Experion™ RNA StdSens Starter Kit

Instruction Manual

Catalog Number

700-7111

BIO·RAD

About This Manual

This manual serves as an instruction manual for the Experion™ RNA StdSens starter kit. General instructions for using the Experion system and the RNA StdSens analysis kit, can be found in the following:

- The Experion RNA StdSens analysis kit instruction manual (bulletin 10000976)
- The Experion system instruction manual (bulletin 10001312)

In addition, a number of tech notes, product information sheets, and flyers provide more details about the features and applications of the Experion system. For a complete, up-to-date list of all Experion literature, visit www.bio-rad.com/experion/

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Notices

Table of Contents

Introduction — Product Description and Components

The Experion™ System and RNA StdSens Starter Kit

The Experion automated electrophoresis system employs LabChip microfluidic technology to automate protein and nucleic acid electrophoresis and analysis. This system integrates separation, detection, and data analysis within a single platform, using smaller sample and reagent quantities than standard analysis methods. The Experion system accomplishes analysis in a single 30 minute, automated step.

The Experion electrophoresis system includes the following components:

• Automated electrophoresis station
• Microfluidic chips
• Priming station
• Vortex station (for RNA and DNA analyses only)
• Reagents (analysis kits and individual components)
• Experion software

The Experion system includes the following components: 1) automated electrophoresis station, 2) priming station, 3) vortex station used for nucleic acid analysis only, 4) system operation and data analysis tools, and 5) analysis kits, which include the (a) chips and (b) reagents for protein (Pro260 kit), standard-sensitivity RNA (StdSens kit), high-sensitivity RNA (HighSens kit), and DNA (DNA 1K and 12K kits) analyses.

At the center of the Experion system is the microfluidic chip, which contains a series of plastic wells bonded over a small glass plate. The glass plate is etched with an optimized network of microchannels, which connect with the base of the plastic wells. The channels are primed with a gel matrix, the samples are applied to the appropriate wells, and the electrophoresis station directs the samples through these microchannels by controlling the voltages and currents that are applied. The microfluidic chip, in conjunction with the Experion reagents, electrophoresis station, and software accomplish separation, staining, destaining, detection, and basic data analysis without any user intervention.

The Experion RNA StdSens starter kit was designed to introduce new users to RNA analysis with the Experion system as well as to enable troubleshooting, performance maintenance, or performance qualification of the Experion system. The starter kit is derived from the Experion RNA StdSens analysis kit, which is used with the Experion system to determine total RNA and mRNA integrity, purity, and concentration at nanogram levels. A second RNA analysis kit (the Experion RNA HighSens analysis kit) is used for analysis at picogram levels. All kits include the Experion RNA ladder, which has been optimized for automated electrophoresis on the Experion system. They also feature the Experion RNA loading buffer, which contains a 50 bp marker that is used for the proper alignment of samples to the RNA ladder. The starter kit contains the same components as the Experion RNA StdSens analysis kit in addition to other supplies that allow a new user to start using the system immediately.

RNA analysis with the Experion system compares favorably with other, more conventional methods for RNA quantitation, such as UV spectroscopy, with regards to assay linearity, reproducibility, and accuracy. However, in contrast with UV spectroscopy, Experion analysis is relatively unaffected by reagents common to RNA preparation. The additional microfluidic separation step also affords the unique ability to visually evaluate the RNA sample for degradation and for the presence of copurifying nucleic acids, such as genomic DNA and tRNA (Woo and Strong 2006)[1].

For more information on RNA analysis using the Experion system, refer to Appendix A of this manual.

Detection and Quantitation Ranges of the Experion RNA Analysis Kits

	RNA StdSens Analysis Kit		RNA HighSens Analysis Kit	
	Total RNA	mRNA	Total RNA	mRNA
Detection range	5–500 ng/µl	25–250 ng/µl	100–5,000 pg/µl	500–5,000 pg/µl
Quantitation range	25–500 ng/µl	25–250 ng/µl	200–5,000 pg/µl	500–1,000 pg/µl

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[1] Woo L and Strong W, RNA quantitation: A comparative analysis using the Experion automated electrophoresis system, RiboGreen reagent, and ultraviolet spectroscopy, Bio-Rad bulletin 5420 (2006)

Kit Components and Storage Conditions

Experion RNA StdSens Starter Kit Components and Storage Conditions

Component	Storage Conditions
Experion RNA StdSens chips, 3	Ambient
Reagents for analysis	
RNA gel, 1 vial, 1,250 µl	4°C
RNA loading buffer, 1 vial, 900 µl	4°C
RNA stain, 1 vial, 20 µl	4°C
Spin filters, 1	4°C
RNA ladder, 1 vial, 20 µl	–70°C
Mouse liver total RNA sample (500 ng/µl), 20 µl	–70°C
DEPC-treated water, 50 ml	Ambient
Experion electrode cleaner, 25 ml	Ambient
Experion cleaning chips, 2	Ambient
Foam swabs, 25	Ambient
Narrow-bore 20 µl pipet tips, RNase-free, 96 (1 rack)	Ambient
0.65 ml microcentrifuge tubes, RNase-free, 50	Ambient
Experion Training Video: Chip Loading (DVD)	Ambient
Instructions	Ambient

Additional Materials and Equipment Required

- Experion automated electrophoresis station
- Experion priming station
- Experion vortex station
- Microcentrifuge (1,000–10,000 x g)
- Aluminum foil
- Heating block or water bath set at 70°C
- Benchtop vortexer
- Extra spin filter for additional filtration steps (optional, catalog #700-7254)
- Ice

The Experion RNA StdSens Starter Kit Tests

The Experion RNA StdSens starter kit includes all the reagents, consumables, and instructions required to perform two tests that demonstrate the performance of the Experion system for RNA analysis. Each test uses various dilutions and preparations of a mouse liver total RNA sample, and each test follows the standard Experion RNA StdSens assay workflow, which is summarized below.

Experion RNA StdSens Starter Kit Tests

Test	Name	Purpose	Description
1	RNA analysis basics	Used for familiarization with the Experion system and for troubleshooting. Demonstrates the basic features, functions, and reproducibility of an Experion RNA StdSens assay	Run an Experion RNA StdSens analysis of 12 replicates of a total RNA sample (100 ng/µl), check the resulting electropherograms for performance, and determine the reproducibility of quantitation
2	Quantitation across a range of concentrations	Used for demonstration of the quantitative dynamic range of the Experion RNA StdSens assay.	Prepare a dilution series of the total RNA sample (25–500 ng/µl) and use this series to analyze the reproducibility of quantitation across the quantitative range

Before running any of the tests:

- Watch the Experion Training Video: Chip Loading (supplied with this kit and available at www.bio-rad.com/experion/)

- Clean the electrodes as detailed in Appendix B. When working with RNA, pay special attention to cleaning the Experion system to ensure the electrodes remain free from RNases. This is particularly true if one system serves many users within a laboratory

Note: If this is the first time the Experion electrophoresis station is being used, refer to Appendix C for instructions on how to prepare the system for use.

Experion RNA StdSens Starter Kit Test Workflow

Set up the electrophoresis station and clean the electrodes

Equilibrate the reagents

Filter the gel and prepare the gel-stain solution

Prepare the samples and the RNA ladder

Prime the chip

Load the chip

Perform the run within 5 minutes of chip loading and analyze the data
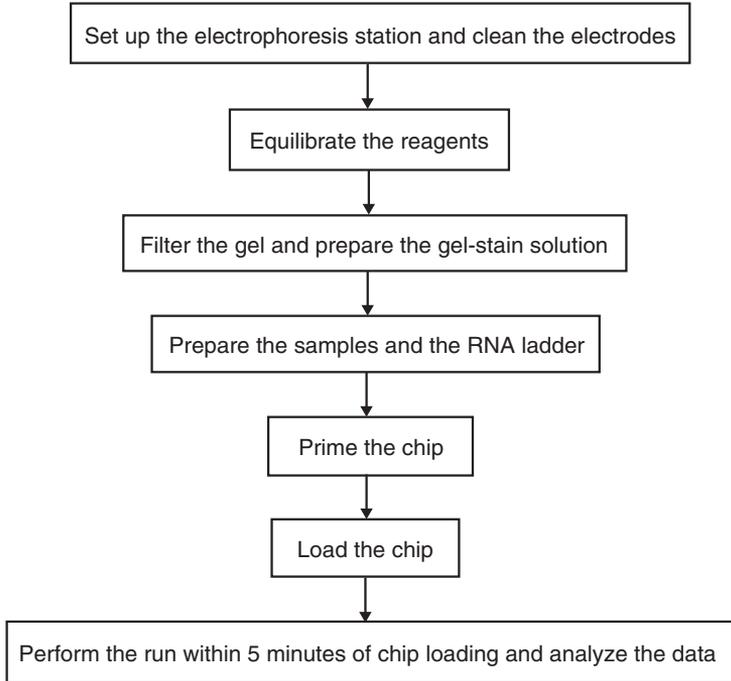
Starter Kit Test 1 — RNA Analysis Basics

1.1 Overview of Test 1

Microfluidic chips use much smaller amounts and volumes of samples and reagents than traditional slab gels. As a result, microfluidic separations are much more sensitive to variability in pipetting technique when compared to traditional electrophoresis methods. Proper pipetting skills and tools are critical to a successful Experion™ analysis.

In this test, you run an Experion RNA StdSens analysis of a single concentration of the total RNA sample, check the resulting electropherograms for proper performance, examine quantitation results, and export the data to determine the reproducibility of the analysis.

Since the use of proper pipet tips can also impact assay performance, use the narrow-bore, RNase-free pipet tips included with the Experion RNA StdSens starter kit (or similar narrow-bore tips) for loading samples and reagents into the microfluidic chip. Narrow-bore tips are required for successful chip loading (other tips can introduce bubbles during chip loading, leading to problems in performing the assay or in the quality of results).

1.2 Assay Procedure

1.2.1 Set Up the Electrophoresis Station and Clean the Electrodes

Note: If this is the first time the Experion electrophoresis station is being used, refer to Appendix C for instructions on how to prepare the system for use.

1. Power on the computer.
2. Power on the Experion electrophoresis station. Push the green button in the center of the front panel. The steady green LED above the button indicates that the unit is on.
3. Launch Experion software. Check the screen to confirm that the instrument and computer are communicating properly.
   When communication has been established:
   • A green dot with the last four digits of the instrument serial number appears in the lower right corner of the main software screen
   • The electrophoresis station icon appears in the upper left corner
   When there is no connection, these indicators are absent and a "disconnected" message appears next to the **Start Run** button ▶ in the upper left corner of the screen. In addition, a grayed-out instrument icon appears in the upper left corner of the software screen.
4. Clean the electrodes as described in Appendix B.

1.2.2 Equilibrate the Kit Reagents

1. Set a heating block or water bath to 70°C. You will use this heating block to denature the samples and the RNA ladder later in the protocol.
2. Remove the kit components from storage.
   • Thaw the following kit components on ice (~15–20 min):
     – Total RNA sample
     – RNA ladder
   • Equilibrate the following kit components to room temperature (~15–20 min):
     – RNA stain (blue cap)

6

– RNA loading buffer (yellow cap)

– RNA gel (green cap)[2]

Notes: Experion RNA stain (blue cap) is light-sensitive and contains DMSO, which is highly hygroscopic. Cap the tube of stain tightly and keep it covered (for example, with foil) at all times to avoid exposure to light.

Inspect the reagents prior to thawing: if the RNA gel (green cap) has been frozen, do not use it.

Do not store reagents at room temperature for more than 2 hr, as this will affect their shelf life.

3. Vortex the contents of each tube and briefly centrifuge to collect the solutions to the bottoms of the tubes. Make sure that the RNA stain solution (blue cap) is completely thawed before proceeding.

4. Prepare 3 µl aliquots of the RNA ladder in RNase-free microcentrifuge tubes. Freeze all aliquots except the one you will be using at –80°C. This ensures integrity of the RNA ladder, a critical component of the RNA StdSens assay.

1.2.3 Filter the Gel and Prepare the Gel-Stain Solution

Notes: The Experion RNA StdSens starter kit includes 1 spin filter, which is sufficient for one preparation of filtered gel (G). You will also use the prepared G in Test 2. The prepared G can be used for up to 4 weeks when stored in the dark at 4°C between each use. The gel can be refiltered once after that to extend its use for another 4 weeks.

Additional spin filter tubes and reagents may also be purchased separately (see Appendix D for ordering information).

This protocol generates enough GS for 3 chips. Increase the amount, if required, by using a 65:1 ratio of gel and stain. **Prepare fresh GS daily**.

1. Filter the gel. Pipet 600 µl RNA gel (green cap) into a spin filter and label the tube with "G" and the date. Centrifuge at 1,500 x g for 10 min. Confirm that all of the gel has passed through the filter and then discard the filter.

2. Prepare the GS. Pipet 65 µl filtered gel into an RNase-free 0.65 ml microcentrifuge tube and label the tube "GS". Add 1 µl RNA stain and vortex. Keep the GS protected from light.

1.2.4 Prepare the Samples and RNA Ladder

1. Once the total RNA sample and RNA ladder have thawed, vortex the tubes briefly and spin down for a few sec. The concentration of the stock total RNA preparation is 500 ng/µl.

2. Denature the RNA sample and the RNA ladder. Transfer 4 µl total RNA sample and 3 µl RNA ladder to separate 0.65 ml RNase-free microcentrifuge tubes labeled "S1" and "L", respectively. Heat the tubes at 70°C for 2 min, and place them on ice immediately after heating. Vortex briefly and spin down. Keep the tubes on ice.

3. Generate the 100 ng/µl sample used in this test by adding 16 µl DEPC-treated water to the sample tube (S1).

4. Vortex to mix the tubes, and spin down.

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[2] If the filtered gel (G) was prepared previously, remove it from storage and equilibrate it to room temperature.

1.2.5 Prime the Chip

Before priming the chip, ensure that all samples, the RNA ladder, and other reagents are prepared and ready to be loaded. If a chip is not used within 5 min of priming and loading, reagents may evaporate, leading to poor results. Use the narrow-bore pipet tips that are supplied with the kit (or equivalent tips) when loading samples or reagents into the chip.

1. Pipet 9 µl GS into the well labeled **GS** (gel priming well) (Figure 1.1). Insert the pipet tip vertically and to the bottom of the well when dispensing. Dispense slowly, and do not expel air at the end of the pipetting step.

Note: Placing the pipet tip at the edge of the well or allowing the gel to slide down the wall of the well may lead to bubble formation at the bottom of the well. Remove any bubbles at the bottom of the well by removing the GS and loading a new aliquot. For help with chip loading, refer to the Experion Training Video: Chip Loading.
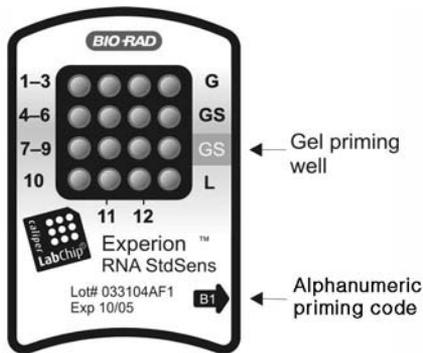

Fig. 1.1. Experion RNA StdSens chip. The locations of the gel priming well (GS) and alphanumeric priming code are indicated.

2. Set the pressure setting to **B** and the time setting to **1**, as specified by the alphanumeric code on the chip (Figure 1.1).

3. Open the Experion priming station by pressing down on the lever on the front.

4. Place the chip on the chip platform, matching the arrow on the chip with the alignment arrow on the chip platform. A post on the chip prevents insertion in the wrong position. Do not force the chip into position.

5. Carefully close the priming station by pressing down on the lid. The lid should snap closed.

6. Press the **Start** button. A "Priming" message appears on the screen of the priming station, the priming station pressurizes, and the timer begins to count down. Priming requires approximately 30 sec to complete. Do not open the priming station during the countdown.

7. An audible signal and "Ready" message indicate that priming is complete. Open the priming station by pressing down on the release lever.

    Note: If the lid sticks, press down on it while pressing down on the release lever.

8. Remove the chip from the priming station, turn it over, and inspect the microchannels for bubbles or evidence of incomplete priming. If the chip is primed properly, the microchannels are difficult to see (it may be helpful to compare a primed chip to a new, unused chip). If you detect a problem, such as a bubble or incomplete priming, prime a new chip.

9. Place the chip on a clean surface for loading.

1.2.6 Load the Chip

1. Pipet 9 µl GS into the other well labeled **GS** (NOT the gel priming well) (Figures 1.1 and 1.2).

2. Pipet 9 µl filtered gel into the well labeled **G** (Figures 1.1 and 1.2).

3. Pipet 5 µl loading buffer (yellow cap) into each sample well (**1–12**) and into the ladder well (**L**).

    Note: Use a new pipet tip for each delivery to prevent contamination of the loading buffer stock. Alternatively, remove 70 µl of loading buffer into an RNase-free tube and pipet 5 µl to each well from this volume.

4. Pipet 1 µl denatured RNA ladder into the well labeled **L**. Every chip must have the RNA ladder loaded into the ladder well labeled **L**.

5. Pipet 1 µl sample into each of the sample wells (**1–12**). Every sample well (**1–12**) must contain solution.

S1	S1	S1	G
S1	S1	S1	GS
S1	S1	S1	GS
S1	S1	S1	L

Fig. 1.2. Chip layout for Experion RNA StdSens starter kit Test 1. S1, sample; GS, gel-stain solution; G, RNA gel; L, RNA ladder.

6. Inspect all wells to make sure that there is no excessive bubble formation from pipetting. Hold the chip above a light background and look down through the wells (Figure 1.3). Dislodge any bubbles at the bottom of the well with a clean pipet tip or by removing and reloading the solution.
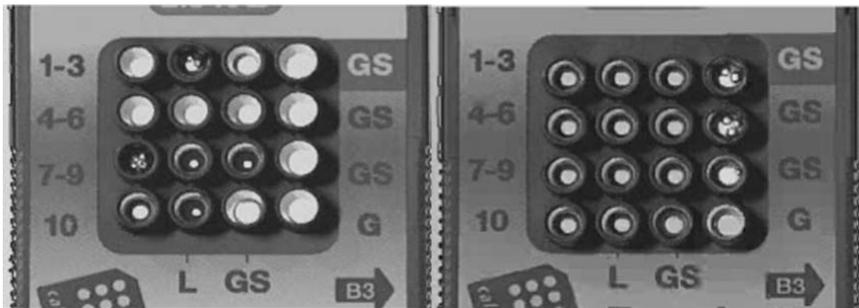
Fig. 1.3. Bubble formation during loading of Experion Pro260 chips. Left, example of bubbles trapped at the bottom of wells. The GS and G wells and sample wells 1, 3, and 4–6 contain no solution. Well 10 is filled properly and has no bubbles; large bubbles have formed at the bottom of wells 8 and 9 and in the ladder well (L). Note the difference in the diameter of the light-colored circles in wells 10 and L. Right, example of bubble formation at the surface of wells. Small bubbles have formed at the surface of the three GS wells on the right side of the chip, and the rest of the wells have no bubbles. Surface bubbles should not cause problems during a run, but bubbles at the bottoms of wells must be removed.

7. Place the chip in the Experion vortex station. Turn on the vortexer, which operates for 60 sec and then automatically shuts off. Remove the chip when the vortexer stops.

8. Place the loaded chip into the Experion electrophoresis station and start the run. Run a loaded chip immediately (within 5 min) or excess evaporation may occur, leading to poor results or a chip performance error.

1.2.7 Run the RNA StdSens Analysis

1. Open the lid of the electrophoresis station by pulling the release latch. Place the primed and loaded chip on the chip platform and close the lid.

2. In the Experion software toolbar, click the **New Run** button [New Run...]. The **New Run** screen opens (Figure 1.4).
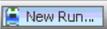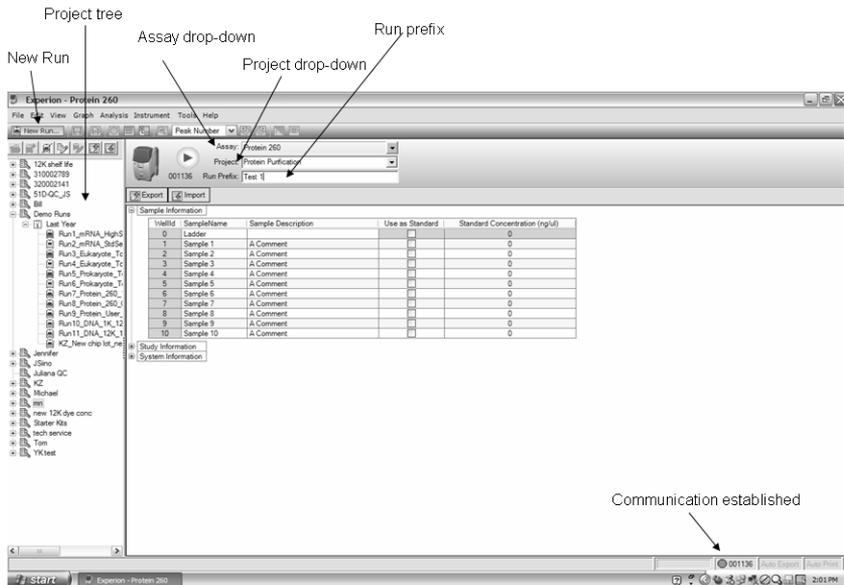
Fig. 1.4. Details of the New Run screen. The green dot in the lower right hand corner indicates that communication between the electrophoresis station and Experion software has been established.

3. From the **Assay** drop-down list, select **StdSens > Eukaryotic total RNA**.

4. Select a project folder for the run from the **Project** drop-down list.

   - Or -

   Create a new project folder by typing a name in the **Project** field or by selecting **File > Project > New**.

   The project folder appears in the project tree after the run has started.

5. Enter a name for the run in the **Run Prefix** field.

6. Click the **Start Run** button ▶ at the top of the window to start the run. The **New Run** dialog opens (Figure 1.5).

11
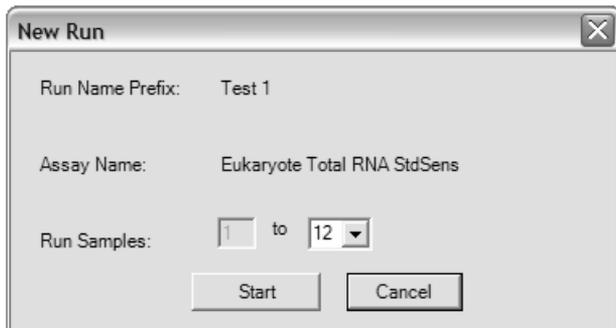
Fig. 1.5. New Run dialog. The Experion system stops analysis when it reaches the number of samples entered.

7. Select the number of samples to be analyzed (12 for both tests).

8. Click **Start** to begin the run. The green LED in the center of the front panel on the electrophoresis station blinks once the run has started.

Warning: Do **not** open the lid of the Experion electrophoresis station until the run is complete. The lid does not lock. Opening the lid prematurely aborts the run.

The system performs a number of checks at the beginning of a run: it confirms that a chip has been inserted, that all wells contain liquid, that electrical connections are made, etc. A calibration counter marks the progress of these calibrations in the upper right corner of the screen.

Note: An "IV Check Error" message indicates the system cannot make electrical contact in one or more of the wells. This often means that either there is a bubble in a well or that a well is empty. If this message appears, abort the run and check the chip for bubbles or empty well(s). Refill the affected well(s), and start the run again.

9. Enter the sample names under **Sample Information**. For example, enter "S1". In addition, add sample details, such as sample concentration. Alternatively, enter this information when the run is complete using the **Run Info** tab.

10. During separation, the sample name is highlighted in gray in the project tree and the electropherogram trace and virtual gel bands appear in real time. By default, the electropherogram of the sample being separated appears in the electropherogram view; in the gel view, the lane corresponding to that sample is outlined in pink and has a dark background. To display the electropherogram from another sample that has already been separated, click on either the sample name in the project tree or on a lane in the virtual gel.

11. When analysis is complete (after approximately 30 min), the instrument beeps and a window opens indicating the end of the run. Select **OK** to the prompt and remove the chip from the chip platform.

12. Clean the electrodes using DEPC-treated water. Add 800 µl DEPC-treated water to a cleaning chip, and insert the cleaning chip into the electrophoresis station for 1 min. Remove the chip and allow the electrodes to dry for 1 min before closing the lid. See Appendix B for more details.

13. Analyze the run and the data.

1.3 Data Analysis

Evaluate the performance of Test 1 and the analysis of the data by Experion software.

1. Evaluate the virtual gel to ensure that all lanes (samples) are visible. Check that the lower marker is present in each sample (indicated by pink triangles) and that it is aligned across all lanes (Figure 1.6).
2. Modify the gel view by changing the fluorescence intensity scale. By default, Experion software displays the virtual gel and electropherograms as "scaled to local", which means the gel lane or trace of each sample is scaled to the highest peak intensity detected in that sample. This is the best method for viewing an electropherogram. To adjust the fluorescence intensity scale so that the virtual gel most closely resembles a gel-based separation, select **Graph > Scale to Global**. The virtual gel appears as it does in Figure 1.6, right panel. All lanes are scaled to the highest peak found across the entire chip, which enables comparison of the concentration differences among the samples in the virtual gel
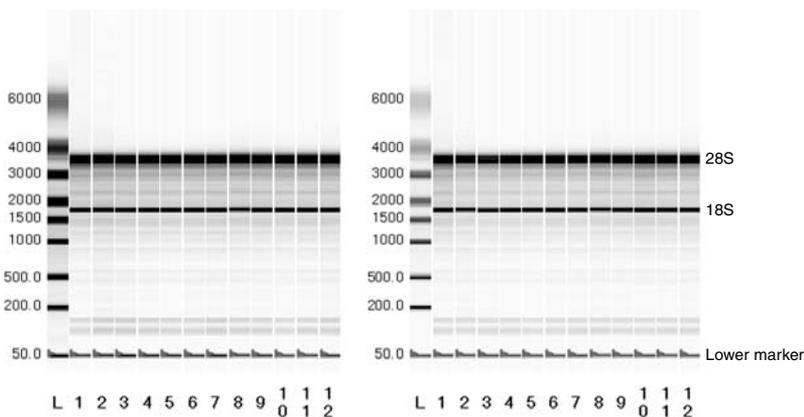
Fig. 1.6. Virtual gel generated by Test 1. Left, virtual gel "scaled to local". Right, virtual gel "scaled to global". L, RNA ladder; lanes 1–12, 100 ng/µl total RNA sample.

3. Evaluate the separation of the RNA ladder (Figure 1.7). This is a critical step in data analysis because much of the automated data analysis performed by Experion software is based on the successful separation of the RNA ladder. To display the ladder electropherogram, click the ladder well in the project tree, or click on the lane labeled **L** in the virtual gel. The electropherogram should resemble the one shown below and should have the following features:

   • A lower marker peak at ~21 sec followed by a set of 8 well-resolved RNA ladder peaks

   • Flat baseline

   • Marker peak at least 5 fluorescence units above the baseline
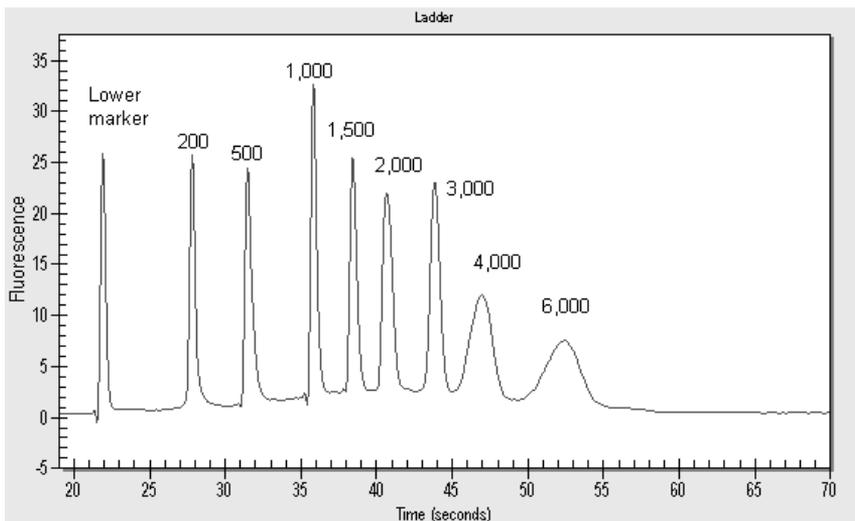
Fig. 1.7. Separation of the RNA ladder (scaled to local). Note the flat baseline and well-resolved peaks. Fragment sizes are given in nucleotides.

4. Examine the separation of the total RNA sample. Click on the sample name in the project tree or on a lane in the virtual gel. The electropherogram appears and should have the following features (Figure 1.8):

- A lower marker peak at ~21 sec, at least 5 fluorescence units above the baseline, and distinguishable from the sample peaks

- Flat baseline

- Well-resolved 18S and 28S rRNA peaks
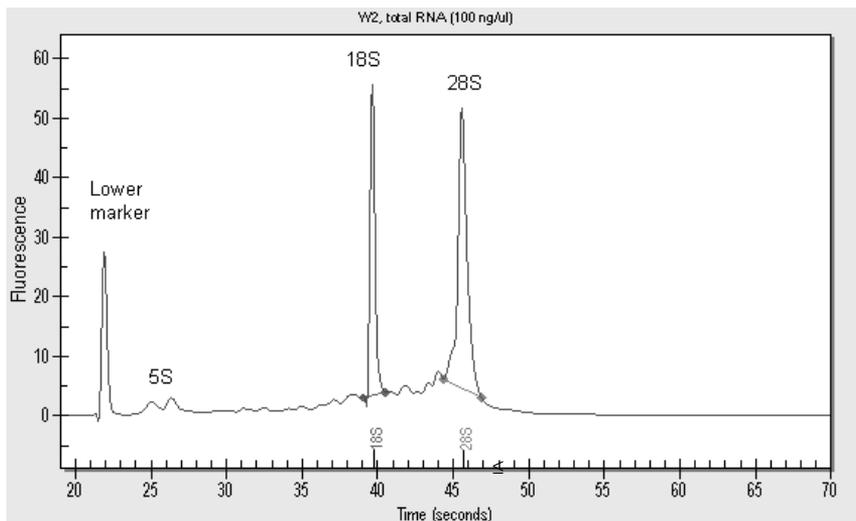
Fig. 1.8. Separation of the 100 ng/µl mouse liver total RNA sample (scaled to local). Results shown are typical for separations of total RNA from eukaryotic sources. The two most prominent peaks, aside from the lower alignment marker, represent 18S and 28S ribosomal RNA (rRNA).

5. Evaluate the data analysis performed by Experion software. Click on the **Results** tab and click on lane 2 in the virtual gel. The Results table for sample 2 opens (Figure 1.9). View the RNA concentration given in the Results table. In the example in Figure 1.9, the total concentration of the sample is 112.14 ng/µl.

Fragment Number	Fragment Name	Start Time	End Time	Area	% of Total Area
1	18S	39.05	40.55	50.00	17.82
2	28S	44.40	46.90	73.62	26.24

RNA Area   280.56
RNA Concentration 112.14 ng/µl
Ratio [28S/18S]   1.47

Fig. 1.9. Results table generated for well 2 in Test 1.

6. To perform a qualitative evaluation of quantitation reproducibility, overlay the electropherograms of replicate samples. For example, in the virtual gel, select lane 1 and hold the Shift key while selecting lane 12. The electropherograms from all 12 wells overlap (Figure 1.10). Tighter overlaps signify better reproducibility.
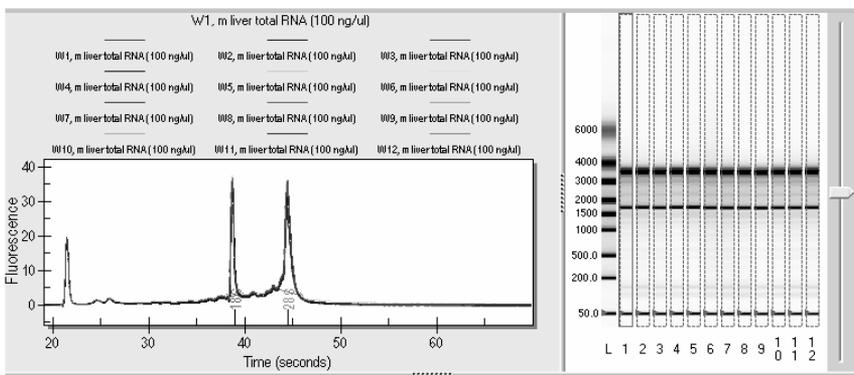
Fig. 1.10. Electropherogram overlay showing the overlay of all 12 of the 100 ng/µl samples.

7. Evaluate the reproducibility of quantitation by calculating the coefficient of variation (%CV), which is defined as the standard deviation/mean x 100. To do this, export the data to a Microsoft Excel or other spreadsheet application either by entering the RNA concentration values manually or by using the data export feature of Experion software. Reproducibility is most strongly influenced by pipetting technique (the ability to reproducibly pipet 1 µl sample and 5 µl loading buffer into each well).

8. To use the export feature of Experion software, select **File > Export Data...** The **Export** dialog opens. Select **Result Table** and **Format as Single Table** and then click **Export**. In the **Save As** window, name the file and designate a folder for it. Click **Save**.

9. Launch Excel software and select **File > Open**. Change **Files of type** to **All**, locate the text file exported in step 8, and select **Open**. The Import wizard opens. Click **Finish**.

10. The data appear in the spreadsheet. Click on cell H1 (Peak Number) and select **Data > Sort** to sort the data by peak number.

11. Insert 3 columns between columns I and J, and label them ***Mean Conc***, ***Stdev***, and ***%CV***, respectively. Use Excel software to determine the mean, standard deviation, and %CV for samples 1–12 (Figure 1.11). The %CV for Test 1 using the mouse liver total RNA sample provided is typically ≤15%

	F	G	H	I	J	K	L
1	Well	Sample Name	Peak Num	RNA Concentration (ng/µl)	Mean	Stdev	%CV
2	1	W1, total RNA (100 ng/ul)	2	117.64			
3	2	W2, total RNA (100 ng/ul)	2	112.14			
4	3	W3, total RNA (100 ng/ul)	2	100.13			
5	4	W4, total RNA (100 ng/ul)	2	126.13			
6	5	W5, total RNA (100 ng/ul)	2	110.04			
7	6	W6, total RNA (100 ng/ul)	2	102.15			
8	7	W7, total RNA (100 ng/ul)	2	105.26			
9	8	W8, total RNA (100 ng/ul)	2	109.28			
10	9	W9, total RNA (100 ng/ul)	2	104.49			
11	10	W10, total RNA (100 ng/ul)	2	117.89			
12	11	W11, total RNA (100 ng/ul)	2	125.69			
13	12	W12, total RNA (100 ng/ul)	2	110.18	111.7517	8.566073	7.7

Fig. 1.11. Excel spreadsheet showing example of calculation of mean, standard deviation, and %CV.

Expected Performance of Test 1[3]. Based on results obtained using 72 samples at 100 ng/µl.

Measurement	Typical Results
Quantitation reproducibility	%CV ≤ 15% for 100 ng/µl total RNA

Quantitation of RNA samples with the Experion system offers reproducibility and accuracy that are comparable to other traditional methods, such as UV spectroscopy. It also offers a level of sensitivity that is similar to that offered by fluorescent probes, such as the RiboGreen reagent (Woo and Strong 2006)[3].

If your results do not appear as they do in these examples, refer to the Troubleshooting section of the Experion RNA analysis kit and Experion system instruction manuals or contact Bio-Rad technical support. Typically, problems with reproducibility are due to problems with pipetting or chip loading. For tips on optimizing your results, refer to the Experion Training Video: Chip Loading and the Essential Practices and Troubleshooting sections of the Experion system instruction manual.

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[3] For more detailed analyses of the expected performance of the Experion RNA StdSens assay, refer to the following Bio-Rad tech notes:

Urban et al., Performance comparison of the Experion automated electrophoresis system and a competing automated system for RNA analysis, Bio-Rad bulletin 5286 (2005)

Woo L and Strong W, RNA Quantitation: A comparative analysis using the Experion automated electrophoresis system, RiboGreen reagent, and ultraviolet spectroscopy, Bio-Rad bulletin 5420 (2006)

Starter Kit Test 2 — Quantitation Across a Range of Concentrations

2.1 Overview of Test 2

In this test, you analyze a dilution series of the total RNA sample to examine the reproducibility of quantitation across the entire quantitation range of the Experion™ RNA StdSens analysis kit (25–500 ng/μl).

Note: The quantitative range of the Experion RNA StdSens analysis kit is different from its detection range: whereas the detection range of the assay for total RNA is 5–500 ng/μl, the quantitative range is 25–500 ng/μl (at the lower end of the quantitative range, the signal-to-noise ratio is lower). Though RNA detection and purity analysis can be performed at concentrations of less than 25 ng/μl using the StdSens assay, quantitation at these low concentrations is not recommended since RNA concentrations of <25 ng/μl are outside the linear range of detection. For quantitation of samples containing 0.1–5 ng/μl RNA, use the Experion RNA HighSens analysis kit.

2.2 Assay Procedure

1. Set up the electrophoresis station, clean the electrodes, thaw and equilibrate the reagents, and prepare the gel (G) and gel-stain solution (GS) as outlined in Sections 1.2.1–1.2.3. If the G has already been prepared, equilibrate it as detailed in Section 1.2.2.

2. Denature the total RNA sample and RNA ladder as described in Section 1.2.4, steps 1–2, using 3 μl RNA ladder and 7 μl total RNA sample (stock preparation). Label the total RNA sample "S1".

3. This test uses a dilution series of the total RNA sample. Label 3 microcentrifuge tubes (S1–S3) and prepare the following dilutions:

   Tube S1 (500 ng/μl):   No additions; this is the stock preparation you denatured

   Tube S2 (100 ng/μl):   8 μl DEPC-treated water + 2 μl total RNA from tube S1

   Tube S3 (25 ng/μl):   6 μl DEPC-treated water + 2 μl total RNA from tube S2

4. Vortex to mix the tubes, and spin down.

5. Prime and load the chip as described in Sections 1.2.5 and 1.2.6 and using the chip layout shown in Figure 2.1.

S3	S2	S1	G
S3	S2	S1	GS
S3	S2	S1	GS
S3	S2	S1	L

Fig. 2.1. Chip layout for Experion RNA StdSens starter kit Test 2. S1–S3, sample numbers; GS, gel-stain solution; G, RNA gel; L, RNA ladder.

6. Run the RNA StdSens analysis as described in Section 1.2.7. Enter the sample names and information appropriate to the chip layout in Figure 2.1.

2.3 Data Analysis

Evaluate the run and the analysis of the data by Experion software.

1. Evaluate the virtual gel to ensure that all lanes (samples) are visible. Check that the lower marker is present in each sample (indicated by pink triangles) and that it is aligned across all lanes (Figure 2.2).

2. Select **Graph > Scale to Global** to adjust the fluorescence intensity scale so that the virtual gel most closely resembles a gel-based separation (Figure 2.2). All lanes are scaled to the highest peak found across the entire chip, which enables comparison of the concentration differences among the samples in the virtual gel.
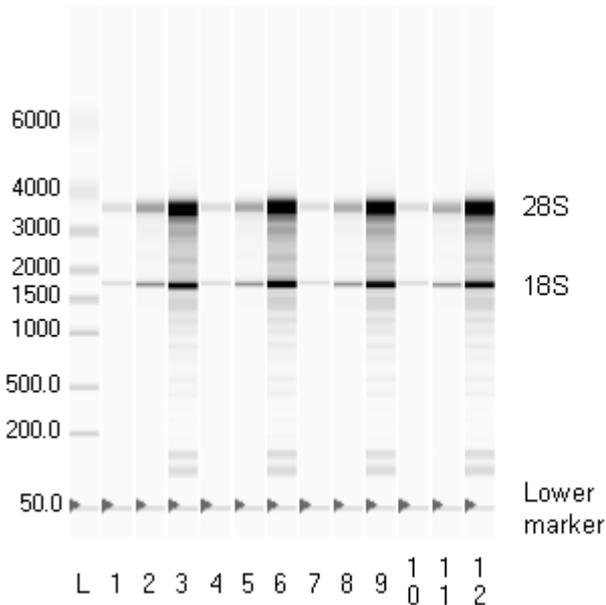

Fig. 2.2. Virtual gel generated by Test 2. L, RNA ladder; lanes 1, 4, 7, and 10, 25 ng/µl total RNA; lanes 2, 5, 8, and 11, 100 ng/µl total RNA; lanes 3, 6, 9, and 12, 500 ng/µl total RNA. In this example, the virtual gel was "scaled to global" to emphasize the differences in sample concentrations.

3. Evaluate the separation of the RNA ladder. This is a critical step in data analysis because much of the automated data analysis performed by Experion software is based on the successful separation of the RNA ladder. To display the ladder electropherogram, click the ladder well in the project tree, or click on the lane labeled **L** in the virtual gel. The electropherogram should resemble the one shown in Figure 1.7.

4. Examine the separation of one of the total RNA samples. Click on a sample name in the project tree or on a lane in the virtual gel. The electropherogram appears and should have the features described for Figure 1.8 (a lower marker peak at ~21 sec that is at least 5 fluorescence units above the baseline and distinguishable from the sample peak, a flat baseline, and well-resolved 18S and 28S rRNA peaks).

5. Evaluate the data analysis performed by Experion software. Click on the **Results** tab and view the RNA concentrations given for each sample in the Results table. See Test 1 for an example.

6. To perform a qualitative evaluation of quantitation reproducibility, overlay the electropherograms of replicate samples. For example, in the virtual gel, select lane 3 and hold the Ctrl key while select lanes 6, 9 and 12. The electropherograms from wells 3, 6, 9 and 12 (500 ng/µl samples) overlap (Figure 2.3). Repeat for the other concentrations. Tighter overlaps signify better reproducibility.
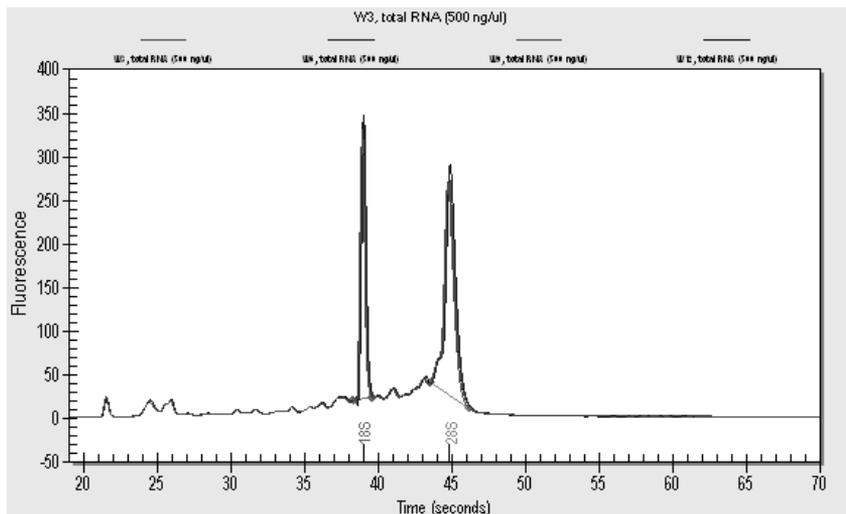
Fig. 2.3. Electropherogram overlay of the 500 ng/µl samples.

7. To perform a quantitative evaluation of reproducibility, calculate the coefficient of variation (%CV), which is defined as the standard deviation/mean x 100. To do this, export the data to a Microsoft Excel or other spreadsheet application either by entering the values manually in Excel or by using the data export feature of Experion software as described in Test 1. The data appear in the spreadsheet after sorting by peak number (Figure 2.4).

	F	G	H	I	
1	Well	Sample Name	Peak Number	RNA Concentration (ng/µl)	
2	1	W1, m liver total RNA (25 ng/ul)	2	32.26	
3	2	W2, m liver total RNA (100 ng/ul)	2	102.83	
4	3	W3, m liver total RNA (500 ng/ul)	2	659.64	
5	4	W4, m liver total RNA (25 ng/ul)	2	30.82	
6	5	W5, m liver total RNA (100 ng/ul)	2	96.63	
7	6	W6, m liver total RNA (500 ng/ul)	2	672.17	
8	7	W7, m liver total RNA (25 ng/ul)	2	33.89	
9	8	W8, m liver total RNA (100 ng/ul)	2	110.59	
10	9	W9, m liver total RNA (500 ng/ul)	2	678.9	
11	10	W10, m liver total RNA (25 ng/ul)	2	31.73	
12	11	W11, m liver total RNA (100 ng/ul)	2	95.61	
13	12	W12, m liver total RNA (500 ng/ul)	2	725.5	

Fig. 2.4. Excel spreadsheet showing Test 2 data sorted by peak number.

8. Insert a column between columns H and I, and label it *Exp Conc* (Expected Concentration). Enter the expected concentration for each well.

9. Click on cell I1 (Exp Conc) and select **Data > Sort** to sort the data by expected concentration (Figure 2.5).

	F	G	H	I	J	
1	Well	Sample Name	Peak Number	Exp Conc	RNA Concentration (ng/µl)	
2	1	W1, m liver total RNA (25 ng/ul)	2	25	32.26	
3	4	W4, m liver total RNA (25 ng/ul)	2	25	30.82	
4	7	W7, m liver total RNA (25 ng/ul)	2	25	33.89	
5	10	W10, m liver total RNA (25 ng/ul)	2	25	31.73	
6	2	W2, m liver total RNA (100 ng/ul)	2	100	102.83	
7	5	W5, m liver total RNA (100 ng/ul)	2	100	96.63	
8	8	W8, m liver total RNA (100 ng/ul)	2	100	110.59	
9	11	W11, m liver total RNA (100 ng/ul)	2	100	95.61	
10	3	W3, m liver total RNA (500 ng/ul)	2	500	659.64	
11	6	W6, m liver total RNA (500 ng/ul)	2	500	672.17	
12	9	W9, m liver total RNA (500 ng/ul)	2	500	678.9	
13	12	W12, m liver total RNA (500 ng/ul)	2	500	725.5	

Fig. 2.5. Excel spreadsheet showing Test 2 data sorted by expected concentration.

10. Insert 3 columns after column J, and label them *Mean Conc*, *Stdev*, and *%CV*. Use Excel software to determine the mean, standard deviation, and %CV for each set of replicates (each concentration examined) (Figure 2.6).

	F	G	H	I	J	V	W	X	
1	Well	Sample Name	Peak Number	Exp Conc	RNA Concentration (ng/µl)	Mean	Stdev	%CV	
2	1	W1, m liver total RNA (25 ng/ul)	2	25	32.26				
3	4	W4, m liver total RNA (25 ng/ul)	2	25	30.82				
4	7	W7, m liver total RNA (25 ng/ul)	2	25	33.89				
5	10	W10, m liver total RNA (25 ng/ul)	2	25	31.73	32.175	1.288733	4.0	
6	2	W2, m liver total RNA (100 ng/ul)	2	100	102.83				
7	5	W5, m liver total RNA (100 ng/ul)	2	100	96.63				
8	8	W8, m liver total RNA (100 ng/ul)	2	100	110.59				
9	11	W11, m liver total RNA (100 ng/ul)	2	100	95.61	101.415	6.898722	6.8	
10	3	W3, m liver total RNA (500 ng/ul)	2	500	659.64				
11	6	W6, m liver total RNA (500 ng/ul)	2	500	672.17				
12	9	W9, m liver total RNA (500 ng/ul)	2	500	678.9				
13	12	W12, m liver total RNA (500 ng/ul)	2	500	725.5	684.0525	28.76113	4.2	

Fig. 2.6. Excel spreadsheet showing example of calculation of mean, standard deviation, and %CV.

11. Plot the results as a function of sample concentration (Figure 2.7). The overall trend should be linear.
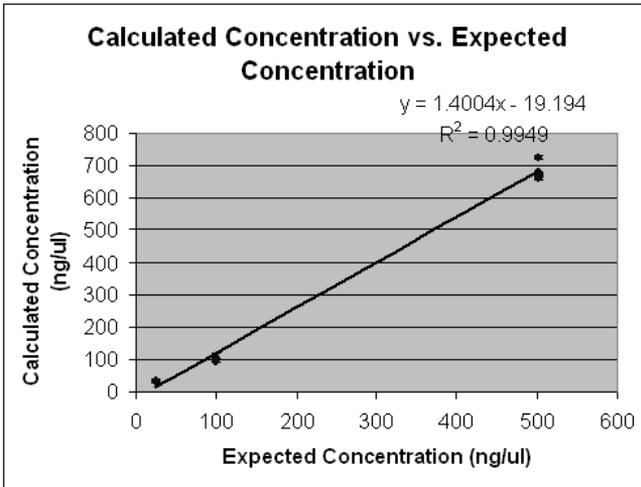
Fig. 2.7. Plot of concentration calculated by Experion software as a function of expected concentration.

Expected Performance of Test 2. Based on results obtained using a total of 6 chips.

Measurement	Typical Results
Quantitation reproducibility	%CV ≤ 20% for total RNA (25–500 ng/µl)

If your results do not appear as they do in these examples, refer to the Troubleshooting section of the Experion RNA StdSens and Experion system instruction manuals or contact Bio-Rad technical support. Typically, problems with reproducibility are due to problems with pipetting or chip loading. For tips on optimizing your results, refer to the Experion Training Video: Chip Loading and the Essential Practices and Troubleshooting sections of the Experion system instruction manual.

Appendix A — How the Experion™ System Works

The Experion electrophoresis system performs electrophoresis of samples within a microfluidic chip. Within each chip, a series of microchannels connects the sample wells to a separation channel and buffer wells. A set of electrodes in the electrophoresis station applies a voltage across the microchannels, causing charged molecules in the samples to migrate into and through the separation channel. Samples are run sequentially, with a sufficient lag between them to prevent cross-contamination. For separation, the microchannels are filled with a proprietary gel-stain solution (GS) that acts as a sieving matrix; therefore, the sample fragments migrate through the separation channel at a rate based on their size and charge. Finally, sample fragments interact with a fluorescent dye during separation and are detected as they pass by a laser and photodiode detector.

RNA analysis is accomplished with the Experion RNA analysis kits. Two RNA analysis kits are available, and they differ in the level of sensitivity that they offer: the Experion RNA StdSens analysis kit is used for the evaluation and quantitation of samples containing as little as 5 ng/µl RNA, and the Experion HighSens analysis kit is used for higher sensitivity evaluation of as little as 100 pg/µl RNA. Both kits use the following steps for RNA analysis:

1. **Preparing the chip (priming and loading)** — Priming fills the microchannels of the microfluidic chip with the GS, which contains both the sieving matrix and fluorescent dye. RNA samples are then prepared and added to the sample wells along with the loading buffer.

2. **Vortexing the chip** — Vortexing ensures that samples and loading buffer are mixed. Vortexing the chip itself reduces the number of sample manipulation and pipetting steps required, improving reproducibility.

3. **Running the chip** — The chip is inserted into the electrophoresis station and, as the instrument lid is closed, the electrodes come in contact with the solution in the wells. Voltage is applied to the sample wells of the chip, causing the charged RNA fragments to migrate into the separation channel. In the separation channel, the RNA fragments separate as they move at different rates through the gel matrix, depending on their size. During separation, the fluorescent dye intercalates into the RNA strands.

4. **Detecting the fragments** — As the fragments migrate towards the end of the separation channel, a laser[4] excites the dye, causing it to fluoresce if it is bound to RNA (excess, free dye does not fluoresce appreciably). A photodiode detects the fluorescence, and Experion software plots the fluorescence intensity versus time to produce an electropherogram and a virtual gel image.

5. **Analyzing the data** — Following separation, Experion software subtracts background noise, removes spikes, identifies and integrates peaks, and assigns their sizes and concentrations. Following analysis, parameters may be changed and the data reanalyzed. The software also assigns 18S and 28S rRNA peaks based on their size and calculates the total concentration of the sample by comparing the area under the sample electropherogram (excluding the lower marker) to the area under the ladder electropherogram.

[4] 635 nm excitation, 685 nm filter collection.

How Experion Software Analyzes RNA

Data Presentation

As the photodiode detects fluorescent signals from the dye-RNA complexes, Experion software converts the signal into an electropherogram (a plot of fluorescence vs. time). Experion RNA electropherograms differ depending on the type of sample analyzed (eukaryotic or prokaryotic total RNA or mRNA), but they generally have the following features:

Sample peak(s) — signal(s) generated by the sample RNA fragment(s)

Lower marker (LM) — signal generated by the internal lower (50 bp) marker, which is included in the loading buffer and used to align the separations across all wells in the chip
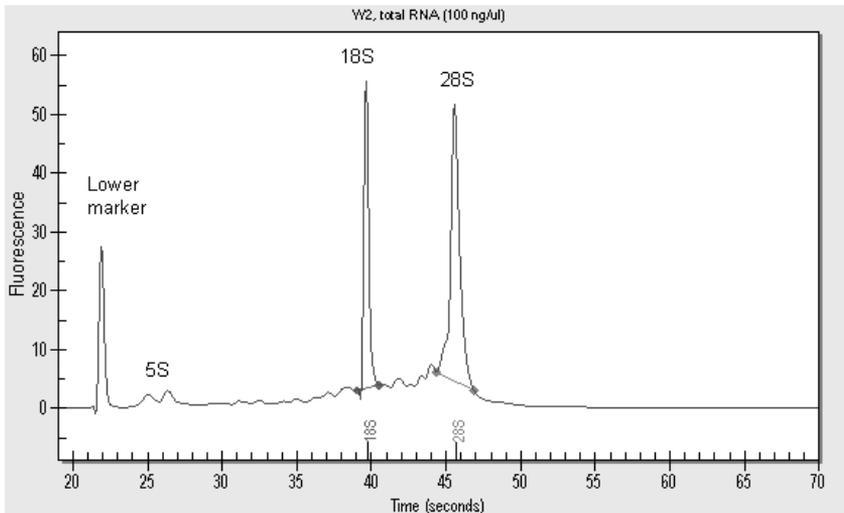

Electropherogram generated by Experion RNA StdSens analysis of eukaryotic total RNA. Results shown are typical for separations of total RNA from eukaryotic sources. The two most prominent peaks, aside from the lower alignment marker, represent 18S and 28S ribosomal RNA (rRNA).
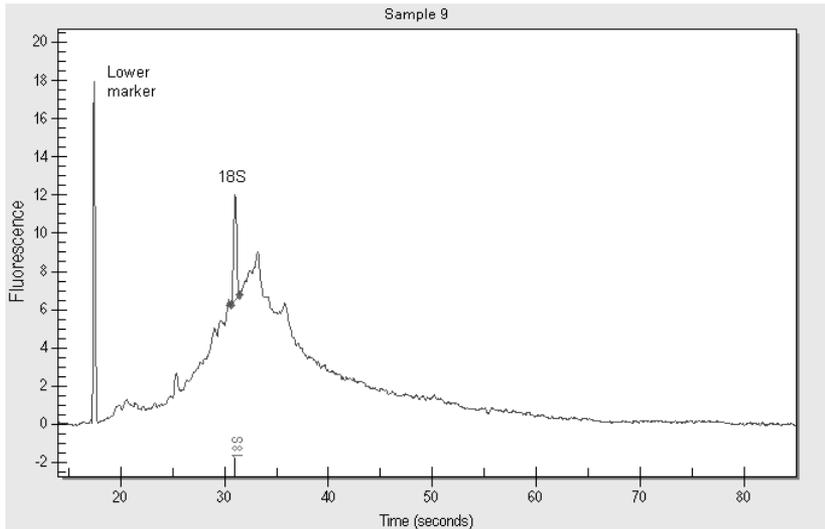
Electropherogram generated by Experion RNA HighSens analysis of mRNA. For mRNA assays, the sample peak is usually broad. Any 18S ribosomal RNA contamination is identified by the software.

Once separation occurs, these data are converted into a gel-like image, or virtual gel. Each lane in the virtual gel corresponds to a different sample. The sample and lower markers seen in the electropherogram also appear in the virtual gel.
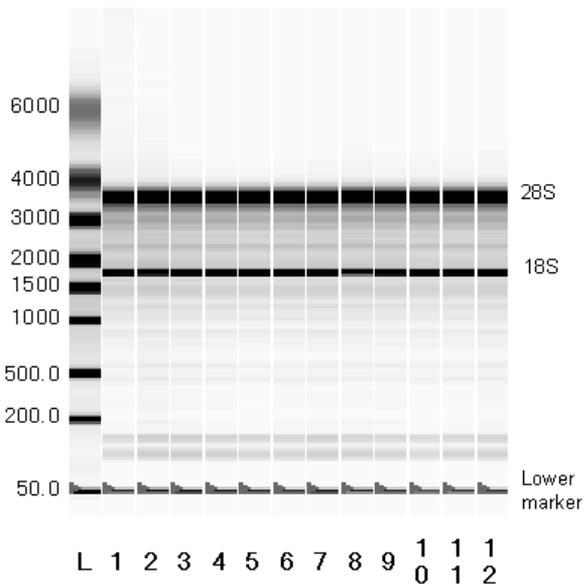
Virtual gel generated by Experion RNA StdSens analysis of eukaryotic total RNA (scaled to local). Pink triangles indicate the positions of the lower marker.

When separation is complete, RNA fragments are identified as peaks in the electropherograms and as bands in the virtual gel. The results of data analysis are also tabulated and presented in a Results table at the end of analysis. The types of data available include the following:

- Concentration (in ng/µl or pg/µl, depending on the kit used)
- % rRNA contamination (mRNA analysis)
- 28S/18S (eukaryotic) or 23S/16S (prokaryotic) ratio (total RNA analysis)
- % of Total area

Fragment Number	Fragment Name	Start Time	End Time	Area	% of Total Area
1	18S	30.60	31.55	6.37	2.11

RNA Area	302.08
RNA Concentration	754.22 pg/µl
rRNA Contamination	2.11 %

Results table generated by RNA HighSens analysis of mRNA. Note that whereas the mRNA assay indicates rRNA contamination, the total RNA assay shows the ribosomal ratio, as shown in Fig. 1.9.

Data Analysis: Normalization, Sizing, and Quantitation

Following separation, RNA fragments are analyzed using one or more of the following:

- An internal marker for normalizing the migration times of samples in different wells
- RNA ladder for determining fragment size (sizing) and concentration (quantitation) of samples

Normalization: Aligning the Peaks

To compensate for small variations in samples or factors influencing separation (pH, dye concentration, porosity of matrix, injection volumes, etc.), Experion RNA analysis uses an internal marker to normalize the migration times between samples. The 50 bp lower marker is included in the RNA loading buffer and, therefore, appears in all samples. Inclusion of this marker and the normalization process ensures that the system software properly identifies peaks. Once aligned, the software then further analyzes the data as described below.

Sizing

The first sample to be analyzed is the RNA ladder, which has been optimized for automated electrophoresis on the Experion system. The RNA ladder contains eight purified 200–6,000 nt RNA fragments. Experion software constructs a standard curve of migration time as a function of size from the RNA ladder separation. It then calculates the size of the RNA fragments from the sample wells by comparing their migration times to the standard curve.

Quantitation

Experion software begins RNA quantitation by comparing the time-corrected area (corrected area) under the electropherogram of a sample (excluding the lower marker) with the corrected area under the electropherogram of the RNA ladder. It then uses this value and the known ladder concentration (160 ng/µl for the RNA StdSens assay, and 1,000 pg/µl for the RNA HighSens assay) to determine the RNA concentration of the sample.

Appendix B — Cleaning the Electrodes

Two types of cleaning procedures ensure the best results from the Experion™ electrophoresis station: 1) a deep-cleaning procedure, which applies electrode cleaner directly to electrode pins using swabs, and 2) a routine procedure, which uses cleaning chips and is performed directly before and after every run.

Method 1: Deep Cleaning

This manual procedure applies Experion electrode cleaner directly to the 16 platinum pins in the electrode manifold. This method is the most stringent method of cleaning the Experion electrode pins and helps to ensure RNase-free operation of the system. The need for this deep cleaning will vary from lab to lab, depending on the frequency of use of the Experion system and on the types of assays (protein, RNA, or DNA analysis) that are performed. Recognize that an Experion electrophoresis station used to analyze protein samples will become contaminated with RNases and should be deep cleaned prior to any RNA analysis.

Perform deep cleaning:

• Prior to first use of the Experion electrophoresis station

• Between a protein and an RNA analysis

• Any time when RNase contamination is suspected or any contamination (for example, salt deposits or other precipitates) is visible on the electrodes

• Whenever a chip has been left in the electrophoresis station for an extended period of time (for example, overnight)

Perform this procedure with the electrode manifold in place (installed in the electrophoresis station). It requires the following supplies, all of which are included in the Experion RNA StdSens starter kit:

• Experion electrode cleaner

• Experion DEPC-treated water

• Foam swabs

Warnings: Keep the chip platform completely dry during this procedure. Cover the chip platform using plastic wrap and a paper towel to protect it from liquid.

1. Ensure that power to the electrophoresis station has been shut off.

2. Add 0.5–1 ml Experion electrode cleaner to a microcentrifuge tube. Insert a swab into the solution until it is saturated with the electrode cleaner.

3. Use the swab to gently scrub each electrode pin one at a time, on all four sides. Do not press too hard on the pins, as they may bend. Finally, clean the tip of each pin. Add more electrode cleaner to the swab as necessary.

   -Or-

   Move the swab up and down, and side to side, 2–3 times along the columns and rows of pins. Finally, clean the tips of each of the pins. Add more electrode cleaner to the swab as necessary.

4. Repeat steps 2 and 3 twice, each time with a fresh swab soaked in DEPC-treated water.

5. Fill a cleaning chip with 800 µl DEPC-treated water and place it in the electrophoresis station for 1 min.

6. Remove the chip and allow the pins to dry completely by leaving the lid open for at least 10 min.

Note: Refer to the Experion system instruction manual for more details and images.

Method 2: Routine Cleaning (RNA Analysis)

When performing RNA analyses, there are two types of routine cleaning procedures: one is used before each run and involves use of Experion electrode cleaner and DEPC-treated water (not required if a deep cleaning has just been performed), and the other is performed after each run and involves a rinse using only DEPC-treated water.

Notes: Label the RNA cleaning chips when you begin: dedicate one for use with DEPC-treated water and the other for electrode cleaner.

If performing an RNA analysis after a protein analysis, perform the deep cleaning procedure described above prior to RNA analysis.

Warning: Never store the cleaning chip inside the electrophoresis station. Store the empty cleaning chip covered to keep the wells clean.

Preparing the Cleaning Chips

If the cleaning chips are being used for the first time, wash them with electrode cleaner to remove any RNase contamination:

1. Fill the two cleaning chips with 800 µl Experion electrode cleaner. Let sit for 5 min.

2. Rinse the chips by treating with 800 µl DEPC-treated water for 1 min.

3. Repeat the DEPC-treated water rinse step 4 more times.

Method 2A: Cleaning Before an RNA Assay

Before each RNA analysis, clean the electrodes using both the Experion electrode cleaner and DEPC-treated water (not required if a deep cleaning has just been performed):

1. Fill a cleaning chip with 800 µl Experion electrode cleaner. Gently tap the side of the cleaning chip to remove any trapped bubbles from the wells. Label this chip "EC" to indicate it contains the electrode cleaner.

Note: Experion electrode cleaner inactivates and removes RNases from the electrode, and the water rinse removes traces of the cleaner from the electrodes to prevent it from affecting the analysis.

2. Open the lid of the electrophoresis station and place the cleaning chip on the chip plat-form.

3. Close the lid and leave it closed for 2 min.

4. Fill a second cleaning chip with 800 µl DEPC-treated water. Label this cleaning chip "DEPC".

5. Open the lid and remove the cleaning chip labeled EC.

6. Insert the "DEPC" chip. Close the lid for 5 min. Remove the chip and discard the washes.

7. Replace the water in the "DEPC" chip and repeat steps 6.

8. Allow the electrodes to dry for 1 min.

9. Close the lid.

10. Replace the water in both cleaning chips after each use to avoid contamination. For storage, remove the water from the cleaning chips and store the chips in a clean location.

Method 2B: Cleaning After an RNA Assay

Following each RNA analysis, rinse the electrodes with DEPC-treated water:

1. Fill the cleaning chip labeled "DEPC" with 800 µl DEPC-treated water. Gently tap the side of the cleaning chip to remove any trapped bubbles from the wells.

2. Open the lid of the electrophoresis station and place the cleaning chip on the chip platform.

3. Close the lid and leave it closed for 1 min.

4. Open the lid and remove the cleaning chip.

5. Allow the electrodes to dry for 1 min.

6. Close the lid.

7. Replace the water in the cleaning chip after use to avoid contamination. For storage, remove the water from the cleaning chip and store the chip in a clean location.

Appendix C — First-Time Use of the Experion™ System

The first time that the Experion electrophoresis station is used, do the following:

1. Establish communication between the software and electrophoresis station.

    a. Power on the computer.

    b. Power on the Experion electrophoresis station by pushing the green button in the center of the front panel. The steady green LED above the button indicates that the unit is on.

    c. Launch Experion software. Click the Experion software icon 🔲 on the desktop or select **Start > All Programs > Bio-Rad Laboratories Experion > Experion**. If a prompt appears asking if Windows should block the program, click **No**.

    The following indicate that communication has been established:

    • A green dot with the last four digits of the instrument serial number in the lower right corner of the main software screen

    • An electrophoresis station icon in the upper left corner

    When there is no connection, these indicators are absent and a "disconnected" message appears next to the **Start Run** button ▶ in the upper left corner of the screen. In addition, a grayed-out instrument icon appears in the upper left corner of the software screen.

2. Confirm that all power saver and/or power options are turned off before starting the assay (refer to the Experion system instruction manual for more details). Do not let the computer "sleep" during a run.

3. Clean the electrode manifold using Experion electrode cleaner and the deep cleaning procedure described in Appendix B.

Appendix D — Ordering Information

Catalog #	Description
700-7001	**Experion™ System**, 100/120 V, for RNA and DNA analysis, includes electrophoresis station, priming station, vortex station, software, USB2 cable, instructions (analysis kits sold separately)
700-7002	**Experion System**, 220/240 V, for RNA and DNA analysis, includes electrophoresis station, priming station, vortex station, software, USB2 cable, instructions (analysis kits sold separately)
700-7111	**Experion RNA StdSens Starter Kit**, includes 3 Experion RNA StdSens chips, 2 cleaning chips, Experion reagents, spin filters, total RNA standard, cleaning swabs, electrode cleaner, tips and tubes to run 3 chips, instructions
700-7110	**Experion Pro260 Starter Kit**, includes 3 Experion Pro260 chips, 1 cleaning chip, Experion reagents, spin filters, IgG protein standard, DTT, cleaning swabs, electrode cleaner, tips and tubes to run 3 chips, instructions
700-7103	**Experion RNA StdSens Analysis Kit for 10 Chips**, includes 10 RNA StdSens chips, 1,250 µl RNA gel, 20 µl RNA StdSens stain, 20 µl RNA ladder, 900 µl RNA StdSens Loading buffer, 2 spin filters
700-7105	**Experion RNA HighSens Analysis Kit for 10 Chips**, includes 10 RNA HighSens chips, 1,250 µl RNA gel, 20 µl RNA HighSens stain, 20 µl RNA ladder, 900 µl RNA HighSens loading buffer, 2 spin filters
700-7153	**Experion RNA StdSens Chips**, 10
700-7154	**Experion RNA StdSens Reagents and Supplies**, for 10 chips, includes 1,250 µl RNA gel, 20 µl RNA StdSens stain, 20 µl RNA ladder, 900 µl RNA StdSens Loading buffer, 2 spin filters
700-7255	**RNA Ladder**, 20 µl
700-7251	**Experion Cleaning Chips**, 10
700-7254	**Experion Spin Filters**, 10
700-7252	**Experion Electrode Cleaner,** 250 ml
700-7253	**Experion DEPC-Treated Water**, 100 ml
700-7264	**Swabs**, 25
VWR87001-688	**VWR barrier pipet tips, sterile** (narrow-bore, 20 µl universal pipet tips, RNase-free) (Purchase directly from VWR Scientific Products)
5395	**Experion Training Video: Chip Loading** (DVD) (also available at discover.bio-rad.com/Experion/)
10000976	**Experion RNA StdSens Instruction Manual**
10001312	**Experion System Instruction Manual**

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