a comment for this experiment	3 5000 n/a No		Add Delete
	Peak Result Peak Calling Result	1 Sample(s) selected	Start Analysis
	Login: 30.06.2022 10:01:27 Name	Developer Mode: DNA QI	Asphere Base: Disconnecte

The **Experiments** panel on the left displays functions for <u>Handling samples and experiments</u>. The middle panel displays several graphical views and result tables. For more information, refer to <u>Viewing sample</u> <u>data</u>.

On the right is the **Parameters** panel, where analysis parameters and report/export options can be specified.

If it is not visible, you can display it using the **View** menu (selecting the menu item **View/Show Analysis Parameters**) or by clicking the ^[] icon at the far right of the view selection bar:

Gel Image Electropherogram Peak Calling Marker

6.4.1 Introduction into analysis

Before (re)analysis of samples can start, the experiment must be <u>loaded</u> in the **Experiment Explorer** on the left and the samples must be <u>visualized</u> in the **Gel view** or **Electropherogram view**. Additionally, the samples that shall be analyzed must be <u>selected for analysis</u>. Refer to <u>Handling samples and experiments</u> and <u>Viewing sample data</u> and its sub sections for more details.

The analysis of samples is performed using a two-step approach. First, peaks are detected in the raw data. As a result, the alignment marker peaks are found and the first, very basic peak properties are available, e.g. peak start and peak end, peak apex as a function of time, as well as peak area. After peak detection, the samples can be viewed and aligned according to the alignment marker peaks found in the <u>Gel view</u> or the <u>Electropherogram overview</u>. Because peak detection is the basis of the analysis, ensure that the peaks and especially the alignment marker peaks have been found correctly. For details, see <u>Peak detection</u>.

In a second step, the peak sizes and peak concentrations are determined by mapping the detected peaks in the sample to peaks of a reference marker which is created based on a size marker that has been used in this or a previous run. To use a size marker from a previous run for analysis, it is important that the same cartridge, method, instrument and alignment marker are used. For size and concentration, see section <u>Size</u> and <u>Concentration determination</u>. The software allows you to combine the first and the second analysis step.

Optionally, a third analysis step is possible. Either <u>Peak Calling</u> or <u>Distribution analysis</u> can be used based on peak detection and size determination. These analyses can be used to check whether the samples meet certain defined expectations.

Follow the instructions in <u>Analysis of DNA samples</u> and <u>Analysis of RNA samples</u> for step-by-step guidance for your specific samples.

One can benefit from fully automated processing by including analysis steps already into the process of data acquisition. Starting with analysis parameters initially provided by the software, customize the parameters such that the analysis gives good and reliable results for your samples. Save them into <u>Profiles</u> and use them to create fully automated customized <u>Process Profiles</u>.

6.4.2 Handling samples and experiments

The **Experiment Explorer** in the **Analysis** environment displays samples in a concise manner and provides functionality to load and manage samples.

After a process is completed, the result data is saved in an experiment. If the **Analysis** environment is selected, the experiment is automatically loaded in the Experiment Explorer.

An experiment groups samples by plates and rows in the order that they were processed. For detailed information, refer to the <u>Run parameters and result structure</u> section.

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Several experiments can be loaded, but only one experiment can be active at a time. The samples in the experiment can be visualized and analyzed. A note for the active experiment can be entered in the note field at the bottom of the explorer.

Customized experiments can be created to combine sample result data from different experiments. Refer to the <u>Customizing experiments</u> section for detailed information.



The Experiment Explorer toolbar has the following buttons:

Load experiment from disk: Refer to the Loading sample data section for detailed information.
Save : Save the active experiment to disk. Refer to the <u>Saving an experiment</u> section for detailed information.
Move/rename : Save the active experiment; it allows the name and directory to be changed. Refer to the <u>Saving an experiment</u> section for detailed information.
Note: This function does not create a copy of the experiment.
Create new experiment : Refer to the <u>Creating a new experiment</u> section for detailed information.

6.4.2.1 Meaning of sample icons

In the **Experiment Explorer**, samples are symbolized by a well and size markers by a ladder

These symbols vary slightly according to the analysis, visualization, and selection status:

Analyzed	Meaning	Not analyzed
• =	Not visualized, not selected.	0
• =	Visualized, not selected.	o 🔳
• =	Not visualized, selected.	•
• =	Visualized, selected.	• ■

The current focus sample has, in addition, a border **L**. This sample is displayed in the Single Electropherogram view and you can inspect its properties.

The **Size Marker** property for a sample can be changed by using the sample context menu. Use the **Size Marker** option to toggle.

A reference marker can be created afterward from samples with the Size Marker property.

6.4.2.2 Loading sample data

To load samples from the disk, proceed as follows:

- 1. Click is at the top of the Experiment Explorer.
- 2. A file dialog appears, showing all experiments that were created in the current run mode (e.g. in DNA mode, you will not see RNA experiments). Navigation starts in the most recently opened directory, or otherwise in the default experiment directory. You can navigate to a different directory.

Note: To open calibration results, select the path ending with "[Calibration Results]"

- 3. Select the experiment from the list by double-clicking it, or clicking it with the mouse and clicking **Load** file.
- 4. The experiment will be loaded and displayed in the Experiment Explorer.

Note: If there were experiments already loaded in the **Experiment Explorer**, the experiment just opened will be displayed as the last experiment, at the bottom. The newly loaded experiment automatically becomes active.

Note: In **DNA** mode, loaded experiments that include peak calling or distribution analyses trigger the software to automatically activate peak calling or distribution analysis features, respectively.

Alternatively, you can open one or more experiments via drag and drop from the Windows Explorer. To do so, navigate to the folder where the experiments to be opened are located and drag and drop them into the **Experiment Explorer**. All experiments dragged and dropped into the **Experiment Explorer** that can be opened in the given mode (e.g. in DNA mode, only DNA experiments can be opened) will be loaded and displayed. Files that cannot be opened by the application will be ignored.

Note: Close experiments after analysis and reporting is completed.

٢	Select experiment to load			
Look in	Dna			▼
Path	C:\ProgramData\QIAGEN\QIAxcel\ScreenGel 2.	.0\Data\Ex	perimer	nt
	Name		Туре	Date Modified
	210519099_2021-05-19_09-31-28.xdrx 210519099_2021-05-23_12-45-56.xdrx 210601D99_2021-06-01_09-23-05.xdrx 210720133_2021-04-22_08-13-23.xdrx		Exp. Exp. Exp. Exp.	28.05.2021 09:56:12 28.05.2021 11:08:34 01.06.2021 09:31:32 28.05.2021 11:07:34
		Load		Cancel

Representation of an experiment on disk

An experiment is represented on disk as a file with the name of the experiment. The file extension depends on the mode used to create the experiment (**.xdrx** for DNA, **.xrrx** for RNA, and for calibration data created during cartridge calibration, **.xcdrx**, and **.xcrrx**, respectively).

Loading experiments of previous software versions

To load an experiment that was created with a previous QIAxcel ScreenGel software version, proceed as described above. The loaded experiments will be automatically converted (migrated) to the newest version.

Note: A migrated experiment cannot be opened by previous software versions any more.

In version 1.0.x of QIAxcel ScreenGel, an experiment was represented on disk as a folder with the name of the experiment. The folder contained a file with the name of the experiment, describing the experiment structure. The extension of this file was dependent on the mode used to create the experiment (**.xdr** for DNA, **.xrr** for RNA, **.xcr** for calibration data created during cartridge calibration). In addition, the folder contained several files, each containing the sample data of a sample row. The file names were extended based on the experiment name.

When an experiment created with QlAxcel ScreenGel software version 1.0.x is loaded to QlAxcel ScreenGel software versions 1.1 or higher, it is automatically converted (migrated) to the newest version. After saving the experiment, the folder with the experiment and sample files is replaced by a single experiment file with the appropriate new file extension (see above).

When a **RNA** experiment created with QIAxcel ScreenGel software version 1.0.x is loaded to QIAxcel ScreenGel software versions 1.1 or higher, the search criteria for the 18S and 28S peak in the reference marker table are automatically converted to a peak calling instruction.

6.4.2.3 Selecting samples

Samples can be selected in the **Experiment Explorer** or in the **Gel Image** view or the **Electropherogram** overview.

For more information about activating an experiment, see the Activating an experiment section.

There are different options to select samples inside the active experiment:



Select an individual sample with a left-click in the **Experiment Explorer.**

Note: Already activated samples can be selected with a left-click on a sample in the **Gel Image** view or **Electropherogram overview**.

▼ C100	607	A9	9_2	010	0-07	7-10	6_1	3-3	4-3	7	۵	×
▼ C100607A99_2010-07-16_13 R1 E1												
	1	2	3	4	5	6	7	8	9	10	11	12
A		•	•	•	•	•	٩	•	•	•	•	•
В	•	•	•	•	•	•		2	•	•	•	•
1												

To select multiple single samples, simultaneously hold the **Ctrl** key and left-click the samples in **Experiment Explorer.**

Note: Already visualized samples can be selected with a left-click on a sample in the **Gel Image** view or **Electropherogram overview**.



To select multiple samples in a rectangular area use the **Experiment Explorer**, simultaneously hold the **Shift** key and — for this example — left-click the samples A3 and B5.

Use the **Experiment explorer** to select all samples in a row, left-click the row letter.

To select multiple rows, hold the **Ctrl** or **Shift** keys simultaneously.

Use the **Experiment Explorer** view to select all of the samples of a plate, right-click the plate name and select the context menu option **Select Whole Plate**.

In the **Experiment Explorer** view all of the samples of the experiment can be selected. Right-click the experiment name and select the context menu option **Select all Samples**

Note: Alternatively all activated samples can be selected via the **Select All** button in the **Gel Image** or **Electropherogram** overview.

Switch the focus by left-clicking a sample. During sample selection, the last sample selected has the focus.

If samples from different experiments need to be selected, create a new customized experiment containing these samples. For more information about customized experiments, see the <u>Customizing experiments</u> section.

6.4.2.4 Selecting samples for analysis or report

After samples have been visualized in Gel Image or Electropherogram view they can easily be selected for analysis. Descriptions are provided below:

View	Single sample selection	Multiple sample selection
Gel Image	Select a single sample by clicking the gel lane header of the sample	Selection of multiple samples can be performed by clicking the gel lane headers using the Shift or Ctrl key (like in the Windows Explorer). Alternatively, you can select all visualized gel lanes by clicking the Select All key in the tool bar.
Single Electropherogram	Select a single sample in the Experiment Explorer. In this view, the visualized sample will be analyzed.	Not possible in this view.
Electropherogram Overview	Select a single sample by clicking the electropherogram of the sample.	Selection of multiple samples can be performed by clicking the electropherograms using the Shift or Ctrl key (like in the Windows Explorer). Alternatively, you can select all visualized electropherograms by clicking the Select All key in the tool bar.
Electropherogram Superposition	Not possible in this view.	Select multiple samples from the Experiment Explorer view using the Shift or Ctrl key.

Note: Additionally, for all views it is possible to select activated samples from the Experiment Explorer. For single sample selection click on the wanted sample position. For selection of multiple samples use the Shift or Ctrl key or the mouse pointer.

6.4.2.5 Expanding and collapsing

For a better overview, you can collapse experiments or plates, regardless of whether the experiment is active or not.

To collapse an experiment:

- Click ▼ to the left of the experiment name. The entire experiment collapses down to the experiment name.
- 2. Click > to expand it again.

Note: You can collapse a single plate inside an experiment by clicking ▼ to the left of the plate name. The plate collapses down to the plate name. Click ► to expand it again.

6.4.2.6 Activating an experiment

Only one experiment can be active at a time. The samples in the active experiment can be visualized and analyzed.

To activate an experiment in the **Experiment Explorer**, right-click the experiment and select **Activate (1)** from the context menu or double-click the experiment header.



The previously active experiment will be deactivated automatically. If this experiment was modified, the system asks whether the changes should be saved or not. Click **Yes** to save or **No** to discard the changes. Select **Cancel** to cancel activation. For more information about saving experiments, see the <u>Saving an</u> <u>experiment</u> section.

Note: Experiments from older software versions cannot be deactivated. Instead, they will be closed if you activate another experiment. Avoid this by saving them in the new format. Note, however, that once saved with the current software version, the experiments can no longer be opened with older software versions.

6.4.2.7 Saving an experiment

To save the active experiment, click the **Save** button at the top of the **Experiment Explorer**.

All modifications made to this experiment will be saved, as specified in the settings. This includes analysis results, as well as information about the configuration of the visualization.

Note: Sample data cannot be saved individually.

If you want to save the active experiment to another directory, use the **Move/rename** button 2. This will open a file dialog for choosing a different directory and experiment name.

Note: To avoid data redundancy, the previous experiment file will be removed.

The same file dialog opens if the experiment was <u>created</u> in the Experiment Explorer and has not yet been saved.

(1)	Select destination folder and experim	ent name.	
Look in	DNA		▼ 💼
Path	C:\ProgramData\QIAGEN\QIAxcel\Scree	enGel 2.0\Data\Experim	ent
	Name		Date Modified
 C2 ⁻ C2 ⁻ C2 ⁻ C2 ⁻ C2 ⁻	10519099_2021-05-19_09-31-28.xdrx 10519099_2021-05-23_12-45-56.xdrx 10601D99_2021-06-01_09-23-05.xdrx 10720133_2021-04-22_08-13-23.xdrx		28.05.2021 09:56:12 28.05.2021 11:08:34 01.06.2021 09:55:14 28.05.2021 11:07:34
Name C	210601D99_2021-06-01_09-23-05		
		Move/Rename	Cancel

The file dialog enables navigation to the directories of the disk, creation of a new directory, and allows the user to choose the new name of the experiment. The appropriate file extension will be appended automatically.

Note: The experiment name is restricted to 40 characters.

Information stored in the experiment

The following information is stored in the experiment:

- Experiment structure
- Visualization information: visualization status of the samples (not for superimposition), sample order in the visualization.

The following information is saved in each sample since it can differ from sample to sample:

- **Run information**: all run parameters, including cartridge information, parameters of the method used, sample injection time, and separation time. The run information is stored immediately after the process has finished and will not be changed later.
- Analysis information, if the sample has been analyzed: includes analysis parameters, reference marker table used during analysis, and peak calling information.

For more details about samples refer to the <u>Inspecting sample properties</u> section. For details about settings and directory configuration, refer to the <u>Settings</u> section.

6.4.2.8 Closing an experiment

You can close an experiment by clicking \times to the right of the experiment name, regardless of whether the experiment is active or not.

If the experiment was modified, the system asks you whether to save the changes or not. Click **Yes** to save or **No** to discard the changes. Select **Cancel** to cancel closing.

For more details about saving experiments, see the Saving an experiment section.

6.4.2.9 Revising sample information

To revise sample information or sample comments after the run, select the sample in the Experiment Explorer, right-click the sample, and select the context menu option **Revise Sample Information** or **Sample Comment**. Enter the new sample information and/or comment in the dialog that appears.

Note: The context menu option **Revise Sample Information or Sample Comment** is enabled only if the user has the right to revise the sample information. The administrator can grant the right in the <u>User management</u>.

Note: The report will indicate that a sample name or comment has been revised.

6.4.2.10 Handling an incomplete experiment

An experiment can be incomplete if the run stopped during data acquisition. This is indicated in the Experiment Explorer with a yellow lock icon. Position the mouse over the lock icon to display a tooltip with further information.

Experiments												Ge	el Ima	ge	Electropherogram				
6 6 🔊 c ⁺														२ 🕅 ⁵ , 👗					
•	▼ C210712R00_2021-07-15_0001 ♀! ×																		
F		1	2	3	4	5	6	7	8	9	10		3 ou	it of	8	runs	s perf	forme	ed successfully.
	А	0	0	0	0	0	0	0	0	0	0	0	0			FU×		6,00	0.
	в	0	0	0	0	0	0	0	0	0	0	0	0			nits IR		5,00	0.
	С	0	0	0	0	0	0	0	0	0	0	0	0			nce U		4,00	0
														-		oresce		3,00	0

You can remove this icon from the experiment (for example, if you could successfully reprocess the missing runs) by right-clicking the experiment name and select the context menu option **Accept Corrupt Experiment**.

Exp	perim	ents	5			Gel Image El	ectrophere		
	6		7				Q ×		
▼ C210712R00_2021-07-15_0001									
•	C210	712	R0	0_2	021	1-07	7-15	Select all Samples	
		1	2	3	4	5	6	Visualize Whole Experiment	
	A	0	0	0	0	0	0	Accept Corrupt Experiment]
	В	0	0	0	0	0	0	·····································	
	С	0	0	0	0	0	0		
								3,000	

Note: This option is only available to users, where the Administrator has granted this right (based on their user role).

The Administrator can grant the right **May accept incomplete experiments** using the **User Management** system.

The report for such a run will contain information about the incomplete process and the acceptance in the report overview section.

6.4.3 Viewing sample data

The Analysis environment offers several options for visualization of raw data and analysis results:

- A Gel image view, which can display several samples see <u>Gel view</u>.
- A Single Electropherogram view, which visualizes a single sample along with its analysis results see <u>Electropherogram view</u>.
- An Electropherogram overview, which visualizes several electropherograms in a gallery see <u>Electropherogram overview</u>.
- An Electropherogram Superposition view, which visualizes several electropherograms in one plot see <u>Electropherogram Superposition view</u>.

All views allow data exploration by zooming and scaling the data.

6.4.3.1 Adding samples to views

Note: A maximum of 97 samples can be visualized at a time in the Gel Image view and Electropherogram overview. If this limit is reached, visualization of more samples is disabled.

Samples can be added to a view by dragging samples from the **Experiment Explorer** to:

- The Gel view
- The Electropherogram overview
- The Electropherogram Superposition view (see description at the end of this section)

Proceed as follows:

- 1. In the Experiment Explorer select the samples to be visualized.
- 2. Left-click the selected samples, drag to the desired view, and drop. All selected samples will be displayed in the selected view.

Note: In Gel view and Electropherogram overview a marker is shown when dropping. All samples will be displayed at the selected position. See also <u>Changing lane order</u>.



Alternatively, samples can be added to the views through the sample context menu of the Experiment Explorer. Use the **Visualize Selected Samples** option to add the selected samples.



Finally, to visualize a plate, right-click on the plate name and select the context menu option **Visualize Whole Plate**. To visualize the whole experiment, right-click the experiment name and select the context menu option **Visualize Whole Experiment**.

Unlike all other views, the **superposition view** does not automatically display all samples selected for visualization.

To add samples to the superposition view:

- 1. In the Experiment Explorer select the samples to be added.
- 2. Open the context menu of the selected samples in the Experiment Explorer.

3. Select the **Superimpose** option.



The selected samples are added to the superposition view. In the **Experiment Explorer**, all displayed samples are marked by a colored rectangle corresponding to their color in the superposition.

Note: Up to 12 electropherograms can be superimposed. A warning will be displayed if the limit is exceeded.

Alternatively, drag and drop up to 12 selected samples from the **Experiment Explorer** to the **superposition view**.

6.4.3.2 Removing samples from views

Samples can be removed from the views by dragging the selected samples from the view back to the **Experiment Explorer**.



Note: You can drop the samples anywhere in the Experiment Explorer.

Alternatively, use the context menu of the samples in the **Experiment Explorer**:

- 1. Select the samples that should be removed from the view.
- 2. Right-click a selected sample and uncheck the Visualize Selected Samples.



Finally, if a whole plate or a whole experiment is visualized, you can remove all the samples from the view. Right-click the plate name or the experiment name and uncheck the **Visualize Whole Plate** or **Visualize Whole Experiment** option from the context menu.

To remove samples from the superposition view, proceed as follows:

- 1. In the Experiment Explorer, select the samples to be removed from the view.
- 2. Open the context menu of the selected samples in the Experiment Explorer.
- 3. Unselect the **Superimpose** option.

6.4.3.3 Exporting view to clipboard

Sample views can be copied to the clipboard:

- 1. Select the gel lanes/electropherograms to be copied (in <u>Gel view</u> or <u>Electropherogram overview</u>).
- 2. Right-click the selected sample and select Copy to clipboard and the desired image resolution.
- 3. Paste the image.

Note: You can use the keyboard shortcut **Ctrl+C** to copy selected images to the clipboard. Using the keyboard shortcut will use the default resolution value (96 dpi).

Note: When you paste the image in an application, make sure that it uses the right resolution. QIAxcel ScreenGel puts the pixel array on clipboard which is adapted for desired resolution (this means that the image has the appropriate pixel size). The application you paste the image into must be able to interpret this information correctly.

Note: The samples that have been copied to the clipboard, have the same image settings as in the visualization view (such as color palette, contrast, noise cutoff, and zoom factor).

Note: In the <u>Single Electropherogram view</u> and <u>Electropherogram Superposition view</u> view the gel lane and C-channel diagram will be copied to the clipboard too if they are displayed. If you hide both gel lane and C-channel diagram, only the samples will be copied to clipboard. You can access the **Copy to clipboard** pop-up menu from gel lane and C-channel diagram as well.



6.4.3.4 Direct printing view

You can directly print images of the selected samples:

- 1. Select the lanes/electropherograms to be printed.
- 2. Right-click on a selected sample and select Print.

This function works in a similar way to the **copying to clipboard** function with default image resolution; the selected samples will be sent to the default printer. They have the same image settings as in the visualization view (such as contrast, noise cutoff, and zoom factor).

The image sent to the printer is identical to the image you would copy to the clipboard; if the gel lane and electrical current curve diagram are displayed in the <u>Single Electropherogram view</u> and <u>Electropherogram</u> <u>Superposition view</u>, they will be printed as well.

6.4.3.5 Result table

The analysis result table of a sample is displayed in the **Analysis** environment below the Electropherogram views or the Gel view. Sample information, sample comment (if entered), position of the sample on the plate, and experiment name / plate ID are displayed at the top of the result table. When an analyzed sample is selected, the results are shown in the result table. The displayed columns depend on the mode. Right-click the header line of the table, select **Show column**, and then select the property you are interested in.

Sample Info:		Position: A	5 Experiment Name
# Size [bp] (Conc. [ng/µl] Saturated		
1 15	Show all Colun	nns	
2 16261	Show Column	×	
	Restore Defau	It Columns ^は	
Peak Result	Smear/gDNA Result	Distribution Analysis Result	

The **Peak Result** table lists the detected peaks of the sample. Use this result table for <u>Standard DNA</u> <u>analysis</u>, for the <u>Fast Analysis of DNA samples</u> and for <u>RNA analysis</u>. Refer to the <u>Peak result columns</u> section for detailed information on available peak properties.

The **Peak Calling Result** table lists the peak calling result of the sample. Use this result table also for <u>RNA</u> integrity analysis. Refer to the <u>Peak calling result columns</u> section for detailed information.

The **Smear/gDNA Result** table (DNA mode) and **Smear Result** table (RNA mode) list the detected smear peaks of the sample and shows properties that are specific for smear peaks. Smear peaks are detected only when the analysis profile option **Smear** or **gDNA** had been selected in the **Analysis Properties** panel at the right of the **Analysis** environment (<u>Smear DNA analysis</u>, <u>gDNA analysis</u>). Refer to the <u>Smear result columns</u> section for detailed information on smear peak properties.

If a distribution analysis was performed, the **Distribution Analysis Result** table is visible instead of the **Peak Calling Result** table. Refer to the <u>Distribution result columns</u> for detailed information.

Note: Peak calling and distribution analysis generate an additional overview result table in the middle panel of the **Analysis** environment. For more information, refer to <u>Peak Calling</u> or <u>Distribution result</u>, respectively. 6.4.3.5.1 Modifying the result table

The information displayed in the result table can be customized for the Electropherogram Single view and overview independently. The customization applies to all further viewing of the results.

Note: In the Electropherogram Superposition view, a small non-customizable result table is displayed.

Adding a column

To add a column:

- 1. Right-click the table header.
- 2. Select the column to be displayed from the Show Column selection.

Note: Select the **Show all Columns** option to display all columns. Select the **Restore Default Columns** option to display the default setting for the application.

Removing a column

To hide a column:

- 1. Right-click the column header.
- 2. Unselect the column from the Show Column selection.

Changing column order

The order of the displayed columns can be changed by dragging a column header to the desired position. While dragging, a marker is displayed that indicates the position of the column when dropping it.

Note: Columns of the peak calling and distribution result tables cannot be reordered.

Resizing column width

The column width can be changed by dragging a vertical cell border in the table header.

Note: Column widths of the peak calling and distribution result tables cannot be changed.

6.4.3.5.2 Interacting with the result table

Selecting and removing a peak

When selecting a peak in the result table, the peak is highlighted in the Electropherogram (Single Electropherogram view and Electropherogram overview). To select a peak in the result table, left-click the peak number in the # column. To unselect the peak, use the **Unselect Peak** context menu option or just click to another cell. To remove the selected peak from the result table, use the **Delete Selected Peak** context menu option. To remove multiple selected peaks from the result table, first select the peaks by holding the **Ctrl** key and left-clicking the corresponding peak numbers in the # column. Alternatively, hold the **Shift** key and left-click the first and last peak numbers in the # column. All peaks in between will be selected. Right-click the selection and click **Delete Selected Peaks**.

Exporting analysis results

The contents of the result table can be exported to other applications using the Windows clipboard. There are several ways to select the table cells:

- Select the cells with the mouse while holding down the left mouse button.
- Click the first column of a row to select a row. You can also select multiple columns using the **Shift** and **Ctrl** key.
- Click the column header to select a column.
- Click the column header of the first column to select all cells.

To copy your selection:

- 1. Select cell(s).
- 2. Right-click a selected cell.
- 3. Click Copy selected cells to clipboard.
- 4. Paste the data to other applications, for example, Microsoft Excel[®].

Note: The result data is exported using the local settings of the Windows system. Make sure that the target application also uses the local settings to interpret the numbers.

6.4.3.5.3 Peak result columns

The following columns are available:

#	Number of peaks in the sample.	
Size [bp]	For DNA mode only. Fragment size in base pairs.	
Size [nt]	For RNA mode only. Fragment size in nucleotides.	
Conc. [ng/µl]	Concentration of the fragment in ng/µl.	
	Note: This value is not calculated for alignment marker peaks.	
--	--	--
Conc. [pg/µl]	For High-Sensitivity experiments only. Concentration of the fragment in $pg/\mu I$.	
	Note: This value is not calculated for alignment marker peaks.	
Saturated	Indicator whether the peak intensity reaches the maximum possible intensity (see <u>Saturated</u> <u>Signals</u> section). The calculated height, area and concentration of saturated peaks are not correct.	
Mol. [nmol/l] Molar concentration (molarity) in nmol/l. The calculation is based on the fragmentation and concentration.		
	Note: This value is not calculated for alignment marker peaks.	
Mol. [pmol/l]	For High-Sensitivity experiments only. Molar concentration (molarity) in pmol/l. The calculation is based on the fragment size and concentration.	
	Note: This value is not calculated for alignment marker peaks.	
S/N	Signal-to-noise ratio of the peak. The noise is approximated with three times the standard deviation of the noise data points from the baseline.	
NA	Normalized area, also called corrected peak area. This is the peak area divided by the migration time of the peak apex.	
Area	Peak area (integration of the area under the signal and above the baseline).	
NA %	Normalized peak area relative to the sum of all normalized peak areas.	
	Note: This value is not calculated for alignment marker peaks.	
Ratio NA	The area ratio with respect to the previous peak. The alignment marker peaks and the first data peak in the table will have an empty field here.	
Height	Height of the peak at its maximum.	
Height %	Peak height relative to the sum of all peak heights.	
	Note: This value is not calculated for alignment marker peaks.	
Res.	The separation resolution relative to the next peak.	
	Note: This value is not calculated for alignment marker peaks.	
FWHM [sec]	The peak width at half the maximum intensity (full width at half maximum), which is a parameter for peak resolution and thus the precision of size calculation.	
Start [min]	Start time/X-value of the peak in minutes.	
Time [min]	Time/X-value for the peak maximum.	
Stop [min]	Stop time/X-value of the peak in minutes.	
Rel. Time	Relative time/X-value for the peak maximum of a sample in relation to alignment markers. This value is mode-dependent:	

In **DNA** mode, in case of 2 alignment marker peaks, the relative position of the peak between lower alignment marker and upper alignment marker is mapped to the interval from 0 to 1, i.e. all peaks have a **Rel. Time** between 0 to 1. The alignment markers have a **Rel. Time** of zero and one, respectively. In case of one alignment marker peak, the relative migration time is calculated as in RNA mode.

In **RNA** mode, the relative position of the peak in relation to the lower alignment marker is calculated, i.e. the alignment marker has a **Rel. Time** of 1 and all other peaks have **Rel. Time** higher than 1.

6.4.3.5.4 Peak calling result columns

Each row in the table corresponds to a sample; the columns **Pos.** and **Sample Info** identify the sample. Please note the **Pos** column is fixed. All following columns are defined by the peak calling instruction. Aggregated columns over all peaks of interest appear when they are selected in the **Calculated Columns** panel. The following columns are grouped by the names of the peaks of interest and show several peak properties. To change the properties listed for each peak of interest, right-click on the table header and select/unselect the property using the **Show Column** option. The change will take effect in the entire table.

Note: If a sample was not analyzed, it is not present in the peak calling result overview.

Parts of the peak calling result table can be copied to the clipboard. Select the cells to be copied and then press **Ctrl+C**. To copy the complete result of a peak calling instruction, right-click in the peak calling result table and select **Copy Peak Calling Result of** [...].

6.4.3.5.5 Smear result columns

The following smear peak properties are available:

#	Number of peak in sample.	
	Note: As alignment marker peaks are no smear peaks, the peak number in this table usually starts with 2.	
Median Size [bp] or [nt]	Determines the fragment size that corresponds to the median of the area of interest. The median is the point in the peak curve, where geometrically the left part of the peak area has the same size as the right part. The value is given in base pairs. For DNA samples units are shown as bp and for RNA samples as nt.	
Conc. Area of Interest [ng/µl] or [pg/µl]	Continuously calculated concentration of the fragments corresponding to the area of interest. Value is given in ng/µl for non-High Sensitivity experiments, and in pg/µl for High Sensitivity experiments.	
Mol. [nmol/] or [pmol/l] Area of Interest	Molar concentration (molarity) of the fragments corresponding to the area of interest. This calculation is based on the continuously calculated fragment size and concentration, not Median Size , and is given in nmol/l for non-High Sensitivity experiments, and in pmol/l for High Sensitivity experiments.	

Start Area of Interest [bp] or [nt]	Start of the area of interest given in bp for DNA or nt for RNA.	
Stop Area of Interest [bp] or [nt]	End of the area of interest given in bp for DNA and nt for RNA.	
% Conc. Area of Interest	Percentage of concentration of fragments in the area of interest relative to the concentration of the smear peak (defined by the start and end of the peak).	
NA Area of Interest	Normalized area based on the area of interest. Area of the smear peak normalized l the migration time.	
% NA Area of Interest	Percentage of the normalized area relative to the normalized area of the smear peak (defined by the start and end of the peak).	

Note: All values listed in the table above refer to the area of interest assigned to a smear peak. Moving the borders of the area of interest of a smear peak triggers the recalculation of the corresponding properties.

In addition, two properties are calculated for the entire sample. The values are displayed just above the smear result table:

Total Concentration [ng/µl] or [pg/µl]	Total concentration of the fragments in the entire sample. The calculation is performed continuously for all data points with a signal above the baseline. The calculation starts after the end of the lower alignment marker peak and ends at the beginning of the upper alignment marker peak. The value is given in ng/µl for non-High Sensitivity experiments, and in pg/µl for High Sensitivity experiments.
Total Molarity [nmol/l] or [pmol/l]	Total molarity of the fragments in the entire sample. The calculation is based on continuously calculated fragment size and concentration for all data points with a signal above the baseline. The calculation starts after the end of the lower alignment marker peak and ends at the beginning of the upper alignment marker peak. The value is given in nmol/l for non-High Sensitivity experiments, and in pmol/l for High Sensitivity experiments.
	If the value is not visible, scroll the horizontal table scroll bar to the right to see the value.

6.4.3.5.6 Distribution result columns

Each row in the table corresponds to a sample, with the columns **Pos.** and **Sample Info** identifying the sample. Columns relevant to the entire sample appear first. Thereafter, columns are grouped according to the names of the defined areas of interest and display several smear peak properties. The remaining columns are then grouped by the name of defined ratios. Find a detailed description in the section <u>Distribution result columns</u>.

To change the properties listed, right-click the table header and select or unselect the property with the **Show Column** option. The change takes effect in the entire table.

Note: If a sample was not analyzed with a smear or gDNA analysis profile, it will not appear in the distribution result overview. If a sample was not analyzed with a reference marker, the distribution values cannot be calculated. In this case, the sample **Quality** is listed as "**Review**." In both cases, follow the instructions in <u>Analysis of DNA samples</u> to check and correct the analysis steps that are required before **Distribution** analysis. Ensure using **Analysis** parameters as described in <u>Smear DNA analysis</u> and <u>gDNA</u> analysis, respectively.

The distribution result table can be copied to the clipboard. To copy the complete result of a distribution profile, right-click in the distribution result table and select **Copy Distribution Result of** [...].

Columns relevant to the entire sample:

Pos	Position on the plate.	
Sample Info	Sample information	
Total Conc. [ng/µl or pg/µl]	Total concentration of the fragments across the entire sample in ng/µl for non- High Sensitivity experiments, and in pg/µl for High Sensitivity. Refer to <u>Smear</u> <u>result columns</u> for details. The column displays "n/a" for size marker samples.	
Total Mol. [nmol/l or pmol/l]	Total molarity of the fragments across the entire sample in nmol/l for non-High Sensitivity experiments, and in pmol/l for High Sensitivity experiments. Refer to <u>Smear result columns</u> for details. The column displays "n/a" for size marker samples.	
Quality	The overall quality assessment of the sample.	
	The column reports "Passed" if all ratio assessments and height checks passed (if applicable). That is, if one sub-assessment failed, the overall quality reported is "Review" as well. The column displays "Not Analyzed" for size marker samples.	

Note: If the sample was not analyzed with a reference marker table, i.e., if the <u>Size and concentration</u> <u>determination procedure</u> was unsuccessful, the **Quality** reported will be "Review," and **Total Conc.** and **Total Mol.** will display "n/a."

Columns relevant to an area of interest (grouped by name of the area of interest):

Mol. [nmol/l or pmol]	Molar concentration (molarity) of the fragments corresponding to the area of interest in nmol/l for non-High Sensitivity experiments, and in pmol/l for High Sensitivity experiments. Refer to <u>Smear result columns</u> for details. The column displays "n/a" for size marker samples.	
Conc. [ng/µl]	Concentration of the fragments corresponding to the area of interest in ng/µl for non-High Sensitivity experiments, and in pg/µl for High Sensitivity. Refer to <u>Smear</u> result columns for details. The column displays "n/a" for size marker samples.	
Size Start [bp or nt]	Start size of the area of interest given in base pairs for DNA or in nucleotides for RNA. The column displays "n/a" for size marker samples.	
	Note: The size may differ slightly from the start size defined in the distribution profile. The area of interest starts at the data point with a size nearest to the defined start size.	
Size End [bp or nt]	End size of the area of interest given in base pairs for DNA or in nucleotides for RNA. The column displays "n/a" for size marker samples.	
	Note: The size may differ slightly from the end size defined in the distribution profile. The area of interest ends at the data point with a size nearest to the defined end size.	
Height [S/N]	Maximum signal height of the area of interest, given as a Signal-to-noise ratio [S, N]. The noise is approximated by taking three times the standard deviation of the noise data points from the baseline. The column displays "n/a" for size marker samples.	
Height Check	The quality assessment of an area of interest with regards to the Height [S/N].	
	The column reports "Not Analyzed" if the height of the area is not to be checked; "Passed" when the checked height of the area of interest is above the defined minimum height for that area; and "Review" when not. The column displays "Not Analyzed" for size marker samples.	

Note: If the sample was not analyzed with a reference marker table, i.e., if the <u>Size and concentration</u> <u>determination procedure</u> was unsuccessful, the **Height Check** reported will be "Not Analyzed" and all other columns will display "n/a."

Note: If the signal in an area of interest does not exceed the baseline at any point (indicating that the area of interest is not present in the sample), the **Height Check** reports "Not Analyzed" when the height of the area is not to be checked, and "Review" when it is to be checked. All other columns display "n/a."

Columns relevant to a ratio (grouped by name of the ratio):

Ratio (Molarity) This column is present only if the calculation basis Molarity has been selected for ratios.

The value of the molarity ratio. That is, the molarity of the area of interest defined as the Numerator divided by Denominator, which may be either the molarity of another area of interest or the Total Molarity. The column displays "n/a" for size marker samples.

Ratio (Concentration)	This column is present only if the calculation basis Concentration has been selected for ratios.	
	The value of the concentration ratio. That is, the concentration of the area of interest defined as the Numerator divided by Denominator, which may be either the concentration of another area of interest or the total concentration. The column displays "n/a" for size marker samples.	
Ratio Quality	The quality assessment of the ratio. The column reports "Not Analyzed" if the quality of the ratio is not to be checked; "Passed" when the value of the ratio is within the defined range; and "Review" when not. The column displays "Review" for size marker samples.	
Numerator	Defined in the distribution profile. This is the area of interest, of which the molarity or the concentration is used as numerator of the ratio, depending on the selected calculation method.	
Denominator	Defined in the distribution profile. Either Total Sample or another area of interest, of which either the molarity or the concentration is used as denominator of the ratio, depending on the selected calculation method.	

Note: If the sample was not analyzed with a reference marker table, i.e., if the <u>Size and concentration</u> <u>determination procedure</u> was unsuccessful, the **Ratio Quality** reported will be "Review" and the **Ratio** will display "n/a."

Note: If the signal of the area of interest used for the numerator does not exceed the baseline at any point (indicating that the area of interest is not present in the sample), the molarity/concentration value is assumed to be "0." If the signal of the area of interest used for the denominator does not exceed the baseline, the column **Ratio Quality** displays "Review." In both cases, the column **Ratio** displays "n/a."

For information about defining the distribution profile, refer to the Modifying a distribution profile section.

6.4.3.6 Gel view

The Gel view displays the Gel images of the samples.

The lanes in the Gel view can be displayed in four view modes:

===	Normal mode	
	Inverted mode	
≡±≣	False color rendering mode	
Gel Image	Electropherogram Peak Calling Marker	
5000 310 310 310 310 310 310 310 310 310	Image: second	
<u>↑</u> -0,14	06 ↓ 3,323 RFU 0 ←1 5,187005 min	Contrast: 1,10

The y-axis on the left-hand side is the main scale for the whole image. When the size scale is selected, it is based on the bands of the reference marker applied to all samples (see sample A1 in the image above).

The y-axis on the right-hand side describes the detected bands of the selected sample only (see above: highlighted with the black border, in the image above sample A7). It is available in the application only, not in report/export images (also not if the **Use Images as Displayed** option is selected).

Above each Gel image the sample position is displayed (e.g. A1, A2, and so on). For a Gel image a tool tip can be displayed by moving the mouse pointer to the sample number. This tool tip has information about the plate, the number of the run, and method that the sample was processed with.

If the mouse pointer hovers over the gel lanes, the values at the current cursor position are shown in the upper left corner of the view.

The <u>result table</u> of one sample is displayed below the Gel view. This sample is marked in the **Experiment Explorer** and outlined with a black border in the Gel view (see sample A7 in the image above). Select the sample to display in the result table by left-clicking it in the **Experiment Explorer** or in the Gel view.

To view a sample in detail, double-click its gel lane. The view changes to Single Electropherogram view. To return to the Gel view, select the **Gel Image** tab again.

You can interact with the Gel view mainly through the buttons above the view:



Toggles zoom mode on/off.

In zoom mode (button pressed), the mouse is used to select a region for zooming into (Rubber band function — see <u>General software usage</u>). In addition, you can use the scroll wheel of the mouse to zoom in and out.

If this button is not pressed, the drag-and-drop function is activated to change the lane order.

Note: The controls in the lower left corner of the Gel view can be used to set the zoom region to absolute values, independently of the zoom mode.

Note: The drag-and-drop function is always possible if the mouse cursor is over the gel lane label, even in the zoom mode (button pressed).



Auto scale.

Resets the zoom region to the overall data range, undoing all zooming.



Undo zoom.

Returns to the previous zoom state.



Toggles a horizontal ruler at cursor position.

To configure the Gel view, click the Image Options button for the following options:

Unit of y-axis Use this option to select the units of the y-axis scale on the left-hand side of the Gel view.

If you select **Size**, the y-axis will show a size scale based on the reference marker. The lanes in the Gel image will be aligned according to the alignment markers.

Note: The size scale can only be created if all selected samples have been run with the same alignment marker, have been analyzed with the same reference marker table, and the alignment markers have been correctly identified. Otherwise, a message will appear instead. In this case, reanalyze the samples with the same reference marker table and ensure that the alignment markers have been correctly identified, or remove from the view the samples that have a different alignment marker or those samples for which alignment markers could not be correctly identified.

If you select **Relative migration time**, the y-axis will show relative migration time. The lanes in the Gel image will be aligned according to the alignment markers.

Note: The relative time scale can only be created if all selected samples have been run and analyzed with the same alignment marker, and the alignment markers have been correctly identified. Otherwise, the lanes cannot be aligned and the y-axis will display a corresponding message. In this case, re-analyze the samples so that the alignment markers are correctly identified, or remove the samples from the view that used a different alignment marker or remove those samples for which the alignment markers could not be correctly identified.

If you select **Absolute migration time**, the lanes are not aligned and the y-axis will show an absolute time scale.

Individual scaling	Select this option to auto-scale the contrast for each gel lane individually.		
	Unselect this option for a comparison of samples.		
Show sample information	Select this option to display sample information at the top of each gel lane.		
Show plate ID	Select this option to display the plate ID at the top of the gel lane.		
Show method	Select this option to display the method applied at the top of each gel lane.		
Show analysis details	Highlight alignment markers	Select this option to highlight the alignment marker bands in green.	
		Note: This highlighting is only possible for analyzed samples if the alignment markers have been correctly identified	
	Show peak size	If this option is selected, the y-axis on the right-hand side will describe the sizes of the detected bands of the selected sample	
		Note: If the sample was not analyzed with a reference marker, the y-axis on the right hand will display "n/a." If the sample was not analyzed, a corresponding message will appear.	
		Note: This option is always selected in the RNA mode.	
	Show median of size	For DNA mode only. Select this option to show the median size of detected smear peaks for the selected sample.	
		Note: If the sample was not analyzed with a DNA smear analysis profile or a reference marker , the y-axis on the right hand will display "n/a." If the sample was not analyzed, a corresponding message will appear.	

Controls for contrast settings are located below the Gel view to the right. The functions are described below:

Contrast	Use the slider to change the contrast according to your needs. The contrast value wi be saved together with the experiment. Thus, you can customize the Gel images independently of other experiments.	
Noise cutoff	Use the slider to customize display of the noise in the signal. Move the slider to the extreme left position to see all signals. Move the slider to the right to suppress small signals, eliminating the noise. As for contrast, the noise cutoff value will be saved together with the experiment.	

6.4.3.6.1 Changing lane order

The gel lanes can be reordered by drag-and-drop. The lane order takes effect for all views where the samples are visualized. The changed order will be saved when saving the experiment.

To change the lane order:

- 1. Select one or more lanes in the Gel view.
- 2. Left-click the header of the selected lane or lanes and drag it to the new position. While dragging, a marker is shown indicating the new position of the lane(s) when dropping.
- Drop the lane(s) when the marker indicates the correct new position. The lane(s) will be placed in that
 position. If more than one lane was selected, the lanes will be inserted in the same order as they were in
 before.

Note: If the zoom button is toggled off, dragging can start also inside a gel lane.

6.4.3.7 Electropherogram view

The Electropherogram view is activated by clicking on the **Electropherogram** tab in the analysis environment. Three Electropherogram views can be selected using the following buttons in the tool bar:



Single Electropherogram view.



Overview mode with several single electropherograms.



Superposition view with several superimposed electropherograms.

The Single Electropherogram view is described in this section. The overview and superposition mode are described in the following sections.



The Single Electropherogram view consists of a plot showing the recorded signal on top and the Gel view representation of the signal below the main plot.

Controls for the zoom region and contrast settings of the Gel view are located below the graphical representations of the signal. The current cursor position is shown in the upper left corner of the view.

The analysis result of the sample is displayed below the Electropherogram view. Refer to section <u>Result table</u> for detailed information about the different types of result tables.

You can interact with the electropherogram through the buttons in the tool bar at the top (all buttons are shown in "**not pressed**" state, left-click a button to press/release).

Q	Toggles zoom mode on/off. For details, see <u>Zooming and scaling</u> .
ж. м	Auto scale. For details, see Zooming and scaling.
5	Undo zoom. For details, see Zooming and scaling.
<u>×</u>	Insert peak. For details, see <u>Adding a peak</u> .
μΣ-Ι	Manual integration of a time range. For details, see <u>Manual range integration</u> .
	Toggles ruler.
•	Activates vertical and horizontal rulers used to compare peak heights and positions.

To configure the Electropherogram view, click the **Image** options button for the following options:

Unit of x-axis	Use this option to select the units of the x-axis scale.		
	If you select Size , the x-axis will show a size scale based on the reference marker, if the sample has been analyzed with a reference marker table and the alignment markers have been correctly identified otherwise, a message will appear.		
	If you select Rel. Time , the x-axis will show relative migration time, if the sample has been analyzed and the alignment markers have been correctly identified otherwise, a message will appear.		
	If you select Abs. Time [mi	in], the x-axis will show an absolute time scale.	
	Note: If the relative time scale is not created, re-analyze the sample so the alignment markers are correctly identified. If the size scale is not created, re-analyze the sample with a reference marker table so that the alignment markers are correctly identified.		
Show gel lane	Select this option, to display the gel representation below the electropherogram in alignment with the x-axis.		
Show electrical current curve	Select this option to display the diagram of the electrical current measured during data acquisition below the Gel representation, if present, in alignment with the x-axis.		
Show analysis details	Select this option to display analysis details for analyzed samples. Additional options can be selected.		
	Mark detected peaks	Select this option to display the apex markers of detected peaks.	
		If the option With label is selected, peak labels are displayed at the apex of each peak.	
		Note: Only peak labels that do not overlap with an adjacent label are displayed. Hover the mouse cursor over the peak apex to get a tool tip with the peak label.	
		Select the label units: Size, or absolute or relative migration time.	
		Note: The label can be displayed as Size only if the sample was analyzed with a reference marker (for details refer to <u>Size</u> and <u>Concentration determination</u>). Otherwise, "n/a" is displayed.	
		A second option Mark median of size is available. If the sample was analyzed with a smear analysis profile , select this option to display the marker with the median size of detected smear peaks. You can switch the corresponding label on and off.	

	Select the option Mark peak start and end to mark the start and end points of detected peaks.
Show areas of interest	If the sample was analyzed with a smear analysis profile , select Show areas of interest to display the areas of interest of detected smear peaks.
Show suspend integration intervals	Select this option to display the suspend integration intervals.
Show threshold	Select this option to display the peak threshold (shown in blue).
	Note: The threshold parameter can be interactively changed by moving the threshold with the mouse. For details, see <u>Modifying</u> <u>the threshold</u> .
Show baseline	Select this option to display the baseline (shown in red).

Below the Gel view at the right, controls for contrast settings are located. The controls are described below:

Contrast	Use the slider to change the contrast according to your needs. Contrast changes apply to gel lanes of all experiments below electropherograms in single view, but do not affect the contrast settings of the Gel overview.
Noise cutoff	Use the slider to customize the display of noise in a signal. Move the slider to the far left position to display all signals. Move the slider to the right to suppress small signals, eliminating noise. As with contrast, changes to the noise cutoff value do not affect the Gel overview.

6.4.3.7.1 Zooming and scaling

The Electropherogram view allows you to explore the data by zooming and scaling.



Toggles zoom mode on/off.

When the button is clicked, the mouse can be used to select a region to zoom into. For information about how to use the Rubber band function, see <u>General software usage</u>. Additionally, you can use the mouse scroll wheel to zoom in and out.



Auto scale.

Resets the zoom region to the entire data range (both in time and RFU dimension), and undoes all zoom operations.



Undo zoom.

Returns to the previous zoom state.

Independent of the zoom mode, all electropherograms can be panned using the axes. You can shift the zoom region horizontally by dragging the x-axis or vertically by dragging the y-axis.





In addition to the tool buttons, the controls in the lower left corner of the **Electropherogram view** can be used to set the zoom region to absolute values.

Note: For RFU values, only the values are permitted that are within the actual values. Values outside the actual value range are not accepted and the background of the input field turns yellow.

Note: For time values, one hour is allowed as the upper range value.

6.4.3.7.2 Peak annotations

The analysis result of a sample is displayed along the raw signal. Each peak has peak markers for start (green), apex (red '+', green for alignment marker peaks), stop (turquoise), and a peak label. In **DNA** mode, if <u>smear</u> or <u>gDNA</u> analysis has been performed, i.e., if the sample has been analyzed with the option **Smear** or **gDNA**, the median of the peak can be marked (with a pink '**x**') instead of the apex



In addition to the peaks, the baseline (in red) and the threshold for peak detection above the baseline (in blue) are displayed in the electropherogram.

Peak markers and units of the peak label can be configured by clicking the Image options button. Whether peak markers, the baseline or the threshold are displayed or hidden can be configured under Image options as well. For details, refer to <u>Electropherogram view</u>.

6.4.3.7.3 Interacting with the result table

The functions are described below:

Selecting a peak	In the electropherogram, left-click the apex marker of a peak. Alternatively, in the result table, left-click the peak number in the # column.
	The peak will be marked with a red circle in the electropherogram and highlighted in the result table
Unselecting a peak	In the electropherogram or in the result table, open the context menu of the selected peak and select the Unselect Peak option.
	The red circle in the electropherogram disappears as well as the highlighting in the result table.
Adding a peak	See <u>Adding a peak</u> for information on how to add a peak. The peak will be added to the result table.
Deleting a peak	See <u>Deleting a peak</u> for information on how to delete a peak.
	The peak will be removed from the result table, and the markers of the peak in the electropherogram disappear.

6.4.3.7.4 Manual range integration

The manual integration of a time range is possible using the **Range Integration Tool**

After clicking the **Range Integration** button, you need to specify a region. To do so, first click the left border and then the right border of the region.



After selecting the region, a dialog is displayed showing the time range borders, the normalized area sum of the region, and the percentage of the normalized area relative to the total normalized area.

Note: The calculated values can be copied to the clipboard by using the context menu of the dialog.

Note: The alignment marker peaks are not included in the normalized area calculation.

6.4.3.8 Electropherogram overview

The Electropherogram overview displays several electropherograms in a gallery view. Twelve samples are always displayed on one page, arranged in a 3 x 4 matrix. It is intended for an overview inspection of the acquired data. In the Electropherogram overview the ruler tool is especially useful. It provides a means to compare peak positions between several samples, both for alignment marker peaks and sample peaks. For analyzed samples, each peak has a peak marker for apex time (sample peaks are red, alignment marker peaks are green).



The zooming and scaling functionality is very similar to the Single Electropherogram view (see <u>Zooming and</u> <u>scaling</u>). The only difference is that zooming affects all electropherograms.

Use the **forward** and **backward** buttons at the bottom of the gallery view to navigate through the pages of the overview. Clicking on a sample in the Experiment Explorer will also navigate to the corresponding electropherogram in the gallery view. Use the scrollbars to navigate through the electropherograms on one page.

To enlarge the overview, collapse the parameters panel on the right side.

A result table displaying information on one sample appears below the Electropherogram overview. Refer to the sections <u>Interacting with the result table</u> and <u>Result table</u> for more information.

To inspect an electropherogram in detail, double-click it. The view changes to Single Electropherogram

view. To return to the overview, click 📖 again.

You can interact with the electropherogram mainly through the buttons in the tool bar:

Q	Toggles zoom mode on/off. For details, see <u>Zooming and scaling</u> .
ĸ	Auto scale. For details, see Zooming and scaling.
ل ك ا	Undo zoom. For details, see <u>Zooming and scaling</u> .
	Toggles ruler.
	Activates vertical and horizontal rulers used to compare peak heights and positions.

To configure the Electropherogram overview, click the Image options button for the following options:

Unit of x-axis	Use this option to select the units of the x-axis.
	If you select Size , the x-axis will show a size scale based on the reference marker. The electropherograms will be aligned according to the alignment markers.
	Note: The size scale can only be created if all selected samples have been run with the same alignment marker, have been analyzed with the same reference marker table, and the alignment markers have been correctly identified. Otherwise, the x-axis will show a corresponding message. In this case, re-analyze the samples with the same reference marker table and ensure that the alignment markers have been correctly identified, or remove the samples from the view that have a different alignment marker or those samples for which alignment markers could not be correctly identified.
	If you select Rel. Time , the x-axis will show relative migration time. The electropherograms will be aligned according to the alignment markers.
	Note: The relative time scale can only be created if all selected samples have been run with the same alignment marker, have been analyzed, and the alignment markers have been correctly identified. Otherwise, the electropherograms will not be aligned and the x-axis will display a corresponding message. In this case, re-analyze the samples so that the alignment markers have been correctly identified, or remove the samples from the view that have a different alignment marker or those samples for which alignment markers could not be correctly identified.
	If you select Abs. Time [min], the electropherograms will not be aligned and the x-axis will show an absolute time scale.
Show sample position	Select this option to display the sample position in the upper left corner of each electropherogram.
Show sample information	Select this option to display the sample information at the top of each electropherogram.
Show plate ID	Select this option to display the plate ID at the top of each electropherogram.
Show method	Select this option to display the method applied at the top of each electropherogram.

	Note: Only one of the options Show sample information , Show plate ID or Show method can be selected at a time.				
Show analysis details	Select this optior be selected.	Select this option to show analysis details for analyzed samples. Additional options can be selected.			
	Mark detected	Select this option to display the apex indicators for detected peaks.			
	peaks	A second option Mark median of size is available. If the samples were analyzed with a smear analysis profile , select this option to display the indicators with the median size of detected smear peaks.			
	Show suspend integration intervals	Select this option to display the suspend integration intervals.			
6.4.3.8.1	Changing order				

Remove samples from the Electropherogram overview by dragging them from the overview into the Experiment Explorer.

Add samples to the Electropherogram overview by dragging them from the Experiment Explorer into the overview. Added samples will be inserted at the end of the gallery.

To change the order of the electropherograms, switch to Gel overview and reorder the samples there. See <u>Changing lane order</u> for details. Then switch back to the Electropherogram overview.

6.4.3.8.2 Interacting with the result table

Below the Electropherogram overview, the result table of one sample is displayed. This sample is marked in the **Experiment Explorer** and outlined with a black border in the gallery overview (see sample B3 in the image below). Select the sample in the **Experiment Explorer** or in the Electropherogram overview by left-clicking it to view its result table.



Inspecting result table of sample A6.

There are additional functions for the result table which can be accessed with a right-click of the mouse. The functions are described below:

Selecting a peak In the electropherogram, left-click on the marker of the peak apex. Alternatively, in the result table, left-click the peak number in the # column.

The peak will be marked with a red circle in the electropherogram and highlighted in the result table.

Unselecting a In the result table, open the context menu of the selected peak and select the **Unselect Peak** option.

The red circle in the electropherogram disappears as well as the highlighting in the result table.

Deleting a peak Select the peak to be deleted in the result table. Open the context menu of the result table and select the option **Delete selected peak**.

The peak will be removed from the result table, the marker of the peak in the Electropherogram overview disappears.

6.4.3.9 Electropherogram superposition view

The Electropherogram Superposition view displays up to 12 electropherograms in one plot. It allows an easy comparison of several measurements.

To discriminate the signals from different samples distinguishable, each sample has a distinct color.



In the lower part of the superposition view, small result tables of all samples are shown along with the corresponding colors. The order of the result tables is the same as the sample order in the Gel view or Electropherogram overview.

You can interact with the electropherogram through the buttons in the tool bar:

l	Toggles	zoom mod	e on/off.	For details	s see	<u>Zooming</u>	and sco	<u>ıling</u> .
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Auto scale. For details see Zooming and scaling.

Undo zoom. For details see Zooming and scaling.

Toggles ruler.

Activates vertical and horizontal rulers used to compare peak heights and positions.

To configure the Electropherogram Superposition view, click the Image options button for the following options:

Unit of x-axis	Use this o	ption to	select the	units o	f the x-axis.

If you select **Size**, the x-axis will show a size scale based on the reference marker. The electropherograms will be aligned according to the alignment markers.

Note: The size scale can only be created if all selected samples have been run with the same alignment marker, have been analyzed with the same reference marker table, and the alignment marker(s) have been correctly identified. Otherwise, the x-axis will display a corresponding message. In this case, re-analyze the samples with the same reference marker table and ensure that the alignment marker(s) have been correctly identified, or remove the samples from the view that have a different alignment marker or those samples for which alignment marker(s) could not be correctly identified.

If you select **Rel. Time**, the x-axis will show relative migration time. The electropherograms will be aligned according to the alignment marker(s).

Note: The relative time scale can only be created if all selected samples have been run with the same alignment marker, have been analyzed, and the alignment marker(s) have been correctly identified. Otherwise, the electropherograms will not be aligned and the x-axis will display a corresponding message. In this case, re-analyze the samples so that the alignment marker(s) have been correctly identified, or remove the samples from the view that have a different alignment marker or those samples for which alignment marker(s) could not be correctly identified.

If you select **Abs. Time** [min], the electropherograms will not be aligned and the x-axis will show an absolute time scale.

Show Select this option to display the diagram of the superimposed electrical current measured during data acquisition below the electropherogram superposition in alignment with the x-axis.

Show size marker peak labels

If this option is selected, and if exactly one size marker is among the superimposed samples, the peak labels will be displayed for detected peaks of the marker sample. Select the units of the peak labels in the drop-down list.

6.4.3.9.1 Adding and removing samples

The superposition view does not automatically display all samples selected for visualization.

To add samples to the superposition view:

- 1. In the **Experiment Explorer** select the samples to be added.
- 2. Open the context menu of the selected samples in the Experiment Explorer.
- 3. Select the **Superimpose** option.

Experiments		Gel Image	Electropherogram
🖻 🖥 🎽 🗗			Q () Ky =
▼ *C210601D99_2021-06-01_09- ▼ C210601D99_2021-06-01_09 1 2 3 4 5 6 7 8 A ■ ● ● ● ● ● ● ● ● ● ● ● ● ● ● ● ● ● ●	23 X R1 E1 9 10 11 12 Visualize Selected S Superimpose Size Marker Remove Analysis D Transfer Analysis In Revise Sample Info	Samples ata structions	e Comment
	Remove Peak Callir	ng Data	
	Transfer Peak Callir	ng Instruction	
	Remove Row		

The selected samples will be added to the superposition view. In the **Experiment Explorer** all samples that are displayed in the superposition view are marked by a colored square according to their color in the superposition.

Note: Up to 12 electropherograms can be superimposed. A warning will be displayed if the limit is exceeded.

Alternatively, drag and drop up to 12 selected samples from the Experiment Explorer to the superposition.

To remove samples from the superposition view, proceed as follows:

- 1. In the Experiment Explorer, select the samples to be removed from the view.
- 2. Open the context menu of the selected samples in the Experiment Explorer.
- 3. Unselect the **Superimpose** option.

6.4.3.10 Inspecting sample properties

To view sample properties:

- 1. Select the sample in the **Experiment Explorer**.
- 2. To view the sample properties, click **Properties** tab.

Analysis	Report	Distribution	Properties X
▼ Sam	ple		
Experime	nt		
MO_V-1	192		
Experime	nt Path		
C:\Progr	amData\QI/	AGEN/QIAxcel\s	ScreenGel 2.
Experime	nt Name / P	late ID	
		5 N I	
Plate Pos	ition	Run Numbe	er.
famala li			
V-1192	1		
Sample (omment		
Run Date 28.12.20	/Time 15 07:56:16	User ID Labor	
Acquisiti	on Rate [Hz]	Rise Time [sec]
8,547		0,3	
Injection	Time [sec]	Separation	Time [sec]
20		206	
Anal	ysis Instru	ctions	
▶ Distr	ibution Pro	ofile	
 Meth 	od		
▶ Cart	ridge		
▶ Platf	orm		

In the **Properties** tab, there are six panels which contain the following information:

• Sample

Contains general information about the sample and the acquisition, for example, the date, experiment ID, and plate ID.

- Analysis Instructions Shows the analysis parameters, alignment marker, and the reference marker used for the analysis.
- Peak Calling Instruction or Distribution profile Shows the peak calling instruction or distribution profile that was last applied to the sample.
- Method Shows the method name and actions used during the acquisition.
- Cartridge

Shows information about the cartridge, such as the cartridge ID and the cartridge type.

• Platform Shows the software version and the instrument information (firmware version and serial number) for the run.

Important: You cannot perform peak calling and distribution analysis in the same experiment. You should either apply peak calling instruction or a distribution profile to a sample, but never both.

6.4.3.11 Saturated signals

Identifying a saturated signal

Overloading a sample by injecting highly concentrated DNA leads to saturation of the detector. Saturated signals are those where the signal level exceeds the maximal detector limit.

A saturated signal usually has a flat top in the electropherogram. These signals are also referred to as "clipped".



Preventing a saturated signal

There are two ways to prevent overloading or saturation of the detector: either dilute the sample (DNA in QX DNA (HS) Dilution Buffer, RNA in RNase-Free water) or inject less sample. A smaller amount can be injected by reducing the injection time

6.4.4 Peak detection



This section describes the peak detection procedure.

Analysis parameters for peak detection are provided in Analysis Profiles.



Alignment marker peaks are marked in green. It is important that the correct peaks are marked as alignment marker peaks because this is the basis for all other analysis functions.

Note: In the image above, the **Analysis** parameter **Alignment Marker Threshold** (see dark blue line in the image above) was greater than the **Threshold** parameter. This leads to a peak in the threshold line at alignment marker positions.

6.4.4.1 Peak detection procedure

To perform peak detection, proceed as follows:

- 1. Load the samples you want to analyze using the **Experiment Explorer**. For details, refer to the <u>Loading</u> sample data section.
- 2. Visualize the samples you want to analyze. See the <u>Viewing sample data</u> section for details on data inspection.
- 3. Select the samples for analysis.
- 4. Open the Analysis parameters panel to be able to specify the analysis properties.

If it is not visible, you can display it using the **View** menu (selecting the menu item **View/Show Analysis Parameters**) or by clicking the ^[] icon at the far right of the view selection bar:

Gel Image Electropherogram Peak Calling Marker

In the **Analysis** properties panel, define the parameters to use for analysis. This can be done by selecting a predefined analysis profile, which can be either a default (e.g. "Default DNA analysis") or a user defined parameter set.

Refer to <u>Analysis of DNA samples</u> and <u>Analysis of RNA samples</u> for getting guidance as to which **Analysis** profile should be selected initially and how to use the analysis options for your specific samples.

Alternatively, a new analysis profile can be created. To modify or create an analysis profile, refer to sections <u>Modifying an analysis profile</u> and <u>Creating a new analysis profile</u>, respectively. The analysis parameters of the selected profile are shown below the profile drop-down list. For general peak detection, no reference marker is required. Check **No Marker** in the **Reference Marker** section, as illustrated in the following figure.

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Analysis Report Peak Calling Properties	×
▶ Mode	
Analysis Properties	
Default DNA Analysis V Save as	
Parameters	
Reference Marker	
No Marker Reference Marker Table	
Marker Table	
	•
1 Sample(s) selected Start Analysis	

Note: If loaded samples have already been analyzed, the analysis parameters from the selected sample can be transferred to the **Analysis** parameters panel. To do this, right-click the sample in the **Experiment Explorer** and select the menu item **Transfer analysis parameters** from the context menu, as shown in the following figure.



Note: The alignment marker of a plate can be changed. To change the alignment marker, select the menu item **Overwrite alignment marker** from the context menu in the **Experiment Explorer** (the mouse cursor should be over the plate name).



The **Overwrite Marker** dialog appears. Select the alignment marker from the drop-down list and confirm your choice by clicking **OK**.

- 5. Start the analysis by clicking the Start Analysis button.
- 6. View the analysis results. For each analyzed sample, the results are shown in tabular form at the bottom of the **Analysis** environment in the **Peak Result** tab (see <u>Result table</u> section) and in the plot of the Single Electropherogram view (see <u>Electropherogram view</u>).

Note: In the case of a smear or gDNA analysis, the **Smear Result** tab at the bottom of the **Analysis** environment shows the calculated properties that are specific to smear peaks. For more details, see the <u>Result table</u> section.

7. Make sure that the right peaks were detected. It is important that the correct peaks are marked as alignment marker peaks (marked with a green '+') because this is the basis for all other analysis functions. If not, <u>adapt the analysis parameters</u> and repeat steps 5 to 7.

Refer to <u>Analysis of DNA samples</u> and <u>Analysis of RNA samples</u> for getting guidance as to which **Analysis** profile should be selected initially and how to use the analysis options for your specific samples.

As a last option, use the **Insert/Delete Peak** options described in the <u>Manually modifying analysis results</u> section.

8. <u>Save the analysis results</u> by clicking the Save button in the Experiment Explorer.

It is possible to automate the analysis (peak detection) during the data acquisition process. To do so, save any changes in the analysis parameters into a <u>customized analysis profile</u>. Make sure that these analysis parameters work for all samples. Then <u>create a Process Profile</u>. In the <u>Process Profile</u> step, select the option **Analysis** in the **Included steps** section. In the <u>Analysis</u> step, select the **customized analysis profile**. Save the Process Profile with these settings for re-use.

6.4.4.2 Modifying an analysis profile

To modify an analysis profile, proceed as follows:

1. Open the Analysis tab at the right side of the Analysis environment.

If it is not visible, you can display it using the **View** menu (selecting the menu item **View/Show Analysis Parameters**) or by clicking the ^[] icon at the far right of the view selection bar:

Gel Image Electropherogram Peak Calling Marker

- 2. Select the analysis profile to be modified in the Profile drop-down list in the Analysis Properties panel.
- 3. Change the analysis parameters as described below.
- 4. To save the modified profile, click the Save as button at the right of the Profile drop-down list.

Note: The parameters of an analysis profile can only be modified by Administrator or Developer users.

Parameters can be changed by:

- Clicking the parameter.
- Changing the value (and unit if possible).
- Clicking OK.

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▼ Paramete	ers		[]	▼ Paramete	ers	
Peak	🔿 Smear 🛛 () gDNA		Peak	⊖ Smear	⊖ gDNA
Smoothing Filter	Start 0,00 min	Window 15 pts		Smoothing Filter	Start 0,00 min	Window 15 pts
Baseline Filter	Start 0,00 min	Window 40 sec		Baseline Filter	Start 0,00 min	Window 40 sec
Minimum Distance	Start 0,00 min	Value 0,25 sec		Minimum Distance	Start 0,00 min	Value 0,25 sec
Suspend Integration	Start 0,00 min	End 0,50 min		Suspend Integration	Start 0,00 min	End 0,50 min
Threshold	Start 0,00 min 2,00 min	Value 2,50 S/N (1) 3,00 S/N		Threshold	Start 0,00 min 2,00 min	Value 2,50 S/N 4,00 S/N
Alignment Marker Threshold	0 S/N			Alignment Marker Threshold	0 s	\$/N
	(Add Delete				Add Delete
Define Parameter Threshold V Start Value 2 2 min 4 S/N				Define Parameter Threshold Start Value 2 min 4 S/N		
	(OK Cancel				OK Cancel

For example, to raise the threshold parameter to 3 S/N starting from 2 minutes, perform the following:

- 1. Click the Add (1) button below the parameters.
- 2. Select the parameter type Threshold (2).
- 3. Adjust the starting time to 2 minutes (3), the threshold value 3 (4), and its unit to S/N (5).
- 4. Accept the changes by clicking **OK**.



Each parameter displayed in the figure above has a corresponding time point which can be adjusted. The only exception is the **Alignment Marker Threshold**.

Note: The time of the initial parameter at 0.0 minutes cannot be changed. The exception is the Suspend Integration parameter. It is off by default but can be activated to ignore a certain time range during peak detection. This range does not necessarily have to start at 0.0 minutes.

The descriptions of all parameters and further instructions are summarized in the table on the next page. In addition, refer to sections <u>Standard DNA analysis</u>, <u>Fast Analysis DNA analysis</u>, <u>Smear DNA analysis</u>, <u>gDNA analysis</u>, and <u>Basic RNA analysis</u> for more information on which parameters are best suited for your samples. More information about the analysis algorithms in QIAxcel ScreenGel and the corresponding parameters can also be found in <u>Appendix D</u>.

Parameter	Description	Adjusting the value		
Peak	This parameter sets establishes the given profile as for sharp peaks.	Select this option if the profile has to be used for <u>Standard</u> <u>DNA analysis</u> , <u>Fast Analysis</u> <u>DNA analysis</u> or <u>Basic RNA</u> <u>analysis</u> .		
Smear	This parameter sets establishes the given profile as a smear profile .	Select this option if the profile has to be used for <u>Smear DNA</u> <u>analysis</u> of DNA libraries or for analysis of RNA FFPE samples.		
gDNA	For DNA mode only. This parameter sets establishes the given profile as a gDNA profile.	Select this option if the profile has to be used for <u>gDNA</u> <u>analysis</u> .		
Baseline Filter	This parameter governs the baseline detection (shown as a red line in electropherograms).	The default value for the Window parameter should be sufficient for most samples.		
	Note: In case of a smear and gDNA analysis, the software detects the baseline automatically and no parameterization by the user is needed. Thus, for smear and gDNA analysis profiles no baseline parameter is shown.	If the baseline starts to follow the course of the data, increase this value. Generally, the value should be set to twice the width of a single peak, but it must not be larger than half of the total migration time.		
		If peak widths differ a lot over time, use distinct values for the Window parameter in different time intervals. For an example, see the "Default RNA QC" analysis parameter settings and <u>Analysis of RNA Samples</u>		

Threshold

The threshold parameter is used during peak detection. Signals that exceed the baseline by the threshold value are detected as peaks. The threshold can be given in RFU, as percentage of the highest signal in the sample, or as a multiple of the (estimated) noise level of the electropherogram.

Note: The software uses the threshold for detection of alignment marker peaks whenever no higher alignment marker threshold is set.

The default setting is a good starting point for most samples. The threshold should be lowered if relevant peaks are below the threshold and not recognized as peaks.

You can specify the threshold using different units. Use the RFU unit to set the threshold to a fixed RFU value. Use the % unit to specify the threshold as a percentage of the highest signal in the sample. Use the S/N unit to specify the threshold as a multiple of the noise level, which is estimated from the sample data.

Note: For smear and gDNA analysis, a higher S/N value is required as threshold.

Note: The threshold parameter can be interactively changed in the Single Electropherogram view. Simply drag the threshold (displayed in blue) with the mouse. When doing so, the sample is re-analyzed using the new threshold (all other parameters are taken from the sample properties).

The default setting is suitable for most samples.

For very noisy data with fronting or tailing increase the value in order to prevent the detection of noise peaks at cluster borders, increase the value.

Minimum Distance

The minimum distance parameter is used during the plausibility check of peaks. It defines the minimum distance two peak clusters must have to be detected as two distinct peaks.

In noisy data with heavy tailing or fronting, noise spikes can cross the minimum peak threshold several times at the border of peaks. Detection of these noise peaks is prevented by this parameter.
Suspend Integration The suspend integration parameter turns off peak detection for a defined period of time. The affected areas are marked with a grey bar in the Single Electropherogram view as well as in the Electropherogram overview. Peaks that overlap with this bar are ignored by the analysis. In the default setting, **Suspend** Integration is turned off, meaning that peak detection is performed on the entire duration of migration time. The time interval for **Suspend** Integration can be specified via absolute or relative migration time. In case the relative time unit is used, alignment marker peaks are not affected by **Suspend** Integration.

Note: The suspend integration interval is shaded gray in the Electropherogram single view and overview.

Note: If relative time is chosen to describe the suspend integration interval, the following constraints apply: in case of 2 alignment marker peaks, all relative times must be between 0 and 1. In case of 1 alignment marker peak, all relative times must be above 1.

The default window width is a good setting for most samples. If two close peaks cannot be resolved, the smoothing filter parameter can be lowered or disabled (i.e. set to 0 pts) to obtain the optimal resolution. One example for disabled smoothing is the **DNA Fast Analysis** profile.

Note: A different smoothing algorithm is applied in RNA mode and for DNA smear analysis or gDNA analysis (Appendix D). The window length parameter of this algorithm is given in seconds.

Smoothing Filter

The smoothing filter parameter defines the smoothing window width, i.e., the strength of the smoothing. The smoothing filter improves the signal-to-noise ratio (S/N) of the data but lowers the resolution.

Alignment Marker Threshold This parameter allows to set a higher threshold for alignment marker peaks. It is useful to avoid that low intensity peaks are detected as alignment marker peaks, e.g. primer dimers before the lower alignment marker peak or noise peaks on the tail of the upper alignment marker. A value between 5 and 10 should be suitable for most samples. If the alignment marker peaks have a lower S/N value, you can lower this threshold, or disable it by setting it to a value below the Threshold parameter.

Note: For smear and gDNA analysis, a higher S/N value is required as alignment marker threshold.

6.4.4.3 Creating a new analysis profile

To create a new analysis profile:

- 1. Open the Analysis tab in the toolbar on the right-hand side of the Analysis environment.
- Select an existing analysis profile from the **Profile** drop-down list. The selected profile serves as a template for the creation of the new profile, or select **NewAnalysisProfile** to start with default parameters.
- 3. Change the analysis parameters according to your needs as described in the <u>Modifying an analysis</u> <u>profile</u> section.
- 4. Store the new profile by clicking the Save as button to the right of the Profile drop-down list.
- 5. Assign a unique name to the profile and click OK.

Note: Only users with user role Developer or Administrator can create new analysis profiles.

6.4.5 Size and concentration determination

The sizes and concentrations of detected peaks in a sample are calculated by referencing them to a sample containing fragments of known size and concentration. The figures below show how a so-called reference marker (middle picture) is used to derive the sizes and concentrations (upper and lower picture). A reference marker can be created in two ways: either a size marker sample is run side by side with the samples that are being analyzed; or a previously saved reference marker is selected in the **Reference Marker** section in the analysis tab. For details, see <u>Creating a reference marker</u>.



Each detected peak is mapped to a corresponding reference marker peak. For peaks without unique corresponding reference marker peaks, the adjacent reference marker peaks are taken as reference (e.g. peaks with known size 310 bp and 603 bp, respectively).



The sizes for the newly detected peaks are either directly transferred from the matching reference marker peaks (15 bp peak) or interpolated using the two adjacent reference marker peaks (562 bp peak interpolated from 310 bp and 603 bp).



Similarly, the concentrations of detected analytes are referred from the known concentrations underlying the corresponding reference marker peaks.

6.4.5.1 Size and concentration determination procedure

To determine size and concentration, proceed as follows:

- 1. Load the samples you want to analyze using the **Experiment Explorer**. For details, refer to the <u>Loading</u> <u>sample data</u> section.
- 2. In the **Analysis** parameters panel at the right side of the **Analysis** environment, define the parameters to use for analysis.

It is possible to <u>re-use</u> the analysis parameters and the reference marker table of an already analyzed sample. This can be helpful if samples have already been analyzed during the data acquisition process, but analysis shall be reviewed or repeated. To re-use parameters, right-click one of the samples in the **Experiment Explorer** and select the **Transfer Analysis Instruction**. The analysis parameters and the reference marker table, if present, from the sample will be transferred to the analysis parameters tab.

Otherwise, select a predefined analysis profile in the **Analysis** properties panel.

Refer to <u>Analysis of DNA samples</u> and <u>Analysis of RNA samples</u> for getting guidance as to which **Analysis** profile should be selected initially and how to use the analysis options for your specific samples.

To modify or create an analysis profile, refer to sections <u>Modifying an analysis profile</u> and <u>Creating a new</u> <u>analysis profile</u>, respectively.

- 3. If the size marker was run side by side with samples, create a new reference marker as described in <u>Creating a reference marker</u>. If samples were to be analysed during data acquisition but either an error message was displayed or the sample results are not as expected, use this procedure to check the created reference marker step by step and recreate the reference marker, if needed. If the reference marker needs to be re-created, all other samples must also be re-analyzed as described in the next steps, using this new reference marker in step 6.
- 4. Visualize the samples you want to analyze. See the <u>Viewing sample data</u> section for details on data inspection.
- 5. Select the samples for analysis.
- 6. Define the reference marker to be used in the **Reference Marker** section of the **Analysis** panel. If you just created or recreated a reference marker in step 2, it is preselected for use. Proceed with step 7. Otherwise, check **Reference Marker Table** and select a previously saved reference marker. Details of the reference marker are shown below the combo-box.

Analysis	Report	Peak Calling	Properties	×				
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▼ Analy	sis Prope	rties						
Default I	DNA Analy	sis V	Save as					
Param	ieters							
▼ Refere	ence Mark	er						
_ O № M	arker							
2 Refer	ence Mark	ker Table	3					
*C1006	07A99_20	10-07-16_13-34	-3. 🔍 🌒					
Marke	r Table							
12 Sample	(s) selecte	ed Sta	rt Analysis 📐	Ť				
			1					

Note: To ensure compatibility, the reference marker drop-down list contains only reference marker tables that match the alignment marker of the selected samples. The drop-down list is marked as invalid and empty if there is no matching reference marker table or no sample has been selected for analysis. In this case, select a sample first. If the drop-down list is still marked as invalid, check the sample alignment marker and correct it if required (refer to <u>Checking alignment marker</u>), or create a new reference marker (refer to <u>Creating a reference marker</u>).

Note: If no sample is selected for analysis, the reference marker drop-down list is empty. Select the samples to analyze before selecting the reference marker. For details, see <u>Selecting samples for analysis or report</u>.

Note: See the table below for an explanation of the color icons next to the **Reference Marker Table** dropdown list:

		The selected reference marker table is fully compatible.
•		The size marker used for the reference marker table was processed within 90 days (DNA)/60 days (RNA) as the sample, and with the same cartridge and method as the sample. For analysis, it is not necessary to run the size marker first. The user can analyze older samples with a newer reference marker as well.
	•	The size marker used for the reference marker table was processed more than 90 days (DNA)/60 days (RNA) earlier or later than the sample, with the same cartridge and method as the sample.
	•	The reference marker table was processed with the same method as the sample, but with another cartridge of the same cartridge type.
		The reference marker table is not compatible with the sample. This can happen if no reference marker matches the cartridge type, method and alignment marker of the sample.
•	It is also shown when the software cannot check the compatibility because no sample has been selected for analysis.	

Important: Selecting the correct size marker will increase the accuracy of the size and concentration determination. Use a marker that contains DNA fragments closest to the size of the DNA fragments of interest. The DNA fragments to be analyzed must fall within the smallest and largest fragment size of the size marker. In addition, the range of the alignment marker must cover the range of the size marker.

- 7. Start the analysis by clicking the Start Analysis button at the bottom of the Analysis Parameters panel.
- 8. View the analysis results. For each analyzed sample, the results are shown in tabular form at the bottom of the Analysis environment in the Peak Result tab. In the case of a smear or gDNA analysis, the Smear/gDNA Result tab shows the calculated properties that are specific to smear peaks. For more details, see the Result table section. The Single Electropherogram view is most suitable for checking peak detection. (see Electropherogram view).

Note: If the samples had been analyzed before with **Peak Calling** or **Distribution** analysis, these results will be automatically updated as well, based on the new calculations for peaks/smear peaks.

In case the sample was analyzed earlier with <u>distribution analysis</u>, the Single Electropherogram view shows smear peaks corresponding to the areas of interest as defined in the <u>distribution profile</u>. In order to see a plot of the peak detection, the distribution analysis needs to be removed temporarily. To do so, right-click the sample in the **Experiment Explorer** to the right and select **Transfer Distribution Profile** to copy the parameters to the **Distribution** panel to the right for later re-use. Then right-click the sample again and select **Remove Distribution Analysis Data** to remove the results of distribution analysis from the sample. The Single Electropherogram View now shows the result of peak detection. It shows the alignment marker peaks (marked green at the apex) and the smear peaks according to the **Smear/gDNA Result** table.

9. Make sure that the right peaks were detected. First of all, it is important that the correct peaks are marked as alignment marker peaks (marked with a green '+') because this is the basis for all other analysis functions. If not, <u>adapt the analysis parameters</u> and repeat the steps 5 to 9.

Refer to <u>Analysis of DNA samples</u> and <u>Analysis of RNA samples</u> for getting guidance as to which **Analysis** profile should be selected initially and how to use the analysis options for your specific samples.

As a last option, use the **Insert/Delete Peak** options as described in the <u>Manually modifying analysis results</u> section.

- In case the distribution analysis needs to be removed temporarily, it has to be repeated manually by using the distribution profile parameters that had been transferred before. Refer to <u>Distribution analysis</u> for instructions.
- 11. Save the analysis results by clicking the Save button in the Experiment Explorer.

It is possible to automate the peak detection and size and concentration determination during the data acquisition process. To do so, save any changes in the analysis parameters into a customized analysis profile. Make sure that these analysis parameters work for all samples and also for the size marker sample. Then <u>create a Process Profile</u>. In the <u>Process Profile</u> step, select the option **Analysis** in the **Included** steps section. In the <u>Analysis</u> step, select the customized analysis profile. If the size marker is to be run side by side with samples, define the size marker position for a run in the right part of the screen in the <u>Run</u> <u>Parameters</u> step, select the option **Run size marker** side by side with sample in the <u>Marker</u> step, and select the alignment marker and the size marker. If instead, a previously created reference marker shall be re-used for several runs, select the option **Reference Marker Table** and select the previously created reference marker shall be re-used potentials, refer to <u>Process Profile</u> options. Save the Process Profile with these settings for re-use. During each run setup, ensure to select the alignment marker in the **Sample Selection** step.

6.4.5.2 Creating a reference marker

To create a new reference marker, a size marker sample with annotated peak times and areas is required. The new reference marker is defined by matching the peaks of the size marker sample to a size marker table (including alignment marker).

Please follow the steps below:

1. In the Experiment Explorer, select the sample that contains the size marker. If it is not marked as a size

marker (or shown as: or), right-click and select the **Size Marker** option from the context menu. The icon chanaes to or .



2. Open the size marker sample in Single Electropherogram view by clicking the Electropherogram tab. If

the size marker sample is already analyzed (or shown as 💻), proceed with step 6.

3. Select a profile in the Analysis Properties toolbar.

Refer to <u>Analysis of DNA samples</u> and <u>Analysis of RNA samples</u> for getting guidance as to which **Analysis** profile should be selected initially and how to use the analysis options for your specific samples.

Note: You can <u>re-use</u> the analysis parameters of an analyzed size marker sample. This can be helpful if the sample has already been analyzed during the data collection process and now the analysis is to be checked or repeated. To do this, right-click the analyzed sample in the Experiment Explorer and select **Transfer Analysis Instruction**. The analysis parameters and the reference marker table, if present, from the sample will then be transferred to the **Analysis Parameters** tab on the right.

- 4. In the Reference Marker box on the Analysis tab, select the No Marker option.
- Start the analysis by clicking the Start Analysis button.
 Note: The result of the analysis is a sample with annotated peak times and areas.
- 6. Make sure that the right peaks were detected. If not, <u>adapt the analysis parameters</u> and re-analyze the size marker sample.

Refer to <u>Analysis of DNA samples</u> and <u>Analysis of RNA samples</u> for getting guidance as to which **Analysis** profile should be selected initially and how to use the analysis options for your specific samples.

As a last option, use the **Insert/Delete Peak** options described in the <u>Manually modifying analysis results</u> section.

- 7. Switch to the **Marker** screen by clicking the **Marker** tab on the top of the middle view.
 - The peak table (with **Rel. Time and NA** columns) of the size marker sample is displayed at the left side of the screen.

Note: The panel is empty if no size marker sample is selected in the **Experiment Explorer**, or if the selected size marker sample is not analyzed. In this case, repeat the steps above first.



Note: Depending on cartridge and method, a size marker results in a different gel trace. As the trace for the size marker (right-hand side) is calculated based on the size marker table, the image may differ from the gel trace from the actual run (left-hand side).

8. In the **Marker** screen, check the Alignment Marker (1) and select the Size Marker (2) used in the size marker sample.

A comparison of the actual sample gel view (left) and the theoretical reference marker gel view (right) is displayed in the middle of the screen.

Note: If the alignment marker is not correct, change it as described in Checking alignment marker.

Note: Creating new size markers is described in <u>Creating a new size marker table</u>.

9. If the number of peaks in the size marker sample on the left matches the number of peaks in the reference marker table on the left (one or two alignment marker peaks plus the size marker peaks), the selected reference marker can be used for further sample analysis. Click **Apply** button (3) at the top of the **Marker** screen to copy the created reference marker to the bottom of the **Marker** panel on the **Analysis** parameters tab.

Optionally, save the reference marker using the **Save as** button at the top of the screen if you want to use the reference marker table for the analysis of further experiments.

Note: The **Apply** and **Save as** buttons are disabled and a warning is displayed at the top of the screen if the reference marker is invalid, e.g. if the number of peaks on the left and on the right sides does not match.

If more peaks than expected were detected in the size marker sample, you can delete them using the **Delete** button below the size marker samples peak table (with **Rel. Time** and **NA**).

Alternatively, switch to the Single Electropherogram view and check whether correct peaks were detected. Use the **Insert/Delete Peak** options described in the <u>Manually modifying analysis results</u> section or change the analysis parameters and re-analyze the size marker sample.

It is possible to automate the creation of the reference marker during the data acquisition process, if the size marker is to be run side by side with the samples. To do this, save all changes to the analysis parameters in a customized analysis profile. Make sure that these analysis parameters also work for the samples.

Then <u>create a Process Profile</u> defining the size marker position for a run in the <u>Run Parameters</u> step and selecting the customized analysis profile in the <u>Analysis</u> step. Save the Process Profile with these settings for re-use.

To continue sample analysis, refer to step 3 of the Size and Concentration determination procedure.

6.4.5.3 Creating a new size marker table

To use a custom size marker, a new size marker table has to be created manually.

To create a new size marker table, proceed as follows:

1. In the Experiment Explorer, select the sample that contains the size marker. If it is not marked as a size

marker (or shown as: or), right-click and select the **Size Marker** option from the context menu that appears. The icon changes to or .

2. Open the Marker screen of the Analysis environment (1).

M500 Alignment Marker M500 QX 15 bp-5 kb Size Marker Save size marker as 201022006 Total Concentration 0 ng/µl Relative Time NA 1,0000 0,010389 0,7206 0,026942 0 0 0,0000 0,007546 Delete 3	larker Creation				
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0,0000 0,007546	0,7206	0,02694		0	
Delete	0,0000	0,00754	*	15	
		Delete			3 Add Dele

3. To create an empty size marker table, select **NewSizeMarker** from the drop-down list (2).

Note: For DNA High-Sensitivity and RNA High-Sensitivity cartridges, it is not possible to create a new size marker. Only a change of total concentration for existing marker is possible.

- 4. Click the Add button (3) below the table to add a new row to the table.
- 5. Define the size and optionally the concentration (4).
- 6. Click **OK** to save the changes to the table (5). Repeat the steps from step 4 to define all rows.
- 7. Save the new size marker by clicking the **Save Size Marker as...** button below the size marker drop-down list (**6**).

Note: A new size marker can only be created by users with the user role Administrator or Developer.

Note: If a sample is selected which is flagged as a size marker, a warning about mismatch of the number of rows be displayed. Ignore the warning and proceed by adding fragment sizes to the size marker table. If you entered all size marker fragments and the warning persists, switch to Single Electropherogram view and check that all peaks have been detected correctly.

6.4.6 Peak Calling

Based on a peak table, which is the result of the sample analysis, a peak calling step can be performed.

Peak calling shows whether corresponding peaks are present in analyzed samples (e.g. because of the same size), and provides peak data for comparison.

To trigger peak calling, the definition of the peaks of interest must be provided by the user first. For this, refer to the <u>Creating a new peak calling instruction</u> section.

To perform a peak calling, proceed as follows:

- 1. Load and activate the experiment for which sample peak calling should be performed.
- If not yet analyzed, analyze the samples of interest as described e.g. in <u>Standard DNA analysis</u> or <u>Basic RNA analysis</u>.

Note: The samples must be analyzed before peak calling.

- 3. Select the visualized samples that the peak calling instruction shall be applied to.
- 4. Open the Peak Calling parameter tab located on the right side of the Analysis environment.

Note: The **Peak Calling** tab is only available if peak calling features are active. Refer to <u>Activating peak</u> <u>calling features</u> for information on how to activate peak calling features.

- 5. Select the peak calling instruction from the **Peak Calling** Instruction drop-down box (1), which contains the set of peaks to search for.
- 6. Start peak calling by clicking the Start Peak Calling button (2) at the bottom of the Peak Calling tab. The peak calling instruction is assigned to the selected samples and the Peak Calling screen in the middle of the Analysis environment is filled with the peak calling result.

Note: The next time you click **Start Peak Calling** with the same unchanged peak calling instruction, the previous peak calling result for that instruction is completely replaced by the new result. If you would like to apply the same peak calling instruction to different data and not overwrite the first set of results, save the instructions under different names, and then apply them subsequently to the different sets of samples.

7. Switch to the **Peak Calling** screen in the middle of the **Analysis** environment to view the result. The structure of the result is described below.

Gel Image	Electropherogram	Peak Calling	Marker							Analysis	Report	t Peak C	alling	Properties	>
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	e size marker samples	KINA Tat_IIIouse_II	unnang V T V F	otal Conc RNA Integr	entration ("Total Conc.") ity Score ("RIS")					Peak Call *my 1 R	ing Instruc NA rat_mo	tion buse_huma		Save as	
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18S 28S	Name	PositionTol1869 nt74700 nt7	.[%] (,00 (,00 (,00 Ratio	Ratio Norm	nalized Area ("Ratio")					□ In O Fi ● Fi	clude size nd centere nd highest Name	marker san ed peak in ir peak in inte	nples nterval erval Position	Tol. [%]	
										185			1869 nt 4700 nt	7,00	
CF_Jurkat	t_RNeasy_Verd_05121	12 R1 E1				1	18S	2	8S	203			Add	Delete	
Pos.	Sam	ple Info	R	IS Ratio	Total RNA Conc. [ng/µl]	Found	Size [nt]	Found	Size [nt]						
A01 Jurk	at_RNeasy_486_1_1		1	0,0 1,9	574,65	Yes	1799	Yes	4516	Name					
A02 Jurk	at_RNeasy_486_1_2		1	0,0 1,9	3 597,82	Yes	1761	Yes	4422	18S					
A03 Jurk	at_RNeasy_400_1_3		1	0,0 1,8	653,86	Yes	1757	Yes	4408	Positio	n		Т	lerance	
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A05 Jurk	at_RNeasy_300_1_5		1	0,0 1,8	506,93	Yes	1834	Yes	4554						
A06 Jurk	at_RNeasy_300_1_6		1	0,0 1,9	521,66	Yes	1829	Yes	4538			ſ	OK	Cancel	
A07 Jurk	at_RNeasy_200_1_7		1	0,0 1,9	319,70	Yes	1869	Yes	4598]
A08 Jurk	at_RNeasy_200_1_8		1	0,0 2,0	317,80	Yes	1881	Yes	4633	▼ Cal	culated C	olumns			1
A09 Jurk	at_RNeasy_100_1_9			8,9 1,9	113,44	Yes	1931	Yes	4841						
A10 Jurk	at_RNeasy_100_1_10			9,5 2,0	132,10	Yes	1937	Yes	4856	Tot	al Concen	itration ("To	tal Conc.')	
A11 Jurk	at_RNeasy_50_1_11			8,0 1,6	67,48	Yes	1945	Yes	4991	N RN	A integrity	Score ("RI	5")		
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										Rauo	18S	•			
<									Þ	11 Samp	le(s) sele	cted	Start Pe	ak Calling	(2

8. To save the result of peak calling, save the experiment by clicking the **Save** button in the **Experiment Explorer**.

Peak calling result

Every sample can be analyzed individually (using peak calling). Therefore, the overview of peak calling results is grouped according to the respective peak calling instruction and named accordingly (see figure above). The result of the peak calling instructions can be collapsed individually. It is structured as follows: at the top the table peak calling instruction, followed by a list of the samples to which the peak calling instruction was applied. The list of samples is then grouped by the plates to which the samples belonged.

Each row in the table corresponds to a sample; the columns **Pos.** and **Sample Info** identify the sample. Please note the **Pos** column is fixed. All following columns are defined by the peak calling instruction. Aggregated columns over all peaks of interest appear when they are selected in the **Calculated Columns** panel. The following columns are grouped by the names of the peaks of interest and show several peak properties. To change the properties listed for each peak of interest, right-click on the table header and select/unselect the property using the **Show Column** option. The change will take effect in the entire table.

Note: If a sample was not analyzed, it is not present in the peak calling result overview.

Parts of the peak calling result table can be copied to the clipboard. Select the cells to be copied and then press **Ctrl+C**. To copy the complete result of a peak calling instruction, right-click in the peak calling result table and select **Copy Peak Calling Result of** [...].

Removing a peak calling result

To remove the result of peak calling, right-click the sample in the Experiment Explorer and select **Remove Peak Calling Data** from the context menu that appears, as shown in the figure below. This menu item is only active if peak calling has been performed for the sample.



It is possible to automate peak calling during the data acquisition process. Ensure you have already automated the <u>Size and concentration determination procedure</u> for your samples using <u>Standard DNA</u> analysis, <u>Fast Analysis DNA analysis</u> or <u>Basic RNA analysis</u>. To automate peak calling, save any changes in the peak calling instructions into a customized peak calling instruction. Make sure that these parameters work for all samples. Then <u>modify the Process Profile</u> you have created to automate the size and concentration procedure for your samples. In the <u>Process Profile</u> step, select the option **Peak Calling** in the **Included** steps section additionally. If distribution analysis features are still active, activate peak calling features now. See the section <u>Activating distribution analysis features</u>. In the <u>Peak Calling</u> step, select the customized peak calling instruction. For further details, refer to <u>Process Profile</u> options. Save the Process Profile with these settings for reuse. During each run setup, ensure selecting the alignment marker in the **Sample Selection** step.

6.4.6.1 Activating peak calling features

Either peak calling or distribution analysis features can be applied to samples of an active experiment.

To switch to the peak calling features if distribution anaylsis is active, proceed as follows for the current experiment:

- 1. Open the **Distribution** tab located on the right side of the **Analysis** environment.
- 2. Click Switch to Peak Calling.



The peak calling result tables replace the distribution result tables and the peak calling parameter panel replaces the distribution parameter panel. The software keeps this setting until an experiment that used distribution analysis is activated or opened.

Note: It is not possible to activate peak calling features if a distribution analysis has already been performed in the current experiment. In this case, <u>remove the distribution analysis data</u> first.

6.4.6.2 Modifying a peak calling instruction

Peak calling instructions contain a set of parameters, defining the peaks to search for (peaks of interest).

Note: Peak calling instructions can only be modified by users with the user role Administrator or Developer.

To modify a peak calling instruction proceed as follows:

- 1. Open the Peak Calling tab at the right of the Analysis environment.
- 2. Select the peak calling instruction to be modified from the Peak Calling Instruction drop-down box (1).
- 3. Change the definitions for the peaks of interest (2) according to your needs as described below (3).

Analysis	Report	Peak C	alling	Properties	×			
Switch to Distribution								
Peak Callin	g Instructio	n	(1)		_			
b3_peaks			▼[Save as				
🔻 Peak	s of Intere	st						
	ude size ma	arker san	nnles					
		anter san	iipico					
Fine	d centered p	beak in ir	nterval					
O Fine	d highest pe	ak in inte	erval					
	Name		Position	Tol. [%]				
15			15 n	t 10,00				
560			560 n	t 10,00				
603 (2		603 n	t 10,00				
peak 50	00		5000 n	t 10,00				
			Add	Delete				
Name								
603								
Position 60	Position 3 Tolerance 603 Size ▼ 10 %							
			OK	Cancel				

Note: The name in the **Peak Calling Instruction** drop-down box will appear with an asterisk to the left, indicating that the table has been changed.

4. Save the modified peak calling instruction by clicking the **Save as** button to the right of the **Peak Calling Instruction** drop-down box.

Peak calling configuration:

Include size marker samples	If this option is chosen, peak calling can be performed on the size marker sample too. Otherwise, peak calling will be skipped for size marker samples.
Find centered / highest peak in interval	If more than one peak is found within the tolerance interval, this setting determines whether the centered peak (nearest to the sought position) or the highest peak is chosen.

To add a peak of interest:

- 1. Click the Add button. A new row with empty fields will appear at the bottom of the table.
- 2. Specify the peak of interest in the edit area below.

Name	Specifies the name of the peak of interest. This will be the column header in the peak calling result table.
	Note: The names of the peaks must be different.
Position	Specifies the position of the peak to search for and the unit of the position (Size, Rel. Time, or Time).
	Note: The positions of the peaks must be different.
	Note: If Rel. Time is chosen to describe the position, the following constraints apply: In case of two alignment marker peaks, all relative times must be between 0 and 1. In case of one alignment marker peak, all relative times must be above 1.
Tolerance	Tolerance for peak searching. In a sample the peak will be considered as detected if its position difference is less than the given tolerance from the defined position.

3. Click **OK** to add the new peak of interest to the table.

Note: The peaks of interest are sorted by position in the peak calling instruction table.

To delete a peak of interest:

- 1. Select the peaks row in the table.
- 2. Click the **Delete** button.

To modify a peak of interest:

- 1. Select the row of the peak of interest to be modified.
- 2. Edit the values of the peak of interest in the edit area below.
- 3. Click OK. The values in the selected row are changed.

Note: The peaks of interest are sorted by position in the peak calling instruction table.

Calculated columns:

In addition to finding the specified peaks, depending on the run mode, the following parameters can be calculated during peak calling:

Total Concentration	For RNA mode only. The total concentration of the RNA over the whole sample.
RNA Integrity Score	For RNA mode only. RNA integrity score (RIS) is a value from 0 to 10, where a value of 10 indicates completely intact RNA. This is especially useful for <u>RNA QC</u> .
	Note: The value is not calculated if the 18S peak was not found.
Ratio Normalized Area	Choose two peaks of interest. If both are found in the sample, the ratio of their normalized areas is calculated. This is especially useful for <u>RNA QC</u> using the 28S and 18S peaks.
Relative Abundance	For each peak of interest, the normalized area relative to the highest normalized area of corresponding peaks over all samples is calculated. (For the sample in which the normalized area of the peak is highest, the relative abundance is 100%. For all other samples, it is the percental fraction of its normalized area relative to the highest value.)

6.4.6.3 Creating a new peak calling instruction

To create a new peak calling instruction:

- 1. Open the Peak Calling tab at the right of the Analysis environment.
- Select a peak calling instruction from the Peak Calling Instruction drop-down box. The selected peak
 calling instruction serves as a template for the creation of the new one. Select NewPeakCallingInstruction
 to start with an empty table.
- 3. Specify the peaks of interest according to your needs as described in the <u>Modifying a peak definition</u> <u>table</u> section.
- 4. Save the new peak calling instruction by clicking the **Save as** button to the right of the **Peak Calling Instruction** drop-down box. Enter a new unique peak calling instruction name and click **OK**.

6.4.6.4 Peak pattern matching

Based on a peak calling step, a peak pattern matching step can be performed. Peak pattern matching shows whether certain patterns of peaks (determined in the peak calling step) are present in analyzed samples.

In order to perform peak pattern matching, proceed as follows:

- 1. Complete steps 1 to 5 described in the Peak Calling section.
- 2. In the Peak Pattern Matching panel, click the Add button to add a new pattern.
- Enter a name for the new pattern and define it. The pattern is defined using the checkboxes that are displayed based on the current peak calling instruction. Check the box for each peak to be included in the pattern. Unchecked peaks will not be included in the pattern.
- 4. Click **OK** to confirm the pattern definition. By clicking **Cancel**, the definition step is canceled and no changes are made.

Note: You can define more than one pattern for a given peak pattern matching step. To add further patterns, repeat steps 2 to 4.

Note: To delete a pattern, click on it and press the Delete button.

5. Start peak calling and switch to the **Peak Calling** screen in the middle of the **Analysis** environment to view the results. For details see steps 6 and 7 of section <u>Peak Calling</u>.

The peak calling result will indicate which pattern matches to each sample. Results are listed in the samplespecific column **Pattern** of the peak calling result table. If the peak calling result of a sample does not match any defined pattern, "**n/a**" will be displayed in the column and the result will be highlighted in yellow.

For details about viewing the **Peak calling** result, refer to section <u>Peak Calling</u>.

Note: Peak pattern matching is an extension to the peak calling instruction and is saved along with the peak calling instruction. It will also appear in any reports, exports, or print outs of the peak calling instruction.

Gel Imag	ge Electropherogram Peak Calling Marker		Analysis Report Peak Calling Properties X
Pe:	ak Calling Result of Imy 1 RNA rat mouse human		Switch to Distribution
	and a second		Peak Calling Instruction
🗌 In	clude size marker samples	Total Concentration ("Total Conc.")	*my 1 RNA rat_mouse_human V Save as
O Fi	nd centered peak in interval	RNA Integrity Score ("RIS")	
() Fi	nd highest neak in interval	Ref. Peak 18S 🔻	Total Concentration ("Total Conc.")
	п. т., д. т. т. р. т.	Patio Normalized Area ("Datio")	RNA Integrity Score ("RIS")
	Name Position Tol. [%]		Ref. Peak 18S V
	1869 nt 7,00		A Ratio Normalized Area ("Ratio")
28S	4700 nt 7,00	18S V both + +	
			285 🔻
		Relative Abundance	Rauo
05.1			
CF_JU	rkat_RNeasy_verd_051212 R1 E1		Relative Abundance
		18S 28S	
Pos.	Sample Info	RIS Ratio Total RNA Conc. [ng/µI] Pattern Found Size [nt] Found Size [nt]	 Peak Pattern Matching
A01	Jurkat_RNeasy_486_1_1	n/a n/a 574,65 n/a No n/a Yes 4516	
A02	Jurkat_Riveasy_406_1_2	100 n/a 669.96 opt/ 199 Voc 1767 No n/a	
A03	Jurkat_RNeasy_400_1_5	10.0 n/a 583.43 only 185 Yes 1791 No n/a	2
A05	Jurkat RNeasy 300 1 5	10.0 1.89 506.93 both Yes 1834 Yes 4554	eN r
A06	Jurkat RNeasy 300 1 6	10.0 1,97 521,66 both Yes 1829 Yes 4538	a s s
A07	Jurkat_RNeasy_200_1_7	10,0 1,91 319,70 both Yes 1869 Yes 4598	5 4 L
A08	Jurkat_RNeasy_200_1_8	10,0 2,04 317,80 both Yes 1881 Yes 4633	both + +
A09	Jurkat_RNeasy_100_1_9	8,9 1,90 113,44 both Yes 1931 Yes 4841	bour + +
A10	Jurkat_RNeasy_100_1_10	9,5 2,03 132,10 both Yes 1937 Yes 4856	Add Delete
A11	Jurkat_RNeasy_50_1_11	8,0 1,63 67,48 both Yes 1945 Yes 4991	
			Pattern Name
			Peaks V
			OK Cancel
•		+	11 Sample(s) selected Start Peak Calling

6.4.7 Analysis of DNA samples

This section describes how to analyze a DNA sample. Instructions are given for four types of analyses: (1) standard analysis of DNA samples, (2) analysis of samples using fast analysis, (3) analysis of samples containing DNA libraries, and (4) analysis of samples containing genomic DNA. For all four samples types, perform steps 1–6 and follow the instructions corresponding to the relevant analysis type.

For detailed and up to date information on DNA cartridges, methods and analysis refer to QIAxcel DNA High-Sensitivity Kit Handbook for High Sensitivity cartridges and the QIAxcel DNA Handbook for all other DNA cartridges, available on www.qiagen.com/.

- 1. Select the DNA mode if this is not already selected. Check the current mode in the bottom right corner of the software screen. Change the mode if another mode than DNA is selected. For information how to change between modes refer to the <u>Mode</u> section.
- 2. Switch to the Analysis environment by selecting the Analysis icon in the main toolbar.
- 3. If the samples you want to analyze are not yet listed in the Experiment Explorer at the left, select the Load experiment icon from the Experiment Explorer's toolbar. If the samples are listed, but not active (grayed out), right click it's experiment name and select Activate. For further details, refer to the Loading sample data or Activating an experiment section.

4. For the **Fast Analysis** of samples, or if the samples to analyze contain DNA libraries, or to perform quality control on gDNA purified with silica-based technologies, or for standard analysis of DNA samples for which size and concentration shall be determined, follow the instructions from the <u>Size and concentration determination procedure</u>.

For information on which **Analysis** profile should be used as a starting point and which analysis parameters are required for your samples, refer to the respective sections: <u>Fast Analysis DNA analysis</u> for **Fast Analysis** cartridge, <u>Standard DNA analysis</u> for standard and high resolution analysis of DNA samples, <u>Smear DNA analysis</u> for samples that contain DNA libraries, and <u>gDNA analysis</u> for samples containing gDNA purified with silica-based technologies. Follow the <u>Size and concentration</u> <u>determination procedure</u> also if samples should have been analyzed during data acquisition process, but either an error message was shown or sample results are not as expected. In this case, use the procedure to check the analysis step by step and correct if needed. Follow the instructions to reuse analysis parameters that have been used for the samples.

If no size calculation shall be performed, follow the instructions of the Peak detection procedure.

- 5. Note: The available profiles depend on the cartridge and only relevant analysis profiles are shown.
- Optional: For the fast analysis of samples, standard analysis or high resolution analysis of DNA samples for which size and concentration have been determined, peak calling can be performed. Follow the instructions described in section <u>Peak Calling</u>.
- 7. For DNA libraries with specific distribution expectations or to asses the quality of a library in routine use, or to assess the quality of gDNA, perform <u>Distribution analysis</u>. Follow the instructions described in <u>Distribution analysis</u>. For information on which **Distribution** profile should be used as a starting point for gDNA samples, refer to section <u>gDNA analysis</u>.

The result table summarize all properties that the QIAxcel ScreenGel calculates for detected peaks. Rightclick the header line of the table, select **Show column**, and then select the property you are interested in. Alternatively, right-click the header line of the table and select **Show all columns**. This option displays all the properties calculated by QIAxcel ScreenGel. For more information, refer to section <u>Result table</u> and sections <u>Standard DNA analysis</u>, <u>Fast Analysis DNA analysis</u>, <u>Smear DNA analysis</u>, and <u>gDNA analysis</u>, respectively.

Note: It is possible to re-use the analysis parameters and the reference marker table of an already analyzed sample. This can be helpful if analysis shall be reviewed or repeated. To do so, right-click one of the samples in the **Experiment Explorer** and select **Transfer Analysis Instruction**. The analysis parameters and the reference marker table, if present, will be transferred to the analysis parameters tab.

6.4.7.1 Standard DNA analysis

During the <u>Size and concentration determination procedure</u> or the <u>Peak detection procedure</u>, select the Default DNA Analysis profile as a starting point. Use the DNA High-Sensitivity Analysis profile, for analysis using the DNA High-Sensitivity Cartridge.

Note: The available profiles depend on the cartridge and only relevant analysis profiles are shown.

Note: The QlAxcel ScreenGel software, version 2.0 and higher, comes with an improved default profile, named **Default DNA Analysis**. Here, to be independent from the highest signal in the sample, in the new profile, the threshold parameter is defined as a multiple of the noise level which is estimated from the sample data.

The <u>Single Electropherogram view</u> is most suitable for checking peak detection. Ensure that the plot shows a red "+" at the peak apex for all peaks between/after the alignment marker peak(s). If a pink "x" is shown instead, click the **Image Options** button of the Electropherogram toolbar and select the options **Mark peak apex** and **With Label**, with **Size** [bp] selected. Click **OK** to close the dialog.

The electropherogram plot of the analyzed samples will show the size of the detected DNA molecules in base pairs above the apex of the corresponding peaks. Peak properties are shown in the **Peak Result** table below the electropherogram. For details, refer to the Result table section. To switch the sample to be shown in the electropherogram, click on the next sample in the **Experiment Explorer** on the left.

Ensure that the peak detection has worked as expected. It is important that the correct peaks have been marked as alignment marker peaks (marked with a green '+'). This is the basis for all further analysis functions. If this is not the case, adapt parameters in the **Analysis** parameters panel on the right side of the **Analysis** environment. To do this, open the **Analysis Properties** section and then open the **Parameters** section. For details on how to change the parameters mentioned below, refer to section <u>Modifying an analysis profile</u>.

Note: Collapse and expand parts of the analysis properties by clicking ▼ and ▶, respectively.

Analysis R	eport P	eak Calling	Properties	×					
▶ Mode									
 Analysis 	 Analysis Properties 								
Default DNA Analysis									
 Parameter 	ers								
Peak	● Peak ○ Smear ○ gDNA								
Smoothing	Start	Win	dow						
Filter	0,00 mii	1	15 pts						
Baseline	Start	Win	dow						
Filter	0,00 mii	ו ו	40 sec						
Minimum	Start	Va	lue						
Distance	0,00 mii	1	2,00 sec						
Suspend Integration	Start	E	nd						
Threshold	Start	Va	lue						
	0,00 mil	1	5,00 S/N						
Alignment Marker Threshold	20	S/N							
		Add	Delete						
Define Parameter Select Parameter									
				•					
1 Sample(s) s	elected	Start	Analysis						

- Ensure that the **Peak** option is selected.
- If the raw data being analyzed is too noisy, increase the Smoothing Filter window. Click this parameter line and set the window size to, for example, 25 pts in the edit area below the parameters list and click OK to confirm. To apply time-dependent smoothing, add one or more smoothing filter parameter lines. To do so, click the Add button, select Smoothing Filter, and set the values for start time and window size. Click OK to confirm this setting.

- If peaks were detected before or after the alignment marker peak, increase the **Alignment marker threshold**. Click on the corresponding field and change the value.
- If peak detection is too sensitive, increase the **Threshold**. Click this parameter line and increase the value in the **Value** field in the edit area below the parameters list, and click **OK** to confirm.
- If in a certain range of the migration time, the peak detection is too sensitive, increase the **Threshold** value in this region. To do this, click the **Add** button below the parameters list and select **Threshold** from the **Define parameter** drop-down list. Set as a starting point, the time at which the region begins for which the threshold is to be increased. Here, define a threshold of, e.g., 7 S/N. Click the **OK** button to confirm this setting. Then, at the end of the affected region, reset the threshold to the default value of e.g., 5 S/N in Default DNA Analysis) by adding a third threshold definition. Set the start time of the last added threshold to the end of the affected region and set the threshold value to that of the first threshold parameter line. Click the **OK** button to confirm this setting.

Click the Start Analysis button again to start a new analysis according to this modified analysis profile.

In some cases, it may be necessary to repeat the adaptations to the analysis profile iteratively. If the peak detection worked as expected with the modified analysis profile, save this analysis profile and try to use this new profile instead of the **Default DNA Analysis** analysis profile for future DNA analyses of similar samples.

Note: Only users with user role **Developer** and **Administrator** can save modified parameters of an analysis profile.

6.4.7.2 Fast Analysis DNA analysis

This analysis profile was created to analyze runs with the **Fast Analysis** cartridge. The **Fast Analysis** cartridge uses a high voltage method for rapid analysis of PCR fragments and the **DNA Fast Analysis** profile can be used as a starting point. For detailed information on the **Fast Analysis** cartridge refer to QIAxcel DNA Handbook available on www.qiagen.com/.

For a run that already contains the size marker, follow the <u>Size and concentration determination procedure</u>. During the procedure, create and save a reference marker from the size marker sample.

If the run contains only the size marker, exit the procedure after saving the reference marker.

If the run also contains samples, continue with the procedure to analyze the sample and use the reference marker you just created and saved. For this purpose, activate the **Reference Marker Table** option in the **Reference Marker** section and select the saved reference marker.

For a run that only contains samples, follow the <u>Size and concentration determination procedure</u>, too. During the procedure, use the **DNA Fast Analysis** analysis profile as a starting point, and in the **Marker** section, activate the **Reference Marker Table** option and select the **previously created reference marker**.

The <u>Single Electropherogram view</u> is most suitable for confirming peak detection. Ensure that the plot shows a red "+" at the peak apex for all peaks between the alignment marker peaks. If a pink "x" is shown instead, click the **Image Options** button of the Electropherogram toolbar and select the options **Mark peak apex** and **With Label**, with **Size** [bp] selected. Click **OK** to close the dialog.

The electropherogram plot of the analyzed samples will show the size of the detected DNA molecules in base pairs above the apex of the corresponding peaks. Peak properties are shown in the **Peak Result** table below the electropherogram. For details, refer to the <u>Result table</u> section. To switch the sample shown in the electropherogram, click on the next sample in the **Experiment Explorer** on the left.

Ensure that the peak detection has worked as expected. It is important that the correct peaks have been marked as alignment marker peaks (marked with a green '+'). This is the basis for all further analysis functions. If this is not the case, adapt parameters in the **Analysis** parameters panel on the right side of the **Analysis** environment. To do this, open the **Analysis Properties** section and then open the **Parameters** section. For details on how to change the parameters mentioned below, refer to section <u>Modifying an analysis profile</u>.

Note: Collapse and expand parts of the analysis properties by clicking ▼ and ▶, respectively.

Analysis	Report	Pea	k Calling	P	roperties	×	
▶ Mode						Â	
▼ Analys	is Proper	ties					
DNA Fas	t Analysis		v s	ave	as		
▼ Parameters							
● Peak ○ Smear ○ gDNA							
Smoothing	Sta	rt	Win	dow			
Filter	0,00	min			0 pts		
Baseline	Sta	rt	Win	dow			
Filter	0,00	min			40 sec		
Minimum	Sta	rt	Value				
Distance	0,00	min	0,25 sec		25 sec		
Suspend	Sta	rt	End				
Integration	0,00	min		0,	50 min		
Threshold	Sta	rt	Value				
	0,00	min		10	0,00 %		
Alignment Marker Threshold	0	s	i/N				
			Add		Delete		
Define Parameter Select Parameter							
1 Sample(s)) selected	1	Start	Ana	ilysis	•	

• Ensure that the **Peak** option is selected.

• If you're analyzing samples, in the **Reference Marker** section, check **Reference Marker Table** and select a previously saved reference marker. If you're creating the reference marker, in the **Reference Marker** section, check **No Marker**.

Analysis	Report	Peak Calling	Properties	×
▶ Mode				•
▼ Analy	sis Prope	rties		
DNA Fa	st Analysis	•	Save as	
► Paran	neters			
▼ Refer	ence Mark	er		1
⊚ No M	larker			Ш
⊖ Refe	rence Mark	ker Table		Ш
Marke	er Table]
1 Sample/	s) selector		Analysis	•
i sample(araciectet	Stan	Analysis	

- If peaks were detected before or after the alignment marker peak, increase the **Alignment marker threshold**. Click on the field and change the value.
- If peak detection is too sensitive, increase the **Threshold**. Click this parameter line and increase the value in the **Value** field in the edit area below the parameters list, and click **OK** to confirm.

- When analyzing **Fast Analysis** data, the default baseline filter window (40 seconds) might be too large. In this case, a notification will suggest to decrease the baseline filter window. To do so, click the **Baseline filter line** and set the **Window** size to 39 in the edit area below the parameters list, and click **OK** to confirm. In most cases, this is sufficient.
- If in a region with many peaks close together, some peaks are not detected, decrease the minimum distance by clicking on the **Minimum Distance parameter line** and setting the distance **Value** from default 0.25 to 0.1 seconds. Click **OK** to confirm this setting. If you do not need minimum distance between peaks, set the minimum distance value to 0 seconds.
- For **Fast Analysis** samples, smoothing should be deactivated as set in the **DNA Fast Analysis** analysis profile. Here, in the **Smoothing Filter parameter line**, the **Window** value is set to 0 pts.

Click the Start Analysis button again to start a new analysis according to this modified analysis profile.

In some cases, it may be necessary to repeat the adaptations to the analysis profile iteratively. If the peak detection worked as expected with the modified analysis profile, save this analysis profile and try to use this new profile instead of the **DNA Fast Analysis** analysis profile for future **Fast Analysis** samples.

Note: Only users with user role **Developer** or **Administrator** can save modified parameters of an analysis profile.

6.4.7.3 gDNA analysis

Use the analysis options described below for analysis of genomic DNA quality.

Note: Make sure to use the 15 bp alignment marker along with gDNA samples. For details, refer to <u>Checking alignment marker</u>.

During the <u>Size and concentration determination procedure</u>, select the **gDNA Analysis** profile as a starting point.

The <u>Single Electropherogram view</u> is most suitable for checking peak detection. Ensure that the plot shows a pink "x" for all smear peaks between the alignment marker peaks. If a red "+" is shown instead, click the **Image Options** button of the Electropherogram toolbar and select the options **Mark median of size** and **With Label**. Click **OK** to close the dialog. The electropherogram plot of the analyzed samples will mark the median of size in base pairs of the corresponding peaks.

After the <u>Size and concentration determination procedure</u>, the results are shown in the **Smear/gDNA Result** table below the electropherogram. Find the size estimation of the gDNA in base pairs in the column **Median Size** [bp]. The concentration estimation of the gDNA is displayed as **Total Concentration** just above the **Smear/gDNA Result** table. For further details, refer to the <u>Result table</u> section.

To switch the selected sample in the electropherogram, click on the next sample in the **Experiment Explorer** on the left.

In case the sample was analyzed earlier with <u>distribution analysis</u>, the Single Electropherogram view shows smear peaks corresponding to the areas of interest as defined in the <u>distribution profile</u>. In order to see a plot of the peak detection, the distribution analysis needs to be removed temporarily. To do so, right-click the sample in the **Experiment Explorer** to the right and select **Transfer Distribution Profile** to copy the parameters to the **Distribution** panel to the right for later re-use. Then right-click the sample again and select **Remove Distribution Analysis Data** to remove the results of distribution analysis from the sample. The Single Electropherogram View now shows the result of peak detection. It shows the alignment marker peaks (marked green at the apex) and the smear peaks according to the **Smear/gDNA Result** table.

Ensure that the peak detection has worked as expected. It is important that the correct peaks have been marked as alignment marker peaks (marked with a green '+'). This is the basis for all further analysis functions. If this is not the case, adapt parameters in the **Analysis** parameters panel on the right side of the **Analysis** environment. To do this, open the **Analysis Properties** section and then open the **Parameters** section. For details on how to change the parameters mentioned below, refer to section <u>Modifying an analysis profile</u>.

Note: Collapse and expand parts of the analysis properties by clicking ▼ and ▶, respectively.

Analysis	Report	port Peak Calling		Properties	×		
▶ Mode							
 Analysis Properties 							
gDNA A	naiysis			ave as			
▼ Paran	neters						
O Peak	⊖ s	mear	⊚ gDNA				
Smoothin	ng S	itart	Win	dow			
Filter	0,0	0 min		10 sec			
Minimum	S	tart	Va	lue			
Distance	0,0	0 min 0 min		10,00 sec			
	10,0	viiiii		50,00 300			
Suspend	on S	tart	Er	nd			
Threshol	a 9	tart	Va				
mesho	0,0	0,00 min 50,00 S/N					
Alignmer Marker Threshol	nt 200 d		S/N				
			Add	Delete			
Define Pa Select F	irameter Paramete	ſ					
			ОК	Cancel	•		
1 Sample(s) selected Start Analysis							

• Ensure that the **gDNA** option is selected.

- If the first detected peak is not the alignment marker peak, as a first step, increase the **Alignment marker threshold**. Click on the field and change the value. If this is not sufficient, click the **Add** button below the parameter list and select **Suspend Integration** from the **Define parameter** drop-down list. Use the **Absolute** time unit and define the starting point as 0 minutes. Set the end point of this parameter to just before the start of the alignment marker peak (e.g. set the end to 2 minutes if the first alignment marker peak appears at 2.5 minutes).
- If signals were incorrectly detected as peaks, increase the threshold. Click this parameter line and
 increase the value in the Value field in the edit area below the parameters list and click OK to confirm.
 Because the smoothing algorithm was optimized for the typical shape of smear data, a high S/N value is
 required as threshold.

Click the Start Analysis button again to start a new analysis using the modified analysis profile.

In some cases, it may be necessary to repeatedly adapt the analysis profile. If the peak detection worked as expected with a modified analysis profile, save and use the modified analysis profile for future DNA analysis of similar samples, instead of using the **gDNA Analysis** profile.

To assess the quality of the gDNA, perform **Distribution analysis**. Select the **Default gDNA Integrity Ratio** profile as a starting point. For information on how to perform distribution analysis, refer to section <u>Distribution analysis</u>. Distribution analysis can only be performed after the <u>Size and concentration</u> <u>determination procedure</u> has been completed successfully.

6.4.7.4 Smear DNA analysis

Use the analysis options described below to obtain basic information about e.g. minimum and maximum fragment sizes, molarity or concentration of DNA libraries samples.

During the <u>Size and concentration determination procedure</u>, select the **NGS Library Analysis** profile as a starting point.

The <u>Single Electropherogram view</u> is most suitable for controlling peak detection. Ensure that the plot shows a pink "x" for all smear peaks between the alignment marker peaks. If a red "+" is shown instead, click the **Image Options** button of the Electropherogram toolbar and select the options **Mark median of size** and **With Label**. Click **OK** to close the dialog. The electropherogram plot of the analyzed samples will mark the median of size in base pairs of the corresponding peaks.

Smear peak properties are shown in the **Smear/gDNA Result** table below the electropherogram. For details, refer to the <u>Result table</u> section.

To switch the sample to be shown in the electropherogram, click on the next sample in the **Experiment Explorer** on the left.

In case the sample was analyzed earlier with <u>distribution analysis</u>, the Single Electropherogram view shows smear peaks corresponding to the areas of interest as defined in the <u>distribution profile</u>. In order to see a plot of the peak detection, the distribution analysis needs to be removed temporarily. To do so, right-click the sample in the **Experiment Explorer** to the right and select **Transfer Distribution Profile** to copy the parameters to the **Distribution** panel to the right for later re-use. Then right-click the sample again and select **Remove Distribution Analysis Data** to remove the results of distribution analysis from the sample. The Single Electropherogram View now shows the result of peak detection. It shows the alignment marker peaks (marked green at the apex) and the smear peaks according to the **Smear/gDNA Result** table.

Ensure that the peak detection has worked as expected. It is important that the correct peaks have been marked as alignment marker peaks (marked with a green '+'). This is the basis for all further analysis functions. If this is not the case, adapt parameters in the **Analysis** parameters panel on the right side of the **Analysis** environment. To do this, open the **Analysis Properties** section and then open the **Parameters** section. For details on how to change the parameters mentioned below, refer to section <u>Modifying an analysis profile</u>.

Note: Collapse and expand parts of the analysis properties by clicking ▼ and ▶, respectively.

Analysis Re	eport Pea	ak Calling	Properties	×				
▼ Mode								
● DNA ○ RNA								
▼ Analysis	Properties							
NGS Library	/ Analysis	▼ [5	Save as					
▼ Paramete	r\$	-						
🔿 Peak 💿 Smear		⊖ gDNA						
Smoothing	Start	Win	dow					
Filter	0,00 min		10 sec					
Minimum	Start	Value						
Distance	0,00 min	3,00 sec						
	7,00 min		20,00 sec					
Suspend Integration	gration Start End							
Threshold	Start	Va						
	0,00 min							
Alignment Marker Threshold	200	S/N						
		Add	Delete					
Define Param	eter							
Select Para	meter							
				•				
1 Sample(s) selected Start Analysis								

- Ensure that the **Smear** option is selected.
- If peaks were detected before the first or after the last alignment marker peak, increase the **Alignment marker threshold**. Click on the field and change the value.
- If signals were incorrectly detected as peaks, increase the threshold. Because the smoothing algorithm was optimized for the typical shape of smear data, a high S/N value is required as threshold.

Click the Start Analysis button again to start a new analysis using the modified analysis profile.

In some cases, it may be necessary to repeatedly adapt the analysis profile. If the peak detection worked as expected with a modified analysis profile, save and use the modified analysis profile for future analysis of similar samples, instead of using the **NGS Library Analysis** profile.

Smear analysis distinguishes samples marked as size marker and does not define smear peaks for these samples. For all other samples, the analysis distinguishes between alignment marker peaks and smear peaks. The software assigns an area of interest to each detected smear peak. The borders of an area of interest are marked by two vertical pink lines and, by default, are the same as the borders of the corresponding peak. In case the detected peak borders do not describe the actual area of interest, move the borders to the correct positions. All properties displayed in the **Smear Result** table refer to the area of interest defined by the vertical pink lines.

Note: Upon clicking the **Start Analysis** button, the borders of the normalized areas will be reset to match the borders of the detected smear peak, even if they were previously corrected manually. Moving the threshold does not affect the borders of the area of interest.



However, for routine use, use **Distribution analysis** instead. There you can define the areas of the distribution you are interested in and use these settings for all samples in one step. For information on how to setup distribution analysis, refer to section <u>Distribution analysis</u>. Distribution analysis can only be performed after the <u>Size and concentration determination procedure</u> has been completed successfully.

6.4.7.5 Distribution analysis

Use this analysis type to assess the quality of nucleic acid libraries or genomic DNA in routine use based on the size distribution.

If the parameters panel is not yet open, open it by clicking the icon on the right-hand corner of the tab:

Gel Image Electropherogram Peak Calling Marker

Note: If peak calling features are still active, activate distribution analysis features now. See the section Activating distribution analysis features.

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To perform the distribution analysis:

1. Before distribution analysis can start, ensure that the <u>Size and concentration determination procedure</u> has been performed using analysis profile parameters as described in the sections <u>Smear DNA analysis</u> or <u>gDNA analysis</u> respectively.

Gel Image Electropherogram Distribution Marker		Analysis F	Report	Distribution	Properties	×
ロ IIII 回 Q の N N X IX 十 Image Options Select All		▼ Mode				
		● DNA C) RNA			
ធ្មី 0.800-		 Analysis 	s Proper	ties		
÷ 0.700		NGS Libra	ry Analys	sis 🔻	Save as	
g 0,600 +		▼ Paramet	ers			
وَّ 0,400-						
0.300		O Peak	⊚ Sme	ear ⊖gDN	A	
		Smoothing	Star	rt Wi	ndow	
		Filter	0,00	min	10 sec	
		Minimum	Star	t V	alue	
Peak Size			0,00 r	nin	3,00 sec	
	-11		7,00 r	nin	20,00 sec	
⊥ -0,087224 0,890524 RFU I→ -0,843570 <i< td=""> 9,279279 min</i<>		Suspend Integration	Star	rt E	End	
Sample Info: Library A1 Position: A1 Experiment Name / Plate ID: 2015-03	-29	Threshold	Star	t V	alue	
Total Conc. [ng/µ] 0,941577101522185 Total Molarity [nmol/I] 5,45663477402509			0,00	min	50,00 S/N	
# Median Size [db] Conc. Area or interest [mg/µ] Moi. Area or interest [mmoi/ŋ] 2 256 0,87 5,18		Alignment Marker Threshold	200	S/N		
				Add	Delete	
		Define Para Select Par	neter ameter	Ŧ		
Peak Result Smear/gDNA Result Distribution Analysis Result		1 Sample(s)	selected	Sta	rt Analysis	

- 2. Open the **Distribution** tab located on the right side of the **Analysis** environment.
- 3. In case the distribution analysis needs to be removed temporarily during the <u>Size and concentration</u> <u>determination procedure</u>, use the distribution profile parameters that had been transferred before they should be visible in the **Distribution** parameters tab at the right. Otherwise, select the distribution profile relevant for the samples from the **Distribution Profile** drop-down box. For gDNA quality analysis, select the **Default gDNA Integrity Ratio profile**. A distribution profile defines the areas of interest that are, for example, expected for the library. For DNA libraries, refer to the section <u>Creating a distribution profile</u> for information on how to create the profile.
- 4. Select the samples to be analyzed with this distribution profile.
- 5. Click the **Start Analysis** button at the bottom of the **Distribution** tab at the right to start the distribution analysis.
- 6. The result for one sample is displayed in the Distribution Analysis Result table below the electropherogram and an overview result table for all samples is available in the Distribution screen in the middle of the Analysis environment. Refer to the section <u>Distribution result</u> for a description of the result structure and to the section <u>Distribution result columns</u> for a description of the available columns.



7. To save the result of the distribution analysis, save the experiment by clicking in the **Experiment Explorer** at the left.

Note: After distribution analysis, the single Electropherogram shows the areas of interest as defined in the **Distribution** profile and the borders of areas of interest cannot be moved anymore.

Note: In some cases, the signal in an area of interest does not exceed the baseline at any point, indicating that the area of interest is not present in a sample.
It is possible to automate distribution analysis during the data acquisition process. Ensure that you have already automated the <u>Size and concentration determination procedure</u> for your samples using <u>Smear DNA</u> analysis or <u>gDNA</u> analysis. To automate also the distribution analysis, save any changes in the distribution analysis parameters into a customized distribution profile. Make sure that these parameters work for all samples. Then <u>modify the Process Profile</u> you have created to automate the size and concentration determination procedure for your samples. In the <u>Process Profile</u> step, select the option **Distribution Analysis** in the **Included** steps section additionally. If peak calling features are still active, activate distribution analysis features now. See the section <u>Activating distribution analysis features</u>. In the <u>Distribution Analysis</u> step, select the customized distribution profile. For further details, refer to <u>Process Profile</u> options. Save the Process Profile with these settings for re-use. During each run setup, ensure to select the alignment marker in the **Sample Selection** step.

6.4.7.5.1 Activating distribution analysis features

Either peak calling or distribution analysis features can be applied to samples of an active experiment.

To activate distribution features if peak calling anaylsis is active, proceed as follows for the current experiment

- 1. Open the Peak Calling tab located on the right-hand side of the Analysis environment.
- 2. Click on Switch to Distribution.



The distribution result tables replace the peak calling result tables and the distribution parameter panel replaces the peak calling parameter panel. The software keeps this setting until an experiment that used peak calling is activated or opened.

Note: It is not possible to activate distribution analysis features if peak calling has already been performed in the current experiment. In this case, <u>remove the peak calling data</u> first.

6.4.7.5.2 Distribution result

In the **Analysis** environment, each sample can be analyzed individually by means of distribution analysis. Therefore, the distribution result overview is grouped by distribution profiles and each group is labeled with the name of the applied distribution profile. The result for each distribution profile can be collapsed individually.

The result of each distribution profile lists the samples to which the profile was applied. This list is organized into groups of samples belonging to the same plate. Thus, in the screenshot below, a second group of samples from another plate might appear within the collapsible area of the distribution profile under a second heading MO_V-1192 R1|E1.

Each row in the table corresponds to a sample, with the columns **Pos.** and **Sample Info** identifying the sample. Columns relevant to the entire sample appear first. Thereafter, columns are grouped according to the names of the defined areas of interest and display several smear peak properties. The remaining columns are then grouped by the name of defined ratios. Find a detailed description in the section <u>Distribution result columns</u>.

To change the properties listed, right-click the table header and select or unselect the property with the **Show Column** option. The change takes effect in the entire table.

Note: If a sample was not analyzed with a smear or gDNA analysis profile, it will not appear in the distribution result overview. If a sample was not analyzed with a reference marker, the distribution values cannot be calculated. In this case, the sample **Quality** is listed as "**Review**." In both cases, follow the instructions in <u>Analysis of DNA samples</u> to check and correct the analysis steps that are required before **Distribution** analysis. Ensure using **Analysis** parameters as described in <u>Smear DNA analysis</u> and <u>gDNA</u> analysis, respectively.

The distribution result table can be copied to the clipboard. To copy the complete result of a distribution profile, right-click in the distribution result table and select **Copy Distribution Result of** [...].

Gel Image	Electropherogram	Distribution	Marker																
▼ Distrib	ution Analysis Result	of [LP ATP BRC	A v21																
MO_V-119	2 R1 E1									-									
				1						Sn	nall (Aol1)						Lib	ary (Aol2)	
Pos.		Sample Info		Total Conc. [ng/µl]	Total Mol. [nmol/I]	Quality	Mol. [nmol/1]	Conc. [ng/µl]	Size Start [bp]	Size End [bp]	Median Size [bp]	Height [S/N]	Height Check	Mol. [nmol/l]	Conc. [ng/µl]	Size Start [bp]	Size End [bp]	Median Size [bp]	
A02	V-1192_2			0,21	4,14	Review	1,41	0,10	50	170	0	4,81	Not Analyzed	0,29	0,04	170	300	0	
A03	V-1192_3			0,95	7,17	Passed	1,10	0,09	50	170	0	13,09	Not Analyzed	4,96	0,76	170	300	0	
A04	V-1192_4			1,88	12,99	Passed	1,29	0,10	50	170	0	13,65	Not Analyzed	10,69	1,64	170	300	0	
A05	V-1192_5			1,74	12,43	Passed	1,59	0,12	50	170	0	15,57	Not Analyzed	9,43	1,44	170	300	0	
A06	V-1192_6			1,67	11,27	Passed	0,47	0,04	50	170	0	8,80	Not Analyzed	10,28	1,57	170	300	0	
A07	V-1192_7			1,47	14,03	Passed	1,91	0,13	50	1/0	0	5,80	Not Analyzed	7,88	1,20	1/0	300	0	
AUC	V-1192_0			1,20	9,40	Passed	0,89	0,07	50	170	0	0,07	Not Analyzed	7,42	1,13	170	300	0	Η.
BOG	SM			1,00	12,15 n/a	Not Analyzed	1,15 n/a	0,09	00 n/a	n/a	n/a	06,1	Not Analyzed	9,00	1,4/	n/2	000 n/a	n/a	H
				100	110	Troci analyzou	100	110	100	100	1 100	10.0	reet/ analyzed	1 100	110	100	100		4
▼ Distrib	ution Analysis Result	of [NewDistributi	onProfile]																
NO 1/ 440	0.041.54																		
WO_V-119	2 RIJEI																		
											Aol								
Pos.		Sample Info		Total Conc.	Total Mol.	Quality	Mol. (nmol/l)	Conc. Ing/ull	Size Start	Size End	Median Size	Height (S/N)	Height Check						
A12	V-1192_12			1,97	13,57	Not Analyzed	11,71	1,80	180	300	236	571,84	Not Analyzed	i					
	_										· · · · ·								
L																			
							_												
<																			•

Note: If the distribution profile was modified but not saved, and then used for a subset of previously analysed samples, two distribution results will be listed with the same distribution profile name. To identify the differences between the used profiles, right-click one of the samples in the **Experiment Explorer** on the left and select the **Transfer Distribution Profile** item from the context menu. Inspect the parameters in the **Distribution** tab on the right. Do the same with a sample from the second distribution result.

6.4.7.5.3 Modifying a distribution profile

A distribution profile specifies the quality criteria for a DNA/RNA library or genomic DNA. The profile defines areas of interest by start and end size, as well as molarity or concentration ratios between defined areas of interest or against the total molarity or total concentration of the sample.

Note: Distribution profiles can only be modified by users with the user role Administrator or Developer.

To modify a distribution profile proceed as follows:

1. Open the **Distribution** tab on the right of the Analysis environment.

Note: The **Distribution** tab is available only if distribution features are active. Refer to <u>Activating distribution</u> <u>analysis features</u> for information on how to activate distribution analysis features.

2. Select the distribution profile to be modified from the Distribution Profile drop-down box.

Analysis	Report	Distribution Properties		×	
	Switc	h to Peak Calling			
Distribution	n Profile				
NGS_Distribution_Analysis V Save as					
Area	s of Intere	st			
Ratio	08				

3. Follow the instructions below to change definitions of areas of interest and/or of ratios as needed.

Note: The name of the profile in the **Distribution Profile** drop-down box will appear preceded by an "*", indicating that the profile has been modified.

4. Save the modified distribution profile by clicking the **Save as** button to the right of the **Distribution Profile** drop-down box.

Note: At least one area of interest must be defined.

Note: Collapse and expand parts of the distribution profile by clicking ▼and ▶, respectively.

To add an area of interest:

1. Click the Add button. A new empty row appears at the bottom of the table.

 Areas of Interes 	t		
Name	Start [bp]	End [bp]	Min Height [S/N]
	0	0	n/a
Small (AoI1)	50	170	n/a
Library (Aol2)	170	300	40
Large (AoI3)	300	2000	n/a
Name		Add	Delete
		Check sm	ear height
Start [bp] End [bp	o] Min)	1 Height 0	[S/N]
		OK	Cancel

2. Define the area of interest in the editable fields:

Name	A unique name for the area of interest. This will be the column header in the distribution result table, grouping all columns relevant to this area.				
	Note: Names of the areas must be unique and can have a maximum length of 20 characters. Total Molarity or Total Concentration cannot be used as a name for an area of interest.				
Start [bp]	Define in base pairs the size where the area of interest starts.				
End [bp]	Define in base pairs the size where the area of interest ends.				
Check smear height	Select this option to check that the signal of the area of interest is above the threshold defined by Min Height (below).				
Min Height	Enabled if the option Check smear height has been selected.				
[S/N]	Defines the threshold for the area of interest as a multiple of the noise level, which is estimated from the sample data.				
	Note: If Check smear height is not selected, "n/a" will appear in the table of areas of interest.				

Note: Areas of interest may not overlap. The start or end field appears yellow in case of overlapping sizes. An area may, however, start at the size where another area ends.

Note: Make sure that size definitions of the areas of interest are in within the sizes of the alignment marker. Either use a suitable alignment marker or adapt the areas of interest. The distribution analysis will fail, otherwise, because all areas of interest are assumed to be within the range of the alignment marker. For gDNA analysis, refer to the **Default gDNA Integrity Ratio** profile for the maximum size.

Example: Use areas of interest to distinguish between wanted (library) and unwanted (small or large artifacts) fragments during a library preparation reaction and use the option **Check smear height** for the library.

3. Click **OK** to add the new area of interest to the table.

Note: The areas of interest are sorted in the table by start size.

Note: At least one area of interest must be defined.

To delete an area of interest:

- 1. Select the corresponding row in the table.
- 2. Click the **Delete** button.

Note: If the deleted area of interest was used in the definition of a ratio, the ratio becomes invalid. Then delete the ratio as well.

To modify an area of interest:

- 1. Select the row of the area of interest to be modified.
- 2. Modify the values of the area of interest in the editable fields.
- 3. Click OK. The values in the selected row are changed.

Note: Areas of interest are sorted by position in the Areas of Interest table.

The QIAxcel ScreenGel allows to define ratios to assess the relationship between wanted and unwanted fragments in a distribution.

To add a ratio:

- 1. Expand the Ratio section.
- 2. Ensure that the Ratios option is selected.
- 3. If the QIAxcel ScreenGel shall include an assessment of the defined ratios into the overall sample quality, select the option Check Quality. In this case, you are forced to enter limits for the ratio value (see below). Select Show Values, otherwise, if you want to see the ratio calculations in the distribution result table without quality assessment based on the ratios.
- Select the calculation basis for the ratios to be defined. It is possible to select Concentration or Molarity. This calculation basis will be used for all ratios.
- 5. Click the Add button. A new empty row will appear at the bottom of the Ratio Definitions list.
- 6. Define the ratio in the editable fields:

Ratio Name	A unique name for the ratio. This will be the column header in the distribution result table, grouping all columns related to this ratio.
	Note: Names of ratios must be unique and have a maximum length of 20 characters. The same name may not be used for ratios and for areas of interest.
Calculate ratio for	Define the nominator and denominator of the ratio by selecting the respective areas of interest from the drop-down boxes. In addition, the denominator drop-down box offers the option to select the Total Sample , which means the Total Molarity or Total Concentration , respectively, depending on the selected calculation method.
Acceptance range	These fields are available only if the option Check Quality has been selected.

Enter a minimum and maximum value in the Min. and Max. fields. The quality of the ratio will be considered as "Passed" for samples where the value of the ratio falls within this range, as "Review" otherwise.

Example: Select **Check Quality** and calculation basis **Concentration**, and define a ratio against the **Total Sample** for an area of unwanted fragments, and enter low values for the acceptance range.

Often, it is an iterative process to find out the acceptance range for a certain type of samples. If you find values that work for your samples, save the changes into this or a <u>new distribution profile</u> for re-use. If the ratio quality is frequently assessed as "Review" although the samples are good enough for downstream application, consider broadening the acceptance range or continuing without a quality check for ratios.

7. Click **OK** to add the new ratio to the list.

Note: Ratios appear in the list in the same order as in the distribution result table.

Note: Ratio names or range values that are too long may not be fully displayed in the list. A tool tip appears with the complete name by hovering the mouse over the truncated name or value.

 Ratios
Ratios
Check Quality O Show Values
Select calculation basis
Molarity V
Ratio Definitions
Small_Total
0 <= Small (Aol1) Total Sample <= 0,3
Large_Total
0 <= Large (Aol3) Total Sample <= 0,2
Add Delete
Ratio Name
Large_Total
Calculate ratio for Acceptance range Min.
Large (Aol3) V 0
Max.
Total Sample V 0,2
OK Cancel

To delete a ratio:

- 1. Select the ratio row in the list.
- 2. Click the **Delete** button and approve the confirmation message.
- 3. If you deleted the last ratio, uncheck the Ratios option at the top of the Ratios section.

To modify a ratio:

- 1. Select the row of the ratio to be modified.
- 2. Modify the values of the ratio in the editable fields.
- 3. Click **OK**. The values in the selected row are changed.

Note: Ratios appear in the list in same order as in the distribution result table.

6.4.7.5.4 Creating a distribution profile

Note: Distribution analysis features must be active to create a distribution profile. Refer to <u>Activating</u> <u>distribution analysis features</u> for information on how to activate distribution analysis features.

To create a new distribution profile:

- 1. Open the **Distribution** tab on the right of the **Analysis** environment.
- Select a distribution profile from the Distribution Profile drop-down box. The selected distribution profile serves as a template for the creation of the new one. Alternatively, select NewDistributionProfile to start with an empty profile.
- 3. Define the distribution profile as needed. Refer to the section Modifying a distribution profile for details.
- 4. Save the new distribution profile by clicking the **Save as** button to the right of the **Distribution Profile** drop-down box. Enter a unique distribution profile name and click **OK**.

Note: Only users with user role Administrator or Developer can create new distribution profiles.

6.4.7.5.5 Removing distribution results

To remove results of a distribution analysis from one or more samples:

- 1. Select the samples in the **Experiment Explorer**. Refer to the section <u>Selecting samples</u> for information on how to select multiple samples.
- 2. Right-click the selection and select Remove Distribution Analysis Data from the context menu.

The distribution analysis results for these samples are cleared from the distribution result tables. The electropherograms of the samples are updated because the areas of interest are no longer defined by a distribution profile.

In the Experiment Explorer, the samples will still appear as "analyzed."

6.4.8 Analysis of RNA samples

of sample.

This section describes how RNA samples can be analyzed. In addition to size and concentration, parameters such as the **285/185 ratio** and **RNA Integrity Score (RIS)** can be calculated. The RIS is a tool for objective assessment of the the quality of eukaryotic RNA samples. Perform the following steps for each analysis. Follow the instructions for the desired analysis type.

For detailed and up-to-date information on RNA methods and analyses, refer to the QIAxcel RNA Handbook for QIAxcel RNA Quality Control Cartridge, available on www.qiagen.com/

- 1. Switch to the Analysis environment by selecting the Analysis icon in the main toolbar.
- Check the current mode in the bottom right corner of the software screen. Switch to RNA mode if not already selected. If DNA mode is selected, change the mode via the Analysis parameters panel (refer to the <u>Mode</u> section).
 Note: Only Developer or Administrator users can modify and save parameters of an analysis profile. Users with user role Operator can select another analysis profile which is more suitable for their type
- 3. If the samples you want to analyze are not yet listed in the **Experiment Explorer**, select the **Load experiment** icon from the **Experiment Explorer** toolbar. If the samples are listed but not active (grayed out), right click its experiment name and select **Activate or double-click the experiment name**. For further details, refer to the <u>Loading sample data</u> or <u>Activating an experiment</u> section.
- 4. To determine the size and concentration of RNA molecules, follow the instructions of the <u>Size and concentration determination procedure</u> and the <u>Peak detection procedure</u>. Refer to section <u>Basic RNA analysis</u> for information on which **Analysis** profile should be used as a starting point and which analysis parameters are required for your samples. Follow the <u>Size and concentration determination procedure</u> or <u>Peak detection procedure</u> also if samples should have been analyzed during data acquisition process, but either an error message has been shown or sample results are not as expected. In this case, use the procedure to check the analysis step by step and correct if needed. Follow the instructions to re-use the analysis parameters that have been used for the samples.
- To calculate the 285/185 ratio and/or the RNA Integrity Score (RIS), perform peak calling. Follow the instructions described in section <u>Peak Calling</u>. For information on which peak calling instruction should be used as a starting point, refer to section <u>RNA integrity analysis</u>.

The result tables summarize all the properties that QlAxcel ScreenGel has calculated for detected peaks. Right-click on the table header, select **Show column**, and then select the property you are interested in. Alternatively, you can right-click the table header and select **Show all columns**. This option displays all the properties calculated by the QlAxcel ScreenGel software. For more information, refer to section <u>Result table</u> and sections <u>Basic RNA analysis</u> and <u>RNA integrity analysis</u>, respectively.

Select **Distribution Analysis** for RNA degradation assessment or other smear-based analysis.

6.4.8.1 Basic RNA analysis

During the <u>Size and concentration determination procedure</u> or the <u>Peak detection procedure</u>, select the **Default RNA QC** analysis profile or the **RNA High-Sensitivity Analysis** (for High-sensitivity experiments) analysis profile as a starting point.

The <u>Single Electropherogram view</u> is most suitable for peak detection. Ensure that the plot shows a red "+" sign at the peak apex for all peaks after the alignment marker peak. If not, click the **Image Options** button of the Electropherogram toolbar and select the options **Mark peak apex** and **With Label**, with **Size [nt]** selected. Click **OK** to close the dialog. To change the sample shown in the electropherogram, click on Next Sample in the **Experiment Explorer** at the left.

Peak properties are shown in the **Peak Result** table below the electropherogram. For details, refer to the <u>Result table</u> section. After the <u>Size and concentration determination procedure</u> is completed, the size of detected RNA molecules in nucleotides is displayed in the Single Electropherogram view.

The concentrations can be examined in the peak result table. If concentrations are not shown by default, right-click on the table header, choose **Show column** and then select the **concentration column**, which displays the concentrations in $ng/\mu l$ for non-High Sensitivity experiments, and in $pg/\mu l$ for High Sensitivity experiments.

Ensure that the peak detection has worked as expected. It is important that the correct peaks have been marked as alignment marker peaks (marked with a green '+'). This is the basis for all further analysis functions. If this is not the case, adapt parameters in the **Analysis** parameters panel on the right side of the **Analysis** environment. To do this, open the **Analysis Properties** section and then open the **Parameters** section. For details on how to change the parameters mentioned below, refer to section <u>Modifying an</u> analysis profile.

Note: Collapse and expand parts of the analysis properties by clicking ▼ and ▶, respectively.

Analysis R	eport	Peak	Calling	Properties	1			
▼ Mode					í			
⊖ DNA ⊚	RNA							
▼ Analysis	Properti	es						
Default RNA QC 🛛 🔻 Save as								
▼ Paramete	ers							
eak	⊖ Smea	Nr.						
Smoothing	Start		Wir	dow				
Filter	0,00 min			10 sec				
Baseline	Start		Wir	dow				
Filter	0,00 m	in		40 sec				
	5,00 m	in		100 sec				
Minimum	Start		Va	lue				
Distance	0,00 m	in		5,00 sec				
Suspend Integration	Start		E	nd	ĺ			
Threshold	Start		Va	lue				
	0,00 m	in		100,00 S/N				
Alignment Marker Threshold	500	Si	N					
			Add	Delete	,			
12 Sample(s)	selected		Sta	rt Analysis	1			

• If the first peak detected is not the alignment marker peak, first increase the **Alignment marker threshold**. Click on the field and change the value.

If this is not sufficient, use the **Suspend Integration** parameter. Click the **Add** button below the parameters list and select **Suspend Integration** from the **Define parameter** drop-down list. In the drop-down list next to **End**, select **Absolute** to use the **Absolute** time unit. Define the starting point as 0 minutes. Set the ending point of this parameter to just before the start of the alignment marker peak (e.g. set the end to 2 minutes, if the first alignment marker peak appears at 2.5 minutes). Click the **OK** button to confirm.

- If the analysis profile is too sensitive and detects noise as peaks, increase the **Threshold** (e.g. from 100 S/N to 150 S/N). Click **OK** to confirm.
- If the baseline follows the 28S peak, increase the baseline filter window size for roughly the last third of the migration time. To add a second interval to the baseline filter parameter, click the **Add** button below the parameters list and select **Baseline Filter** from the **Define parameter** drop-down list. Then, e.g. set the start to 6 minutes and the window size to 80 seconds. Click the **OK** button to confirm.

Click the Start Analysis button again to start a new analysis according to this modified analysis profile.

In some cases, it may be necessary to iteratively repeat the adaptations to the analysis profile. If peak detection worked as expected with the modified analysis profile, save this analysis profile to use it for future RNA analysis as an alternative to the **Default RNA QC** or the **RNA High-Sensitivity Analysis** analysis profile.

6.4.8.2 RNA integrity analysis

This section provides instructions to investigate the integrity of RNA molecules. The RNA integrity check can be done with or without a size marker. Both approaches are described below.

Note: When analysing RNA Integrity Score (RIS) with the QIAxcel RNA High Sensitivity Kit, samples must be in the concentration range of $1-50 \text{ ng/}\mu\text{L}$.

If the parameters panel is not yet open, open it by clicking on the icon appearing on the right:

Gel Image	Electropherogram	Peak Calling	Marker	
		-		

Performing quality control of RNA with a size marker

- 1. Ensure that the <u>Size and concentration determination procedure</u> has been performed using analysis profile parameters, as described in the section <u>Standard RNA analysis</u>.
- Switch to the Peak Calling tab of the parameters panel and select one of the provided peak calling instructions: Default RNA prokaryote v2, Default RNA rat_mouse_human v2 or RNA High-Sensitivity Quality Control
- 3. To determine the RNA integrity, select the RNA Integrity Score option and choose the 18S peak. To examine the 28S-to-18S-ratio in one sample, select the Ratio Normalized Area option and choose the 28S and the 18S peak. For prokaryotic RNA, choose the 23S and 16S peak, respectively. For detailed information on how to define peak calling instructions, refer to Modifying peak calling instructions.

Ŀ

Analysis	Report	Pea	k Calling	Properties	×
	Swite	ch to D)istribution)	
eak Callin	g Instructio	on			_
RNA High	-Sensitivity	Qual	ty C 🔻	Save as	J
▼ Peak	s of Intere	est			4
	ude size m	arker	samples		
⊖ Find	centered	neak i	n interval		
Find	highest p	eak in	interval		
	Name		Position	Tol. [%]	
185			1869 nt 4700 ot	25,00	
203			470011	23,00	
			Add	Delete	
Name					
Position			т	plerance	
	Size		Ŧ	%	
			OK	Cancel	
▼ Calc	ulated Col	lumns			
Tetal	Concerts	ation (Talal Cone		
RNA	Integrity S	Score ("RIS")	'	
Re	Peak	18.5	T		
	Nemala		T CONTRACT		
✓ Ratic	o reormaliz	ed Are	a ("Ratio")		
Ratio	28 S	•			
Flatto	18 S				
Rela	tive Abund	lance			
					,

- 4. In the **Gel Image** overview, select all samples.
- 5. Click the Start Peak Calling button appearing at the bottom of the Peak Calling tab.

6. Inspect the Peak Calling result in the Peak Calling Screen.

Gel Image	Electropherogram	Peak Calling Marker
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Each row in this table represents the peak calling result for one sample. The RNA integrity score is given in the column **RIS**. The 28S-to-18S-ratio, is given in the column **Ratio**. In addition, the total concentration of RNA is given in the column **Total RNA concentration**. Furthermore, it is indicated for the 18S and 28S peak whether they are found based on the criterion specified in the peak calling instruction. For detailed information on the peak calling result table, refer to <u>Peak Calling</u>.

Performing quality control of RNA without size marker

- 1. Ensure that the <u>Peak detection procedure</u> has been performed using analysis profile parameters as described in the section <u>Basic RNA analysis</u>.
- Switch to the Peak Calling tab of the parameters panel and select one of the provided peak calling instructions: Default RNA prokaryote v2, Default RNA rat_mouse_human v2 or RNA High-Sensitivity Quality Control.
- 3. As the analysis was performed without the reference marker, adapt the peak calling instruction to search for peaks based on relative migration time instead of size. To do this, click on the row of the 18S peak and update the position criterion in the parameter area below. Set the **Position** value to an average of the relative migration time displayed in the result tables for the 18S peak of the selected RNA samples and change the unit in the drop-down list from **Size** to **Rel. Time**. Confirm this setting by clicking the **OK** button.

Adjust the tolerance parameter, if the relative migration times differ from sample to sample. Do the same for the 28S peak.

For prokaryotic RNA, change the 23S and 16S peak, respectively. For detailed information on how to define peak calling instructions, refer to section <u>Modifying peak calling instructions</u>.

- To determine the RNA integrity, select the RNA Integrity Score option and choose the 18S peak. To examine the 28S-to-18S-ratio in one sample, select the Ratio Normalized Area option and choose the 28S and the 18S peak.
- 5. In the Gel Image overview, select all samples.
- 7. Click the Start Peak Calling button appearing at the bottom of the Peak Calling tab on the right.
- 8. Inspect the peak calling result in the peak calling result overview.



Each row in this table represents the peak calling result for one sample. The RNA integrity score is given in the column **RIS**. The 28S-to-18S-ratio, is given in the column **Ratio**. In addition, the total concentration of RNA is given in the column **Total RNA concentration**. Furthermore, it is indicated for the 18S and 28S peak whether they are found based on the criterion specified in the peak calling instruction. For detailed information on the peak calling result table, refer to <u>Peak Calling</u>.

6.4.9 Manually modifying analysis results

The QIAxcel ScreenGel algorithms analyzes the data fully automated. However, a user can only influence the automated analysis through the analysis parameters. After an analysis, the results can be modified manually by the user.

6.4.9.1 Modifying the threshold

In the Single Electropherogram view (of up to 12 samples), the threshold parameter can be interactively changed by moving the threshold with the mouse. When doing so, the sample is re-analyzed using the analysis parameters of the sample properties and the new threshold value.

6.4.9.2 Deleting a peak

Deleting a peak is only possible in the Electropherogram view only.

To delete a peak:

- 1. Switch to the Electropherogram view.
- Right-click the top of the peak in the electropherogram and select **Delete Peak** from the context menu. The peak will be deleted.

To delete multiple peaks:

- 1. Holding the **Ctrl**-key, select multiple peaks by left-clicking the top of the peak.
- Right-click on the electropherogram and select **Delete Selected Peaks** from the context menu. The peaks will be deleted.

Alternatively, peaks can be deleted from the result table. To do this, chose the peak from the result table you want to delete. Right-click on the peak and select **Delete Peak** from the context menu.

Note: After a new analysis, deleted peaks may be detected again depending on the analysis parameters. To delete the peaks again, repeat the steps above.

6.4.9.3 Adding a peak

Adding a peak is only possible in the Electropherogram view only. To add a peak:

- 1. Switch to the Electropherogram view.
- 2. Right-click the electropherogram and select Insert Peak from the context menu.
- 3. A marker is displayed that moves along the signal while the mouse is being moved. Move the marker to the left border of the peak area (1) that is to be added. Left-click to set the left border.
- 4. Accordingly, left-click the peak that is to be added to set the right border of the peak area (2). If a peak can be found in the marked area, the peak is added to the result table and the visualization is updated.



Note: Adding a peak before the first alignment marker peak or after the second alignment marker peak is only possible if the signal rises above the alignment marker threshold in the corresponding region.

Note: If the sample was analyzed with smear analysis, a new smear peak can be added as described above. The newly inserted peak may not overlap with the area of interest of another peak.

6.4.9.4 Removing analysis results

Removing the analysis result from samples is only possible using the Experiment Explorer.

To remove the analysis result from samples:

- Select the samples in the Experiment Explorer. Refer to the <u>Selecting samples</u> section for information on how to select multiple samples.
- 2. Right-click the selection and select Remove Analysis Data from the context menu.



The result tables for these samples become empty. The representation of the sample in Gel image and Electropherogram view is updated: all peak annotations as well as baseline and threshold lines disappear.

If peak calling or distribution analysis was performed on the samples, the peak calling or distribution result tables for these samples also become empty and the samples are removed from the peak calling or distribution overview tables.

Note: To restore peak calling or distribution analysis results, re-analyze the samples with an analysis profile and reference marker, and repeat the peak calling or distribution analysis.

Refer to the sections <u>Peak Calling</u> and <u>Removing distribution results</u> for information on how to remove peak calling results or distribution analysis results independently.

6.4.9.5 Checking alignment marker

To check the alignment marker of a sample:

- 1. Select the sample in the Experiment Explorer.
- From the Analysis Instructions panel, view the sample properties at the right side of the Analysis environment. Refer to <u>Inspecting sample properties</u> for detailed information on how to view the properties.

3. Check if the alignment marker of the sample is correct. If not (or no marker is selected), the alignment marker of a plate can be changed. In the Experiment Explorer, right click the plate name. Select the Overwrite alignment marker context menu option. Select the correct alignment marker in the Overwrite Marker dialog that appears and confirm by clicking OK.

Note: The alignment marker can be changed for size marker samples in the **Marker** tab as well. Select the alignment marker of interest from the alignment marker drop down list.

Note: Upon switching from one alignment mode to another (two alignment marker peaks vs. one alignment marker peak), the analysis data of the plate will be removed and the samples must be analyzed again.

Note: To ensure analysis compatibility, the reference marker drop-down list contains only reference marker tables that match the alignment marker of the samples.

6.4.9.6 Modifying areas of interest

In the Single Electropherogram view, if the sample was analyzed with a smear analysis profile and at least one smear peak was detected, the borders of the area of interest for each peak can be interactively changed. To do so, hold left-click and drag the pink vertical lines indicating the area borders to the desired position. The lines may be moved independently of any peak start or end point.

Note: Area of interest may not overlap. Thus, it is not possible to drag a vertical line into the area of interest of another peak.

After moving a border, the properties corresponding to the given area of interest are recalculated using the newly defined borders.

6.4.9.7 Reusing prior analysis parameters

To re-use analysis parameters of an analyzed sample:

1. Right-click a sample in the **Experiment Explorer**.



2. To transfer previously used parameters (for the analysis of the sample) to the **Analysis tab**, select one of the following options:

Transfer Analysis Instructions	The analysis profile will be transferred to the Analysis tab of the toolbar. Additionally, it's the parameters used for and the reference marker table used for <u>size</u> and concentration determination, if present from the sample, will be transferred to the Analysis tab of the toolbar.
	Note : This option is only available if the sample has been analyzed.
Transfer Peak Calling Instruction	The peak calling instruction that has been used with this sample for <u>Peak Calling</u> will be transferred to the Peak Calling tab of the toolbar.
	Note : This option is only available if peak calling has been performed on the sample.
Transfer Distribution Profile	The distribution analysis profile that has been used with this sample for <u>Distribution analysis</u> will be transferred to the Distribution tab of the toolbar.

Note: This option is only available if distribution analysis has been performed on the sample.

Note: It is not possible to perform peak calling and distribution analysis in the same experiment. Either a peak calling instruction or a distribution profile is applied to a sample; but never both.

6.4.10 Customizing experiments

Experiments are created automatically from every process. Additionally, you can create and modify experiments. This enables you to compare samples from different experiments and different processes.

Note: Creating and modifying customized experiments requires the user role Developer or Administrator.

A customized experiment may contain single rows or groups of rows, as well as plates. However, within the experiment, all rows will be grouped in plate(s). Each time a row or plate is added to an experiment, you get a copy of it. This means that the source experiment, from which the row/plate originates, will remain unaffected by all the changes within the target experiment.

Note: You can only modify the composition of customized experiments. The composition of an experiment, which is the automatic result of a process, cannot be modified, see image below.

Note: Run results are marked with a padlock icon, while Custom experiments are not marked.



For general information about handling experiments and the **Experiment Explorer**, see the <u>Handling samples</u> <u>and experiments</u> section.

Note: Different cartridges types cannot be combined together in one experiment.

6.4.10.1 Creating a new experiment

To create a new customized experiment:

- 1. Click the folder icon in the **Experiment Explorer**.
- 2. Enter a unique name for the new experiment into the edit field then click **OK**.

Note: Once you entered the experiment name, it cannot be changed later.

3. A new empty experiment will be created and will be displayed at the bottom of the Experiment Explorer.

If the new experiment is outside the view, use the scrollbar of the **Experiment Explorer**. The new experiment will be activated automatically. If the previously active experiment was modified, the system asks you whether to save the changes or not. For more details about activating and deactivating, see the <u>Activating an experiment</u> section.

For information on how to collect samples for the new experiment, refer to the <u>Modifying an experiment</u> section.

Note: The composition of this experiment can be modified at any time, in contrast to experiments automatically generated by a process.

Note: New experiments can only be created by users with the user roles Developer or Administrator users.

6.4.10.2 Modifying an experiment

If a customized experiment should be modified, ensure that the experiment is activated. For more details about activating an experiment, see the <u>Activating an experiment</u> section.

Note: Only the plate composition of customized experiments can be modified. If an experiment is the (automatic) result of a process, the plate composition cannot be modified, therefore, the context menu options described below are disabled.

To modify an experiment:

- 1. Load the source experiment that contains the samples. For more details about loading, see the <u>Loading</u> <u>samples</u> section.
- 2. Create or activate the experiment that is to be customized in the Experiment Explorer.
- Expand the source experiment so that the required plates and rows are visible. The experiment can be expanded without activating. For more details about expanding, see the <u>Expanding and collapsing</u> section.
- 4. Modify the experiment as required:
 - To add a plate from the source experiment to the active experiment, right-click the plate and select Copy Plate to Active Experiment from the context menu. A copy will be added to the bottom of the active experiment.



Note: The complete plate will be copied to the experiment, but the original stays unchanged. Any further modification of the copy does not affect the original plate.

• To remove a plate from the active experiment, click the **Close** button of the plate. The plate will disappear from the experiment.





Adding a row is similar to adding a plate. Place the mouse cursor over the row to add to the active experiment, then select the Copy to active experiment from the context menu. To preserve the context of the row, a copy of its plate will be created and added to the active experiment containing a copy of the selected row. If you add another row from the same plate to the active experiment, a copy of the row will be created and added to the copy of the plate inside the active experiment. By simultaneously holding the Shift or Ctrl key during selection, multiple rows of the same plate can be added.



Note: A row can be located in the experiment only once.

Note: Individual samples cannot be added to the active experiment. Add the complete row instead, but visualize the required samples only.

 To remove a row from the active experiment, right-click the row letter and select the **Remove Row** from the context menu. The row will be removed from the active experiment. If the plate contains only one row, you cannot remove it; instead, please remove the plate.



6.4.11 Report/Export

The Analysis environment provides a flexible and powerful tool for reporting (as **RTF** or **PDF**) and exporting data (as **XML**).

For convenient re-use, corresponding report and export configurations can be stored in Report/Export Profiles. Thus, both reporting and data export may form a fully automated workflow by including a Report/ Export Profile to a Process Profile.

You can individually define which information should be included in the report and/or export. The Report and Export Options can be customized to your needs.

Each report may contain two main sections:

- An Overview section for information about all samples, such as Gel Image Overview, Electropherogram Overview, etc
- A Sample Detail section for detailed information about each particular sample

In the Sample Details section, information is reported for each sample and grouped in the following subsections:

- Sample Header
- Sample Graphics
- Result Table
- Run Parameter
- Analysis
- Reference Marker

The order of the samples is the same as in the experiments view.

For further details on customizing reports and export, see the Modifying a report/export profile section.

6.4.11.1 Generating a report

The basic procedure of manually generating a report in the **Analysis** environment is as follows:

- Load the samples using the Experiment Explorer. For further details on loading sample data, see section Loading sample data.
- Inspect the samples before reporting. See the <u>Viewing sample data</u> section for more details on data inspection. For this step, the **Gel view** or the **Electropherogram** overview are especially useful.
- Select the samples you would like to report.
 For further details on selecting samples, see section <u>Selecting samples for analysis or report</u>
- 4. Select the report settings and start reporting.

The report toolbar can be found on the right-hand side of the Analysis environment.

Select the **Report** tab of the toolbar.

Select a predefined **Report/Export** profile. The report settings of the selected profile are shown below the drop-down list. For details, refer to the <u>Report options</u> section.

Make sure that at least one of the report formats **PDF** or **RTF** is selected. Additionally, ensure that at least one of the options (**Overview** or **Sample Details**) is selected. If not, select another predefined **Report/Export** profile or modify the settings as required.

Specify the directory where the report files will be saved (refer to section <u>Modifying a report/export profile</u> for information on how to specify the report directory).

To start reporting, click Start Report/Export at the bottom of the Report tab.

Note: The button Start Report/Export is disabled if no samples are selected in the view (see Step 3).

The report files will be generated automatically and stored in the specified directory.

If you have selected the **Print report** option, the report will be printed on the default printer.

Note: Ensure that you have defined a default printer on your system.

5. Inspect the report files.

If the options **Display Report** and **PDF** were selected, the generated report will be opened automatically in the default PDF reader.

6.4.11.2 Report options

The report/export settings are divided into two groups:

- Report Options
- Export Options

Note: You can collapse and expand each group by clicking ▼ and ▶ to the left of the group name.

Report Options
▼ Report Options
Directory
Directory
N\QIAxcel\ScreenGel 2.0\Data\Report
Save to origin experiment directory
Format
PDF RTF
Options
Display Report Print Report
Use Images as Displayed
Overview
Sample Details

The options are described below:

Directory	Select the directory in which the generated report file should be stored by clicking . In the dialog that appears, navigate to the correct directory and click OK .
Save to origin experiment directory	Check this option to save the report in the same directory as the experiment. If this option is checked, any selected path in Directory will be ignored.
PDF	A file in PDF format will be generated.
RTF	A file in RTF format will be generated. You can select both formats.
Display Report	If you select this option and the format option PDF is selected, the generated report will be displayed automatically using the default PDF reader.
Print Report	If you select this option the generated report file(s) will automatically be sent to the default printer.
	Note: Ensure that you have a default printer on your system.
Use Images as Displayed	Use this option to obtain images that look like the presentation in the QIAxcel ScreenGel software in terms of alignment, scales, labels, zoom, etc.
	Note: This option is only available in the analysis environment. It may not be used in a Process Profile.
Overview	The overview lists the configurable general options for the report. Activate the checkboxes for options you would like to use.
Sample Details	Use the checkboxes to activate the content of the sample section you wish to report.

Note: If either PDF or RTF format is enabled, the report/export profile is valid only if the Overview or Sample Details checkbox is additionally selected.

Overview section

If the **Overview** option is selected, the following information is automatically included in the overview section of the report:

Report Date	Date and time of report generation.
Generated by QIAxcel ScreenGel	Software version which was used for creation of the report
Experiment Name	Name of the experiment the sample data belong to.
Cartridge ID	ID(s) of the cartridge(s) that the samples were processed with.
Instrument ID	Serial number of the instrument(s) used to process the samples.

Cartridge Expiration Status	This information is automatically included if the cartridge expiration date has passed.	
Calibration Status	Calibration status of the cartridge(s) at process time.	
Data Acquisition Status	Only in case of an incomplete experiment.	
	Gives information about planned and performed runs (e.g. 2 out of 8 runs performed successfully).	
Corrupt Experiment Accepted by	Only in case of an incomplete experiment, which was accepted by a user.	
	Name of the user who has accepted the experiment.	
Corrupt Experiment Accepted on:	Only in case of an incomplete experiment, which was accepted by a user.	
	Date and time when the experiment was accepted.	

Select the following optional information that you would like to include into the overview section:

✓ Overview	✓ Overview
Overview Comment Plate Comment Reported by Experiment Plate Sample List or Information Peak Calling Result Table Overal Result Table Overal Result Table Get Image Overview Electropherogram Superposition View	 ✓ Overview Experiment Plate Comment Reported by Experiment Path Sample List Lot Information ✓ Peak Calling Result Table Result Table Columns ✓ Sample Information ✓ Found Size Concentration [pg/µl] Molarity [pmol/l] ✓ Calculated Columns (if present) Peak Calling Instruction Table Overall Result Table Overall Smear Result Table
	Gel Image Overview
	Electropherogram Overview
	Electropherogram Superposition View



The optional fields are described next.

Note: If a result table option but no result table column is selected, the check box is highlighted in yellow, and the report/export profile is invalid.

Experiment Plate Comment	Experiment plate comment.				
Reported by	The ID of the user	who generated the report/export.			
Experiment Path	The path where the	The path where the experiment is stored.			
Sample List	All samples of a plate are listed with position, sample information and comment. If the experiment contains samples from different plates, a sample list is included for each plate.				
Lot Information	The entered lot numbers will be included.				
Distribution Analysis Result Table	Use this option for a distribution analysis to include the distribution analysis result overview table. Each row in the table represents a sample.				
	Note: This option is only available if distribution analysis features are active. To activate distribution analysis follow the instructions in <u>Activating distribution analysis features</u>				
	Additional option	Additional options to select are:			
	Show Profile Properties	Include the parameters of the distribution profile that was applied to the samples.			
	Sample Information	Include the Sample Information column.			
	Total Concentration	Include the column Total Concentration of the sample.			
	Total Molarity	Include the column Total Molarity of the sample.			
	Sample Quality	Include the Sample Quality column. This is the overall quality assessment of the sample, generated in the distribution analysis.			
	Aol Concentration	Include the Concentration column for each area of interest.			
	Aol Molarity	Include the Molarity column for each area of interest.			
	Aol Height	Include the Height column for each area of interest.			
	Aol Height Check	Include the Height Check column for each area of interest. This is the quality assessment of the height of the area of interest.			
	Ratio	Include the Ratio column for each defined ratio. Based on the selected calculation method, the column will show molarity or concentration ratio.			
	Ratio Quality	Include the Ratio Quality column for each defined ratio. This is the quality assessment of the ratio.			

Note: If a report includes too many areas of interest or ratios, the table is split between two areas of interest or ratios. Sample information, total concentration, total molarity and sample quality columns appear only in the first table. The remaining tables are marked with "(continued)" in the header.

Note: If samples are selected to which different distribution profiles were applied, a new table is generated for each distribution profile. Furthermore, if samples from different plates are selected, a new table is generated for each plate.

Note: Cells with the value "Review" are highlighted in the report.

Peak Calling The peak calling result overview table will be included. Each row in the table represents a sample. Additional options can be selected (see picture above).

Note: This option is only available if peak calling features are active. To activate peak calling features follow the instructions in <u>Activating peak calling features</u>

 Peak Calling Instruction
 Select this option to include the peak calling instruction table to the peak calling result overview table.

 Sample Information
 Select this option to include the Sample Information column in the peak calling result overview table.

- Found Select this option to include the **Found** column for each peak of interest to the peak calling result overview table.
- Size Select this option to include the **Size** column for each peak of interest in the peak calling result overview table.
- Concentration Select this option to include the **Concentration** column for each peak of interest in the peak calling result overview table.
- Molarity Select this option to include the **Molarity** column for each peak of interest in the peak callina result overview table.

Note: Calculated columns (such as Ratio, RIS) are automatically included, if they are part of the applied peak calling instruction.

Note: If there are too many peaks of interest, the table is split between two peaks of interest, The sample info and the calculated columns appear only in the first table, the following tables are marked with "(continued)" in the header.

Note: If samples to which different peak calling instructions were applied are selected, a new table starts for each peak calling instruction. Additionally, if samples from different plates are selected, a new table starts for each plate.

Overall Select this option to include a table that presents size and/or concentration and/or Result Table molarity results from all samples of a plate. A sub-table is included in the overall table for each selected sample. Sample position on the plate and, if available, sample information appear as the header of each sub-table. Each sub-table has as columns the result values that were selected for reporting. Each row then lists a detected peak and the corresponding results (size and/or concentration and/or molarity).

Note: If samples from different plates are selected, a new table starts for each plate.

Overall Smear Result Table	For a smear or gDNA analysis, use this option to include an overview smear result table, including selected smear result properties.				
	If samples from different plates are selected, a new table starts for each plate. Each row in the table represents a sample.				
	Select each table column to specify the result values to display: Median Size, Conc. Area of Interest, Mol. Area of Interest, % NA Area of Interest, or % Conc. Area of Interest.				
	Note: The columns of th for Result Table (below).	Note: The columns of the Smear Result Table can be selected and reordered as described for Result Table (below).			
	Select the options Total Concentration and Total Molarity to include the values for total concentration or total molarity of the sample.				
Gel Image	Select this option to incl	ude the Gel image of all selected samples.			
Overview	Click the Image options button to specify the appearance of the Gel Image overview. Image options are described below.				
	Note: The Image options button is disabled if the Use Images as Displayed option is selected.				
	Note: The Gel image in the report will have the left-hand y-axis on both sides, even if the Use Images as Displayed option is selected.				
	Unit of the y-axis	If you select Size , the y-axis will show a size scale based on the reference marker on both sides of the Gel image, even if the Use Images as Displayed option is selected. The lanes in the Gel image will be aligned according to the alignment markers.			
		Note: The size scale can only be created if all selected samples have been run with the same alignment marker, have been analyzed with the same reference marker table, and the alignment markers have been correctly identified. Otherwise, a corresponding message will be displayed.			
		If you select Relative migration time , the y-axis will show relative migration time. The lanes in the Gel image will be aligned according to the alignment markers.			
		Note: The relative time scale can only be created if all selected samples have been run with the same alignment marker, have been analyzed, and the alignment markers have been correctly identified. Otherwise, the lanes cannot not be aligned and the y-axis will display a corresponding message.			
		If you select Absolute migration time , the lanes will not be aligned and the y-axis will show an absolute time scale.			
	Zoom to alignment Marker	Select this option to scale the lanes so that all lanes start with the lower alignment marker band and end with the upper alignment marker band (data outside of the alignment markers is truncated).			

			Note: This is only possible if all samples are analyzed.
	Indivic	dual scaling	Select this option to auto-scale the contrast for each gel lane individually.
	Show inform	sample ation	Select this option to include the sample information in each lane.
	Show	plate ID	Select this option to display the plate ID , the repeat number, and the entry number of each lane.
	Show	method	Select this option to display the applied method above the sample lane.
	Gel Lo	yout	Use this option to determine the gel layout.
			Select Standard Layout to order the lanes from left to right, as they are ordered in the view.
			Select Horizontal plate layout to order the lanes starting with A1 at the upper left and ending with H12 at the lower right.
			Select Vertical plate layout to order the lanes starting with H1 at the upper left and ending with A12 at the lower right.
			Select Reverse vertical plate layout for the vertical plate layout but starting with A1 at the upper left and ending with H12 at the lower right.
			Select Split plate layout to split the vertical plate layout. The left portion will start with H1 at the upper left and end with A6 at the lower right. The right portion will start with H7 at the upper left and end with A12 at the lower right.
			Note: Positions for samples not selected remain empty.
	Lanes	per Row	Use this option to specify the number of lanes per row: 4, 6, 8, 12, 16, 24, 32, 36, or 48.
			Note: This option is only available if Standard layout is selected in the Gel Layout option.
Rows per Pag		per Page	Use this option to specify the number of rows to be included on one report page (a page split is introduced after the specified number of rows).
			Note: This option is not available if Standard layout is selected in the Gel Layout option.
	Analysis details		Select Highlight alignment markers to highlight in green the bands of the alignment marker peaks.
Electropherogram Overview	m	Select this option t The order is the sa 12 electropherogra	o include an electropherogram overview of all selected samples. me as in the view. One gallery overview page with no more than ams will always be printed on one report page.

Click on the **Image** options button to specify the appearance of the reported electropherogram overview. Image options are described below.

Note: The **Image** options button is disabled if the **Use Images as Displayed** option is selected.

Unit of x-axis	If you select Size , the x-axis will show a size scale based on the reference marker. The electropherograms will be aligned according to the alignment markers.		
	Note: The size scale can only be created if all selected samples have been run with the same alignment marker, have been analyzed with the same reference marker table, and the alignment markers have been correctly identified. Otherwise, the x-axis will display a corresponding message.		
	If you select Relative migration time , the x-axis will show relative migration time. The electropherograms will be aligned according to the alignment markers.		
	Note: The relative time scale can only be created if all selected samples have been run with the same alignment marker, have been analyzed, and the alignment markers have been correctly identified. Otherwise, the electropherograms will not be aligned and the x-axis will display a corresponding message.		
	If you select Absolute migration time , the electropherograms will not be aligned and the x-axis will show an absolute time scale.		
Zoom to alignment marker	Select this option to scale the electropherograms so that all electropherograms start with the lower alignment maker peak and end with the upper alignment marker peak (data outside of the alignment markers is truncated).		
	Note: This is only possible if all samples are analyzed.		
Individual scaling	Select this option to auto-scale the y-axis for each electropherogram individually.		
Show sample position	Select this option to display he sample position at the top of each electropherogram.		
Show sample information	Select this option to display the sample information at the top of each electropherogram.		
Show plate ID	Select this option to display the experiment name/plate ID at the top of the electropherogram.		
Show method	Select this option to display the applied method at the top of each electropherogram.		
Show	Select this option to show analysis details for the analyzed samples.		
analysis details	Select Mark detected peaks to display the apex markers of detected peaks.		

		Note: A second option Show median of size is available if the sample was analyzed with a smear analysis profile . Select this option then to display the markers with the median size of the detected smear peaks.	
		Select Show suspend integration intervals to display the suspend integration intervals.	
Electropherogram Superposition view	Use this option to include the superposition electropherogram of all selected samples which have the superimposed flag set.		
	Note: The superposition view is restricted to 12 samples. In process environment and in batch processing, the superposition is created only if the experiment contains 12 samples or less.		
	Note: Use the Use Images as Displayed option to report the electropherogram superposition as displayed.		
	Click the Image options button to specify the appearance of the reported electropherogram superposition. Image options are described below.		
	Note: The Image options button is disabled if the Use Images as Displayed option is selected.		
	Unit of x-axis	If you select Size , the x-axis will show a size scale based on the reference marker. The electropherograms will be aligned according to the alignment markers.	
		Note: The size scale can be only created if all superimposed samples have been run with the same alignment marker, have been analyzed with the same reference marker table, and the alignment markers have been correctly identified. Otherwise, the x-axis will show a message, instead.	
		If you select Relative mirgration time , the x-axis will show relative migration time. The electropherograms will be aligned according to the alignment markers.	
		Note: The relative time scale can only be created if all superimposed samples have been run with the same alignment marker, have been analyzed, and the alignment markers have been correctly identified. Otherwise, the electropherograms will not be aligned and the x-axis will show a message, instead.	
		If you select Absolute migration time, the electropherograms will not be aligned and the x-axis will show an absolute time scale.	
	Zoom to alignment marker	Select this option to scale the electropherograms, so that the electropherograms start with the lower alignment maker peak, and end with the upper alignment marker peak, if available (data outside of the alignment markers is truncated)	
		Note: This is only possible if all samples are analyzed.	
Show electrical current curve	Select this option to include a diagram of the superimposed electrical current measured during data acquisition. The electrical current curve will appear below the electropherogram superposition, aligned with the x-axis.		
-------------------------------------	---		
Show size marker peak labels	If this option is selected, and if exactly one size marker is among the superimposed samples, the peak labels will be displayed for the detected peaks of the marker sample. Select the units for the peak labels in the drop-down list.		

Note: In **RNA** mode, if you want the 28S/18S ratio or the RIS to be included in the report, select the overview option **Peak Calling Result Table** (refer to <u>Analysis of RNA samples</u> for information on how to analyze RNA samples).

Sample details section

The following information is included automatically for each sample:

Information	Description	Report section
Experiment Name/ Plate ID	Name/ID of the experiment/plate the sample data belong to.	Sample header
Position	Sample position on the plate.	Sample header
Run Number	If the same sample was processed multiple times, this number identifies the run that produced the sample data (for detailed information, refer to <u>Run parameters and</u> <u>result structure</u>).	Sample header
Run Entry Number	The run entry number of the plate (for detailed information, refer to <u>Run parameters and</u> <u>result structure)</u> .	Sample header
Run Date	Date and time the process was started.	Run

Select the following optional information that you want to include in the sample section:

✓ Sample Details
Start Results on New Page
Experiment Plate Comment
Run Information
Method Information
Cartridge Information
Sample Information
Sample Comment
Analysis Parameter
Reference Marker Table
Single Electropherograms
Single Gel Images
Lot Information
Result Table
Peak Calling Result Table
Smear Result Table

Note: If a result table option but no result table column is selected, the check box is marked yellow, and the report/export profile is invalid.

The optional fields are described below:

Start Results on New Page	If you select t a new page.	this option, the results for each sample will start on
Experiment Plate Comment	If you select this option, the experiment plate comment is included in the Sample Header section.	
Run Information	If you select this option, the following information is included in the Run section of the sample:	
	Rise time	The parameter for the applied Bessel filter (see also <u>Settings)</u> .
	Applied	The applied sample injection time.
injectio time	injection time	Note: This time can differ from the methods sample injection time if there was another sample injection time specified in the Process Profiles run parameters.(see <u>Selecting run parameters</u>).

	Applied separation time	The applied separation time during method run.	
		Note: This time can differ from the methods separation time if the user adjusted the separation time during run.	
	Processed by	The ID of the user who processed the sample.	
	Instrument ID	Serial number of the instrument the sample was processed with.	
Method Information	If you select this option, the following information is included in the Run section:		
	Applied method	Name of the method the sample was processed with.	
	Method injection time	Sample injection time as it was defined in the method.	
	Method injection voltage	Sample injection voltage as it was defined in the method.	
	Method separation time	Separation time as it was defined in the method.	
	Method separation voltaae	Separation voltage as it was defined in the method.	
Cartridge Information	If you select this option, the following information will be included in the Run section:		
	Cartridge ID	ID of the cartridge the sample was processed with.	
	Cartridge Calibration Status	Calibration status of the cartridge at process time: " OK ", " Not calibrated ", or " Conditional OK ." For information on calibration status, refer to section <u>Running the calibration wizard</u> .	
	Cartridge Expiry Date	The expiration date of the cartridge that was used for sample processing.	
Sample Information	The sample i section.	nformation is included to the Sample Header	
Sample Comment	The sample o	comment is included to the Sample Header section.	

Analysis Parameter	The analysis in the report	parameters applied to the sample will be included section Analysis .		
Reference Marker Table	The reference included.	e marker table used during analysis will be		
Single Electropherograms	Use this option	on to include the electropherogram.		
	Select Use In displayed.	nages as Displayed to print the electropherogram as		
	Click the Image options button to specify the appearance of the reported single electropherograms. Image options are described below.			
	Note: If Use Images as Displayed is selected, the gel lane and the electrical current are combined with the electropherogram if they are visible in the view.			
	Note: The Im Displayed οι	age options button is disabled if the Use Images as ption is selected.		
	Unit of x- axis	If you select Size , the x-axis will show a size scale based on the reference marker.		
		Note: The size scale can only be created if the sample has been analyzed with a reference marker table, and the alignment markers have been correctly identified. Otherwise, the x-axis will display a corresponding message.		
		If you select Relative migration time , the x-axis will show relative migration time.		
		Note: The relative time scale can only be created if the sample has been analyzed, and the alignment markers have been correctly identified. Otherwise, the x-axis will display a corresponding message.		
		If you select Absolute migration time [min], the x-axis will show an absolute time scale.		
	Zoom to Alignment Marker	Select this option to scale the electropherograms so that the electropherograms start with the lower alignment maker peak and end with the upper alignment marker peak (data outside of the alignment markers is truncated).		
		Note: This is only possible if all samples are analyzed.		
	Individual Scaling	Select this option to auto-scale the y-axis for each electropherogram individually.		

Show gel lane	Select this option to display the Gel representation below the electropherogram, aligned with the x- axis.	
Show Electrical Current	Select this option to display a diagram of the electrical current measured during data acquisition below the electropherogram or below the gel lane.	
Select Show analysis details to enable the following configuration options:		
Mark detected	Select this option to display the apex markers for detected peaks.	
peaks	If the option With label is selected, peak labels are displayed at the apex of each peak.	
	Note: Only peak labels that do not overlap with an adjacent label are displayed. Hover the mouse cursor over the peak apex to get a tool tip with the peak label.	
	Select the label units: size, absolute or relative migration time.	
	Note: The label can be displayed as Size only if the sample was analyzed with a reference marker (for details refer to <u>Size and Concentration</u> <u>determination</u>). Otherwise, "n/a" is displayed.	
	Note: A second option Mark median of size is available. If the sample was analyzed with a smear analysis profile , select this option to display the marker with the median size of detected smear peaks. You can switch the corresponding label on and off.	
	Select the option Mark peak start and end in order to mark the start and end points of detected peaks.	
Show areas of interest	For DNA mode only. If the sample was analyzed with a smear analysis profile , select Show areas of interest to display the areas of interest of detected smear peaks.	
Show suspend integration interval	Select this option to display the suspend integration intervals.	

	Show threshold	Select this option to display the peak threshold.
	Show baseline	Select this option to display the baseline.
Single Gel View	Use this optic samples.	on to report individual gel lanes for all selected
	Click the Imc reported sing below.	ige options button to specify the appearance of the le Gel images. Image options are described
	Note: The Ime Displayed op	age options button is disabled if the Use Images as ation is selected.
	Unit of y- Axis	If you select Size , the axis will show a size scale based on the sample peaks, if the sample has been analyzed with a reference marker table and the alignment markers have been correctly identified. Otherwise, the axis will display a corresponding message.
		If you select Rel. Time , the axis will show relative migration time, if the sample has been analyzed and the alignment markers have been correctly identified. Otherwise, the axis will display a corresponding message.
		If you select Abs. Time [min], the axis will show an absolute time scale.
	Zoom to Alignment Marker	Select this option to scale the lanes so that the lanes start with the lower alignment maker band and end with the upper alignment marker band (data outside of the alignment markers is truncated).
		Note: This is only possible if all samples are analyzed.
	Individual Scaling	Select this option to auto-scale the contrast for each gel lane individually.
	Analysis details	Select Highlight alignment markers to highlight in green the bands of the alignment marker peaks.
Lot Information	Select this op	tion to include the entered lot numbers.
Result Table	If you select t the report. Ac table (see be	this option, the peak result table will be included in dditional options appear to configure the result low).

Peak Calling Result Table	Select this option to include the peak calling result table in every sample section. Additional options can be selected.		
	Note: This option is only available if peak calling features are active.		
	Sample Information	Select this option to include the Sample Information column in the peak calling result overview table.	
	Found	Select this option to include the Found column for each peak of interest to the peak calling result overview table.	
	Size	Select this option to include the Size column for each peak of interest in the peak calling result overview table.	
	Concentrat ion	Select this option to include the Concentration column for each peak of interest in the peak calling result overview table.	
	Molarity	Select this option to include the Molarity column for each peak of interest in the peak calling result overview table.	
	Note: Calcule included if the	ated columns (such as Ratio, RIS) are automatically ey are part of the applied peak calling instruction.	
	Note: If a rep split between calculated co tables are mo	port includes too many peaks of interest, the table is a two peaks of interest. Sample information and the plumns appear only in the first table. The remaining arked with "(continued)" in the header.	
Smear Result Table	For a smear or gDNA analysis, use this option to include a smear result table. The following columns can be selected: Median Size, Conc. Area of Interest, Mol. Area of Interest, Start Area of Interest, End Area of Interest, % Conc. Area of Interest, NA Area of Interest, and % NA Area of Interest. Refer to <u>Smear</u> <u>result columns</u> for further details.		
	Note: The co and reordere	lumns of the Smear Result Table can be selected ad as described for the Result Table (see below).	
	Select the op include the ve sample.	tions Total Concentration and Total Molarity to alues for total concentration or total molarity of the	
Distribution Result Table	Use this optic distribution a	on for a distribution analysis to include the inalysis result table for a sample.	
	Note: This op features are a	otion is only available if distribution analysis active.	
	Additional of	ptions to select are:	

Show Profile Properties	Include the parameters of the distribution profile that was applied to the sample.	
Sample Information	Include the Sample Information column.	
Total Concentrat ion	Include the column Total Concentration of the sample.	
Total Molarity	Include the column Total Molarity of the sample.	
Sample Quality	Include the Sample Quality column. This is the overall quality assessment of the sample generated in the distribution analysis.	
Aol Concentrat ion	Include the Concentration column for each area of interest.	
Aol Molarity	Include the Molarity column for each area of interest.	
Aol Height	Include the Height column for each area of interest.	
Aol Height Check	Include the Height Check column for each area of interest. This is the quality assessment of the height of the area of interest.	
Ratio	Include the Ratio column for each defined ratio. Based on the selected calculation method, the column will show molarity or concentration ratio.	
Ratio Quality	Include the Ratio Quality column for each defined ratio. This is the quality assessment of the ratio.	
Note: If a report includes too many areas of interest or ratios, the table is split between two areas of interest or ratios. Sample information, total concentration, total molarity and sample quality columns only appear in the first table. The remaining tables are marked with "(continued)" in the table beader.		

Note: In **RNA** mode, if you want the 28S/18S ratio or the RIS to be included in the report, select the overview option **Peak Calling Result Table** (refer to <u>Analysis of RNA samples</u> for further information).

Select the following optional information that you want to include in the peak result table:

~	Resu	IIt Table
		Result Table Columns
	R	Size
	Ľ	Concentration [ng/µl]
		Molarity [nmol/l]
		Saturated
		Signal To Noise
		NA
		Area
		Normalized Area Percentage
		Ratio Normalized Area
		Height
		Height Percentage
		Resolution
		FWHM
		Start [min]
		Time [min]
		Stop [min]
		Rel. Time

Note: If the **Result Table** option is selected, but no result table column is selected, the **Result Table** check box is marked yellow, and the report/export profile is invalid.

The peak result table options are described below:

Size	Adds the result table column Size [bp] or Size [nt] for DNA or RNA mode, respectively.
Concentration	Adds the result table column Conc. [ng/µl] or [pg/µl] corresponding ti the unit used in the result table. If you select this option, the total concentration is added automatically as last value of the column.
Molarity	Adds the result table column Mol. [nmol/l] or [pmol/l] corresponding ti the unit used in the result table.
Height Percentage	Adds the result table column Height % .
Normalized Area	Adds the result table column NA .
Normalized Area Percentage	Adds the result table column NA %.
Signal to Noise	Adds the result table column S/N (Signal-to-noise ratio).
FWHM	Adds the result table column FWHM [sec] (full width at half maximum).
Resolution	Adds the result table column Res .
Ratio Normalized Area	Adds the result table column Ratio NA .
Time	Adds the result table column Time .
Rel.Time	Adds the result table column Rel. Time .
Start	Adds the result table column Start .
Stop	Adds the result table column Stop .
Height	Adds the result table column Height .
Area	Adds the result table column Area .

Note: Refer to section <u>Peak result columns</u> for more information about the columns.

Note: You can specify the field order of the result table in the report by drag and drop. Left-click on a column name and drag it to the new position.

6.4.11.3 Exporting data

To export data in the Analysis environment:

- 1. Load the samples using the **Experiment Explorer**. For more details on loading sample data, see the Loading sample data section.
- 2. Inspect the samples before exporting. For more details on data inspection, see the <u>Viewing sample data</u> section.

Note: The Gel view or the Electropherogram overview are both useful for this step.

3. Select the samples you would like to export. For more details on selecting samples, see the <u>Selecting</u> <u>samples for analysis or report</u> section.

The **Report** tab is located on the right side of the toolbar. You need to select a predefined report/export profile. The export settings of the selected profile are shown below the report settings, for more details, refer to the <u>Export options</u> section. Ensure that at least one of the export options is selected. If not, select another predefined report/export profile or modify the settings as required. Specify the directory in which the export files will be saved, for information on how to specify the export directory, refer to the <u>Export options</u> section.

 Select the Export options in the Report tab. To start exporting, click the Start Report/Export at the bottom of the Report tab.

Note: The **Report** tab is located on the right side of the toolbar. Ensure that at least one of the **Export** options or a predefined Report/Export Profile is selected. The **Export** options of the profile are shown below the **Report** options. For more details, refer to the <u>Export options</u> section. Specify the directory in which the export files will be saved. For information on how to specify the export directory, refer to the <u>Export options</u> section.

Note: The Start Report/Export button is disabled if no samples are selected in the view, see Step 3.

Export files will be generated automatically and stored in the specified directory.

6.4.11.4 Export options

The report/export settings are divided into two groups:

- Report Options
- Export Options

Note: You can collapse and expand each group by clicking ▼ and ▶ to the left of the group name.

Collapse the report options or scroll down to see the export options.

Export Options

 Export Options 	▼ Export Options
Directory NQIAxcel\ScreenGel 2.0\Data\Export Save to origin experiment directory XML Export XML Export XML Raw Data Export Image Options Use Images as Displayed Gel Image Overview Electropherogram Overview Electropherogram Superposition View Single Electropherograms Single Gel Images	Directory N\QIAxcel\ScreenGel 2.0\Data\Export Save to origin experiment directory XML Export Additional Output Formats (0) No additional Output DistributionAnalysis_CSV_Export PeakCalling_CSV_Export ResultTable_CSV_Export Gel Image Overview Gel Image Overview Electropherogram Overview Single Electropherograms Single Gel Images

The options are described below:

Directory	Select the directory that appears, navi	Select the directory to which the export files will be saved by clicking . In the dialog hat appears, navigate to the correct directory and click OK .				
Save to origin experiment directory	Check this box to selected, any selec	ck this box to save the report in the same directory as the experiment. If this option is cted, any selected path in Directory will be ignored.				
XML Export	If you select this op all general experin electropherogram	ption, an exp nent and san raw data. If	port file in XML format will be generated. This file contains nple details of the selected samples, except the samples are analyzed, it contains the analysis results.			
	An additional optic two formats. You c	on to select o can select all	additional output formats appears. The software provides of them.			
	DistributionAnalysi port	s_CSV_Ex	Use this option to export the distribution analysis overview result table in .csv format.			
	PeakCalling_CSV Export		Use this option to export the peak calling result table with all selected peak properties in .csv format.			
	ResultTable_csv_ta	ıble	Use this option to export the analysis results in .csv format (similar to the file format that was exported by the BioCalculator 3.2 Software).			
	SmearResultTable_ rt	_CSV_Expo	Use this option to export the smear result overview table in .csv format.			
	Note: You can cus export file and stor Script processing (style sheets, the co	tomize your re them in the (e.g. VB- or J onfigured exp	own output formats. Define xslt scripts based on the .xml e %DATA_DIR%\ReportExportProfile\XSLT_Export directory. ava-Script) within a XSLT style sheet is supported. In XSLT port directory is available as the parameter exportPath.			
	Note: To open gen and select Unicode Use General as the decimal separator values are not liste Excel and select th (UTF-8) format, whi	nerated .csv e (UTF-8) as e column dat in numbers d ed in columns ne correct de ich may resu	files in Excel, use the Import Data From Text/CSV option the file origin format. Fields are delimited by commas (,). a format. In the advanced options select period (.) as the and make sure not to use a thousands separator. If the s, highlight all cells, then use the Text to Columns function in limiters. This option does not allow selecting the Unicode It in unwanted characters in the column headers.			
XML Raw Data Export	If you select this op in XML format.	ption, electro	opherogram raw data of the selected files will be exported			
	An additional option one format.	on to select o	additional output formats appears. The software provides			
	RawData_CSV_ Export	Use this op (similar to t Software).	tion to export the electropherogram raw data in .csv format he file format which was exported by the BioCalculator 3.2			

Note: You can customize your own output formats. Define xslt scripts based on the xml raw data export file and store them in the %DATA_DIR%\ReportExportProfile \XSLT_RawExport directory. Script processing (e.g. VB- or Java-Script) within a XSLT style sheet is supported. In XSLT style sheets, the configured export directory is available as the parameter exportPath.

The image options are described below:

Use Images as Displayed	Use this option to obtain images that look like to the presentation in the QIAxcel ScreenGel software in terms of alignment, scales, labels, zoom, etc.				
	Note: This option Process Profile.	n is only available in the analysis environment, it may not be used in a			
Gel Image	Use this option t	o export the Gel image of all selected samples.			
Overview	Click on the Ima overview. Image	ige options button to specify the appearance of the exported Gel Image options are described below.			
	Note: The Image selected.	e options button is disabled when the Use Images as Displayed option is			
	Unit of y-axis	If you select Size , the y-axis will show a size scale based on the reference marker on both sides of the Gel image, also if the Use Images as Displayed option is selected. The lanes in the Gel image will be aligned according to the alignment markers.			
		Note: The size scale can only be created if all selected samples have been run with the same alignment marker, have been analyzed with the same reference marker table, and the alignment markers have been correctly identified. Otherwise, a corresponding message will appear.			
		If you select Rel. Time , the y-axis will show relative migration time. The lanes in the Gel image will be aligned according to the alignment markers.			
		Note: The relative time scale can only be created if all selected samples have been run with the same alignment marker, have been analyzed, and the alignment markers have been correctly identified. Otherwise, the lanes cannot not be aligned and the y-axis will show a corresponding message.			
		If you select Abs. Time [min], the lanes will not be aligned and the y-axis will show an absolute time scale.			
	Zoom to Alignment Marker	Select this option to scale the lanes, so that the lanes start with the lower alignment maker band, and end with the upper alignment marker band, if available (data outside of the alignment markers is truncated).			
		Note: This is only possible if all samples are analyzed.			
	Individual scaling	Select this option to auto-scale the contrast for each gel lane individually.			
	Show sample information	Select this option to include the sample information in each lane.			
	Show plate ID	Select this option to display the plate ID, the repeat number, and the entry number of each lane.			
	Show method	Select this option to display the applied method above the sample lane.			

Gel Layout	Select this option to determine the gel layout.
	Select Standard Layout to order the lanes from left to right, as they are ordered in the view.
	Select Horizontal plate layout to order the lanes starting with A1 at the upper left and ending with H12 at the lower right.
	Select Vertical plate layout to order the lanes starting with H1 at the upper left and ending with A12 at the lower right.
	Select Reverse vertical plate layout for the vertical plate layout but starting with A1 at the upper left and ending with H12 at the lower right.
	Select Split plate layout to split the vertical plate layout. The left portion will start with H1 at the upper left and end with A6 at the lower right. The right portion will start with H7 at the upper left and end with A12 at the lower right.
	Note: Positions for samples not selected remain empty.
Lanes per Row	Use this option to specify the number of lanes per row: 4, 6, 8, 12, 16, 24, 32, 36, or 48.
	Note: This option is only available if Standard layout is selected in the Gel Layout option.
Rows per File	Use this option to specify the number of rows to be included in one file (the image is split into several files according to the specified number of rows).
	Note: This option is not available if Standard layout is selected in the Gel Layout option.
Highlight alignment markers	Select this option to highlight in green the bands of the alignment marker peaks.

Electropherogram Overview	Select this opti samples. The c (at most 12 ele	on to export the electropherogram overview for all selected order is the same as in the view. One gallery overview page actropherograms) will always be exported to one file.
	Click on the In Electropherogr	nage options button to specify the appearance of the exported am overview. Image options are described below.
	Note: The Imag option is select	ge options button is disabled if the Use Images as Displayed red.
	Unit of x- axis	If you select Size , the x-axis will show a size scale based on the reference marker. The electropherograms will be aligned according to the alignment markers.
		Note: The size scale can only be created if all selected samples have been run with the same alignment marker, have been analyzed with the same reference marker table, and the alignment markers have been correctly identified. Otherwise, the x-axis will show a corresponding message.
		If you select Rel. Time , the x-axis will show relative migration time. The electropherograms will be aligned according to the alignment markers.
		Note: The relative time scale can only be created if all selected samples have been run with the same alignment marker, have been analyzed, and the alignment markers have been correctly identified. Otherwise, the electropherograms will not be aligned and the x-axis will show a corresponding message.
		If you select Abs. Time [min], the electropherograms will not be aligned and the x-axis will show an absolute time scale.
	Zoom to Alignment Marker	Select this option to scale the electropherograms so that the electropherograms start with the lower alignment maker peak, and end with the upper alignment marker peak (data outside of the alignment markers is truncated).
		Note: This is only possible if all samples are analyzed.
	Individual Scaling	Select this option to auto-scale the y-axis for each electropherogram individually.
	Show sample position	Select this option to show the sample position at the top of each electropherogram.
	Show sample information	Select this option to show the sample information at the top of each electropherogram.

	Show plate ID	Select this option to show the experiment name/plate ID at the top of the electropherogram.
	Show method	Select this option to show the applied method at the top of each electropherogram.
	Show analysis	Select this option to show analysis details for the analyzed samples.
	details	Select Mark detected peaks to show the apex markers of detected peaks.
		Note: A second option Show median of size is available. If the sample was analyzed with a smear analysis profile , select this option to display the markers with the median size of detected smear peaks.
		Select Show suspend integration intervals to display the suspend integration intervals.
Electropherogram Superposition view	Use this option have the "supe	to export a superposition view of all selected samples which rimposed" flag set.
	Note: The super environment and experiment con	prposition view is restricted to 12 samples. In the process and in batch processing, the superposition is only created if the atains 12 samples or less.
	Note: Use the U electropherogra	Jse Images as Displayed option to export the am superposition as displayed.
	Click the Imag electropherogre	e options button to specify the appearance of the reported am superposition. Image options are described below.
	Note: The Imag option is select	ge options button is disabled if the Use Images as Displayed red.
	Unit of x- axis	If you select Size , the x-axis will show a size scale based on the reference marker. The electropherograms will be aligned according to the alignment markers.
		Note: The size scale can only be created if all superimposed samples have been run with the same alignment marker, have been analyzed with the same reference marker table, and the alignment markers have been correctly identified. Otherwise, the x-axis will show a message, instead.
		If you select Rel. Time , the x-axis will show relative migration time. The electropherograms will be aligned according to the alignment markers.

		Note: The relative time scale can only be created if all superimposed samples have been run with the same alignment marker, have been analyzed, and the alignment markers have been correctly identified. Otherwise, the electropherograms will not be aligned and the x-axis will show a message, instead.
		If you select Abs. Time [min], the electropherograms will not be aligned and the x-axis will show an absolute time scale.
	Zoom to alignment marker	Select this option to scale the electropherograms, so that the electropherograms start with the lower alignment maker peak, and end with the upper alignment marker peak, if available (data outside of the alignment markers is truncated).
		Note: This is only possible if all samples are analyzed.
	Show electrical current curve	Select this option to include a diagram of the superimposed electrical current measured during data acquisition. The electrical current curve will appear below the electropherogram superposition, aligned with the x-axis.
	Show size marker peak labels	If this option is selected, and if exactly one size marker is among the superimposed samples, the peak labels will be displayed for the detected peaks of the marker sample. Select the units for the peak labels in the drop-down list.
Single Electropherograms	Select this opti separate files.	on to export the electropherograms for all selected samples in
	Use the Use Im displayed.	ages as Displayed option to export the electropherogram as
	Click on the In single electrop	nage options button to specify the appearance of the exported herograms. Image options are described below.
	Note: If the Use electrical curre in the view.	e Images as Displayed option is selected, the gel lane and the nt are combined with the electropherogram if these are visible
	Note: The Imag option is select	ge options button is disabled if the Use Images as Displayed red.
	Unit of x- axis	If you select Size , the x-axis will show a size scale based on the reference marker.
		Note: The size scale can only be created if the sample has been analyzed with a reference marker table, and the alignment markers have been correctly identified. Otherwise, the x-axis will show a corresponding message.
		If you select Rel. Time , the x-axis will show relative migration time.

	Note: The sample ha been corre correspon	relative time scale can only be created if the is been analyzed and the alignment markers have ectly identified. Otherwise, the x-axis will show a ding message.		
	lf you sele time scale	ct Abs. Time [min], the x-axis will show an absolute .		
Zoom to Alignment Marker	Select this electrophe peak, and available truncated).	Select this option to scale the electropherograms, so that the electropherograms start with the lower alignment maker peak, and end with the upper alignment marker peak, if available (data outside of the alignment markers is truncated).		
	Note: This	is only possible if all samples are analyzed.		
Individual scaling	Select this electrophe	option to auto-scale the y-axis for each program individually.		
Show gel lane	Select this electrophe electrophe	Select this option to export the gel lane combined with the electropherogram (the gel lane is below the electropherogram).		
Show electrical current curve	Select this the electro	Select this option to export the electrical current curve below the electropherogram or below the gel lane.		
Select Show ar samples:	nalysis deta	ils to enable the following options for analyzed		
Mark detected	peaks	Select this option to display the apex markers for detected peaks.		
		If the option With label is selected, peak labels are displayed at the apex of each peak.		
		Note: Only peak labels that do not overlap with an adjacent label are displayed. Hover the mouse cursor over the peak apex to get a tool tip with the peak label.		
		Select the label units: size, absolute or relative migration time.		
		Note: The label can be displayed as Size only , if the sample was analyzed with a reference marker (for details refer to <u>Size and Concentration</u> <u>determination</u>). Otherwise, "n/a" is displayed.		

				Note: A second option Show median of size is available. If the sample was analyzed with a smear analysis profile , select this option to display the markers with the median size of detected smear peaks. You can switch the corresponding label on and off.		
				Select the option Mark peak start and end to mark the starting and ending points of detected peaks.		
		Show areas	s of interest	If the sample was analyzed with a smear analysis profile , select Show areas of interest to display the areas of interest of detected smear peaks.		
		Show suspe integration	end intervals	Select this option to display the suspend integration intervals.		
		Show threst	nold	Select this option to display the peak threshold.		
		Show base	line	Select this option to display the baseline.		
Single Gel	Use this optic	nis option to individually export the gel lanes for all selected samples.				
Images	Click on the l images. Imag	l mage option Je options ar	ecify the appearance of the exported single Gel elow.			
	Note: The Ime selected.	age options	button is disab	led if the Use Images as Displayed option is		
	Unit of y-axis		If you select Size , the axis shows a size scale based or sample peaks, if the sample has been analyzed with a marker table and the alignment markers have been cor identified. Otherwise, the axis will show relative migrat if possible.			
			If you select I if the sample have been co absolute time	Rel. Time , the axis will show relative migration time, has been analyzed and the alignment markers prrectly identified. Otherwise, the axis will show an scale.		
			If you select / scale.	Abs. Time [min], the axis will show an absolute time		
			Note: This op option is sele	tion is not available if the Use Images as Displayed cted.		
	Zoom to Alig Marker	nment	Select this op the lower alig alignment ma truncated).	tion to scale the lanes so that the lanes start with gnment maker band, and end with the upper rker band (data outside of the alignment markers is		
			Note: This is	only possible if all samples are analyzed.		

Individual Scaling	Select this option to auto-scale the contrast for each gel lane individually.
Show analysis details	Select Highlight alignment markers to highlight in green the bands of the alignment marker peaks.

The image export settings are described below. They appear if at least one image type is selected for export.

File Type Select the file type for the image files.

Resolution Select the resolution for the image files; the available resolutions are: 75 dpi (dots per inch), 150 dpi, 300 dpi, and 600 dpi. For small bands, use 150 dpi.

6.4.11.5 Modifying a Report/Export Profile

To modify a Report/Export Profile:

- 1. Open the **Report** tab.
- 2. From the Report/Export Profile drop-down list, select the profile you want to modify.
- 3. Change the profile options according to your needs.

See the <u>Report options</u> and <u>Export options</u> sections for more detailed information.

Note: The system notifies modifications on the profile by displaying a **"*"** at the beginning of the profile name.

4. To save the modified Report/Export Profile, click Save as, then click OK in the Save Profile dialog.

Analysis	Report	Peak Calli	Peak Calling		ropert	ies	×
Report/Ex	port Profile						
*NewReportExportProfile			•		Save	as	N
				_			N۲

Note: If you want to use the modified report/export options only once, click **Start Report/Export** without saving the modifications. However, your changes will be lost when you select another Report/Export profile.

Note: Report profiles provided by QIAGEN cannot be modified. However, to save your modifications, enter a new unique profile name in the **Save Profile** dialog. A new profile will be created containing your modifications. This is only possible for users with the **Developer** role.

6.4.11.6 Creating a new Report/Export Profile

Note: Only users with the Developer and Administrator user role can create a new report/export profile.

To create a new report/export profile proceed as follows:

- 1. Open the **Report** tab.
- 2. Select a report/export profile.

Select the profile from the **Report/Export Profile** drop-down list. The selected profile serves as a template for the creation of the new profile.

Note: To create a report/export profile from scratch, select NewReport/ExportProfile. The system will show *NewReport/ExportProfile after selection.

3. Set the profile options according to your needs. See the <u>Report options</u> and <u>Export options</u> sections for detailed information about report/export options.

Note: The system notifies modifications on the profile by displaying a "*" at the beginning of the profile name.

4. Save the modified report/export profile under a new name.

Click the Save as button and enter a new unique profile name in the Save Profile dialog that appears.

Analysis	Report	Peak Callii	Peak Calling		×
Report/Exp	port Profile	-			
*NewReportExportProfile			•	Save as	N
					NC.

6.5 Service

The **Service** environment provides functionalities for cartridge calibration, troubleshooting and maintenance tools for the QIAxcel Connect instrument.

On the left side of the **Service** environment, a **Status Information** panel is displayed (see <u>Status Information</u> panel for more information).

2 QIAxcel ScreenGel		D X
File View Help	Service Configuration	, Notifications
Status Information	Calibration System Check Maintenance Terminal Logs	
	System Check Instructions	
Instrument Status Cartridge Status Cartridge D Cartridge D Cartridge D Cate Status Calibration Runs Calibration Runs Left 101 5 Expiration Status Valid Not OK Calibration Details	Complete Check Detector Test Filter Check Movement Check Leakage Test Image: Test The tests will take approximately 4.5 hours. Press the "Start" button to run the tests. Start	
	Login: 30.06.2022 12:03.03 Name: Administrator Mode: DNA QIAsph	ere Base: Connected

Note: The Terminal and Logs screens of the Service environment may be accessed by service personnel only and are not described in this manual.

6.5.1 Calibrating a cartridge

Each new cartridge requires intensity calibration before the first sample analysis. The calibration normalizes the intensities of each capillary by applying a factor to each subsequent run. In this way, the natural intensity variations between individual capillaries are corrected. Each cartridge intensity calibration is stored in a single file named **<cartridge-id>_<instrument-id>.xcc**. This file is saved in the directory %DATA_DIR% \CartridgeCalibrationData\.

Note: To inspect calibration data, switch to the analysis environment and click the **Load experiment** button of the **Experiment Explorer**. In the file dialog that appears, select the path ending with "[**Calibration results**]."

Note: If a different computer is used to run a new experiment, the calibration file has to be transferred to the new computer. Otherwise, a recalibration of the cartridge is required.

6.5.1.1 Running the calibration wizard

Intensity calibration of the cartridge is performed in the Calibration screen of the Service environment.

To start the calibration:

- Load the QX Intensity Calibration Marker onto the buffer tray position MARKER2 as described in the respective QIAGEN kit handbook.
- 2. Open the calibration dialog by clicking the Start calibration button.

Note: The total run time for the calibration routine is about 16 minutes.

3. Confirm that the **QX Intensity Calibration Marker** is loaded. Optionally, enter the **lot ID**. Start the calibration by clicking **OK**.

	Calibration Marker
-	to buffer tray position:
	 MARKER 2
	Calibration Marker Lot ID

Note: If the **QX Intensity Calibration Marker** is not loaded correctly in the buffer tray position, cancel the dialog. Load it again and repeat step 1.

4. Once the calibration is complete, the calibration results are displayed as a table next to the Gel/ Electropherogram view. The result table shows the area, calibration factor, and the result (Passed or Failed) for each channel.

Calibratio	on	Syster	n Checl	k M	aintenar	nce	Termina		Logs							
	٩	к <u></u>)		¥ \$												
	C1	C2	C3	C4	C5	C6	C7	C8	C9	C10	C11	C12				
													Cartr C210	idge ID 0622001		
													Ch.	Area	Cal. Factor	Result
													1	0.0048	0.2545 0.3949	Passed Passed
									_				3	0.0050	0.1987	Passed
													4	0.0048	0.2240	Passed
													5	0.0050	0.2029	Passed
													6	0.0049	0.2546	Passed
													7	0.0050	0.1967	Passed
													8	0.0050	0.2114	Passed
													9	0.0052	0.2575	Passed
													11	0.0051	0.2394	Passed
													12	0.0001	0.2430	Passed
													12	0.0040	0.2072	1 assea
															Ļ	
	Estima	ated Time	Remain	ning	_				C	alibratio	n runs f	inished.			Finish	Calibration

Note: A successfully calibrated cartridge should have a normalized area in a range as described in the respective kit handbook.

Note: If one or more channels fail, the calibration process should be repeated. If the issue persists, please contact QIAGEN Technical Services or refer to the **Troubleshooting** section in the handbook supplied with the QIAxcel Kit you are using.

5. Click **Finish Calibration** to accept the calibration result. The cartridge is now calibrated.

Note: Discard the calibration result if one or more channels fail.

Note: A **Developer** user can accept calibration results even if one or more channels have failed. In this case, the calibration status will be marked as **Conditional OK**. Only users with the **Developer** user role can start processes using cartridges with a calibration status **Conditional OK**.

6.5.1.2 Recalibrating a cartridge

To recalibrate a cartridge, repeat the procedure detailed in <u>Running the calibration wizard</u>. The calibration results of previous calibration procedures are discarded when recalibrating a cartridge.

Note: It is possible to calibrate a cartridge that has no calibration runs left. In this case, three regular runs are used instead of one calibration run.

6.5.2 System check

The **System Check** screen of the **Service** environment provides several procedures for detecting problems with the QIAxcel Connect instrument:

• Complete Check

The complete check performs all the system checks listed below.

Detector Test

The detector test checks the raw count of all 12 channels.

• Filter Check

The filter check detects blockage of the purge filter.

Movement Check

The movement test verifies that the sample tray can be moved properly and that the position sensors work as expected.

• Leakage Test

The leakage test checks for N₂ leakage.

Note: The test results are stored in the directory %DATA_DIR%\SystemTest\.

Note: The troubleshooting folder functionality is described in the Troubleshooting folder and log files section.

6.5.2.1 Complete check

A Complete Check performs all available system checks: Detector Test, Filter Check, Movement Check and Leakage Test.



Note: The complete check takes approximately 4.5 hours.

6.5.2.2 Detector test

The **Detector Test** verifies that the detectors of all 12 channels of the QIAxcel Connect instrument work as expected. It may be performed when one or more channels in a new cartridge do not provide data signals and the baseline is flat.

Perform the **Detector Test** by following the instructions shown on the screen. The test takes only several seconds. At the end of the test, the acquired data is shown, and the results are stored.

Note: If not instructed otherwise, perform this test with a cartridge inserted and latched.

Calibration System Check	Maintenance Terminal Logs										
System Check Instructions											
Complete Check Detector Test	Detector Test Results The acquired values shou Otherwise contact QIAGE	Id be greater than or equal to 200.									
Filter Check	The following values were	acquired during the detector test:									
Movement Check	C01 C02 C03 C04 C05										
Leakage Test	2108 6465 9395 21458 27463 4892 128 30990 4960 9701 5316 27600 21039 13049 4501 3	109/6 155/9 11401 25806 32129 10385 23258 16793 19881 16678 16405 5568 16713 3399 31941 18523 6035 18353 760 16236 2983									
	22373 28026 27069 27425 12123 24701 29287 18377 25144 20315 2	18602 19764 1764 6622 7939 6859 30073 28176 19780 29860 2747 21052 4890 11344									
	4944 25518 29540 19199 21604 7973 27990 23019 11297 21836 12116 244 16057 6704 872 2	23112 10840 32033 17222 24329 1044 9000 851 8663 7408 30313 23969 4415 11779 26390 26496 1937 6239 21545 21148 3194									
	18027 4645 3122 24909 18277 10042 2824 5965 18331 719	5967 18859 4671 1985 12015 505 20495 19087 11816 7681 23800 6464 19156 19713									
	26178 31023 25874 2009 12000 31584 28716 27707 16488 9157 2 3851 17793 26388 8029 8918	931 2 21089 7811 25422 140 28770 28466 11468 21263 18181 27414 8829 26410 18780 22184 12572 19425 10658 16609 31115									
	29698 26083 14942 24003 29572 1 32740 30863 30808 14099 3658	19237 28547 24277 8463 31105 25533 6019 9563 17721 17629 15594 29131 13885 20758									
	28528 6847 8576 9701 30377 9059 32370 14892 2366 6775	12977 23313 22076 10951 12850 18236 5866 7145 9969 2995 28682 24220 1494 9802									
		ОК									
	Finished! The detector test has finis	hed and the results are saved.									
		Restart	•								

6.5.2.3 Filter check

In rare instances, the purge filter that sits inside the QIAxcel Connect instrument may become blocked with gel carryover. This blockage can be diagnosed using the **Filter Check**, which takes about two minutes. At the end of the check, the filter status (**Passes** or **Failed**) is shown and the results are stored.

Note: A cleaning tissue is needed for this check.

To perform the Filter Check, follow the instructions shown in the wizard:

Calibration System Check	Maintenance	Terminal	Logs				
System Check Instru	ctions						
Complete Check Detector Test	0	Filter Check The instrume	c ent filter is c	checked in this test	t.		
Filter Check	>					Start	R
Movement Check							
Leakage Test							
							•

6.5.2.4 Movement check

The **Movement Check** verifies that the tray motors and tray sensors work as expected. This test is fully automated and requires no user interaction except removing the cartridge. It takes several seconds. At the end of the test, a test summary (**Passed** or **Failed**) is shown and the results are stored.



6.5.2.5 Leakage test

The **Leakage Test** checks for leakages in the N_2 tubing of the QIAxcel Connect instrument. This fully automated test takes approximately 240 minutes. After the test, the leakage rate is calculated if a leakage is detected and the results of the test are stored.



6.5.3 Maintenance

The **Maintenance** screen of the **Service** environment provides several maintenance and troubleshooting procedures for the QIAxcel Connect instrument and cartridges:

- Motor teaching (for service personnel only; not described here)
- Purge
- Long purge
- Empty N₂ bottle
- Instrument ID
- Troubleshooting folder

6.5.3.1 Purge

The **Purge** method allows the user to quickly remove air bubbles from the gel cartridge and to clean the capillaries. The process is fully automated and takes less than a minute.



6.5.3.2 Long purge

The **Long Purge** method allows the user to remove air bubbles from the gel cartridge and to clean the capillaries more thoroughly. The process is fully automated and takes about 3 minutes.



6.5.3.3 Empty N2 bottle

To remove a pressurized N_2 cylinder, the remaining pressure has to be released first. The **Empty N₂ Bottle** function purges the nitrogen, which takes about a minute.

Note: Please remove the QIAxcel gel cartridge before performing this action.



Important: Repeat this operation until no hissing sounds can be heard at the end of the purge operation.
6.5.3.4 Setting the instrument ID

Setting the **QIAxcel instrument ID** (serial number) is only possible if the instrument is connected and the instrument ID has not been set yet. The instrument ID can be found on a label at the rear side of the instrument housing.

The instrument ID is required to identify each instrument and to check that a cartridge is calibrated on the particular instrument being used (see <u>Calibrating a cartridge</u>).

Important: Make sure you have entered the correct instrument ID before clicking the **Apply** button. Once set, the instrument ID cannot be changed again, except by a QIAGEN service personnel.

Se	rvice Configuration		
	Calibration System Check	Maintenance	Terminal Logs
	Maintenance Instruction	s	
	Motor Teaching		Instrument ID
	Purge	0	Instrument is not connected.
	Long Purge		
	Empty N2 Bottle		Note: Once set, the instrument ID cannot be changed again, except by QIAGEN service personnel.
	Instrument ID		Apply Cancel

6.5.3.5 Troubleshooting folder and log files

The **Troubleshooting** folder function allows export of log files and test results produced within a specified time period.

To export log files or test results:

- 1. Switch to the Service environment.
- 2. Select the Maintenance or System Check screen.

3. Click the **Troubleshooting folder** button at the bottom of the screen. This opens the **Troubleshooting Setup** dialog:

i Troubleshootin	ng Setup
Log Period	
Start Date End Dat 06.09.2021 15 07.09.2	e 021 15
Results	Logging Information
Detector Test	Main 🔺
Filter Test	Error
Leakage Test	Ivd
Sensor / Movement Test	✓ Instrument
•	User
·	· <u> </u>
	OK Cancel

- 4. Select the Start and End Date during which test results were stored or a problem occurred. **Start Date**: yesterday, and **End Date**: today is preselected.
- 5. In the **Results** column, select the test results to be exported.
- 6. In the Logging Information column, select all necessary log file types.
- 7. Click **OK**.

The exported files are stored in the Troubleshooting directory defined in the Settings.

8. To navigate to the **Troubleshooting** directory, select the **Open Data Directory > Troubleshooting** option from the **File** menu.



9. Select the just created Troubleshooting_..._.zip file.

Refer to the <u>Settings</u> section for information about log file levels.

6.6 Configuration

Application settings, users, profiles, and QIAsphere settings are managed in the **Configuration** environment: **Note**: The QIAsphere section is only available for users with **Administrator** user role.

QIAxcel ScreenGel File View Help	<u> </u>	*	1	¢۵				 2	َ× ُ
Process	Analysis	Service	Confi	iguration				Notifica	ations
Settings Profile Manager	User Manager	QIAsphe	re						
Settings Scope									
 Use global settings Use private settings 									
Settings									
21 CFR Part 11 Support	AutoLock 1800 sec		Ma	ximum num 3	nber o	of failed logins			Í
Log Files	Maximum log 10 MBy	g file length te	Ma	ximum log	files (per type			
	Log Type	Log File Ler	vel	Main Log Le	evel	Error Log Leve	el		
	Instrument	Warning	▼ E	Error	•	Error	•		
	IVD Demistration	Information	-	nformation	•	Error	*		
	Persistence	None	-	ntormation	•	Error	-		
	Oiasohere	Information	-	Vone	-	Error	· · ·		
	Service	None	- I	nformation	•	Error	•		
	User	None	- I	nformation	•	Error			
	View	None	-	None	•	Error	•		
	Other	None	- E	Error	-	Error	•		
Generate Experiment Name / Plate ID	✓ Cartridge ✓ Date		Time Counte	Exan	mple 120A9	19_2022-06-30_	12-14-39		
Sample Selection	Enable in	put fields for B	luffer lo	ot IDs					
								ОК	Cancel
							Login: 30.06.2022 12:03:03 Name: Administrator Mode: DNA QIA:	sphere Base: C	Connect

6.6.1 Settings

The **Configuration** environment of the QIAxcel ScreenGel software provides several settings options as described below.

Note: Global settings options are read-only for **Operator** users. Refer to the <u>User roles</u> section for more information.

Use global settings	The global settings option is used to share both general settings and profiles among several users.
Use private settings	When using private settings, both the general settings and the created profiles of the user are private.
	Note: The private settings option can be activated only if the user has the right to maintain private settings (see <u>User management</u>).

The settings are grouped according to functionality. The following table gives an overview of the sections:

21 CFR Part 11 Support	Controls functionality required by 21 CFR Part 11 (FDA guidelines on electronic records). Select this option to enable auto-lock and to limit the number of failed logins.				
	Option	Description			
AutoLock The system au specified peric unlock, the use section for mo		The system automatically locks the QIAxcel ScreenGel after a specified period of idle time, except when a process is running. To unlock, the user must log in again. Refer to the <u>User authentication</u> section for more information about how to unlock.			
		Note: Idle time means time without user action or instrument action.			
	Maximum number of failed logins	After the specified number of failed logins, the system automatically locks the user account. It can only be unlocked by the administrator.			

Log files The QIAxcel ScreenGel writes all log messages to one main log file. In addition, separate log files can be created for individual log message types (IVD, Instrument, etc.) and for errors. The language for all log files is English. An exception is the separate IVD log file (audit trail). If the software language is other than English, the file exists twice, one in English, and one in the selected language.

Start with the levels as shown in the following image:

Maximum log	g file length	N	Maximum log files per type			
10 MBy	te		2			
Log Type	Log File Lev	/el	Main Log Lev	vel	Error Log Lev	/el
IVD	Information	-	Information	-	Error	•
Instrument	Verbose	-	Information	-	Error	•
User	None	-	Information	-	Error	•
View	None	-	Information	-	Error	•
Presentation	None	-	Information	-	Error	•
Service	None	-	Information	-	Error	•
Persistence	None	-	Information	-	Error	•
Other	None	•	Information	•	Error	•

The size of a log file is limited; this size is configured by the Maximum log file length. To always have the latest log files available, a new file will be created when the maximum size has been reached. The number of log files to be kept is configured by the Maximum log files per type. When, for a particular log file type, the maximum number of files has been reached, the oldest file of that type will be deleted.

The verbosity increases from Critical to Verbose. When Critical is selected, only critical log messages are written to the log file. When Verbose is selected, all log messages are written to the log file.

Note: The **Verbose** level can decrease the performance of the QIAxcel ScreenGel software. Keep the information as short as possible for each log and file type. Start with the levels as shown in the image above.

The main log file gives an overview of the last actions in the software. The verbosity of the main log file is set in the **Main Log Level** column of the above table. This describes how much information of a log type will be added to the main log file.

The verbosity of the individual log files is set in the **Log File Level** column. Increase the log level for special types of error tracking.

The error log file gives an overview of the last errors. The verbosity of the error log file is set in the **Error Log Level** column of the table shown above. This describes how much information of a log type will be added to the error log file. Typically, only the verbosities **Error** and **None** are used for the error log file.

Log Type	Description					
IVD	Audit trail. If this is also included in the main log file, it helps to understand the last actions.					
Instrument	In case of communication problems, it can be useful to increase the Log Fil Level to Verbose for a certain time span and to reproduce the situation. We these settings, the Instrument log file reaches the maximum log file length ve soon (after one run). Therefore, disconnect from the instrument just after the situation occurred, export the log files, and return to verbosity Warning. Us verbosity Error in the columns Main Log Level and Error Log Level to keep the overview character of the main log file and the error log file.					
User	Traces the user management actions.					
View	Information from the graphical user interface of the software.					
Presentation	Information from the graphical user interface of the software.					
Service	Information from the software logic.					
Persistence	Information about loading and saving. Increase verbosity of the Log File Level if there are problems with loading and saving experiments or profiles.					
Other	All other information.					
Refer to the <u>Trouk</u>	oleshooting folder and log files section for information how to export log files.					
When enabled, c	lefines how automatically generated plate IDs are constructed.					
The following opt	tions define construction of the plate ID.					
Generate Experim Name / Plate ID	Image: Cartridge ID Image: Time Example Image: Date O Counter 091120A99_2021-05-07_13-23-43					

Option

Description

Cartridge ID

If this option is selected, the generated plate ID will contain the **Cartridge ID** at the start.

Date	If this option is selected, the generated plate ID will contain the date of the data acquisition. If Cartridge ID is included, the date will be placed next to the Cartridge ID .
Time or Counter	Specifies how the uniqueness of the plate ID is achieved: with the time of the process start (including hours, minutes, and seconds) or with the counter incremented for the next plate ID generation.
	Note: If the plate ID is edited during process setup after automatic generation, the time will not reflect the exact process start time, but will reflect the time the plate ID was generated during process setup.

The **Example** field shows what the generated plate ID may look like.

Sample Specifies which optional information can be entered at the Sample Selection screen of the process wizard.

	Option	Description				
	Enable input fields for Buffer lot IDs	Select this option only, if you want to provide lot IDs for Buffers . If this option is not selected, you cannot provide lot IDs for Buffers during process setup.				
	Enable input fields for Marker lot IDs	Select this option only, if you want to provide lot IDs for Markers . If this option is not selected, you cannot provide lot IDs for Markers during process setup.				
	Provide Sample Information	Select this option, if you usually provide sample information. Even if this option is not selected, you can provide sample information, but you have to select the Provide Sample Information option at the Sample Selection screen of the process wizard manually.				
	Prevent modification of imported sample information	Select this option if you want <u>sample information</u> from imported rack files to be read-only .				
Instrument	Option	Description				
manomeni	Latch cartridge automatically	If this option is selected, the system latches the cartridge automatically after a cartridge is inserted and the cartridge door is closed.				
	Perform calibration automatically	If this option is selected, the system performs cartridge calibration automatically, if the inserted cartridge has not yet been calibrated.				
	COM Channel	Specifies the COM channel the QIAxcel Connect instrument is connected to. After login, the system automatically tries to connect to the QIAxcel Connect instrument using the specified COM port .				

Rise Time The rise time is a parameter of the **Bessel** filter applied to the raw data during acquisition. This parameter should not be changed unless you know the function of the **Bessel** filter and the effects of changes to the rise time parameter.

Directories Specifies the directories for acquired raw data and all other application data.

The base directory for the directories listed below is initially (i.e., after the QIAxcel ScreenGel installation) the **Application Data** directory which is specified during the <u>Installation of the</u> <u>ScreenGel Software</u>. All these directories are configurable.

The following directories can be specified:

Directory name	Description
Export	Default export directory. This is the default directory in which export files are generated, unless a different directory is chosen in the export profile.
Report	Default report directory. This is the default directory in which report files are generated, unless a different directory is chosen in the report profile.
Header & footer	Directory for report header and footer. During report generation, the system looks here to find a header and footer to be included in the report.
Sample information	Default directory for importing or exporting sample information (see <u>Running a process</u>). An example import rack file can be found in the default directory %DATA_DIR %\SampleInfo.
Troubleshooting	Directory where files created by the <u>Troubleshooting folder</u> are stored.
Experiment	Directory that contains 3 sub-directories: DNA and RNA . The experiment data files (both acquired as a process run result or combined in the Analysis environment) are saved by default in these sub-directories (according to the application mode), unless a different directory was chosen in the Process Profile.
Local Experiment	During a process, the experiment is saved in this directory. It is hard-coded to %DATA_DIR%\LocalExperiment. After the process has finished, the experiment is moved to the experiment path defined in the Process Profile. Only if this transfer fails (e.g. if it is a remote drive, and someone has tripped over the network cable), the experiment can still be found in the Local Experiment directory.

	ExportTransform	ationScripts	This directory contains XSLT scripts for transforming export XML files. XSLT is a declarative language that enables transformation of the export XML file into a different format, for example, another XML, HTML, or plain text. Some example XSLT files can be found in the default directory %DATA_DIR%\ReportExportProfile\XSLT_Export, for creating files in .csv format from the export.
	ExportRawTrans s	formationScript	This directory contains XSLT scripts for transforming raw data export XML files. An example XSLT file can be found in the default directory %DATA_DIR%\ReportExportProfile \XSLT_RawExport, for creating a file in .csv format from the raw data export.
Acoustic Feedback	Option	Description	
	Enable the selection of a file for Play sound when	Browse and is finished. Ve	select here the file which should be played when experiment alid file formats are .mp3 , .wma , and .wav .

Note: The directories above can be opened in the Windows Explorer directly from QIAxcel ScreenGel Software through the **File/Open Data Directory** menu item.

Most settings changes take immediate effect after clicking **OK**. The following table lists settings that require additional actions:

Generate Experiment Name/Plate ID section	Active after next login.
Instrument-COM channel	Active after reconnection with instrument.
	Note: After clicking OK , the system asks you to reconnect to the new COM channel.
Directories	Next login.

experiment ends

Changing the language

For your convenience, the QIAxcel ScreenGel software graphical user interface (GUI) is available in different languages (ES, JP, DE, IT, FR) in addition to the original English.

Effort has been made to provide accurate translation of the GUI, however please consult the English user manual for comprehensive instructions and support.

To view or to change the languages supported by the software:

- 1. In the File menu select the Language option.
- 2. Select the language in the drop-down box.
- 3. Close and restart the software.

6.6.2 Profile management

Several dialog boxes of the QIAxcel ScreenGel allow the creation of new profiles, for example, analysis profiles, size marker tables, and reference marker tables. The **Profile Manager** lists all profiles grouped by profile type. Profile details can be printed and profiles can be deleted using the **Profile Manager**.

naly	vsis Profile		
	Name	Created	Modified
1	Default DNA Analysis	22.02.2022 23:16:47	25.01.2022 08:33:01
1	Default RNA QC	22.02.2022 23:16:47	25.01.2022 08:33:11
1	DNA Fast Analysis	22.02.2022 23:16:47	25.01.2022 08:33:28
	DNA High Resolution Analysis	22.02.2022 23:16:48	25.01.2022 08:34:08
	DNA High-Sensitivity Analysis	22.02.2022 23:16:48	25.01.2022 08:39:20
	DNA Large Fragment Analysis	22.02.2022 23:16:48	25.01.2022 08:39:30
	DNA QC	22.02.2022 23:16:48	25.01.2022 08:39:47
	gDNA Analysis	22.02.2022 23:16:48	25.01.2022 08:40:52
	NGS Library Analysis	22.02.2022 23:16:48	25.01.2022 08:41:19
	QC Instrument High-Sensitivity	22.02.2022 23:16:49	25.01.2022 08:41:27
i I	RNA High-Sensitivity Analysis	22.02.2022 23:16:47	25.01.2022 08:41:33

Note: Profiles that are marked with a padlock symbol are default profiles provided by QIAGEN. These profiles cannot be deleted.

6.6.3 User management

Administrators have access to the User Manager tab, in which user accounts can be modified:

	>>>> 📈	is Service	ر Configurati	on									
QIAGEN ettings Pro	ofile Manager User Mana	ger QIAsph	here										
User ID	Role	First Name	Last Name	DNA	RNA	Private Settings	Skip Run Checks	Revise Sample Informatio	on Accept incomplete experiments	Last Password Change	Created	Modified	
dmin	Administrator	Admin	Admin	~	~					27.08.2021 10:38:10	27.08.2021 10:37:38	27.08.2021 10:45:01	
dministrator	Administrator			~	~	¥		¥		01.01.1980 00:00:00	21.02.2021 00:00:00	27.08.2021 10:22:21	
aneDoe	Developer	Jane	Doe	~	~					27.08.2021 10:50:34	27.08.2021 10:49:56	27.08.2021 10:50:34	
ohnDoe	Operator	John	Doe	~	~					27.08.2021 10:51:35	27.08.2021 10:51:00	27.08.2021 10:51:35	
] Show deacti	ivated user accounts (4)	Define user acc	count									0	4
		User ID			Initia	al password	First name	Mod	e Options DNA 🗌 Activated 3				
		Role			Con	firm password	Last name	F	RNA Allow private settings				
				v	r -				May skip confirmation	of run checks			
									May revise sample info	rmation after run			
									May accept incomplete	experiments			
												2 Apply	Ca

Adding users

To add a new user:

- 1. Click the Add (1) button.
- 2. Specify a user ID not already in use, a user role, and a password. For details about user roles, refer to the <u>User roles</u> section.

Note: The password must contain one upper-case character, one lower-case character, and one digit. The minimum length of the password is eight characters.

- 3. Set the allowed run modes and user options.
- 4. Click the Apply (2) button.

Note: You can create several user accounts for one person by giving the accounts different user IDs. This may be useful to distinguish different roles.

Editing user accounts

To edit an existing user account:

- 1. Select the user account to be modified.
- 2. The details are displayed in the section at the bottom of the dialog.
- 3. Modify the parameters as required.

4. Click the Apply (2) button.

Note: In the **User Manager**, the user name, role, password, and authorizations of existing user accounts can be changed. The user ID (login name) is the unique identifier of a user and cannot be changed.

Deactivating user accounts

User accounts cannot be deleted, but they can be deactivated if users no longer work with QIAxcel ScreenGel data.

To deactivate a user account:

- 1. Select the user account to be deactivated from the list of users.
- 2. Uncheck the Activated (3) option in the user settings. Deactivated users cannot log in anymore.
- 3. Click the Apply (2) button.

Note: Only activated users are shown in the user table. To show deactivated users, check the **Show** deactivated user accounts (4) box below the user table.

User options

Allow private settings	Allows the user to maintain private settings.
May skip confirmation of run checks	On the Run Check page of the process wizard, the user may skip the confirmation of check messages.
May revise sample information after run	Enables the user to Revise sample information in the <u>Experiment Explorer</u> .
May accept incomplete experiments	If an experiment has run incompletely, the user can mark it as complete in the <u>Experiment</u> <u>Explorer</u> , overriding all warnings.

6.6.4 QIAsphere

The QIAxcel Connect can be connected to the **QIAsphere**® digital platform, enabling **real-time status monitoring** via the **the QIAsphere** App and reception of **Notification** messages and updates.

This configuration requires connecting the QIAxcel Connect via the computer to your network as well as the QIAsphere Base of the Connectivity Package. Follow the instructions from the QIAsphere User Manual (**www.qiagen.com/qiasphere**) to connect the QIAsphere Base to the same local network the QIAxcel Connect is connected to.

Note: The **QlAsphere** tab in Configuration environment is only available for the Administrator. Refer to the <u>User roles</u> section for more information.

To connect the QIAxcel ScreenGel application to the QIAsphere Base, first connect the QIAxcel ScreenGel application with QIAxcel Connect instrument. QIAsphere Base Host Address (1) and Password (3), need to be entered in the corresponding fields in the **QIAsphere** tab of the **Configuration** environment. User Name (2) will be automatically filled with used instrument ID.

Settings	Profile Manager	User Manager	QIAsphere				
5) 🖌 Ena	ble QIAsphere Integra	ation					
QIAsph User Na Passwo	ere Base Host Address ime rd	https://10.100.67 qiaxcel-50003 ••••••	162/gen-instr-api	0)		
Choose Certifica Valid th	certificate ate name ru	CA3EFF097C6C0 QIAsphere-Base- 9/10/2050 4:29:05	D18B3E83BC0AC9BC CA PM	B152E6CA648			
						6 Apply	Cancel

The validity of the entered information can be verified by clicking the Test Connection button (4).

When the connection cannot be established after clicking the **Test Connection** button, a corresponding error message is displayed giving an information for the possible cause of failure.

The **Test Connection** button remains inactive when specified settings are invalid (an invalid setting is marked with a standard warning yellow background color).

When the settings are valid and the **Enable QIAsphere Integration** checkbox (5) is checked, the user needs to click the **Apply** button (6) to enable communication of QIAxcel ScreenGel with the QIAsphere Base. The communication can be stopped by unchecking the **Enable QIAsphere Integration** checkbox and clicking **Apply** button.

When the QIAsphere Integration is ON, QIAsphere Base status is displayed in the status bar of QIAxcel ScreenGel as one of the following:

- Connected when Configuration event was successfully sent
- Disconnected when integration is ON but instrument is not connected to Screengel
- Error when integration is turned ON but configuration event could not be sent or heartbeat cannot be delivered

When the QIAsphere Integration is ON and connection is successfully established, the availability status of QIAxcel Connect instrument is sent to QIAsphere Base as Heartbeat event every 60 seconds and the reception of Notifications inclusive of available download files (QIAxcel ScreenGel update or **Profiles**) is enabled.

6.6.4.1 Notifications

The Notifications section in QIAxcel ScreenGel displays messages from QIAGEN sent via QIAsphere. Further, the Notifications allow to download QIAxcel ScreenGel software updates and new **Profile** files from the QIAsphere cloud:

- Alignment Marker
- Analysis Profile
- Peak Calling Instruction
- Calibration Process Profile
- Cartridge Type
- Distribution Profile
- Interpolation Profile
- Method
- Process Profile
- Report Export Profile
- Size Marker

The Notifications can be accessed by clicking the Notifications button on the right side of the Main toolbar.





New incoming Notifications are marked by a red dot on the Notifications button. The red dot contains number indicator of unread received Notifications. After opening a notification, the number value in the indicator will decrease by one.



Notifications can be expanded or collapsed by clicking on the arrow symbol (1) on the right side of the notification or on the title. To delete read Notifications, press the red "x" icon (2) at right top of notification.



Download Notifications provide an opportunity to download software updates and new **Profile** files. To start the software update process or to download new file, maximize the respective notification and press **Download (1)**. The file download starts after a few seconds. The download status is displayed in the notification (2 and 3). After a successful download, the file could be installed by pressing **Update** (4).



In case of software update, a pop-up window appears with information to start software update, application need to be close. Then, the installation wizard will automatically start with instruction on screen.

In case of new files, a pop-up window appears with information about uploaded files, and request to restart the application to use new files.

Download Notifications display the download status via the following icons:

Notification contains content to download

Download in progress

Download canceled or failed

Download successful

Update completed

7 Maintenance procedures

The following maintenance procedures must be carried out to ensure reliable operation of the QIAxcel Connect:

- Cleaning of the QIAxcel Connect
- Minor corrective maintenance

Following these procedures ensures that the QIAxcel Connect is free of dust and liquid spills.

Important: Disconnect the line power cord from the power outlet before servicing.

Servicing

The QIAxcel Connect is supplied with a warranty that lasts for 1 year, starting from the date of shipment. The warranty includes all repairs due to mechanical breakdown. Application development, software upgrades, accessories, and disposable items are not included in the warranty.

QIAGEN offers comprehensive Service Support Agreements, including Warranty Extensions, Full Cover Support Agreements, and instrument/application training, including on-site installation. Service Support Agreements maximize productivity and ensure high performance from your instrument. In addition, service histories are fully documented and all parts are certified and guaranteed.

Contact your local QIAGEN Field Service Specialist or your local distributor for more information about flexible Service Support Agreements from QIAGEN.

WARNING/	Risk of personal injury and material damage [W12]
	The covers of the QIAxcel Connect must be removed only by qualified personnel appropriately trained by QIAGEN.
	Servicing of the QIAxcel Connect must be performed only by representatives trained and authorized by QIAGEN.

7.1 Cleaning of the QIAxcel Connect

Cleaning agents

CAUTION	Damage to the instrument [C2]
\wedge	Do not use bleach, solvents, or reagents containing acids, alkalis, or abrasives to clean the QIAxcel Connect.

	Risk of fire	[W13]
<u> </u>	Do not allow cleaning fluid or of electrical parts of the QIAxcel O plate drawer that no liquid is sp	econtamination agents to come into contact with the Connect. Take special care while cleaning the sample illed into the inside of the instrument.

WARNIG	Risk of electric shock	[W14]				
<u> </u>	Do not open any panels on the QIAxcel Connect other than described in this user manual.					
	Risk of personal injury and mate	erial damage.				
	Only perform maintenance that	s specifically described in this user manual.				

Wipe the instrument only with a damp cloth.

The sample plate holder inside the instrument should be cleaned occasionally using a moist, soft cloth.

Wash the buffer tray in warm water using a mild detergent, rinse thoroughly with deionized water and let it dry before filling with fresh buffer. The buffer tray should be cleaned before using a new QIAxcel gel cartridge.

Note: Do not use disinfectants to clean the buffer tray.

Additionally, the following disinfectants and detergents are recommended for cleaning the QIAxcel Connect instrument.

Note: If you want to use disinfectants different from those recommended, ensure that their compositions are similar to those described below.

General cleaning of the QIAxcel Connect:

- Mild Detergents (e.g. Mikrozid[®] Liquid or Wipes)
- 70% ethanol (except for the blue vision panel in the Sample door of the QIAxcel Connect)
- Disinfectants (e.g. DECON-QUAT $^{\textcircled{R}}$ 100 or gigasept $^{\textcircled{R}}$ instru AF)

Disinfection:

Ethanol-based disinfectants can be used for disinfection of surfaces: e.g. 70% ethanol (Carl Roth GmbH + Co. KG), or Mikrozid[®] Liquid or Wipes (Schülke & Mayr GmbH).

Note: Ethanol-based disinfectants can be used for disinfection of surfaces, such as the worktable: e.g. Mikrozid Liquid, Mikrozid Wipes. Do not use ethanol-based disinfectants to clean the blue vision panel in the Sample door of the QIAxcel Connect.

Disinfectants based on glyoxal and quaternary ammonium salt can be used for the cleaning of all surfaces of the QIAxcel Connect, e.g. 10 g glyoxal, 12 g lauryldime-thylbenzylammonium chloride, 12 g

myristyldimethylbenzylammonium chloride, and 5 to 15% nonionic detergent per 100 g liquid, Lysetol[®] AF (Gigasept[®] Instru AF in Europe, cat. no. 107410, or DECON-QUAT[®] 100, Veltek Associates, Inc., in the USA, cat. no. DQ100-06-167-01).



Risk of fire or explosion [W16]
When using ethanol or ethanol-based liquids on the QIAxcel Connect, handle such liquids carefully and in accordance with the required safety regulations. If liquid has been spilled, wipe it off and allow flammable vapors to disperse.

Toxic fumes	[W17]
Do not use bleach to disinfect u	sed labware.

General instructions

- Do not use spray bottles to spray cleaning or disinfectant liquids onto surfaces of the QIAxcel Connect.
- If solvents or saline, acidic, or alkaline solutions are spilt on the QIAxcel Connect, wipe the spilt liquid away immediately.
- Follow manufacturer's safety instruction for handling cleaning agents.
- Follow manufacturer's instruction for soaking time and concentration of the cleaning agents.

Important: Immersing for longer than the recommended soak time can harm the instrument.

Note: Disinfection reagents shall be distributed equally on the instrument surface and drops shall be avoided

Ensure that no liquid comes in contact with the Laptop and the display. Liquid may be drawn through the dust protection sealing by capillary forces and cause malfunction of the display.

7.2 Minor corrective maintenance

This section makes you familiar with the activities which may be required to maintain the QIAxcel Connect or correct a failure.

- 7.2.1 Fuse replacement and cleaning
- 1. Switch off the QIAxcel Connect using the main power switch.
- 2. Unplug the power cord from the power outlet and from the rear of the instrument.
- 3. Use a small flat-bladed screwdriver to remove the fuse-carrier assembly located above the area where the cord connector enters into the inlet.



4. Replace the fuse.

Important: Replace only with time-delay T 4 A H (250 V) fuses, marked "T4AH250V" cat. no. 9241502 and cat. no. 9019762. If replacement fuses are not available, contact QIAGEN Technical Services.

- 5. Re-insert the fuse-carrier assembly and plug in the power cord.
- 6. Turn on the power and check the instrument for normal operation, i.e., check that the status changes in the status panel (see section <u>Operating the QIAxcel Connect</u>). If the instrument does not operate normally, or continues to blow fuses, unplug the system and contact QIAGEN Technical Services.

7.2.2 N2 cylinder replacement

N₂ cylinders should be changed when one or both low pressure warnings are shown in the <u>Status</u> Information panel.

To remove the old N₂ cylinder:

Note: To remove a pressurized N₂ cylinder, empty the N₂ bottle first (see <u>Empty N₂ bottle</u>).

1. Open the sample door and remove the buffer tray.

Note: Do not open the sample door, if the cartridge door is open. This may damage the instrument. Please close the cartridge door before opening the sample door.

- 2. Gently pull up on the N_2 cylinder port. Do not pull past the detent hole.
- 3. Turn the cylinder counter-clockwise to allow the remaining N_2 to gradually leak out.



Note: Dispose of empty N_2 cylinders as recyclable steel material according to your local regulations.

Insert a new N₂ cylinder:

4. Screw a new, unpunctured N₂ cylinder into the cylinder port in a clockwise direction.

Important: Only use N₂ cylinders provided by QIAGEN (cat. no. 929705).

5. Turn until the needle inside the port punctures the N₂ cylinder.

Note: Do not overtightened. The cylinder should only be finger-tight.

6. Gently push down on the N_2 cylinder until it is in the stowed (down) position.

7.2.3 Alternate N2 supply

The QIAxcel Connect external N_2 port (located at the rear of the instrument) can be connected to clean, non-condensing compressed nitrogen. This external N_2 source can be used instead of the internal N_2 cylinder. The regulated N_2 minimum input pressure must be 50 psi (345 kPa) and the maximum input pressure should not exceed 75 psi (517 kPa).

To connect an external N₂ source, use the supplied urethane tubing (2 mm inner diameter x 3.18 mm outer diameter) rated for 150 psi. Additional tubing can be purchased separately (cat. no. 9018435).

Connect the urethane tubing from the output of the external N_2 source to the QIAxcel Connect by firmly inserting the tubing into the fitting located on the rear panel of the instrument.

Turn on the external N_2 source pressure, and set the input pressure to 50–75 psi (345–517 kPa).

7.2.4 Blocked purge filter

Before a new sample run, fresh gel is purged from the gel reservoir of the cartridge into the capillaries by applying pressure through the purge line and purge port of the instrument. In rare cases when the QIAxcel gel cartridge was not stored in an upright position and equilibration conditions were not maintained, gel may run into the purge line of the instrument. A purge filter that is placed in the instrument prevents gel from running into the instrument's pressure valves. Gel in the filter or purge line will dry and thus block the filter or purge line.

To diagnose a blocked purge filter or purge line, the **Filter Check** with the QIAxcel ScreenGel software can be performed. If the filter is blocked, a replacement of the purge filter is required. In very rare cases the purge line needs to be rinsed with water to resolve gel debris blocking the purge line tubing.

Note: Replacement of the purge filter and cleaning of the purge line is only required when the filter test failed and tubing is clogged.

Note: It is not required to replace the purge filter or cleaning the purge line as a preventive maintenance.

Performing a Filter Check

- 1. Switch on the QIAxcel Connect.
- 2. Launch the QIAxcel ScreenGel.

- 3. Go to the **System Check** tab in the service environment and perform the guided **Filter Check** (see <u>Filter check</u>).
- 4. If Filter Check fails, replace the filter and test again.

Replacement of purge filter

- 1. Open the service door.
- 2. Remove the purge filter.

Note: Do not pull on the tubing.



- 3. Place a new purge filter and attach tightly to the connectors.
- 4. Perform the Filter Check again.

Note: If the Filter Check fails again, contact QIAGEN Technical Services.

Additional filters can be ordered from QIAGEN.

9021980 Set, Purge Filters (10), QX; please contact QIAGEN Technical Services.

8 Troubleshooting

This section provides information about what to do if an error occurs when using the QIAxcel Connect and QIAxcel ScreenGel.

If you need to contact QIAGEN Technical Services about an error, note down the steps leading to the error and the information from any dialog that appears. This will help the QIAGEN Technical Service Specialist to resolve the error.

Refer to the <u>Troubleshooting folder and log files</u> section for detailed information on how to retrieve log files and system test results.

8.1 System setup

Comments	and	suggestions
Comments	unu	Juggeshons

Instrument does not turn on

a) Power cord not Check the power connection.

b) Fuse blown Replace the fuse.

Computer software cannot communicate with system

a)	USB cable is not connected between PC and system	Check the connection of the USB cable. To retry the connection to the instrument, click the I icon in the software in the <u>Process</u> environment.
b)	Incorrect COM channel settings	Check the COM channel setting (see section <u>Settings</u>). At the bottom of the settings screen, click OK to confirm the changes. Click Yes in the message shown to
		connect again to the newly selected COM channel. If the ficon is visible, the instrument is ready to operate. If the problem persists, change the log level of the instrument log to verbose (see section <u>Settings</u> for information how to do this). Close the software, restart it, and login. Wait until the message appears as "the instrument is not available." Then collect the troubleshooting folder as described in the section <u>Troubleshooting folder and log files</u> . Ensure to check the boxes in the list at the right to include the logging information. Send the file created to QIAGEN Technical Services.
c)	Software cannot detect the COM port.	Check the user permission for the registry entries that are concerned the COM ports. The pertained registry entry is typically named HKEY_LOCAL_MACHINE \SYSTEM\ControlSet001\Enum\ACPI\PNP0501\XXXXXX\Device Parameters. Make sure that user Everyone has at least read permission for this entry, otherwise the COM port scan procedure (which is invoked at start of the application) might fail if the QIAxcel ScreenGel is being operated by a user who does not have administrator permissions.

		Comments and suggestions			
d)	Firmware update is not successful	The QIAxcel ScreenGel software comes with an up-to-date instrument software for instruments with serial number 50000 and higher. If the update process could not be completed, the QIAxcel Connect instrument cannot connect to the QIAxcel ScreenGel software any longer. In this case, the software provides an option to repeat the update process. Ensure that the cable between the instrument and the computer is connected correctly. Turn on the instrument. Restart the QIAxcel ScreenGel and login. Wait until the field is conchanges to field. In this case, the instrument is ready to operate again. Otherwise, a message appears that the instrument is not available. Click the Troubleshoot button. In the next message, click Retry instrument update . Wait for the Updating Instrument dialog. Wait a few minutes until the update is complete and the instrument is ready to operate. Otherwise, if the field is con is visible and the instrument is ready to operate. Otherwise, if the field is con is visible, change the log level of the instrument log to verbose (see section <u>Settings</u> for information how to do this) and repeat the steps to update the instrument again. Then collect the troubleshooting folder as described in the section Troubleshooting folder and log files. Ensure to check the boxes in the list at the right to include the logging information. Send the file created to QIAGEN Technical Services.			
QL	QIAxcel ScreenGel Software does not start				

a)	QlAxcel ScreenGel software not installed	Install the QIAxcel ScreenGel software.
b)	Old version of Microsoft	The QIAxcel ScreenGel software operates with Windows 7 (32 and 64 bit) and Windows 10 (64 bit) (see section <u>Computer and software</u> for more details).

8.2 Operation

Windows

		Comments and suggestions
N ₂	cylinders do not last long	
a)	Air leak	Double check the N ₂ cylinder to make sure it is tight.
b)	Internal leak if N ₂ cylinder last only a couple of days	Contact QIAGEN Technical Services.
Sa	mples tray does not move	
a)	Transport lock is in place	Remove the transport lock.
b)	Sample door is not closed	Close the sample door.
Ca	nnot insert cartridge into system	

		Comments and suggestions
Cartridge installed backwards		Make sure the cartridge front cover faces the front of the system and check that the purge cap seal has been removed.
Ca	rtridge cannot be latched	
a)	N ₂ pressure low	Replace the N ₂ cylinder.
b)	Cartridge door is open	Close the cartridge door.
c)	Cartridge smart key not inserted	Insert the smart key.
Ca	nnot run system	
a)	Sample door and/or cartridge door are open	Close the sample door and/or cartridge door.
b)	Cartridge smart key not detected	Insert the cartridge smart key
c)	Pressure low	Replace the N ₂ cylinder
QI	Axcel ScreenGel Software is slow	
a)	Too many experiments open in Experiment Explorer	Close some of the opened experiments and then restart the QIAxcel ScreenGel. Try to keep the number of opened experiments low.
b)	Too many applications running concurrently.	Close all unnecessary applications.

Problems when files are/shall be located on the network folder

a) Network folder is not visible in the file dialog	If the network folder is not visible in the software although it has been mapped, e.g. by an administrator, try the following to make the mapping visible for all Windows users. Delete the existing mapping. Then use the net use command together with a UNC name to map the network location. For example, in the command prompt, type the following command and press Enter: net use \\< computername >\< sharename > /user:< username >
	Ask your local IT for assistance, if required.
b) It takes too long to open a network folder	If you are using only one of the sub-folders of the network folder, map directly to the sub-folder instead of mapping the entire network folder. Use the net use command as described above.

8.3 DNA applications

Comments and suggestions		
DNA peaks signal variation in channels		
New cartridge requires calibration	Calibrate the cartridge (see section <u>Calibrating a cartridge</u>).	

Slow peak migration time in channels

Comments and suggestions

a)	Air bubbles in gel cartridge capillaries	Perform a purge or 3-minute purge (see <u>Maintenance</u>) to remove air bubbles, or change the QX Separation Buffer.		
b)	Current too low due to low temperature	Increase the room temperature.		
c)	Current too low due to partially dry capillary tips	Perform a purge or 3-minute purge to clean the capillary tips (see <u>Maintenance</u>).		
We	ak signal of the QX Aligni	nent Marker		
Deg Alig	raded/old QX Inment Marker	Change the QX Alignment Marker.		
No	No signal in channels			
a)	No samples injected	Check the volume of sample (6µl minimum volume)		
b)	Blocked capillary channel	Purge the instrument to remove the blockage (see <u>Maintenance</u>). If this does not work, replace the cartridge.		
c)	Broken capillary or optical fiber in cartridge	Replace the cartridge.		
d)	Light source problem (LEDs off)	Check if all LEDs are illuminated. Contact QIAGEN Technical Services.		
e)	No purging of the cartridge/purge filter blocked	Perform a Filter Check.		

Loss of resolution in channels

a)	Old gel in some channels or clogged channels	Perform a purge or 3-minute purge to fill the channels with new gel, or repeat the purge method several times to remove clogging (see <u>Maintenance</u>).
b)	Saturated signals	Reduce the sample injection time or select a more suitable method.
c)	Expired cartridge	Use a new cartridge.
Bro	ad or saturated DNA pea	k/band
a)	DNA concentration too high	Dilute the DNA solution in QX DNA Dilution Buffer. Reduce the injection time. Choose a method for high DNA concentration samples (see <u>Appendix B</u>).

b) Broken channel with Call QIAGEN Technical Services and replace cartridge. high background

DNA signal too low

		Comments and suggestions
a)	DNA concentration too low	Increase the sample injection time. Choose a method for low DNA concentration samples (see <u>Appendix B</u>).
b)	Salt concentration too high in sample solution	Desalt or dilute the DNA samples to reduce the salt concentration and enhance DNA injection.
Sep	paration current (μA) too lo	w
a)	QX Separation Buffer is contaminated	Replace the QX Separation Buffer.
b)	Blocked capillary channel	Replace the QIAxcel gel cartridge.
Sep	paration current (μΑ) too hi	gh
a)	QX Separation Buffer contaminated	Replace the QX Separation Buffer.
b)	Salt concentration too high in sample solution	Desalt or dilute the DNA samples in QX DNA Dilution Buffer.
Dat	a squeezed together on G	el Image folder
a)	Extra peaks before the first peak or after the last peak were detected	Repeat the analysis as described in <u>Analysis of DNA samples</u> . During the procedure, follow the instructions to increase the Alignment Marker Threshold.
b)	Wrong peaks were identified as the first and last peaks for alignment	Repeat the analysis as described in <u>Analysis of DNA samples</u> . During the procedure, follow the instructions to increase the Alignment Marker Threshold.
c)	Upper alignment markers were missing or below the threshold setting	Repeat the analysis as described in <u>Analysis of DNA samples</u> . During the procedure, follow the instructions to decrease the Alignment Marker Threshold. Replace the new markers for the next run.
d)	Sample signals too strong and cause the alignment below the threshold setting	Dilute the samples or use the H Methods (see <u>Appendix B</u>) for sample analysis.
Dai	a not aligned	
a)	Upper marker missing	Change to a new alignment marker.
b)	Migration problem due to air bubbles	Purge and run the sample again.

		Commenis and suggestions
c)	Migration problem due to low temperature	Increase the separation time.
d)	Start Analysis button in the "Analysis" environment not clicked	Click the Start Analysis button to perform analysis.
e)	Samples with different alignment markers visualized	Remove samples with a different alignment marker from the view.
f)	Alignment markers not identified correctly	Repeat the analysis as described in <u>Analysis of DNA samples</u> . During the procedure, follow the instructions to adapt the analysis parameters.
No	size calling in the result to	able
a)	The reference marker is turned off	Repeat the analysis as described in <u>Analysis of DNA samples</u> . Select the right marker table.
b)	Peak detection failed	Repeat the analysis as described in <u>Analysis of DNA samples</u> . During the procedure, ensure that peaks are detected correctly, especially the alignment marker peaks.
Wr	ong size calling	
a)	Wrong reference marker table used	Repeat the analysis as described in <u>Analysis of DNA samples</u> . Select the right marker table.
b)	Reference marker table setting problems	Repeat the analysis as described in <u>Analysis of DNA samples</u> . Make sure that the correct alignment marker is set (refer to <u>Checking alignment marker</u>). Make sure the number of peaks in the Rel. Time and size (bp) columns are equal in the reference marker table.
No	size scale in Gel image	
a)	Data not aligned	Remove samples with a different alignment marker from the view. Repeat the analysis as described in <u>Analysis of DNA samples</u> . Make sure all alignment marker peaks have been identified correctly.
b)	Samples analyzed with different reference marker tables	Repeat the analysis as described in <u>Analysis of DNA samples</u> using the same reference marker table for all samples.
Rec	quired scale type not creat	ted
a)	Samples not analyzed	Click the Start Analysis button to perform the analysis.
b)	Samples with different alignment markers visible	Remove samples with a different alignment marker from the view.

Comments and suggestions

		Comments and suggestions
c)	No size determination	Repeat the analysis as described in <u>Analysis of DNA samples</u> using the same reference marker table for all samples.

Upper alignment markers resolved in double peaks

High-salt sample Dilute the sample or change the parameter setting.

8.4 RNA applications

		Comments and suggestions
Ali	gnment is off	
a)	The first peak is below the threshold	Repeat the analysis as described in the <u>Analysis of RNA samples</u> section. During the procedure, follow the instructions to decrease the Alignment Marker Threshold.
		Contamination, for example by salt, may result in aberrant migration of individual lanes which may be too large for proper alignment. Use less sample or perform additional cleanup of RNA samples that exhibit delayed migration.
b)	Degraded/old calibration marker	Replace the calibration marker.
Ree	quired scale type not crea	nted
a)	Samples not analyzed	Click the Start Analysis button to perform the analysis.
b)	Samples with different alignment markers visible	Remove the samples with a different alignment marker from the view.
c)	No size determination	Repeat the analysis as described in the <u>Analysis of RNA samples</u> section using the same valid reference marker table for all samples.
The	ratio of rRNA (28S/18S) is too low
	Degraded RNA samples	Change the RNA sample.
The	signal of RNA sample is	s too low
	Degraded RNA samples	Change the RNA samples.
Bro	oad RNA peaks	
a)	Degraded RNA samples	Change the RNA samples.
b)	Incomplete RNA denaturation process	Repeat the RNA denaturation process.

		Comments and suggestions		
c)	Aged buffer	Replace the separation buffer every 25 runs.		
On	ly 185 RNA peak detecte	d		
Inc	omplete RNA	Repeat the RNA denaturation process.		
denaturation process		RNA degradation also causes the the 28S peak to disappear first.		
Calibration marker signal variation in channels				
a)	New cartridge requires calibration	Calibrate the cartridge (see section <u>Calibrating a cartridge</u>).		
b)	Cartridge intensity calibration is incorrect	Calibrate the cartridge (see section <u>Calibrating a cartridge</u>).		
c)	Degraded/old QX Calibration Marker	Change the QX Calibration Marker.		
Delayed peak migration in some channels				
a)	Air bubbles in capillaries	Use the Long purge option to remove air bubbles (see <u>Maintenance</u>).		
b)	Air bubbles in calibration marker or RNA sample	Remove the air bubbles from the vials.		

9 Glossary

vials

Term	Description
Analysis profile	Analysis profiles contain settings for sample data analysis. It can be part of Process Profiles.
	It contains no information about a matched reference (size) marker table, to be not depending on the method and the alignment marker, the size marker was processed with.
Baseline	A line in the electropherogram that is a base for analysis calculations. In electropherograms with sharp peaks, the objective of the baseline is to effectively remove the peaks but accurately follow any other disturbances, e.g. baseline drift and shifts, in the electropherogram. In smear analysis the baseline serves as a zero- line.

Term	Description
Buffer tray	The buffer tray is a removable plastic tray that is inserted in the buffer tray holder of the QIAxcel Connect and holds QX Separation and Wash Buffers and QX Intensity and Alignment Markers.
Buffer tray holder	The buffer tray holder is located inside the QIAxcel Connect under the sample door. The buffer tray, which holds QX Separation and Wash Buffers and QX Intensity and Alignment Markers, is placed in the buffer tray holder prior to sample analysis.
Cartridge door	The door that allows the QIAxcel gel cartridge to be loaded into the QIAxcel Connect. This door should remain closed during operation of the QIAxcel Connect.
Channel(s)	Each QIAxcel gel cartridge has 12 channels (capillaries) through which the samples for analysis pass.
Distribution profile	Parameters for distribution analysis with certain areas of interest. An area of interest is defined by a name and a size range. Molarity ratios can be defined between the molarities of two areas of interest, or between the molarity of an area of interest and the total molarity of the sample. For each molarity ratio an acceptance range can be defined.
Environment	The QIAxcel ScreenGel software is composed of four environments providing different functionality: Process, Analysis, Service and Configuration.
Method	A method is the collection of commands and parameters that are applied to an individual sample run. A number of default methods are preinstalled.
Minimum distance	An analysis parameter which specifies the minimum distance two peak clusters must have to be detected as two distinct peaks.
Mineral oil	For covering solutions and/or samples to prevent evaporation.
Peak	A local maximum value of a curve.
Position	An area of a rack/plate that can contain something. Examples of positions include the wells of a microplate or slots in the sample plate holder.
Power switch	A button located at the front of the QIAxcel Connect in the bottom-right corner. It allows the user to switch the QIAxcel Connect on and off.
Process Profile	This defines settings for data acquisition, analysis and report/export. Defines all parameters for the electrophoresis run.
Profile	Profiles combine settings and parameters and can be used for different purposes .
	There are analysis profiles, distribution profiles, Process Profiles, peak calling instructions and report/export profiles.
Purge port	The purge port is located inside the QIAxcel Connect instrument and aligns with the purge hole of the QIAxcel gel cartridge.
QIAsphere	Cloud solution that gathers data sent from QIAsphere Base devices and allows to display them in the Web and Mobile Applications.

Term	Description
QIAsphere Base	The QIAsphere Base is a small device that can be placed in the customers Network and enables a secure transfer of data from QIAGEN instruments to the Cloud and QIAsphere app.
QX Alignment Marker	Allows sample migration time to be calibrated.
QX DNA or RNA (HS) Dilution Buffer	Allows dilution of concentrated samples. QX DNA or RNA HS Dilution Buffer is provided with the respective QIAxcel DNA or RNA High-Sensitivity Kit.
QX (HS) Separation Buffer	Enables separation of DNA or RNA molecules in the QIAxcel gel cartridge. QX HS Separation Buffer is provided with the QIAxcel DNA and RNA High-Sensitivity Kit.
QX DNA or RNA Size Marker	Enables creation of a reference marker table allowing DNA/RNA size and/or concentration determination.
QX (HS) Wash Buffer	For washing the capillary tips to prevent cross-contamination. QX HS Wash Buffer is provided with the QIAxcel DNA and RNA High-Sensitivity Kit.
QX (HS)Intensity Calibration Marker	Allows calibration of the signal intensity for each new gel cartridge. QX HS Intensity Calibration Marker is provided with the QIAxcel DNA and RNA High-Sensitivity Kit.
Reference marker	A reference marker is a size marker with fragments of known size and concentration. It is used for size and concentration determination of samples.
Report/export profile	A report / export profile contains all information needed to create a report and to define the export. It can be saved for further use. It can be part of Process Profiles.
RFU	relative fluorescence unit
RpR	Runs per Row
Sample door	The door that provides access to the sample plate holder and the buffer tray holder. This door should remain closed during operation of the QIAxcel Connect.
Sample plate holder	The sample plate holder is located inside the QIAxcel Connect under the sample door. 96-well plates or sample strips containing the samples for analysis are placed in the sample plate holder.
Service door	The service door allows access to the purge filter and a manometer.
Smart key	This key attached to the QIAxcel gel cartridge holds information about the cartridge (i.e., cartridge identifier, calibration status, number of runs). The smart key should be inserted into the smart key socket on the QIAxcel Connect to enable sample analysis.
Smart key socket	The smart key socket allows the QIAxcel Connect instrument to read and display the information on the smart key.
10 Appendices

The appendices contain technical data, QIAxcel methods and accessories, descriptions of the data analysis algorithms, warranty terms and translations of safety information.

10.1 Appendix A

Technical data

QIAGEN reserves the right to change specifications at any time.

Environmental conditions

Operating conditions	
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Power	100–240 V AC, 50–60 Hz, 100 VA
Fuse rating	4 A (250 V) time-delay fuse (for 100–240 V AC)
Overvoltage category	II
USB connection	USB1, 5V, 500 mA
Air temperature	15–30°C (59–86°F)
Relative humidity	10–75% (noncondensing)
Altitude	Up to 2000 m (6500 ft.)
Place of operation	For indoor use only
Pollution level	2
Environmental class	3K2 (IEC 60721-3-3)
Transportation conditions Air temperature	–25°C to 60°C (–13°F to 140°F) in manufacturer's package
Relative humidity	Max. 75% (noncondensing)
Storage conditions Air temperature	15°C to 30°C (59°F to 86°F) in manufacturer's package
Relative humidity	Max. 75% (noncondensing)

Mechanical data and hardware features

Dimensions (door closed)	Width: 372 mm (14.64 in.) Height: 408 mm (16.06 in.) Depth: 572 mm (22.44 in.)
Dimensions (doors open)	Width: 372 mm (14.64 in.) Height: 599 mm (23.58 in.) Depth: 572 mm (22.44 in.)
Mass	32 kg (70,55 lb.)
Capacity	Up to 96 samples per run
Software	Default protocols are provided with the QIAxcel ScreenGel Software supplied with the QIAxcel Connect. Updated protocols are available from www.qiagen.com/QIAxcelConnect .

Waste Electrical and Electronic Equipment (WEEE)

This section provides information about disposal of waste electrical and electronic equipment by users.

The crossed-out wheeled bin symbol (see below) indicates that this product must not be disposed of with other waste; it must be taken to an approved treatment facility or to a designated collection point for recycling, according to local laws and regulations.

The separate collection and recycling of waste electronic equipment at the time of disposal helps to conserve natural resources and ensures that the product is recycled in a manner that protects human health and the environment.



Recycling can be provided by QIAGEN upon request at additional cost. In the European Union, in accordance with the specific WEEE recycling requirements and where a replacement product is being supplied by QIAGEN, free recycling of its WEEE-marked electronic equipment is provided.

To recycle electronic equipment, contact your local QIAGEN sales office for the required return form. Once the form is submitted, you will be contacted by QIAGEN either to request follow-up information for scheduling collection of the electronic waste or to provide you with an individual quote.

FCC Declaration

The "United States Federal Communications Commission" (USFCC) (in 47 CRF 15. 105) declared that the users of this product must be informed of the following facts and circumstances.

This device complies with part 15 of the FCC. Operation is subject to the following two conditions: (1) This device may not cause harmful interference, and (2) this device must accept any interference received, including interference that may cause undesired operation.

This Class B digital apparatus complies with Canadian ICES-003.

The following statement applies to the products covered in this manual, unless otherwise specified herein. The statement for other products will appear in the accompanying documentation.

NOTE: This equipment has been tested and found to comply with the limits for a Class B digital device, pursuant to part 15 of the FCC Rules. These limits are designed to provide reasonable protection against harmful interference in a residential installation. This equipment generates, uses and can radiate radio frequency energy and, if not installed and used in accordance with the instructions, may cause harmful interference to radio communications. However, there is no guarantee that interference will not occur in a particular installation. If this equipment does cause harmful interference to radio or television reception, which can be determined by turning the equipment off and on, the user is encouraged to try to correct the interference by one or more of the following measures:

- Reorient or relocate the receiving antenna.
- Increase the separation between the equipment and receiver.
- Connect the equipment into an outlet on a circuit different from that to which the receiver is connected.
- Consult the dealer or an experienced radio/ TV technician for help.



QIAGEN GmbH, Germany is not responsible for any radio television interference caused by unauthorized modifications of this equipment or the substitution or attachment of connection cables and equipment other than those specified by QIAGEN GmbH, Germany. The correction of interference caused by such unauthorized modification, substitution or attachment will be the responsibility of the user.

Declaration list of China RoHS (SJ/T 11364)

The environment friendly use period of the QIAxcel Connect instrument is 25 years. The marking for the Restricted Use of Hazardous Substances in Electronic and Electrical Products shown in table below:

Part Name - 部件名称 Toxic or hazardous Substances and Element - 有毒或有害的物质成分

Lead (Pb) 铅	Mercur y (Hg) ≖	Cadmiu m (Cd)	Hexavalent Chromium	Polybrominat ed biphenyls	Polybromina ted diphenyl	
	水	钠	(Cro+) 六价铬	(PBB) 多溴联苯	(PBDE) 多溴联苯醚	
0	0	0	0	0	0	
	•					
Х	0	0	0	0	0	
0	0	0	0	0	0	
				-	-	
Х	0	0	0	0	0	
Х	0	0	0	0	0	
			•	•	•	
0	0	0	0	0	0	
		-				
0	0	0	0	0	0	
0	0	0	0	0	0	
This table is prepared in accordance with the provisions of SJ/T 11364. O: Indicates that this toxic or hazardous substance contained in all of the homogeneous materials for this part is below the limit requirement in GB/T 26572.						
O:代表用于此部件的所有同类型的包含该种有毒或者有害物质的材料均在GB/T 26572 的规定界限以下 X:Indicates that this toxic or hazardous substance contained in at least one of the homogeneous materials used for this part may be above the limit requirement in GB/T 26572. X:代表用于此部件的至少一种此类型的包含该种有毒或者有害物质的材料可能在GB/T						
	Lead (Pb) 铅 〇〇 〇〇 〇〇 〇〇 〇〇 〇〇 〇〇 〇〇 〇〇 〇〇 〇〇 〇〇 〇〇	Lead (Pb) 铅 Mercur y (Hg) 汞 O O O O X O X O X O X O X O X O X O X O X O O O X O O O O O O O O O O O O O O O X O X O O O O O O O O O Image: Second Contract Contraction of the standown of the stand	Lead (Pb) 铅 知Mercur y (Hg) 汞Cadmiu m (Cd) ශOOOOOOXOOOOOXOOXOOXOOXOOO <td>Lead (Pb) 铅Mercur y (Hg) 束Cadmiu m (Cd) 偏Hexavalent Chromium (Cr6+) 六价铬0000X00000000000X00X00X00X00X000<td>Lead (Pb) 铅 Mercur y (Hg) 素 Cadmiu m (Cd) 領 Hexavalent Chromium (Cr6+) 六价格 Polybrominat ed biphenyls (PBB) 多 溴 联 素 0 0 0 0 0 0 X 0 0 0 0 0 X 0 0 0 0 0 X 0 0 0 0 0 X 0 0 0 0 0 X 0 0 0 0 0 X 0 0 0 0 0 X 0 0 0 0 0 X 0 0 0 0 0 X 0 0 0 0 0 X 0 0 0 0 0 X 0 0 0 0 0 X 0 0 0 0 0 X 0 0 0 0 0 X 0 0 0 0 0</td></td>	Lead (Pb) 铅Mercur y (Hg) 束Cadmiu m (Cd) 偏Hexavalent Chromium (Cr6+) 六价铬0000X00000000000X00X00X00X00X000 <td>Lead (Pb) 铅 Mercur y (Hg) 素 Cadmiu m (Cd) 領 Hexavalent Chromium (Cr6+) 六价格 Polybrominat ed biphenyls (PBB) 多 溴 联 素 0 0 0 0 0 0 X 0 0 0 0 0 X 0 0 0 0 0 X 0 0 0 0 0 X 0 0 0 0 0 X 0 0 0 0 0 X 0 0 0 0 0 X 0 0 0 0 0 X 0 0 0 0 0 X 0 0 0 0 0 X 0 0 0 0 0 X 0 0 0 0 0 X 0 0 0 0 0 X 0 0 0 0 0 X 0 0 0 0 0</td>	Lead (Pb) 铅 Mercur y (Hg) 素 Cadmiu m (Cd) 領 Hexavalent Chromium (Cr6+) 六价格 Polybrominat ed biphenyls (PBB) 多 溴 联 素 0 0 0 0 0 0 X 0 0 0 0 0 X 0 0 0 0 0 X 0 0 0 0 0 X 0 0 0 0 0 X 0 0 0 0 0 X 0 0 0 0 0 X 0 0 0 0 0 X 0 0 0 0 0 X 0 0 0 0 0 X 0 0 0 0 0 X 0 0 0 0 0 X 0 0 0 0 0 X 0 0 0 0 0 X 0 0 0 0 0	

Declaration of conformity

Name and address of the legal manufacturer

QIAGEN GmbH QIAGEN Strasse 1 40724 Hilden Germany

An up-to-date Declaration of Conformity can be requested from QIAGEN Technical Services.

10.2 Appendix B

QIAxcel Connect methods

A number of default methods are available for each QIAxcel Kit. Note that only the methods available for the particular cartridge type can be selected.

Note: If you require a custom method to be created, please contact QIAGEN Technical Services.

DNA/RNA methods

Each method name (normal sensitivity) is an acronym, providing information on the specific QIAxcel Kit/ QIAxcel gel cartridge being used, the sample injection time and voltage, and the separation time and voltage, as described below.



Please find below a complete list of methods available for each QIAxcel gel cartridge type and information regarding their associated fragment sizes and best resolution.

QIAxcel DNA High Resolution methods

The QIAxcel DNA High Resolution gel cartridge is designed for high-resolution (3–5 bp) genotyping, highresolution multiplex PCR, and AFLP/RFLP analysis of less than 20 fragments. The gel cartridge can separate fragment sizes from 15 bp to 10 kb. The resolution depends on the fragment size and the selected method:

	Fragment size 100–500 bp	500 bp – 1 kb	1–5 kb	5–10 kb
Method	Best resolution			
0M400*	20 bp	100 bp	500 bp	N/A
0M500* 0L500 [†]	10 bp	50 bp	200 bp	N/A
0H500 [‡]				
0M800* 0H800 [‡]	3–5 bp	N/A	N/A	N/A
0M1200* 0L1200 [†] 0H1200 [‡]	N/A	N/A	500 bp – 1 kb	1– 1.5 kb

Guidelines for method selection using the QIAxcel DNA High Resolution Kit

* We recommend the 0M400, 0M500, 0M800, and 0M1200 methods for DNA concentrations of 10–100 ng/µl (e.g. PCR products amplified from genomic DNA with 30–40 cycles).

 \dagger We recommend the OL500 and OL1200 methods for DNA concentrations of <10 ng/µl.

 \ddagger We recommend the OH400, OH500, OH800, and OL1200 methods for DNA concentrations of >100 ng/ µl (e.g. high-yield PCR products).

M Methods

We recommend the OM400, OM500, OM800, and OM1200 methods for DNA concentrations of 10–100 $ng/\mu l.$

Method	Sample injection voltage (kV)	Sample injection time (s)*	Separation voltage (kV)	Separation time (s)
0M400	5	10	6	400
0M500	5	10	5	500
0M800	5	10	3	800
0M1200	5	10	3.5	1200

* Sample injection time can be adjusted (minimum 1 s; maximum 60 s). Reduce the injection time if the signal is saturated, as indicated by peaks with flat tops. Increase sample injection time if the signal is below the default threshold setting of 5%.

L Methods

We recommend the OL500 and OL1200 methods for DNA concentrations of <10 $ng/\mu l$.

Method	Sample injection voltage (kV)	Sample injection time (s)*	Separation voltage (kV)	Separation time (s)
0L500	8	20	5	500
OL1200	8	20	3.5	1200

* Sample injection time can be adjusted (minimum 1 s; maximum 60 s). Reduce the injection time if the signal is saturated, as indicated by peaks with flat tops. Increase sample injection time if the signal is below the default threshold setting of 5%.

H Methods

We recommend the OH500, OH800, and OH1200 methods for DNA concentrations of >100 $ng/\mu l$.

Method	Sample injection voltage (kV)	Sample injection time (s)*	Separation voltage (kV)	Separation time (s)
0H500	2	20	5	500
0H800	2	20	3	800
0H1200	2	20	3.5	1200

* Sample injection time can be adjusted (minimum 1 s; maximum 60 s). Reduce the injection time if the signal is saturated, as indicated by peaks with flat tops. Increase sample injection time if the signal is below the default threshold setting of 5%.

Note: Unpurified PCR products contain dNTPs and primers, which contribute to the optical density (OD) and can cause overestimation of DNA concentration.

QIAxcel DNA Screening methods

The QIAxcel DNA Screening Kit is designed for low-resolution (>20 bp) genotyping, low-resolution multiplex PCR, single PCR screening, plasmid DNA digestion checking, plasmid and oligo DNA quantity checking, and gDNA quality checking. The gel cartridge can separate the fragment size range from 15 bp to 5 kb. The resolution depends on the fragment size and the method chosen to run the assay:

	Fragment siz <500 bp	e 500 bp – 1 kb	1–5 kb			
Method	Best resolution	on				
AM320*	20 bp	100 bp	500 bp			
AM420* AL420 [†] AH420 [‡]	20 bp	100 bp	500 bp			
APH600 APL600	Uncut plasm	id DNA checking				
AM900	gDNA quali	aDNA quality checking				

Guidelines for method selection using the QIAxcel DNA Screening Kit

* The AM320 and AM420 methods are recommended for DNA concentrations 10–100 ng/µl (e.g. PCR products [30–40 cycles] amplified from genomic DNA).

† The AL420 method is recommended for DNA concentrations <10 ng/µl.

‡ The AH420 method is recommended for DNA concentrations >100 ng/µl (e.g. high-yield PCR products).

M Methods

The AM320 and AM420 methods are recommended for DNA concentrations 10–100 ng/µl, while method APH600 is recommended for purified high-copy plasmid DNA (50–300 ng/µl) in elution buffer. Method AM900 is recommended for a visual quality check of gDNA purified using silica-based methods.

Method	Sample injection voltage (kV)	Sample injection time (s)*	Separation voltage (kV)	Separation time (s)
AM320	5	10	6	320
AM420	5	10	5	420
APH600	1	5	6	600
AM900	2	40	3.5	900

L Methods

The AL420 method is recommended for DNA concentrations <10 ng/ μ l, while method APL600 is recommended for purified low-copy plasmid DNA (<50 ng/ μ l) in elution buffer.

Method	Sample injection voltage (kV)	Sample injection time (s)*	Separation voltage (kV)	Separation time (s)
AL420	8	20	5	420
APL600	3	5	6	600

* Sample injection time can be adjusted from 5 to 40 seconds to obtain optimal signals. Reduce the injection time if the signal is saturated, as indicated by peaks with flat tops. Increase sample injection time if the signal is below the default threshold setting of 7%.

H Methods

The AH420 method is recommended for DNA concentrations >100 ng/µl.

Method	Sample injection voltage (kV)	Sample injection time (s)*	Separation voltage (kV)	Separation time (s)
AH420	2	20	5	420

* Sample injection time can be adjusted from 5 to 40 seconds to obtain optimal signals. Reduce the injection time if the signal is saturated, as indicated by peaks with flat tops. Increase sample injection time if the signal is below the default setting of the threshold of 7%.

Note: Unpurified PCR products contain dNTPs and primers, which contribute to the optical density (OD) and can cause overestimation of DNA concentration.

QIAxcel DNA Fast Analysis methods

The QIAxcel DNA Fast Analysis gel cartridge is designed for rapid analysis of PCR fragments. The gel cartridge can separate fragments ranging from 15 bp to 3 kb in size.

Note: The QIAxcel DNA Fast Analysis Cartridge, QX DNA Size Marker 50 bp -1.5 kb, and the corresponding methods are not suitable for concentration determination.

Method	Sample injection voltage (kV)	Sample injection time (s)*	Separation voltage (kV)	Separation time (s)
DM80 v2.0 [†]	15	8	15	80
DM150	10	10	10	150
DM190	10	15	10	190

Methods for use with the QIAxcel DNA Fast Analysis Cartridge

* Sample injection time can be adjusted (minimum 1 s; maximum 40 s). To obtain optimal signals, we recommend using a minimum of 5 s and a maximum of 15 s.

† DM80 v2.0 includes a longer purge time between runs and thus improves the background level.

QIAxcel RNA Quality Control methods

The QIAxcel RNA Quality Control gel cartridge is designed for quality control of total RNA, cRNA, fragmented RNA, and single-stranded cDNA.

- Method CM-F-RNA is recommended for use with fragmented RNA and fragmented DNA at a concentration of 250–500 ng/µl.
- Method CM-RNA is recommended for use with total RNA at a concentration of 300–1000 ng/µl or cRNA at a concentration of 100–500 ng/µl.
- Method CL-RNA is recommended for use with total RNA at a concentration of 50–300 ng/ μ l or cRNA at a concentration of <100 ng/ μ l.

Note: Total RNA and cRNA concentrations >1 μ g/ μ l should be diluted to 1 μ g/ μ l in sterile DEPC water before denaturing.

Method	Sample injection voltage (kV)	Sample injection time (s)*	Separation voltage (kV)	Separation time (s)
CM-F-RNA	7	20	3	600
CM-RNA	5	20	6	600
CL-RNA	8	20	6	600

* Sample injection time can be adjusted from 5 to 40 seconds to obtain optimal signals. Reduce the injection time if the signal is saturated, as indicated by peaks with flat tops. Increase sample injection time if the signal is below the default threshold setting of 7%.

QIAxcel DNA High-Sensitivity method

The QIAxcel DNA High-Sensitivity Gel Catridge is designed for low concentration sample.

Method	Sample injection voltage (kV)	Sample injection time (s)*	Separation voltage (kV)	Separation time (s)
DNA High- Sensitivity	5	10	5	420

QIAxcel RNA High-Sensitivity methods

The QIAxcel RNA High-Sensitivity Gel Catridge is designed for low concentration sample.

- Method RNA High-Sensitivity low is recommended for samples with very low concentration, applying a higher injection time.
- Method RNA High-Sensitivity medium is recommended for samples with low concentration.

Method	Sample injection voltage (kV)	Sample injection time (s)*	Separation voltage (kV)	Separation time (s)
RNA High- Sensitivity low	8	20	6	600
RNA High- Sensitivity medium	8	5	6	600

10.3 Appendix C

QIAxcel accessories

Product	Contents	Cat. no.
QIAxcel Kits		
QIAxcel DNA High-Sensitivity Kit (1200)	QIAxcel DNA High-Sensitivity Cartridge, Buffer, Mineral Oil, QX HS Calibration Marker, QX DNA HS Size Marker (100 bp - 1 kb), QX DNA HS Alignment Marker, 12-Tube Strips	929012
QIAxcel RNA High-Sensitivity Kit (1200)	QIAxcel RNA High-Sensitivity Cartridge, Buffer, Mineral Oil, QX HS Calibration Marker, QX RNA HS Size Marker (200–6000 nt), QX DNA HS Alignment Marker, 12-Tube Strips	929112
QIAxcel DNA High Resolution Kit (1200)	QIAxcel DNA High Resolution Cartridge, Buffers, Mineral Oil, QX Intensity Calibration Marker, 12-Tube Strips	929002
QIAxcel DNA Screening Kit (2400)	QIAxcel DNA Screening Cartridge, Buffers, Mineral Oil, QX Intensity Calibration Marker, 12-Tube Strips	929004
QIAxcel DNA Fast Analysis Kit (3000)	QIAxcel DNA Fast Analysis Cartridge, Buffers, Mineral Oil, QX Intensity Calibration Marker, QX DNA Size Marker 50 bp – 1.5 kb, QX Alignment Marker 15 bp/3 kb, 12-Tube Strips	929008
QIAxcel RNA QC Kit v2.0 (1200)	For 100 runs of 12 samples: QIAxcel RNA Quality Control Cartridge, Buffers, Mineral Oil, QX Intensity Calibration Marker, QX RNA Alignment Marker, QX RNA Size Marker 200–6000 nt, QX RNA Denaturation Buffer, 12-Tube Strips	929104
DNA size markers		
QX DNA Size Marker 100 bp – 2.5 kb (50 µL)	50 µl DNA size marker with 14 fragments: 100 bp – 2.5 kb	929559
QX DNA Size Marker 25–500 bp v2.0	050 μl DNA size marker with 17 fragments: 25–500 bp	929560
QX DNA Size Marker 50–800 bp v2.0	050 µl DNA size marker with 11 fragments: 50–800 bp	929561
QX DNA Size Marker 50 bp–1.5 kb (100 µL)	DNA size marker with fragments of 50, 100, 200, 300, 400, 500, 1000, and 1500 bp	929554
QX DNA Size Marker Large-Fragment Kit	Ready-to-use DNA size marker with fragments of 1, 3, 5, 10, and 20 kb; concentration 22.75 ng/µl	929710
RNA size markers		
QX RNA Size Marker 200–6000 (20 μL)	RNA size marker with 8 fragments: 200–6000 nt; concentration: 60 ng/µL	929580

Alignment markers

QX Alignment Marker 15 bp/600 bp (1.5 mL)	Alignment marker with 15 bp and 600 bp fragments	929530
QX Alignment Marker 15 bp/1 kb (1.5 mL)	5Alignment marker with 15 bp and 1 kb fragments	929521
QX Alignment Marker 15 bp/3 kb (1.5 mL)	5Alignment marker with 15 bp and 3 kb fragments	929522
QX Alignment Marker 15 bp/10 kb (1.5 mL)	Alignment marker with 15 bp and 10 kb fragments	929523
QX Alignment Marker 15 bp/5 kb (1.5 mL)	5Alignment marker with 15 bp and 5 kb fragments	929524
QX Alignment Marker 50 bp/1 kb (1.5 mL)	5Alignment marker with 50 bp and 1 kb fragments	929526
QX Alignment Marker 50 bp/5 kb (1.5 mL)	5Alignment marker with 50 bp and 5 kb fragments	929529
QX RNA Alignment Marker (1.5 mL)	1.5 mL RNA alignment marker	929510
Calibration marker		
QX Intensity Calibration Marker (600 µL)	600 µL QX Intensity Calibration Marker	929500
Buffers		
QX DNA Dilution Buffer (15 mL)	15 mL QX DNA Dilution Buffer	929601
QX DNA HS Dilution Buffer (100 mL)	100 mL QX DNA HS Dilution Buffer	929612
QX RNA Dilution Buffer (15 mL)	15 mL QX RNA Dilution Buffer	929602
QX RNA HS Dilution Buffer (100 mL)	100 mL QX RNA HS Dilution Buffer	929613
QX RNA Booster Buffer (100 mL)	100 mL QX RNA Booster Buffer	929614
QX Separation Buffer (40 mL)	40 mL QX Separation Buffer	929603
QX FA Separation Buffer (40 mL)	40 mL QX Separation Buffer; for use with QIAxcel DNA Fast Analysis Cartridges	929606
QX RNA Denaturation Buffer (2 x 1.4 mL)	2 x 1.4 mL QX RNA Denaturation Buffer	929607
QX Wash Buffer (40 mL)	40 mL QX Wash Buffer	929604
QX Mineral Oil (50 mL)	50 mL QX Mineral Oil	929605

Accessories

QX Cartridge Stand

QX Cartridge Stand

929701

QX Cartridge Stand Cover	QX Cartridge Stand Cover	929707
QX Cartridge Stand (with cover)	Cartridge stand with cover for use with the QIAxcel system	929708
QX Buffer Tray	QX Buffer Tray	929702
QX 0.2 mL 12-Tube Strip (80)	80 x QX 0.2 mL 12-Tube Strips	929703
QX Colored 0.2 mL 12-Tube Strip (80)	80 x QX Colored 0.2 mL 12-Tube Strips	929704
QX Nitrogen Cylinder (6)	6 x QX Nitrogen Cylinder	929705
QX 0.2 mL 12-Tube Strip Caps (80)	80 strip caps for use with QX 0.2 mL 12-Tube Strips	929706

For up-to-date licensing information and product-specific disclaimers, see the respective QIAGEN kit handbook or user manual. QIAGEN kit handbooks and user manuals are available at <u>www.qiagen.com</u> or can be requested from QIAGEN Technical Services or your local distributor.

10.4 Appendix D

DNA analysis algorithm description

Summary

The figure below shows a flow diagram of the algorithm. The first step of the data analysis is smoothing. Then, an iterative approach is used to construct the baseline. The objective of this baseline construction is to effectively remove the peaks but accurately follow any other disturbances, e.g. baseline drift and shifts, in the electropherogram. The next step is the cluster detection. Clusters are defined as data subsets in the electropherogram where there is a certain minimal difference in signal between the actual data and the constructed baseline. In this step, no distinction is made between a single peak or multiple not baseline separated peaks. The next step is to split clusters that contain more than one peak. Finally, the actual peak detection is performed, in which peak apex, start point, end point and area are determined.



Smoothing

Smoothing is performed to increase the signal-to-noise ratio (S/N) of the data.

For DNA data the smoothing is performed with a Savitzky-Golay filter using a second order polynomial. For the data smoothing of smear DNA samples and RNA samples a weighted average filter is applied.

Baseline construction

The baseline construction is based on a moving median filter. The median of a dataset is defined as the center point of a dataset sorted in ascending order. A moving median filter processes the input data by replacing each data point by the median of a data subset centered around that datapoints.

If the original dataset is an electropherogram, the electrophoretic peaks will be sorted towards the high end because they have a higher intensity than the noise peaks. This means that as long as the size of the moving median filter (Filter Size) is at least twice the baseline peak width, datapoints of that peak will never reach the center (median) of the data subset, and will therefore be removed.



Mathematically the construction of the baseline using a moving median filter can be described as follows:

$$B(i) = Median_{x \to -r}^{i+r} (E(x))$$

in which B(i) represents the constructed baseline, E(i) the electropherogram, and r the filter rank.

An exception is the analysis of smear DNA and gDNA samples. The baseline for smear DNA analysis is constructed by taking the straight line that results from interpolating between the average signal values of ranges specified at the beginning and at the end of the electropherogram. The baseline for gDNA analysis is constructed by taking a horizontal straight line that starts from the average signal values at the beginning of the electropherogram.

Cluster detection

After the baseline construction, the actual peak detection is started. The first step of this process is called cluster detection. Clusters are defined as regions, i.e. data subsets, in the electropherogram where there is a significant difference between the raw data and the constructed baseline (see figure below). The parameter used to find these regions is the Threshold. The threshold is an imaginary line parallel to the constructed baseline, which marks the minimal height a peak must have (in reference to the constructed baseline) to be detected.



The identified regions can contain either one peak (peaks 1 and 2) or multiple, not baseline separated, peaks (peaks 3 and 4). In either case the region is called a cluster. Initially, the cluster start- and end points are marked as the points where the raw datapoints and the Minimum Peakheight threshold line cross.

The iteration process

As pointed out before, the successful removal of peaks during the baseline construction is dependent on the baseline filter size and peak width. In some cases it is not easy, or even impossible, to find a baseline filter size for optimal removal of all peaks in the electropherogram. This is especially the case in electropherograms with a wide variety of peak widths (requiring a large filter size on a very unstable baseline (requiring a small filter size to accurately preserve these relative high frequency baseline disturbances). The following Figure shows the effect of a too small filter size on the results of the baseline construction.



In the first pass, peak 1 is removed, but in peaks 2 and 3, the constructed baseline starts following the peak. Without any further modifications to the algorithm, this would result in inaccurate peak integration. To solve this problem the algorithm was made iterative. This means that multiple passes through the baseline construction and cluster detection are made, improving the baseline in each pass. The whole iteration process is based on the fact that even though the baseline construction during the first pass was not optimal, normally most of the clusters are still detected.

The principle of the iteration process is that the cluster information obtained in the previous pass is used to optimize the baseline construction in the current pass. In every additional pass, similar to the first pass, a moving median filter is used for the baseline construction. However, before determining the baseline, a check is made for overlaps between the clusters detected in the previous pass and the current filter position. If an overlap is detected, those points are deleted from the sorted data subset, leaving virtually only datapoints corresponding to baseline points in the original electropherogram. The value for the baseline is now determined by taking the median of this new shortened data subset.

The figure of pass 2 shows the new baseline calculated after two passes. The baseline under peak 2 is now correct but the baseline under peak 3 is still incorrect. Note, however, that the cluster start- and end points in peak 3 have shifted considerably closer to the correct position. A third pass therefore will remove a larger part of the peak from the sorted data subset leading to an improved baseline as shown after pass 3.

Cluster splitting

During the cluster detection, clusters with one or several peaks are determined. In cluster splitting step splits clusters with more than one peak into several clusters. This facilitates the peak detection in the next step. The algorithm used is very similar to the cluster detection algorithm. Cluster splitting is performed by iteratively increasing the threshold until clusters with more than one peak decompose to several clusters.

Note: During **Smear Analysis** the goal is to detect smear peaks. As these peaks are relatively broad, the cluster splitting step is omitted during smear peak detection.

Peak detection

After the cluster splitting, each cluster contains exactly one peak. The apex of the peak is determined by looking up the maximum data point in the cluster. Then, the exact start- and end points of the peak are determined. Two criteria are used for this purpose; the first criterion locates the start- and end points of a cluster at the first point (in both directions, starting from the center of the cluster) were the raw data and the constructed baseline cross each other. The second criterion is used to detect cluster borders in case of overlapping peaks (where the raw data and baseline do not cross). In this case the start- and end points of a peak are located at the points where the absolute value of the first derivative remains below a certain threshold for several datapoints. Starting at the center of the cluster, the actual start- and end points of a cluster are found at the first points where one of these two criteria is met.

Note: During the peak detection, peaks that are too close to each other are merged (based on the **Minimum distance** parameter). To ensure detection of overlapping peaks, make sure that the value of the **Minimum distance** is less than the distance between two peaks. Lower the **Minimum distance** if necessary, especially if your peaks are very sharp and closely resolved.

Note: During Smear and gDNA Analysis alignment marker peaks are detected as normal peaks and smear peaks are detected as smear peaks. Each smear peak has an assigned Area of Interest. The borders of the Area of Interest are initially set to the borders of the detected peak.

10.5 Appendix E

Liability clause

QIAGEN shall be released from all obligations under its warranty in the event repairs or modifications are made by persons other than its own personnel, except in cases where the Company has given its written consent to perform such repairs or modifications.

All materials replaced under this warranty will be warranted only for the duration of the original warranty period, and in no case beyond the original expiration date of original warranty unless authorized in writing by an officer of the Company. Read-out devices, interfacing devices and associated software will be warranted only for the period offered by the original manufacturer of these products. Representations and warranties made by any person, including representatives of QIAGEN, which are inconsistent or in conflict with the conditions in this warranty shall not be binding upon the Company unless produced in writing and approved by an officer of QIAGEN.

10.6 Appendix F

Informations de sécurité

Avant d'utiliser le QIAxcel Connect, il est impératif de lire attentivement ce manuel et de porter une attention particulière aux informations de sécurité. Afin de garantir un fonctionnement de l'appareil en toute sécurité et de maintenir l'appareil en bon état de marche, il est impératif de suivre les instructions et les informations de sécurité fournies dans le présent manuel d'utilisation.

Les types d'informations de sécurité suivants sont fournis tout au long du manuel.



La formule **« WARNING » (DANGER)** est utilisée pour avertir des situations pouvant occasionner des dommages corporels à l'utilisateur ou à d'autres personnes.

Les détails sur ces circonstances sont donnés dans un encadré semblable à celui-ci.

	Le terme « CAUTION » (AVERTISSEMENT) est utilisé pour signaler les situations susceptibles de provoquer des détériorations de l'instrument ou d'autre matériel.
<u> </u>	Les détails sur ces circonstances figurent dans un encadré semblable à celui-ci.

Les conseils donnés dans ce manuel ont pour but de venir compléter les exigences de sécurité habituelles en vigueur dans le pays de l'utilisateur et non de s'y substituer.

Utilisation appropriée

WARNING/ CAUTION	Risque de dommages corporels et matériels [W1] L'utilisation non convenable du QIAxcel Connect peut causer des blessures ou des détériorations de l'instrument.
	Le QIAxcel Connect ne doit être utilisé que par du personnel qualifié qui a été formé de façon appropriée.
	Seul un ingénieur du service après-vente QIAGEN est autorisé à effectuer des travaux d'entretien sur le QIAxcel Connect.

Procéder à la maintenance comme décrit dans la section

« <u>Maintenance Procedures</u> » (Procédures de Maintenance). QIAGEN facture les réparations rendues nécessaires suite à une maintenance inappropriée.



Risque de dommages corporels et matériels [W2]

Le QIAxcel Connect est trop lourd pour être soulevé par une personne. Pour éviter des dommages corporels ou matériels, ne pas soulever l'instrument tout seul.

WARNING	Risque de dommages corporels et matériels [W3]
	Ne pas essayer de bouger le QIAxcel Connect pendant son fonctionnement.

En cas d'urgence, éteignez le QIAxcel Connect à l'aide de l'interrupteur d'alimentation situé à l'arrière de l'appareil et débranchez le câble d'alimentation de la prise de courant.

CAUTION	Détérioration de l'appareil [C1]
	Si « Pressure 1» indique « Low » (bas) augmentez la pression du système avant d'utiliser la commande « unlatch » (décrocher). En enlevant la Cartridge et la décrochant à pression réduite, l'appareil peut se déteriorer.

CAUTION	Détérioration de l'appareil [C2]
\wedge	Ne pas utiliser d'eau de Javel, de solvants ou de réactifs contenant des acides, alcalins ou abrasifs pour nettoyer le QIAxcel Connect.

CAUTION	Détérioration de l'appareil [C3]
	Eviter de renverser de l'eau ou des substances chimiques sur le QIAxcel Connect. Tout dommage causé par de l'eau ou des produits chimiques mettra fin à la garantie.

CAUTION	Détérioration de l'appareil [C4]
	Ne pas empiler les instruments et ne pas placer de produits sur le QIAxcel Connect.

Sécurité électrique

Avant l'entretien, débranchez le cordon d'alimentation de la prise de courant.

CAUTION	Endommagement des composants électroniques [C5]
	Avant de mettre l'instrument sous tension, veiller à utiliser la bonne tension d'alimentation. L'utilisation d'une tension d'alimentation incorrecte risque d'endommager les composants électroniques. Pour connaître la tension d'alimentation recommandée, consulter les spécifications indiquées sur la plaque signalétique de l'instrument.

WARNING	Danger électrique [W4]
	Utiliser uniquement le câble fourni avec le QIAxcel Connect.

WARNING	Risque d'électrocution [W5]
\wedge	Toute interruption du conducteur de protection à l'intérieur ou à l'extérieur de l'instrument, ou
	déconnexion du raccord du conducteur de protection (terre) peut rendre l'instrument dangereux. Il est interdit d'interrompre volontairement ce conducteur.
	Présence de tensions mortelles dans l'instrument
	Lorsque l'instrument est relié au secteur, les raccords peuvent être sous tension, et des parties sous tension peuvent être découvertes en ouvrant des capots ou en retirant des pièces (à l'exception de celles auxquelles il est possible d'accéder manuellement).

Afin que le QIAxcel Connect fonctionne de manière satisfaisante et en toute sécurité, conformez-vous aux conseils suivants :

- Le câble d'alimentation doit être relié à une prise d'alimentation disposant d'un conducteur de protection (terre/masse).
- Ne réglez ni ne remplacez aucune pièce interne à l'appareil.
- Ne faites pas fonctionner l'appareil si des capots ou des pièces ont été retirés.
- Si du liquide a été renversé à l'intérieur de l'appareil, éteignez-le et débranchez-le de la prise d'alimentation, puis contactez le support technique de QIAGEN.

• Si vous remplacez les fusibles, ne les remplacer qu'avec des fusibles du même type et équivalent en voltage, comme indiqué sur le fusible.

Si l'appareil devient dangereux sur le plan électrique, empêchez d'autres membres du personnel de l'utiliser et contacter le support technique de QIAGEN ; l'appareil peut être dangereux électriquement si :

- celui-ci ou le câble d'alimentation semble endommagé ;
- il a été stocké dans des conditions défavorables pendant une longue période ;
- il a subi des chocs sévères durant le transport.

Environnement

Conditions de fonctionnement

WARNING	Atmosphère explosive [W6]
	Le QIAxcel Connect n'est pas conçu pour fonctionner dans une atmosphère explosive.

WARNING	Risque d'explosion [W7]
$\mathbf{\Lambda}$	Le QIAxcel Connect a été conçu pour l'utilisation des réactifs et substances fournis par les QIAGEN QIAxcel Kits. L'utilisation des réactifs et substances autres que celles indiquées peut entrainer un risque d'incendie ou d'explosion.

CAUTION	Détérioration de l'appareil [C6]
	La lumière directe du soleil peut décolorer des parties de l'instrument et endommager des parties en plastique. Placer le QIAxcel Connect en dehors de la lumière directe du soleil.

CAUTION	Déterioration de la Cartridge [C7]
	Lors de l'utilisation de la cartouche de Gel, ne pas la laisser plus de 15 minutes hors de la position d'arrêt (« Wash Park ») du compartiment des liquides. Au-delà de cette période, les extrémités des capillaires risquent de se dessécher et d'influencer le bon fonctionnement de la Cartridge. Des capillaires desséchés mettront fin à la garantie.
	Les extrémités des capillaires sont en verre et sont très fragiles.
	Faites attention à ne pas heurter une surface dure. Cela pourrait les casser et influencer le bon fonctionnement de la Cartridge. Des extrémités de capillaires cassées mettront fin à la garantie.

CAUTION	Déterioration de la Cartridge [C8]
	Si vous utilisez moins de 12 échantillons, les puits vides doivent être remplis de QX DNA (HS) Dilution Buffer ou QX RNA (HS) Dilution Buffer. Si les puits restent vides, les capillaires non-utilisés peuvent s'abîmer.

CAUTION	Détérioration de la Cartridge [C9]
\wedge	L'exposition à la lumière solaire directe peut provoquer le blanchiment de la Cartridge et des réactifs à l'intérieur et détériorer les pièces en plastique.
	La Cartridge doit être tenue à l'abri de la lumière directe du soleil.

Produits chimiques

WARNING	Substances chimiques dangere-uses [W8]
\wedge	Certaines substances chimiques utilisées avec cet instrument peuvent être dangere- uses ou peuvent le devenir après que le protocole ait été effectué.
	Toujours porter des lunettes de protection, deux paires de gants et une blouse de laboratoire.
	La personne responsable (par exemple le Chef du laboratoire) doit prendre les précautions nécessaires pour assurer la sécurité de l'environnement du poste de travail et pour être sûr que les opérateurs de l'instrument sont suffisamment formés et non exposés à des quantités dangere-uses de substances toxiques (chimiques ou biologiques) comme défini dans « Material Safety Data Sheets (MSDS) » ou des documents « OSHA, ACGIH ou COSHH ».
	L'évacuation des vapeurs et déchets doit être conforme à tous règlements et dispositions légales - au plan national, départemental et local - concernant la santé et la sécurité.

- * OSHA: Occupational Safety and Health Administration (United States of America).
- † ACGIH: American Conference of Government Industrial Hygienists (United States of America).
- ‡ COSHH: Control of Substances Hazardous to Health (United Kingdom).

Risque de feu [W9]

Lors du nettoyage du QIAxcel Connect avec un désinfectant à base d'alcool, laisser la porte du QIAxcel Connect ouverte pour permettre aux vapeurs inflammables de s'évaporer.

Mise au rebut des déchets

Les consommables et flacons utilisés peuvent contenir des agents chimiques dangereux. Ces déchets doivent être convenablement collectés et mis au rebut conformément aux règles de sécurité locales.

Pour savoir comment mettre au rebut le QIAxcel Connect, voir l'annexe A (« Appendix A »).

Dangers mécaniques

La porte à cartouche et la porte à échantillons du QIAxcel Connect doivent être fermées à tout moment pendant l'usage de l'instrument.

WARNING	Eléments mobiles [W10]
$\mathbf{\Lambda}$	Afin d'éviter tout contact avec les éléments mobiles du QIAxcel Connect lorsqu'il est en marche, toujours fermer les portes de l'instrument pour les échantillons et pour la Cartridge.
	Si les détecteurs ne fonctionnent pas correctement, contacter le Support Technique QIAGEN.

Symboles sur le QIAxcel Connect

Symbole	Emplacement	Description
CE	Plaquette à l'arrière de l'appareil	Marquage CE
	Plaquette à l'arrière de l'appareil	Marquage CSA pour le Canada et les Etats-Unis
UK CA	Plaquette à l'arrière de l'appareil	Marquage UKCA pour la Grande-Bretagne (Angleterre, Pays de Galles, Écosse)
	Plaquette à l'arrière de l'appareil	Fabricant légal
X	Plaquette à l'arrière de l'appareil	Symbole DEEE
25	Plaquette à l'arrière de l'appareil	Marquage RoHS pour la Chine (restriction de l'utilisation de certaines substances dangere- uses dans les équipements électriques et électroniques)
<u>ک</u>	Plaque à l'arrière de l'appareil	Marque RCM pour l'Australie et la Nouvelle-Zélande
i	Plaquette à l'arrière de l'appareil	Lire le mode d'emploi.

Symbole	Emplacement	Description
	Plaquette à l'arrière de l'appareil	Date de fabrication
<u> </u>	Arrière de l'instrument à la prise électrique	Attention, risque de choc électrique
	Avant de l'instrument à l'interrupteur de marche/arrêt	MARCHE/ARRÊT (bouton poussoir)

CAUTION	Endommagement des composants électroniques [C10]
	Utiliser uniquement le câble fourni avec le QIAxcel Connect.

WARNING	Risque de surchauffe [W11]
	Afin de garantir une bonne ventilation, laisser un dégagement d'au moins 10 cm sur les côtés et à l'arrière du QIAxcel Connect. Les fentes et les ouvertures qui assurent la ventilation du QIAxcel Connect ne doivent pas être obstruées.

WARNING/	Risque de dommages corporels et matériels [W12]
	Les capots du QIAxcel Connect peuvent être retirés uniquement par du personnel qualifié correctement formé par QIAGEN.
	L'entretien du QIAxcel Connect doit être effectué uniquement par des représentants formés et autorisés par QIAGEN.

WARNING	Risque d'incendie [W13]
	Ne pas laisser le liquide de nettoyage ou les agents de décontamination entrer en contact avec les pièces électriques du QIAxcel Connect. Lors du nettoyage du Sample Plate Drawer, veiller impérativement à ce qu'aucun liquide ne soit déversé à l'intérieur de l'instrument.

WARNING	Risque de décharge électrique [W14]
\wedge	N'ouvrir aucun panneau sur le QIAxcel Connect autre que ceux décrits dans le présent manuel d'utilisation.
	Risque de dommages corporels et matériels.
	Effectuer uniquement la maintenance spécifiquement décrite dans le présent manuel d'utilisation.

WARNING	Risque d'explosion [W15]
	Lors du nettoyage du QIAxcel Connect avec un désinfectant à base d'alcool, laisser les vapeurs inflammables s'évaporer.

WARNING	Risque d'incendie ou d'explosion [W16]
	En cas d'utilisation d'éthanol ou de liquides à base d'éthanol sur le QIAxcel Connect, manipuler ces liquides avec prudence en observant les règles de sécurité exigées. En cas de déversement de liquide, l'essuyer et laisser les vapeurs inflammables s'évaporer.

WARNING	Vapeurs toxiques [W17]
	Ne pas utiliser de produit à base d'eau de Javel pour désinfecter les accessoires de laboratoire usagés.

10.7 Appendix G

Sicherheitshinweise

Vor der Inbetriebnahme des QIAxcel Connect sollten Sie dieses Handbuch sorgfältig durchlesen – beachten Sie insbesondere die Sicherheitshinweise. Die Gebrauchsanweisungen und Sicherheitshinweise im Handbuch müssen befolgt werden, um einen sicheren Betrieb des Geräts zu gewährleisten und das Gerät in einem sicheren Zustand zu erhalten.

In diesem Handbuch werden die folgenden beiden Kategorien von Sicherheitshinweisen verwendet:

	"WARNING" (WARNUNG) weist auf Situationen und Umstände hin, die zu einer Verletzung des Benutzers oder anderer Personen führen können.
	Nähere Angaben zu der Art der Gefährdung und der Vermeidung solcher Situationen werden in einem Textfeld wie diesem neben der Warnung gemacht.

Der Begriff "CAUTION" (ACHTUNG) weist Sie auf Situationen hin, in denen das Gerät oder andere Geräte beschädigt werden könnten.
Nähere Einzelheiten über diese Situationen werden in einem Textfeld wie diesem beschrieben.

Die in diesem Handbuch enthaltenen Hinweise stellen eine Ergänzung und keinen Ersatz der üblichen Sicherheitsanforderungen dar, die im jeweiligen Land gelten.

Sachgemäße Handhabung

	Verletzungsgefahr und Beschädigung des Gerätes [W1] Die unsachgemäße Bedienung des QIAxcel Connect kann zu einer Verletzung des Benutzers oder zur Beschädigung des Gerätes führen.
	Die Bedienung des QIAxcel Connect darf nur durch qualifiziertes Personal, das entsprechend geschult wurde, erfolgen.
	Die Wartung des QIAxcel Connect darf nur durch Mitarbeiter des QIAGEN Kundendienstes durchgeführt werden.

Führen Sie alle Wartungsarbeiten gemäß den Anweisungen in Abschnitt "<u>Maintenance</u> <u>Procedures</u>" (Wartungsarbeiten) dieses Handbuchs durch. QIAGEN stellt alle Reparaturen in Rechnung, die nachweislich auf eine inkorrekte Wartung zurückzuführen sind.



Verletzungsgefahr und Beschädigung des Gerätes [W2]

Der QIAxcel Connect ist zu schwer um von einer Person gehoben zu werden. Um Verletzungen des Benutzers oder eine Beschädigung des Gerätes zu vermeiden ist davon abzusehen, das Gerät alleine zu heben.

WARNING

Verletzungsgefahr und Beschädigung des Gerätes [W3]

Den QIAxcel Connect während eines Laufes nicht bewegen.

Schalten Sie im Notfall den QIAxcel Connect aus (der Netzschalter befindet sich auf der Geräterückseite), und ziehen Sie den Netzstecker aus der Steckdose.

CAUTION	Beschädigung des Gerätes [C1]
	Wenn die Statusanzeige Pressure 1, « Low » (niedrig) anzeigt, erhöhen Sie den Systemdruck bevor Sie unlatch (entsperren) auslösen. Das Herausnehmen der Kartusche und das Entsperren bei niedrigem Druck kann das Gerät beschädigen.

CAUTION	Beschädigung des Gerätes [C2]
	Verwenden Sie keine Bleichmittel, Lösungsmittel oder säure-, laugen- oder scheuermittel-haltige Reagenzien zur Reinigung des QIAxcel Connect.

CAUTION	Beschädigung des Gerätes [C3]
	Vermeiden Sie es, Wasser oder Chemikalien auf dem QIAxcel Connect zu verschütten. Durch verschüttetes Wasser oder verschüttete Chemikalien verursachte Geräteschäden sind nicht durch die Garantie abgedeckt.

CAUTION	Beschädigung des Geräts [C4]
$\mathbf{\Lambda}$	Stapeln Sie keine Geräte aufeinander und stellen Sie keine Gegenstände auf den QIAxcel Connect.

Schutz vor Stromschlag

Ziehen Sie die Netzanschlusskabel aus den Steckdosen, bevor Sie Wartungsarbeiten am Gerät vornehmen.

CAUTION	Beschädigung von elektronischen Bauteilen [C5]
	Stellen Sie vor dem Einschalten des Geräts sicher, dass die korrekte Versorgungsspannung verwendet wird. Eine falsche Versorgungsspannung kann Schäden an der Elektronik hervorrufen. Überprüfen Sie die empfohlene Versorgungsspannung anhand der technischen Daten auf dem Typenschild des Geräts.

WARNING	Stromschlaggefahr [W4]
A	Verwenden Sie ausschließlich das im Lieferumfang des QIAxcel Connect enthaltene Kabel.

WARNING	Gefährdung durch Elektrizität [W5]
	Jede Unterbrechung des Schutzleiters (Erdungs- bzw. Masseleiter) im Gerät oder außerhalb des Geräts und jede Abtrennung des Schutzleiters am Anschluss der Netzleitung erhöht die Gefahr eines Stromschlags.
	Eine absichtliche Unterbrechung der Schutzleiterverbindung ist verboten.
	Gefährliche Spannung im Gerät
	Auch in ausgeschaltetem Zustand kann an einigen Stellen im Gerät Netzspannung anliegen, wenn das Gerät am Stromnetz angeschlossen ist. Das Öffnen oder Entfernen von Gehäuseteilen kann diese stromführenden Teile freilegen.

Um einen zufriedenstellenden und sicheren Betrieb des QIAxcel Connect zu gewährleisten, befolgen Sie bitte die nachstehenden Hinweise:

- Die Geräte-Netzkabel müssen an Wechselstrom-Steckdosen mit Schutzleiter (Erdungs-/Masseleiter) angeschlossen werden.
- Nehmen Sie im Geräteinneren keine Einstellungen an Teilen vor und wechseln Sie keine Teile aus.
- Nehmen Sie das Gerät nicht in Betrieb, wenn Abdeckungen oder Teile entfernt worden sind.
- Falls Flüssigkeit auf dem Gerät verschüttet wird und in das Gerät läuft, dann schalten Sie es sofort aus, trennen Sie es von der Netzspannung (Stecker ziehen!) und setzen Sie sich mit dem Technischen Service von QIAGEN in Verbindung.
- Beim Austausch der Netzsicherung ersetzen Sie diese nur durch eine desselben Typs und der Stromstärke, die auf dem Etikett/Typenschild angegeben ist.

Falls die elektrische Sicherheit bei der Bedienung des Geräts nicht mehr gewährleistet werden kann, muss das Gerät gegen unbefugte oder unabsichtliche Benutzung gesichert werden. Kontaktieren Sie anschließend den Technischen Service von QIAGEN. Die elektrische Sicherheit des Geräts ist nicht mehr gegeben, wenn:

- das Gerät oder das Netzkabel beschädigt erscheint;
- das Gerät längere Zeit unter ungünstigen Bedingungen Gelagert wurde;
- das Gerät unsachgemäß transportiert worden ist.

Umgebungsbedingungen

Betriebsbedingungen

WARNING	Explosionsfähige Atmosphären [W6]
	Der QIAxcel Connect darf nicht in explosionsfähigen Atmosphären betrieben werden.

WARNING	Explosionsgefahr [W7]
$\mathbf{\Lambda}$	Der QIAxcel Connect ist ausschließlich mit Reagenzien und Substanzen aus den QIAGEN QIAxcel Kits zu benutzen. Die Benutzung von anderen Reagenzien oder Substanzen kann Feuer oder eine Explosion auslösen.

CAUTION	Beschädigung des Gerätes [C6]
$\mathbf{\Lambda}$	Direktes Sonnenlicht kann Teile des Gerätes bleichen und Plastikteile schädigen. Der QIAxcel Connect darf nicht ins direkte Sonnenlicht gestellt werden.

CAUTION	Beschädigung der Cartridge [C7]
	Die Gel Cartridge sollte nicht länger als 15 Minuten außerhalb der Parkposition (« Wash Park ») des Solution Trays aufbewahrt werden. Wird dieser Zeitrahmen überschritten, trocknen die Spitzen der Kapillaren aus. Ausgetrocknete Kapillarspitzen sind nicht durch die Garantie abgedeckt.
	Die Kapillarspitzen sind aus Glas und sehr zerbrechlich. Achten Sie darauf, die Spitzen nicht auf harte Oberflächen aufzusetzen. Dadurch können die Kapillaren brechen und damit die Funktion der Cartridge beeinträchtigen. Zerbrochene Kapillarspitzen sind nicht durch die Garantie abgedeckt.

CAUTION	Beschädigung der Cartridge [C8]
	Wenn weniger als 12 Proben verarbeitet werden, füllen Sie die leeren Probenbehältern mit QX DNA (HS) Dilution Buffer oder QX RNA (HS) Dilution Buffer. Andernfalls können Schäden an den kapillaren Kanälen entstehen.
CAUTION	Beschädigung der Cartridge [C9]
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\wedge	Direktes Sonnenlicht kann zum Ausbleichen der Cartridge und der darin befindlichen Reagenzien führen und Schäden an Kunststoffteilen verursachen.
	Die Cartridge muss vor direkter Sonneneinstrahlung geschützt werden.

Chemikalien

WARNING	Gefährliche Chemikalien [W8]
	Einige der in Verbindung mit diesem Gerät verwendeten Chemikalien sind gesundheitsgefährdend oder können nach Beendigung eines Protokoll-Laufs gesundheitsgefährdend werden. Es sollten immer Sicherheitsbrille, zwei Paar Handschuhe und ein Laborkittel getragen werden. Der Betreiber der Anlage ist für die Gewährleistung der Sicherheit am Arbeitsplatz verantwortlich. Er hat sicherzustellen, dass die Bediener des Gerätes ausreichend geschult sind und nicht gesundheitsgefährdenden Konzentrationen toxischer Substanzen (chemischer oder biologischer) gemäß « Material Safety Data Sheets (MSDS) » oder « OSHA, ACGIH oder COSHH », ausgesetzt sind.
	Bei der Behandlung von Abluft und bei der Abfallbeseitigung sind alle gesetzlichen Regelungen zur Gesundheit und Sicherheit auf nationaler, regionaler und lokaler Ebene zu berücksichtigen.

- * OSHA: Occupational Safety and Health Administration (United States of America).
- † ACGIH: American Conference of Government Industrial Hygienists (United States of America).
- ‡ COSHH: Control of Substances Hazardous to Health (United Kingdom).

WARNING	Feuergefahr [W9]
	Beim Reinigen des QIAxcel Connect mit einem auf Alkohol basierenden Desinfektionsmittel muss die Tür des QIAxcel Connect offen gelassen werden, damit die entzündbaren Dämpfe entweichen können.

Entsorgen von Abfällen

Benutzte Verbrauchsartikel und Behälter könnten gefährliche Chemikalien enthalten. Derartige Abfälle müssen gesammelt und gemäß den Geltenden kommunalen Sicherheitsbestimmungen entsorgt werden.

Weitere Informationen zur Entsorgung vom QIAxcel Connect finden Sie im Anhang A ("<u>Appendix A</u>").

Gefahren durch mechanische Teile

Die Klappen für den Proben- und Cartridge-Einsatz des QIAxcel Connect müssen während des Betriebs geschlossen sein.



Symbole auf dem QIAxcel Connect

Symbol	Fundstelle	Beschreibung
CE	Plakette auf der Rückseite des Gerätes	CE-Zeichen
	Plakette auf der Rückseite des Gerätes	CSA-Zeichen für Kanada und die USA
UK CA	Plakette auf der Rückseite des Gerätes	UKCA-Kennzeichen für Großbritannien (England, Wales, Schottland)
	Plakette auf der Rückseite des Gerätes	Hersteller i.S.d. Gesetzes
X	Plakette auf der Rückseite des Gerätes	WEEE-Zeichen
25	Plakette auf der Rückseite des Gerätes	RoHS-Kennzeichen für China (Einschränkungen in Bezug auf den Gebrauch bestimmter Gefahrstoffe in Elektro- und Elektronikgeräten)

Symbol	Fundstelle	Beschreibung
\bigotimes	Plakette auf der Rückseite des Gerätes	RCM-Zeichen für Australien und Neuseeland
i	Plakette auf der Rückseite des Gerätes	Gebrauchsanweisung beachten.
	Plakette auf der Rückseite des Gerätes	Herstellungsdatum
<u> </u>	Geräterückseite beim Netzeingang	Vorsicht, Stromschlaggefahr
	Gerätevorderseite auf dem Ein/Aus- Schalter	EIN/"AUS" (Druckschalter)

CAUTION

Beschädigung von elektronischen Bauteilen [C10]

Verwenden Sie ausschließlich das im Lieferumfang des QIAxcel Connect enthaltene Kabel.

WARNING	Überhitzungsgefahr [W11]	
	Vergewissern Sie sich, dass zu den Seiten und der Rückwand des QIAxcel Connect ein Mindestabstand von 10 cm eingehalten wird, damit eine ausreichende Belüftung des Geräts gewährleistet ist. Schlitze und Öffnungen, die der Be- und Entlüftung des QIAxcel Connect dienen, dürfen nicht abgedeckt werden.	

WARNING/	Gefahr von Personen- und Sachschäden [W12]
	Die Abdeckungen des QIAxcel Connect dürfen nur durch qualifiziertes, sachgerecht von QIAGEN geschultes Personal entfernt werden.
	Die Wartung des QIAxcel Connect darf nur durch von QIAGEN geschulte und autorisierte Vertreter durchgeführt werden.

WARNING	Brandgefahr [W13]
	Achten Sie darauf, dass die elektrischen Bauteile des QIAxcel Connect nicht in Kontakt mit Reinigungsflüssigkeit oder Dekontaminationsmitteln kommen. Achten Sie insbesondere bei der Reinigung der Sample Plate Drawer darauf, dass keine Flüssigkeitsspritzer in das Gerät gelangen.

WARNING	Stromschlaggefahr [W14]
	Es dürfen ausschließlich die in diesem Benutzerhandbuch genannten Abdeckungen des QIAxcel Connect geöffnet werden.
	Gefahr von Personen- und Sachschäden
	Es dürfen nur Wartungsarbeiten ausgeführt werden, die in diesem Benutzerhandbuch konkret beschrieben sind.

WARNING
Λ

Explosionsgefahr [W15]

Achten Sie bei der Reinigung des QIAxcel Connect mit Desinfektionsmittel auf Alkoholbasis darauf, dass entzündbare Dämpfe sich verflüchtigen können.

WARNING	Brand- oder Explosionsgefahr [W16]
	Bei der Verwendung von Ethanol oder Flüssigkeiten auf Ethanolbasis am QIAxcel Connect müssen diese Flüssigkeiten vorsichtig und in Übereinstimmung mit den erforderlichen Sicherheitsbestimmungen gehandhabt werden. Nehmen Sie verschüttete Flüssigkeit auf und achten Sie darauf, dass entzündbare Dämpfe sich verflüchtigen können.

WARNING	Giftige Dämpfe [W17]
	Verwenden Sie zum Desinfizieren von gebrauchtem Labormaterial keine Bleichmittel.

10.8 Appendix H

License terms

DotNetZip, Microsoft Enterprise Library, and WPF Toolkit

QIAxcel ScreenGel uses the DotNetZip library version 1.9.1.8, the Enterprise Library 6.0, and the WPF Toolkit 3.5, which are licensed under the Microsoft Public License (Ms-PL).

Microsoft Public License (Ms-PL)

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10.9 Appendix I

Document Revision History

Date	Description
August 2023	Removal of obsolete information, improvement of descriptions, specification
	of concentration range for application of the RNA Integrity Score added, list
	update of QIAxcel accessories, and update of Appendix H (License terms).
August 2022	New QIAsphere Connectivity feature and improvement of the Analysis
	settings. Screenshots were updated.
February 2022	Initial Release of the QIAxcel Connect System User Manual.

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