3730xl DNA Analyzer

3730xl Data Collection Software 5 with Windows™ 10 Operating System

Catalog Number A39079

Pub. No. 100077622 Rev. A

Note: For safety and biohazard guidelines, see the "Safety" appendix in the *3730xl DNA Analyzer with 3730xl Data Collection Software 5 User Guide* (Pub. No. 100077621). Read the Safety Data Sheets (SDSs) and follow the handling instructions. Wear appropriate protective eyewear, clothing, and gloves.

This Quick Reference assumes that a Thermo Fisher Scientific technical representative has installed the instrument, that spatial and spectral calibrations have been run, and that an install check has been run.

Product description

The Applied Biosystems[™] 3730xl DNA Analyzer with 3730xl Data Collection Software 5 is a fluorescence-based DNA analysis instrument. It uses capillary electrophoresis technology with 48 or 96 capillaries.

Power on the computer and the instrument

- 1. Power on the monitor and computer.
- 2. In the Log On to Windows dialog box:
 - a. In the User Name field, enter your user name.
 - **b.** In the **Password** field, enter your password.
 - c. Click OK.
- 3. Close the oven door.
- 4. Close the stacker door.
- 5. Close the instrument door.

Fill the water and buffer reservoirs

- 1. Press the **Tray** button to bring the autosampler to the forward position.
- 2. Wait for the autosampler to stop moving and for the green status light to illuminate before you open the instrument door.
- **3.** Unplug the buffer reservoir. Remove the buffer, water, and waste reservoir assemblies from the instrument.

4. Disassemble each reservoir assembly, empty the contents of the reservoirs into an aqueous waste container. Clean each reservoir using deionized water.

Note: Clean the buffer jar, and water, waste, and buffer reservoirs weekly in warm water (≤40°C), followed by a rinse with deionized water.

5. Dry the reservoirs using lint-free wipes, then refill and reassemble the reservoirs.

Note: Add 80 mL 1X run buffer to the buffer reservoir. Add 80 mL high-quality deionized water to the water and waste reservoir assemblies.

- **6.** To prevent damage to the capillary array, inspect each reservoir to ensure that the septa are aligned correctly and fit snugly.
- **7.** Dry the exterior of the reservoirs using lint-free wipes, then place the reservoirs into the instrument.



1 Buffer reservoir

- 2 Water reservoir
- ③ Waste reservoir

Fill the anode buffer jar

Replace the anode buffer at the following times.

- Before each group of scheduled runs, or at least every 48 hours
- After you replenish polymer
- After you replace the capillary array
- After you run the Bubble Remove wizard
- Every time that you change the buffer reservoir

IMPORTANT! The operations list above introduce polymer into the anode buffer jar.



When you replace the 1X run buffer, do not add fresh buffer to old buffer. Discard old buffer, then add fresh buffer.

IMPORTANT! Using old 1X buffer can lead to loss of resolution and data quality.

- 1. Remove the anode buffer jar by pulling it down, then twisting it slowly.
- 2. Empty, then rinse the anode buffer jar using deionized water, then 1X run buffer.
- 3. Add 67 mL 1X run buffer to the jar.
- **4.** Put the anode buffer jar on the instrument with the overflow hole facing you.
- 5. Verify that the electrode is immersed in the buffer.

Check the system

1. Check for bubbles at any point where the pump channels join.

Note: If bubbles are present, replace the buffer if required, then run the **Bubble Remove Wizard**.

2. Ensure that the active calibration is appropriate for the dye set and capillary length you are running.

Connect the instrument to your Thermo Fisher Connect account

- 1. Open the Data Collection Software.
- In the navigation pane of the Data Collection Software, select
 ▲GA Instruments >
 ≣ ga3730 >
 A Thermo Fisher
 Connect.
- **3.** Enter the **User ID** and **Password** for your Thermo Fisher Connect account, then click **Link Account**.

Create a results group (using defaults)

A **Results Group** determines how data is processed, how data files are named, and where data files are saved.

- In the navigation pane of the Data Collection Software, select
 ▲ GA Instruments ▶ □ Results Group.
- 2. Click New.
- 3. In the General tab, enter a Results Group Name.
- 4. Click the Analysis tab, then select the Analysis Type.

For information on autoanalysis, see the documentation for the secondary analysis software.

5. Click OK.

Create a plate record

Note: This section assumes that an instrument protocol and analysis protocol have been created. For more information, see the *3730xl DNA Analyzer with 3730xl Data Collection Software 5 User Guide* (Pub. No. 100077621).

- In the navigation pane of the Data Collection Software, select
 ▲ GA Instruments ▶ ga3730 ▶ Plate Manager.
- 2. Click New.

You can also import a plate record. See "Import a plate record".

3. Enter the plate information in the **New Plate Dialog** box, then click **OK**.

IMPORTANT! If you are creating the plate record to run in auto mode, scan or enter the barcode of a physical plate to link it to this plate record.

- 4. Enter a sample name in the **Sample Name** field, then press **Enter**.
- 5. In the **Results Group** field, select the field, select ∨, then select the **Results Group** from the previous section.
- **6.** In the **Instrument Protocol** field, click the field, select **∨**, then select a protocol.
- **7.** For the remaining fields, select click the field, select ∨, then select an appropriate item for your application.

The settings in these lists are provided by the secondary analysis software. For information on these settings, see the software help in the secondary analysis software.

- **8.** Fill in the remaining rows by manually entering and selecting information, by using **Fill Down** commands, or by copy/pasting.
- 9. Click OK.

Import a plate record

- 1. In the **Plate Manager**, click **Import**, then navigate to the plate record TXT file to import.
- 2. Select the file of interest, then click **Open**.

A progress screen opens. Click **OK** when the import is complete.

Prepare and load sample plates

- 1. Pipet samples into the plate according to the plate layout.
- 2. Briefly centrifuge the plate.

3. Ensure that each sample is positioned correctly in the bottom of its well.

IMPORTANT! If the contents of any well contain bubbles or are not found at the bottom of the well, briefly centrifuge the plate, remove the plate from the centrifuge, and ensure that each sample is positioned correctly in the bottom of its well.



- **4.** Store the plate on ice, protected from light, until you prepare the plate assembly and load the plate in the instrument.
- **5.** Prepare the plate assembly on a clean, level surface. Wear gloves when handling septa. Do not heat plates that are sealed with septa.



Figure 1 Septa assembly

- \bigcirc Plate retainer
- Plate septum
- (3) Septum-sealed sample plate
- ④ Black plate base
- (5) Assembled components

IMPORTANT! Ensure that the holes of the plate retainer and the septa are aligned. The plate can damage the array if the retainer and the septum holes are not aligned.

WARNING! Use only black plate bases with septa-sealed plates. If you are using MicroAmp[™] Fast 96-Well Reaction Plates (0.1 mL), use only blue plate bases and matching retainer.



Figure 2 Heat-sealed assembly

- 1 Plate retainer
- (2) Heat-sealed sample plate
- (3) Gray plate base
- (4) Assembled components

WARNING! Use only gray plate bases with heat-sealed plates. If you are using MicroAmp[™] Fast 96-Well Reaction Plates (0.1 mL), use only dark green plate base and matching retainer.

6. Ensure that the retainer clip is flush with the plate base. A simple way to ensure that they are flush is to run your finger along the edge.

Run the instrument in auto mode

- 1. Select ▲GA Instruments ▶ 📰 ga3730 ▶ Instrument name ▶ ■ Run Scheduler.
- 2. At the top of the screen, select **Instrument** → **Instrument** *name*, then select **Auto**.

Note: The Instrument menu is available only when the Run Scheduler is displayed.

3. Load the plates into the input stack in the stacker.

The plates will be run in the order in which they are placed in the stacker. The bottom plate runs first.

4. Click 🍉 (Run).

As the plates are moved to the autosampler, plate barcodes are scanned and their plate records are used for the run.

IMPORTANT! Ensure that barcodes have been scanned or manually entered for each plate. If the plate barcode scanned by the instrument does not match a barcode in the plate record, the plate will be put in the **Output Stack**.

During an auto run:

- No plates are listed in the Input Stack
- The currently running plate is listed in the **Auto Sampler** field
- Completed plates are listed in the Output Stack

Run the instrument in manual mode

- Select ▲GA Instruments ▶ ∑ ga3730 ▶ Instrument name ▶
 Run Scheduler.
- 2. At the top of the screen, select **Instrument →** *Instrument name* **Run Mode**, then select **Manual**.

Note: The **Instrument** menu is available only when the **Run Scheduler** is displayed.

3. Click **Search A...** [Search plate(s) and add to the input stack] to search for plate records.

The Add Plates to Input Stack dialog box opens.

- Click Find All or type the name of the plate or scan the Plate ID, then click Search.
- 5. Select the plate records to add, then click Add.
- 6. When the needed plate records are listed, click Done.
- 7. As needed, click **Up**, **Down**, or **Remove** to organize the list of plate records in the **Input Stack** list.
- **8.** Load the plates in the In Stack in the order to match the corresponding plate record. The bottom plate runs first and uses the first plate record in the list.

IMPORTANT! The order of the plate records must match the stack order of the plates in the In Stack. If the order does not match, data files will contain the wrong plate record information.

9. Click 🕨 (Run).

During a manual run, the following steps occur.

- Plates are listed in the Input Stack
- The currently running plate is listed in the **Auto Sampler** field
- Completed plates are listed in the **Output Stack**

Controlling the run

You can use the toolbar at the top of the **Data Collection Software** window to control the run.

| То | Click |
|--|-------|
| Start the run | |
| Stop the current run | |
| Stop after the current run | |
| Skip to next run (stops the current run and begins next scheduled run) | + |
| Pause the current run | |
| Resume after pause (start the next scheduled run) | |

Monitor the status of the run

1. Select ▲GA Instruments > 📰 ga3730 > Instrument name > ■ Run Scheduler.



- Input stack—In manual run mode, lists the plate records for the plates to be run. In auto run mode, lists "Unknown".
- (2) Output stack—Validated plates. Plates that pass validation are listed as OK and will be run. Plates that fail validation are listed as Validation failed and will not be run.
- (3) Autosampler—The plate currently in the autosampler. The runs for the current plate are listed under Current Runs.
- ④ **Current runs**—All injections set up for the plate in the plate record
- (5) Plate—The wells to be sampled when the selected injection is run are .
- 2. Monitor the System status indicator in bottom left of screen.
 - —Instrument is running
 - Itashing—Check the Event Log.
 - Instrument has stopped



Monitor a run from InstrumentConnect

- 1. Sign in to **thermofisher.com/cloud**.
- 2. Click **Connect Your Lab**, then click **(**) to access InstrumentConnect.
- 3. Click the instrument to display instrument status.

View data from a completed run (Run History)

You can view the injection run history for processed plates that are stored in the database.

- 1. Select AGA Instruments > 📰 ga3730 > 🛄 Run History.
- 2. Search for the plate record of interest.
- **3.** Select the run of interest, then select any of the icons that are listed below.

| Run History views | lcon |
|-----------------------------|------|
| EPT Viewer | 44 |
| Spatial Calibration Viewer | 777 |
| Capillary Viewer | |
| Array Viewer | |
| Spectral Calibration Viewer | |
| Reextraction | 1 |

Secondary analysis software

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

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

Thermo Fisher Cloud secondary analysis apps

| Analysis | Арр | Description |
|-------------------|--|---|
| Sequencing | Quality Check (QC) module | Automatically checks sequence trace quality. |
| | <mark>∧</mark> ~ | • Provides a results summary that is based on quality parameter settings. |
| | | Auto-flags lower-quality traces for further inspection. |
| | Variant Analysis (VA) module | Finds variants in samples that are sequenced on Applied Biosystems[™] genetic analyzers. |
| | VA | Reports variants at genomic coordinates. |
| En (| EL. | Allows export of variant calls in standard Variant Call Format. |
| | Next-generation Confirmation (NGC) module | Confirms next-generation sequencing (NGS) variants using CE technology. |
| | • Allows visualization of the variants that are detected by both NGS and CE platforms. | |
| | ar 🔪 | Allows export of confirmed variants in standard Variant Call Format. |
| Fragment analysis | Sizing Analysis Module Peak Scanner™ Software | Performs peak sizing. |
| Microsa | <mark>∧</mark> @ | |
| | Microsatellite Analysis Software | Analyzes a mixture of DNA fragments, separated by size, and determines |
| | MSA | microsatellite alleles present in the sample. |

Desktop secondary analysis software for Windows[™] 10 operating system

IMPORTANT! Older versions of the desktop secondary analysis software do not run on the Windows¹ 10 operating system. Older versions may or may not be able to analyze data files that are generated by the 3730xl Data Collection Software 5. Contact Support for information on obtaining the latest versions of software if you did not buy these applications with the instrument.

| Analysis | Software | Minimum version required |
|-------------------|---|--------------------------|
| Sequencing | (Required) Sequencing Analysis Software | 7 |
| | SeqScape [™] Software | 4 |
| | Variant Reporter™ Software | 3 |
| Fragment analysis | (Required) GeneMapper [™] Software | 6 |

Daily maintenance

| Task | Frequency |
|---|--------------------------|
| Ensure that adequate levels of liquid are in the buffer, waste, and water reservoirs. | Before each run |
| Ensure the plate assemblies are properly assembled. | Before each run |
| IMPORTANT! The holes in the plate retainer must align with the holes in the septa, or the capillary tips will be damaged. Make sure the retainer clips are flush with the sides of the plate and with the plate base. A simple way to ensure that they are flush is to run your finger along the edge. | |
| Ensure the plate assemblies are properly positioned on the plate deck. Plates should sit snugly on the deck. | Before each run |
| IMPORTANT! Do not use warped plates. | |
| Check the level of buffer in the anode buffer jar. | Before each run |
| Ensure that the overflow hole is facing toward the front of the instrument and is not blocked. | |
| Replace the 1X run buffer in the buffer jar and the buffer reservoir on the instrument, and the water in the water reservoir. Ensure that the outside of the assemblies are dry. | Every 48 hours |
| When you replace the 1X run buffer, do not add fresh buffer to old buffer. Discard old buffer, then add fresh buffer. | |
| Check for bubbles in the pump block, lower polymer block, interconnect tube, polymer supply tube, and channels. | Daily or before each run |
| Remove all bubbles using the Bubble Remove wizard. | |
| Check the loading-end header of the capillary array. Ensure that the capillary tips are not crushed or damaged. | Daily or before each run |
| Check the level of polymer in the bottle to ensure that the volume is sufficient for runs. | Daily or before each run |
| Clean the instrument surfaces. | Daily |
| Check for leaks around the array knob, interconnect tubing nuts, and check valve. | Daily |

AAA

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The information in this guide is subject to change without notice.

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| Revision | Date | Description |
|----------|----------------|---------------|
| A | 1 October 2018 | New document. |

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