applied biosystems

CarrierMax™ SMN1/SMN2 Reagent Kit

USER GUIDE

for use with:
3500/3500xL Genetic Analyzer
SeqStudio™ Genetic Analyzer
CarrierMax™ Software
GeneMapper™ Software

Catalog Number 952363
Publication Number MAN0018882
Revision C.0

For Research Use Only. Not for use in diagnostic procedures.

ThermoFisher SCIENTIFIC

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

DISCLAIMER: TO THE EXTENT ALLOWED BY LAW, THERMO FISHER SCIENTIFIC INC. AND/OR ITS AFFILIATE(S) WILL NOT BE LIABLE FOR SPECIAL, INCIDENTAL, INDIRECT, PUNITIVE, MULTIPLE, OR CONSEQUENTIAL DAMAGES IN CONNECTION WITH OR ARISING FROM THIS DOCUMENT, INCLUDING YOUR USE OF IT.

Revision history: Pub. No. MAN0018882

Revision	Date	Description
C.0	21 July 2022	Update to the Note in Figure 4.
B.0	15 March 2022	Updated the control DNA for the kit.
A.0	22 September 2020	New user guide for the CarrierMax™ SMN1/SMN2 Reagent Kit.

Contents

1 Product information

IMPORTANT! Before using this product, read and understand the information in the "Safety" appendix in this document.

Product description

The Applied Biosystems™ CarrierMax™ SMN1/SMN2 Reagent Kit uses multiplex PCR amplification of genomic DNA and capillary electrophoresis to detect deletions in exon 7 that result in truncated SMN protein. In addition, the kit detects 2 variants associated with the silent carrier (2+0) haplotype. The results are processed with GeneMapper™ Software in conjunction with complimentary Applied Biosystems™ CarrierMax™ Software to consolidate data and simplify carrier classifications in SMN1 carrier screening research.

Contents and storage

Table 1 CarrierMax™ SMN1/SMN2 Reagent Kit (Cat. No. 952363)

Contents	Amount	Storage
CarrierMax™SMN1/SMN2 Reagent 2xPCR Reaction buffer	1 mL	–25°C to –15°C. Protect from light[1].
CarrierMax™ SMN1/SMN2 Reagent CNV Primers Mix	25 µL	
CarrierMax™ SMN1/SMN2 Reagent SNP Primers Mix	25 µL	
CarrierMax™ SMN1/SMN2 Reagent Taq DNA Polymerase	40 µL	
CarrierMax™ SMN1/SMN2 Reagent CM500 Size Standard	40 µL	
CarrierMax™ SMN1/SMN2 Reagent Nuclease-free Water	1 mL	

[1] The kit is stable for one year when stored at –25°C to –15°C. Do not freeze, then thaw, more than 5 times.

Required materials not supplied

Unless otherwise indicated, all materials are available through **thermofisher.com**. "MLS" indicates that the material is available from **fisherscientific.com** or another major laboratory supplier.

Table 2 Materials for generating PCR products

Item	Source
Instruments and equipment	
One of the following thermocyclers:[1] • Veriti™ Thermal Cycler • ProFlex™ 96-well PCR System • GeneAmp™ PCR System 9700	Contact your local sales office.
Benchtop microcentrifuge	MLS
Vortex mixer	MLS
Adjustable micropipettors	MLS
Controls	
CarrierMax™ SMN Control DNA Kit	952445
Plates and other consumables	
PureLink™ Genomic DNA Mini Kit or equivalent[2]	K1820-00
MicroAmp™ Optical 96-Well Reaction Plate with Barcode	4306737
MicroAmp™ Clear Adhesive Film, or equivalent	4306311
Aerosol-resistant pipette tips	MLS
Other plastic consumables	**thermofisher.com/plastics**

[1] You can use an equivalent thermal cycler. Optimize the protocols for other thermal cyclers.
[2] The MagMAX™ DNA Multi-Sample Ultra 2.0 Kit (Cat. No. A36570) can be used for automated sample preparation.

Table 3 Materials for capillary electrophoresis

Item	Source
Instruments and equipment	
One of the following genetic analyzers: • 3500/3500xL Genetic Analyzer with: – 3500 Data Collection Software (Recommended: v3.3; Windows™ 10 operating system) • SeqStudio™ Genetic Analyzer with: – (Recommended) SeqStudio™ Plate Manager	Contact your local sales office.

Table 3 Materials for capillary electrophoresis *(continued)*

Item	Source
Biosafety cabinet	MLS
Benchtop microcentrifuge	MLS
Vortex mixer	MLS
Adjustable micropipettors	MLS
General reagents	
Hi-Di™ Formamide	4311320
CarrierMax™ A5D Matrix Standard Kit	952364
Consumables and reagents for the 3500/3500xL Genetic Analyzer	
Septa Cathode Buffer Container (for the 3500 series Genetic analyzers)	4410715
Septa for 3500/3500xL Genetic Analyzers, 96 well	4412614
Anode Buffer Container	4393927
Cathode Buffer Container	4408256
Polymer, POP-7™ (96)	A26073
Conditioning reagent	4393718
Capillary array, 8-Capillary, 50-cm	4404685
Capillary array, 24-Capillary, 50-cm	4404689
Retainer & Base Set (Standard) for 3500/3500xL Genetic Analyzers, 96 well	4410228
Retainer & Base Set (Standard) for 3500/3500xL Genetic Analyzers, 8 tube	4410231
Consumables and reagents for the SeqStudio™ Genetic Analyzer	
Septa for SeqStudio™ Genetic Analyzer, 96 well	A36541
Septa for SeqStudio™ Genetic Analyzer, 8 strip	A36543
SeqStudio™ Cartridge or SeqStudio™ Cartridge v2	A33671 or A41331
Cathode Buffer Container	A33401
Reservoir Septa	A35640
Plates and other consumables	
MicroAmp™ Optical 96-Well Reaction Plate with Barcode	4306737
MicroAmp™ Optical 96-Well Reaction Plate	N8010560

Table 3 Materials for capillary electrophoresis *(continued)*

Item	Source
MicroAmp™ Clear Adhesive Film, or equivalent	4306311
Nonstick, RNase-Free Microfuge Tubes, 1.5 mL	AM12450

Table 4 Materials to analyze data

Item	Source
GeneMapper™ Software	Contact your local sales office.
CarrierMax™ Software	Complimentary download from **thermofisher.com/carriermax-smn1**.

Workflow

System workflow

Chapter 2, Prepare for capillary electrophoresis

Chapter 3, Prepare samples and run the PCR

Chapter 4, Analyze the results

2 Prepare for capillary electrophoresis

3500/3500xL Genetic Analyzer

Perform spectral calibration

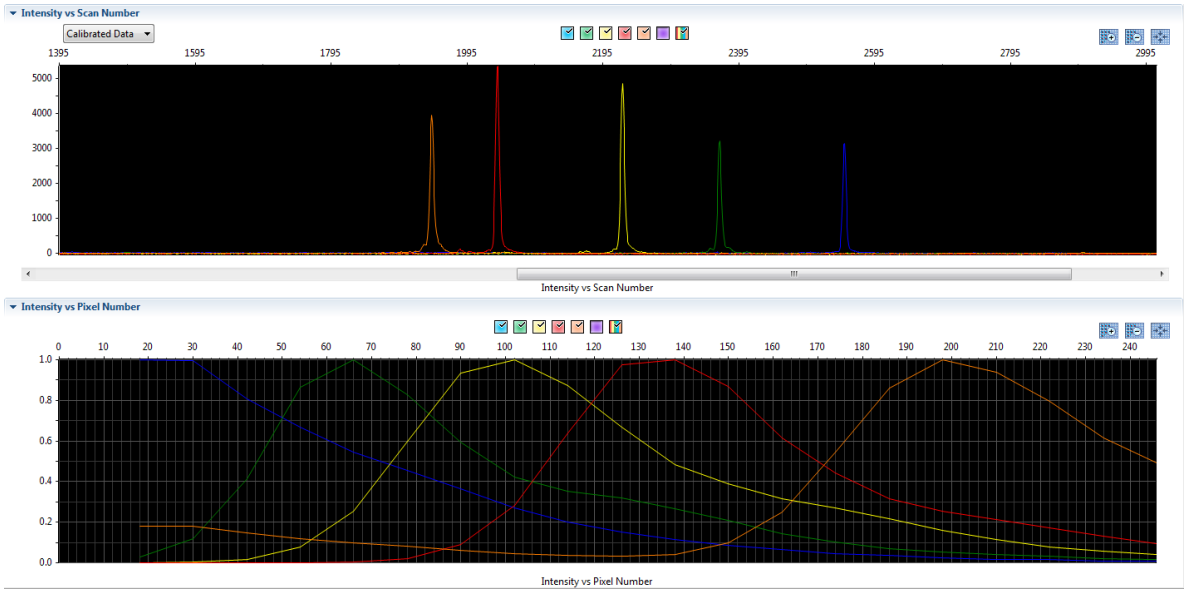
Before using the CarrierMax™ SMN1/SMN2 Reagent Kit for the first time, perform a spectral calibration with the CarrierMax™ A5D Matrix Standard.

Spectral calibration is required each time a new array is installed.

1. (First time only) Create an **A5D Matrix Standard** dye set in the 3500 Series Data Collection Software.
   a. Go to **Library**, select **Dye Set**, then click **Create**.
   b. Go to the **Dye Set Name**, then enter **A5D**. Choose **Matrix Standard** in **Chemistry**.
   c. Go to **Dye Set Template**, then select **E5 Template**.
   d. Click **Save**.
2. Perform a spectral calibration using the CarrierMax™ A5D Matrix Standard Kit (Cat. No. 952364).

   IMPORTANT! A spectral calibration must be performed for the A5D Dye Set to run the assay.

   See the *CarrierMax™ A5D Matrix Standard Kit Product Information Sheet* (Pub. No. MAN0018893). The following figure is an example of a passing 5-dye spectral calibration.



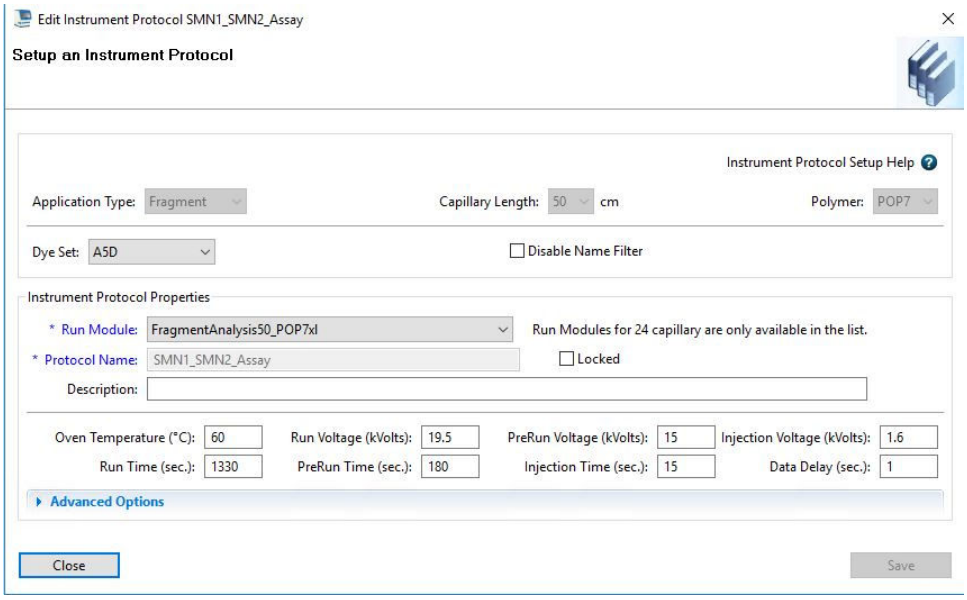
Electrophoresis software setup

(Recommended) Ensure that your genetic analyzer is running on the Windows™ operating system with 3500 Data Collection Software v3.3 or v4.

The following instructions describe setting up an assay. For more detailed information on the instrument procedures, see the documents listed in Appendix C, “Documentation and support”.

Create an instrument protocol

1. Navigate to the **Instrument Protocols** library.
2. Click **Create**.
3. Specify the settings listed below.
   - **Capillary length—50cm**
   - **Polymer—POP7**
   - **Dye Set—A5D**
   - **Run Module—FragmentAnalysis50_POP7xl**
   - **Protocol Name—SMN1_SMN2_Assay**
   - **Oven Temperature (°C)—60**
   - **Run Time (sec)—1,330**
   - **Run Voltage (kVolts)—19.5**
   - **PreRun Time (sec)—180**
   - **PreRun Voltage (kVolts)—15**
   - **Injection Time (sec)—15**
   - **Injection Voltage (kVolts)—1.6**
   - **Data Delay (sec)—1**

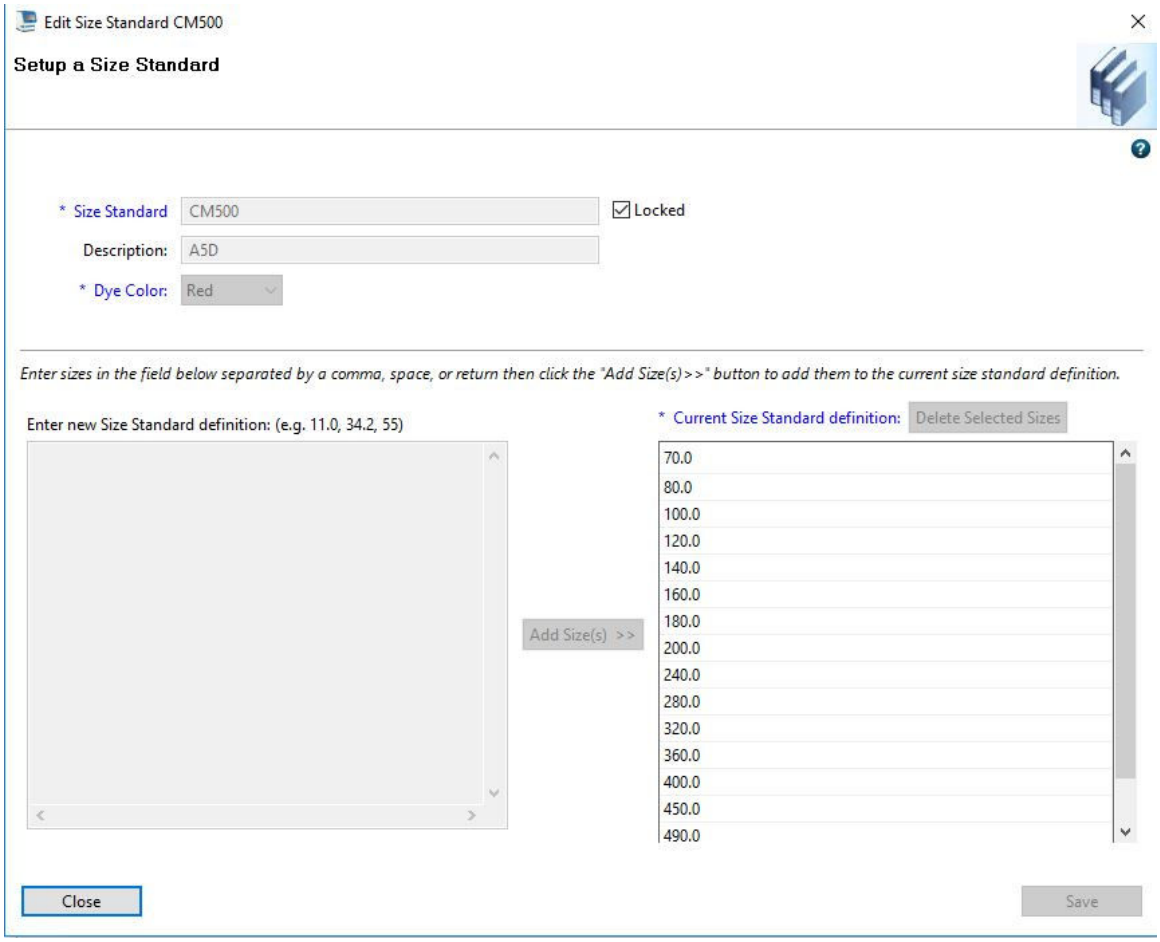


4. Click **Save**.

Create the size standards

A size standard defines the sizes of known fragments. It is used to generate a standard curve. The standard curve is used to determine the sizing of unknown samples. When you create a sizecalling (fragment analysis) protocol, you add a size standard to the protocol.

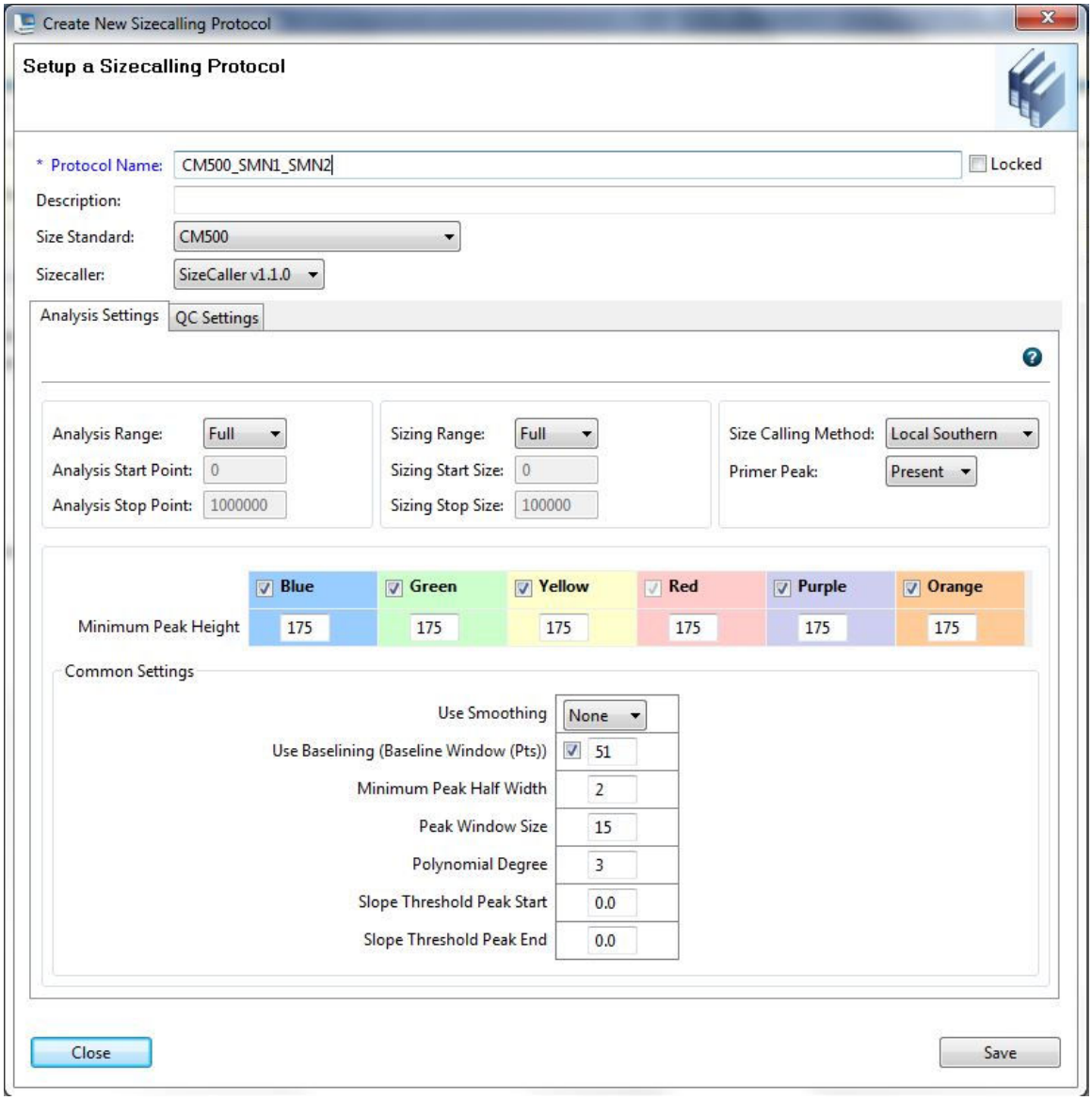
1. Navigate to the **Size Standards** library.
2. Click **New**.
3. Specify the settings listed below.
   - **Size Standard name**—CM500; Select **Locked**.
   - **Description**—A5D
   - **Dye Color**—Red
   - **Size Standards** (list in left text box)—70, 80, 100, 120, 140, 160, 180, 200, 240, 280, 320, 360, 400, 450, 490, 500



4. Click **Save**.

Create the sizecalling protocol

1. Navigate to the **Sizecalling Protocols** library.
2. Click **Create**.
3. Specify the settings listed below.
   - **Protocol Name**—**CM500_SMN1_SMN2**
   - **Sizecaller**—**SizeCaller v1.1.0**
   - **Analysis Range and Sizing Range**—**Full**
   - **Size Calling Method**—**Local Southern**
   - **Primer Peak**—**Present**
   - **Minimum Peak Height**—175 (all)
   - **Use Smoothing**—None
   - **Use Baselining (Baseline Window) (Pts)**—51
   - **Minimum Peak Half Width**—2
   - **Peak Window Size**—15
   - **Polynomial Degree**—3
   - **Slope Threshold Peak Start/End**—0.0 (both)



4. Click **Save**.

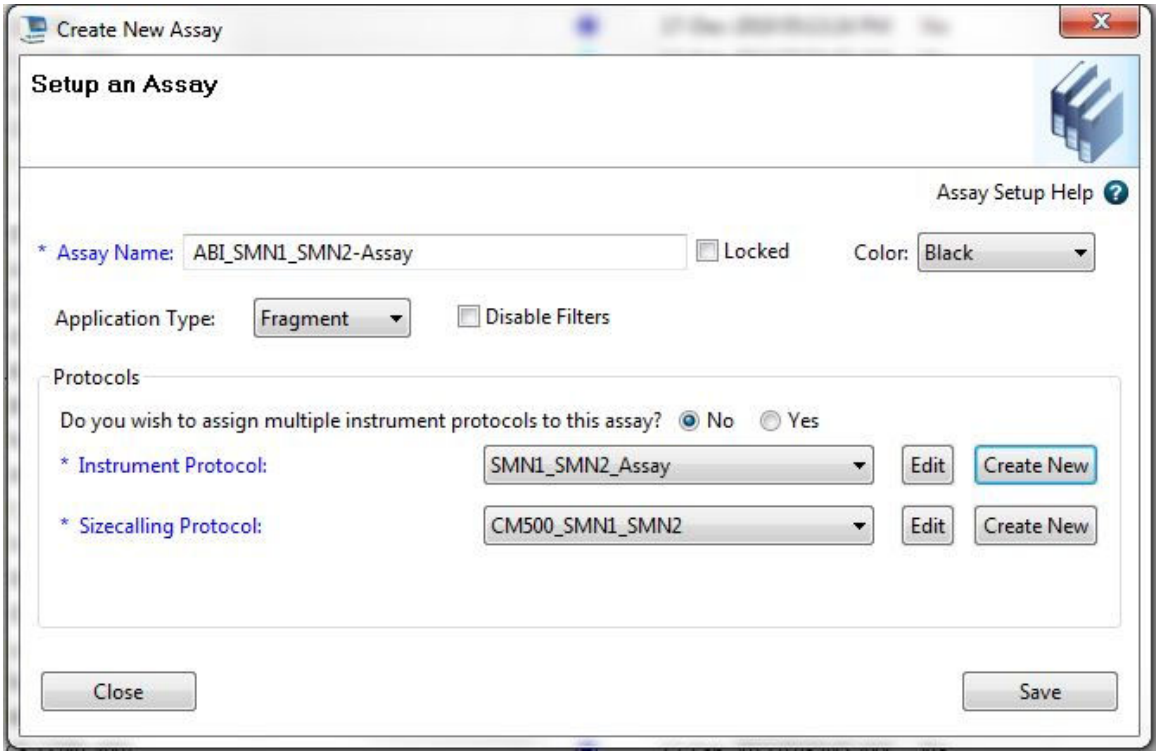
Create the Assay

1. Navigate to the **Assays** library.

   The list of items in the library may be filtered based on the library filtering user preference. Click **Disable Filters** to show all items in the list.

2. Click **Create**.

3. Specify the settings.
   - **Assay Name**—ABI_SMN1_SMN2_Assay
   - **Color**—Black
   - **Application Type**—Fragment
   - **Do you wish to assign multiple protocols to this assay?**—No
   - **Instrument Protocol**—**SMN1_SMN2_Assay** (select from the dropdown list)
   - **Sizecalling Protocol**—**CM500_SMN1_SMN2** (select from the dropdown list)



4. Click **Save**.

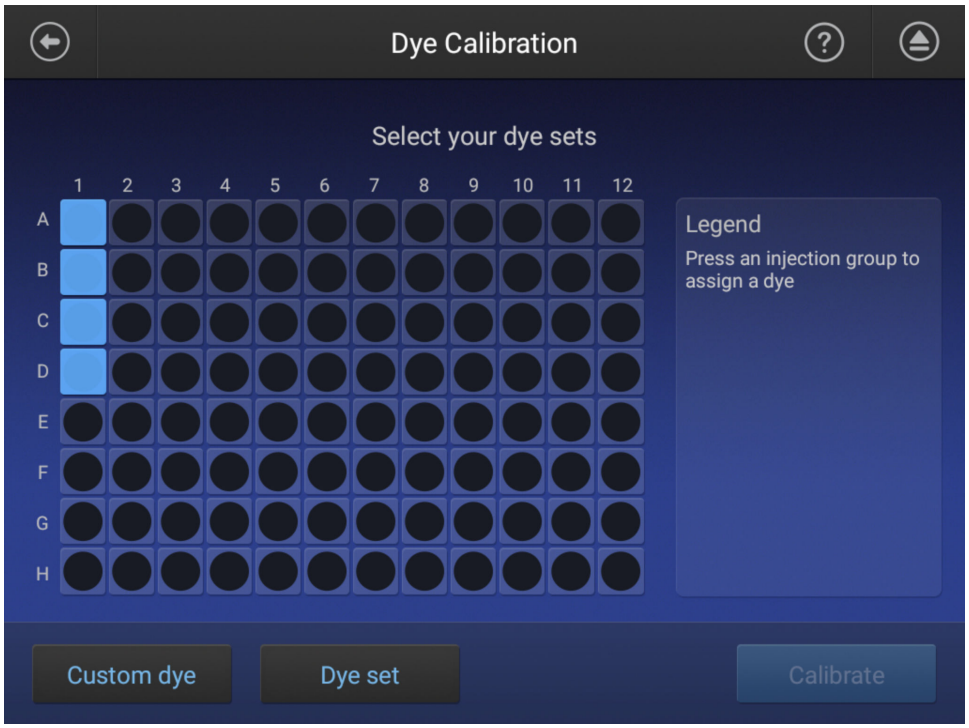
SeqStudio™ Genetic Analyzer

Perform a system dye calibration

A system dye calibration requires ~30 minutes to complete.

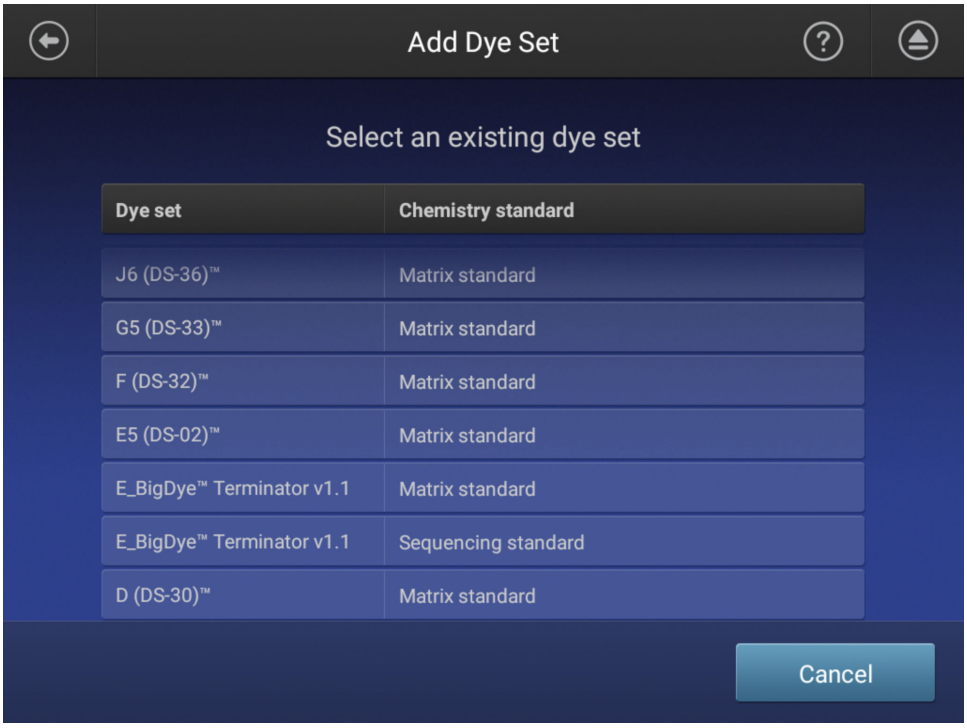
Prepare the dye set calibration standards and plate as described in the *CarrierMax™ A5D Matrix Standard Kit Product Information Sheet* (Pub. No. MAN0018893).

1. In the home screen, tap ⚙ **Settings** ▸ **Maintenance and Service** ▸ **Calibration** ▸ **Dye Calibration**.
2. Tap the injection group for the dye set in the plate, then tap **Custom dye**.

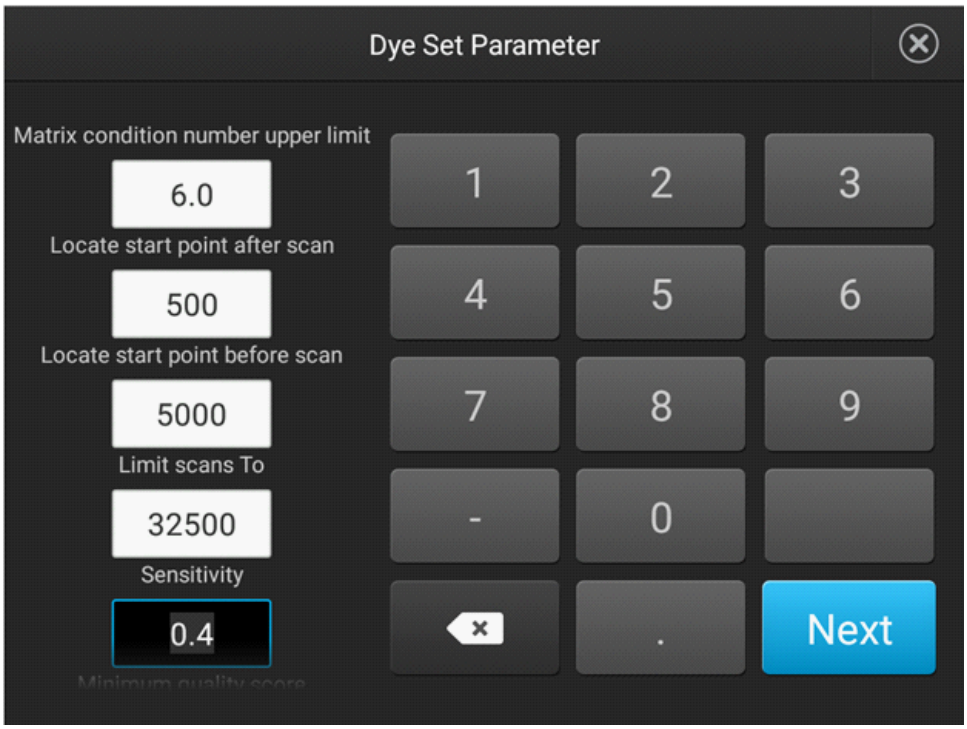


3. In the **Dye Set** screen, tap **Add**.

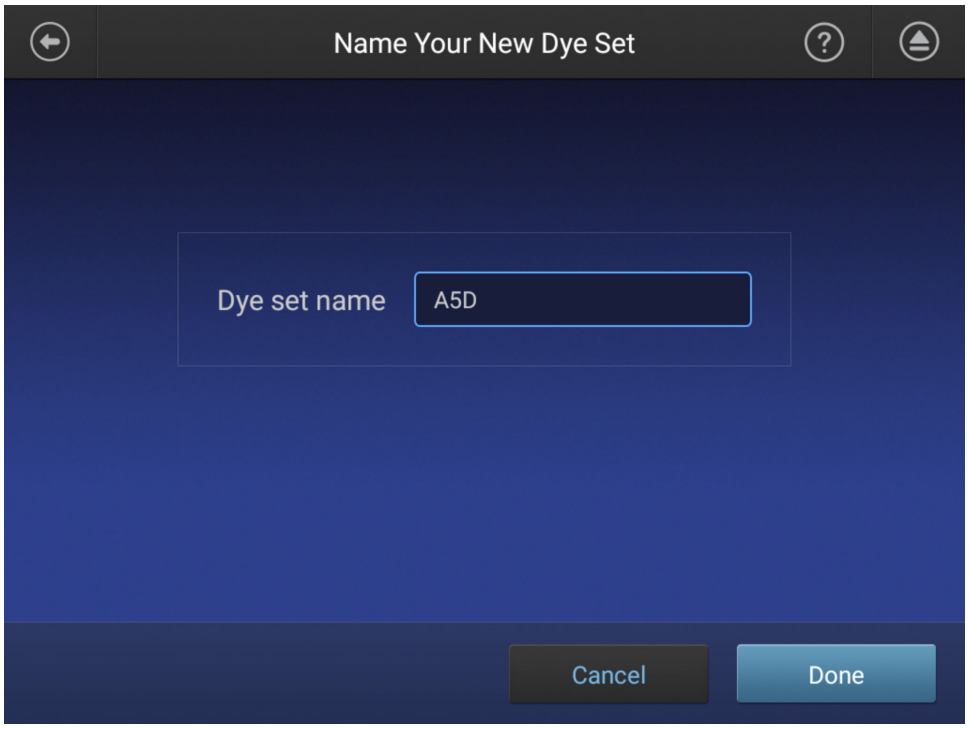
4. Select E5 (DS-02) as a starting template.



5. In the **Dye Set Parameter** screen, tap **Next**.



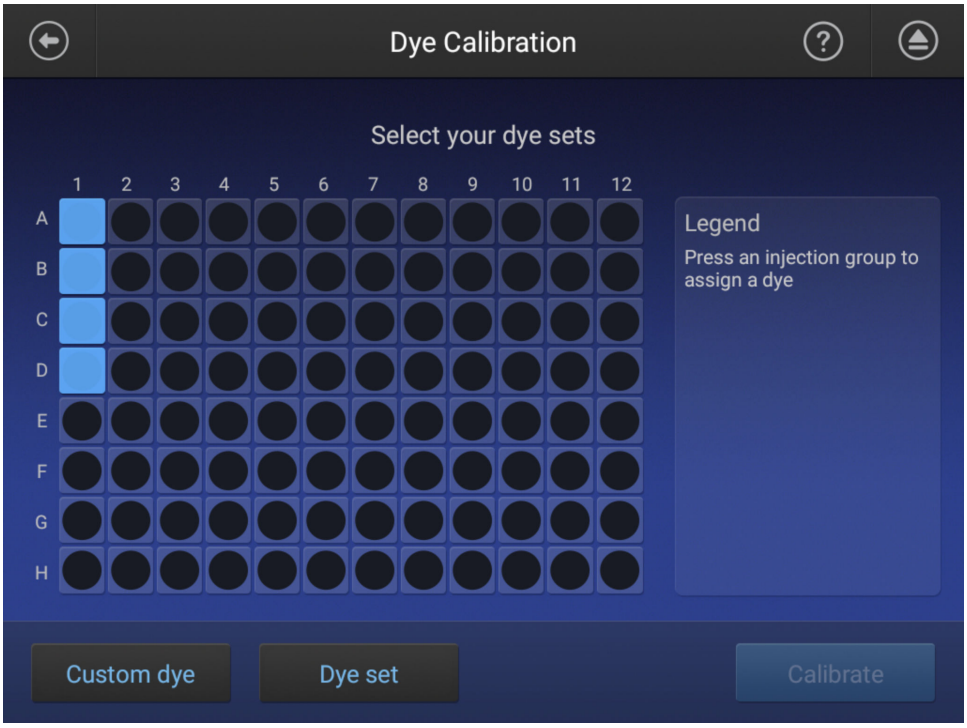
6. Set the **Dye set name** to **A5D**, then click **Done**.



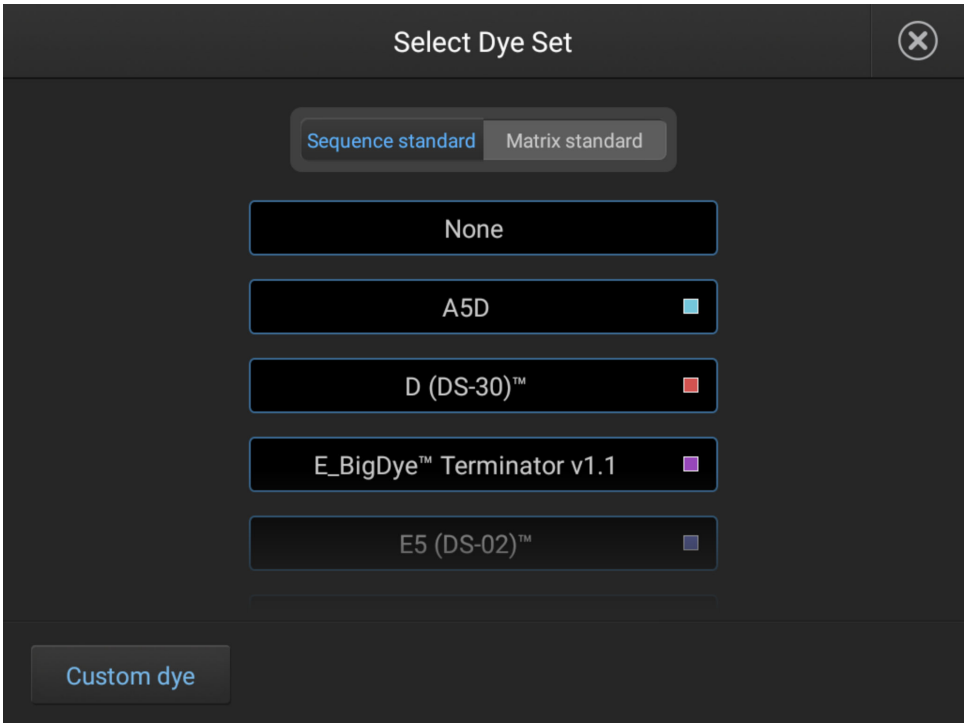
A5D is then listed in the **Dye Set** list.

7. Return to the home screen, tap **Settings** ▸ **Maintenance and Service** ▸ **Calibration** ▸ **Dye Calibration**.

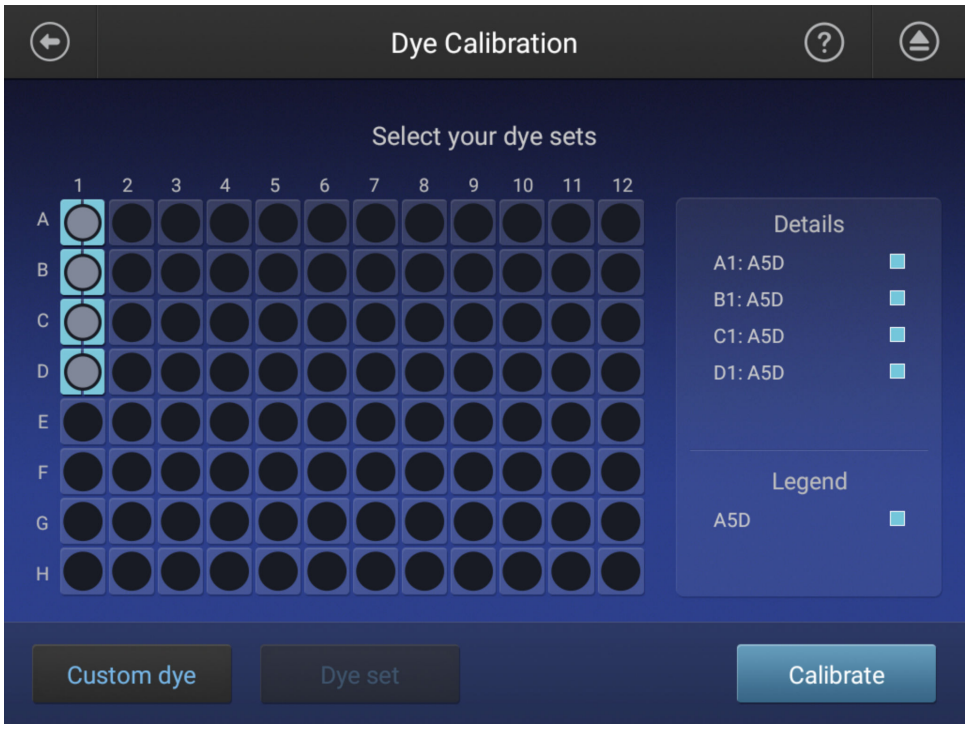
8. Highlight the injection group to assign a dye, then tap **Dye set**.



9. Tap **Matrix Standard**, then select the **A5D** matrix standard.



10. Tap **Calibrate**.



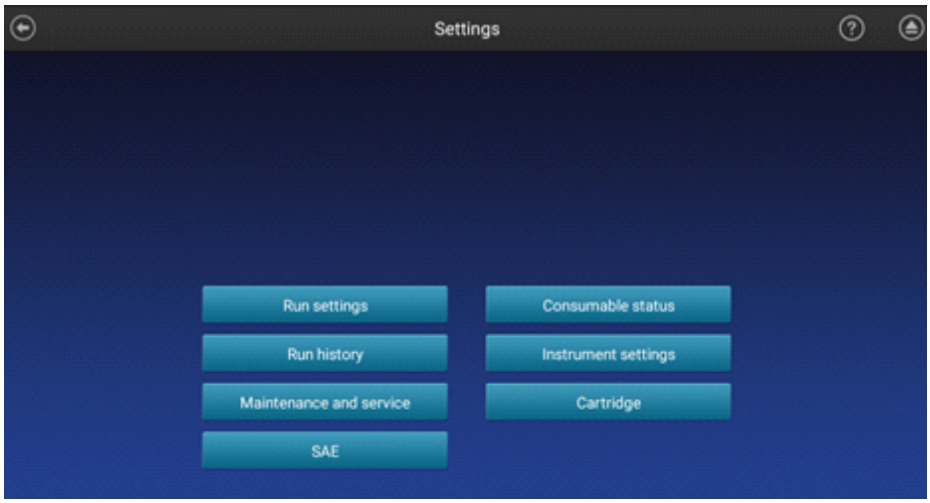
The calibration run starts.

IMPORTANT! If the dye calibration fails:

- The results of the calibration are not saved, and the calibration plate is not moved to **Run History**.
- The instrument does not allow you to rerun the plate setup for a failed calibration. Close the calibration screen, then start a new calibration.

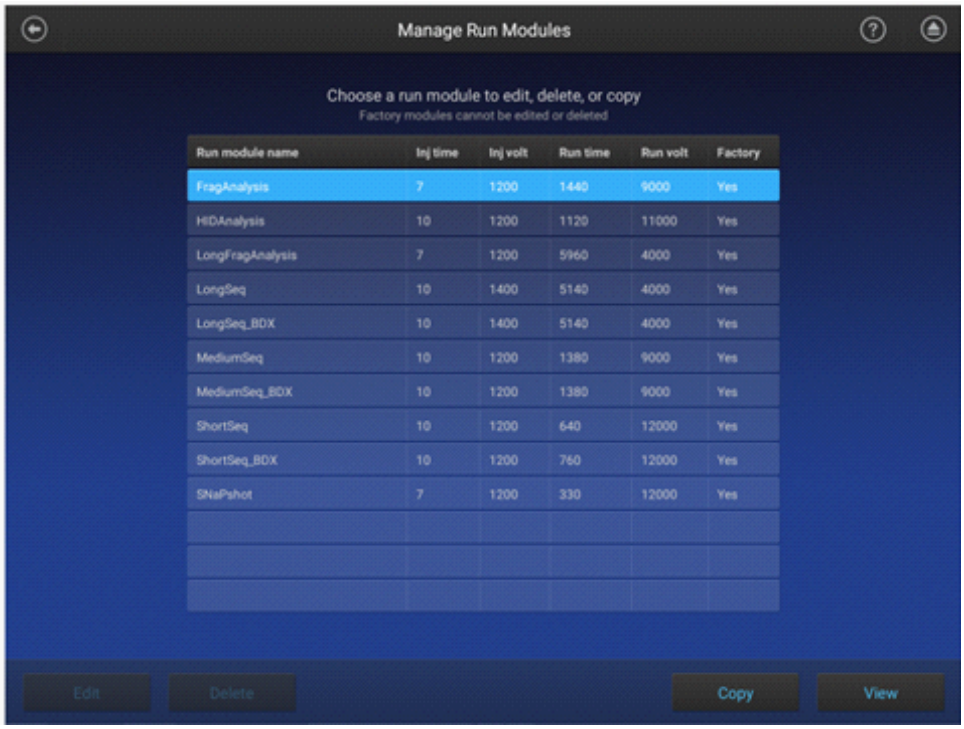
Create the CarrierMax™ SMN1/SMN2 Reagent Kit run module

1. On the SeqStudio™ Genetic Analyzer, go to **Settings** ▸ **Run Settings**.



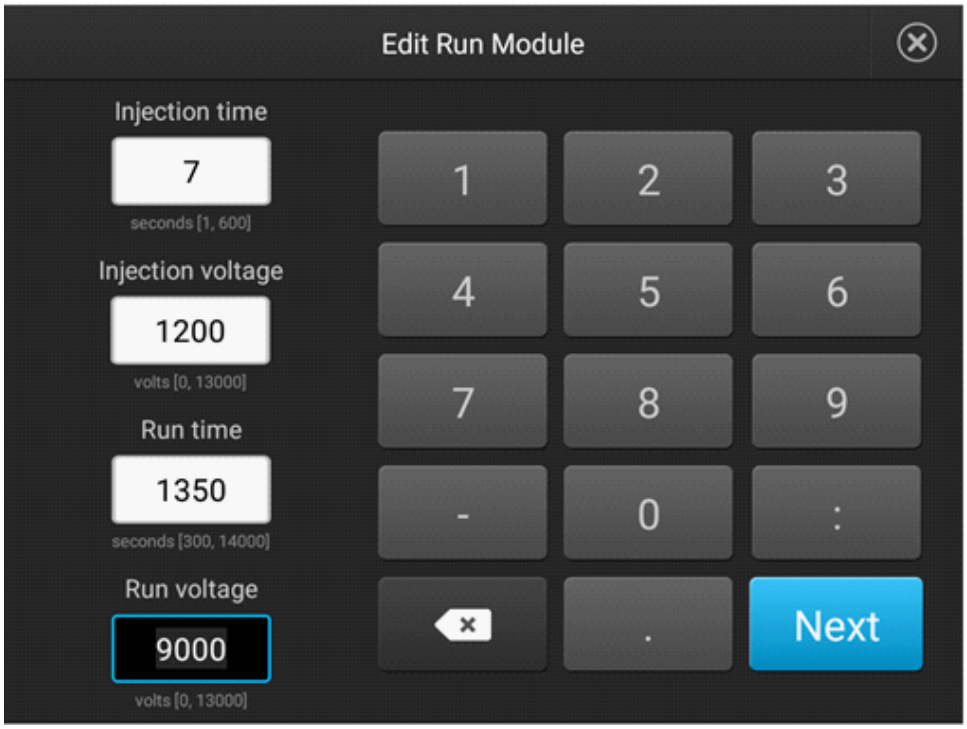
2. Select **Run modules** ▸ **FragAnalysis**, then tap **Copy**.

3. Select the default **FragAnalysis** run module (or a user-created run module), then tap **Copy**.



An editable copy of the **FragAnalysis** run module is created.

4. In the **Edit Run Module** screen enter the following parameters.
   - Injection Time—7 seconds
   - Injection Voltage—1,200 volts
   - Separation (Run) Time—1,350 seconds
   - Separation (Run) Voltage—9,000 volts

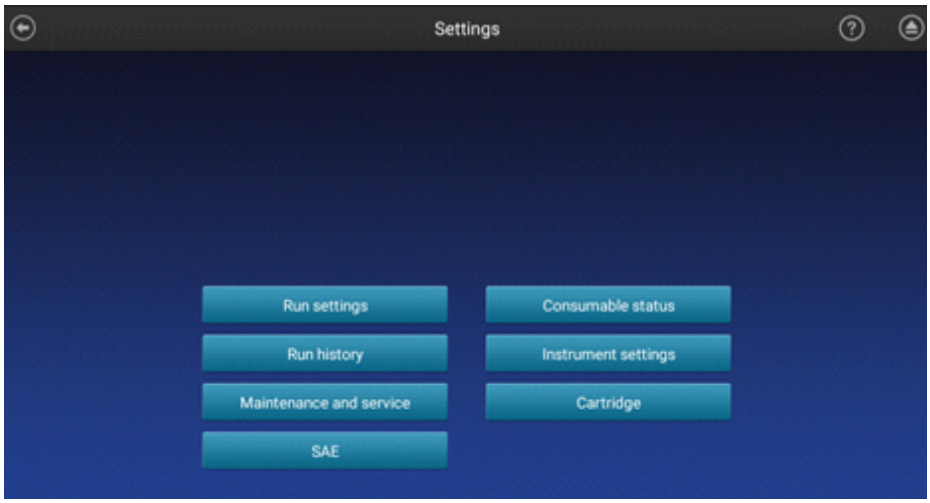


5. Tap **Next**, then change the **Run Module Name** to **SMN_Assay**.

6. Tap **Done**.

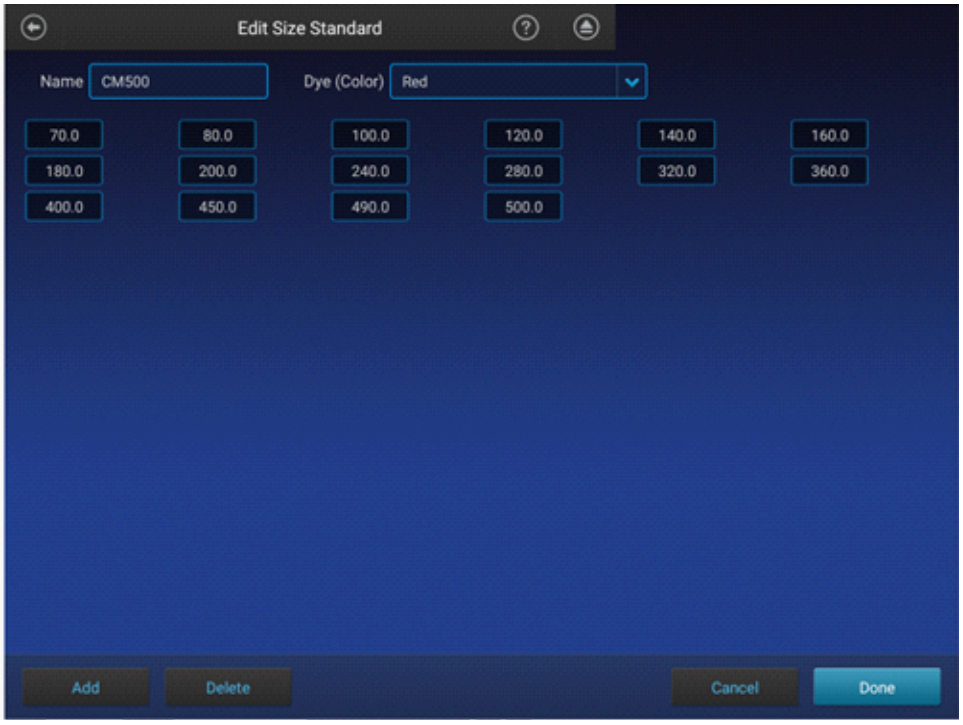
Create the CarrierMax™ SMN1/SMN2 Reagent Kit Size Standards

1. On the SeqStudio™ Genetic Analyzer, go to **Settings** ▸ **Run Settings**.



2. Select **Size standard** ▸ **GS500ROX**, then tap **Copy**.
   An editable copy of the size stanadard is created.

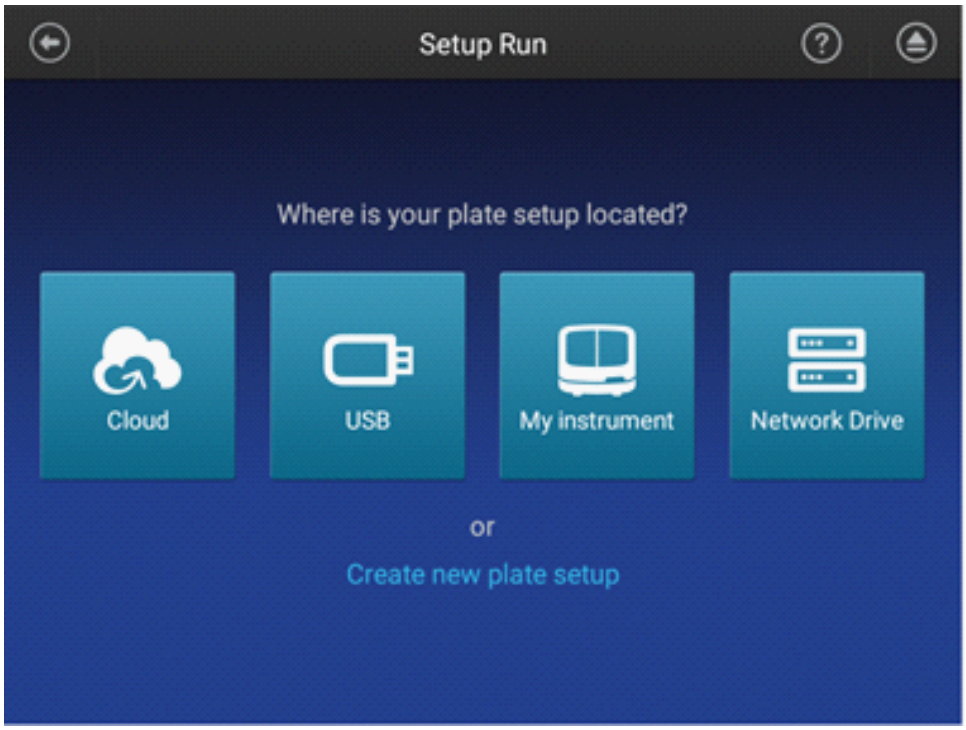
3. In the **Edit Size Standard** screen enter the following parameters.
   - **Name—CM500**
   - **Dye (Color)—Red**
   - **Fragment sizes—70, 80, 100, 120, 140, 160, 180, 200, 240, 280, 320, 360, 400, 450, 490, 500**



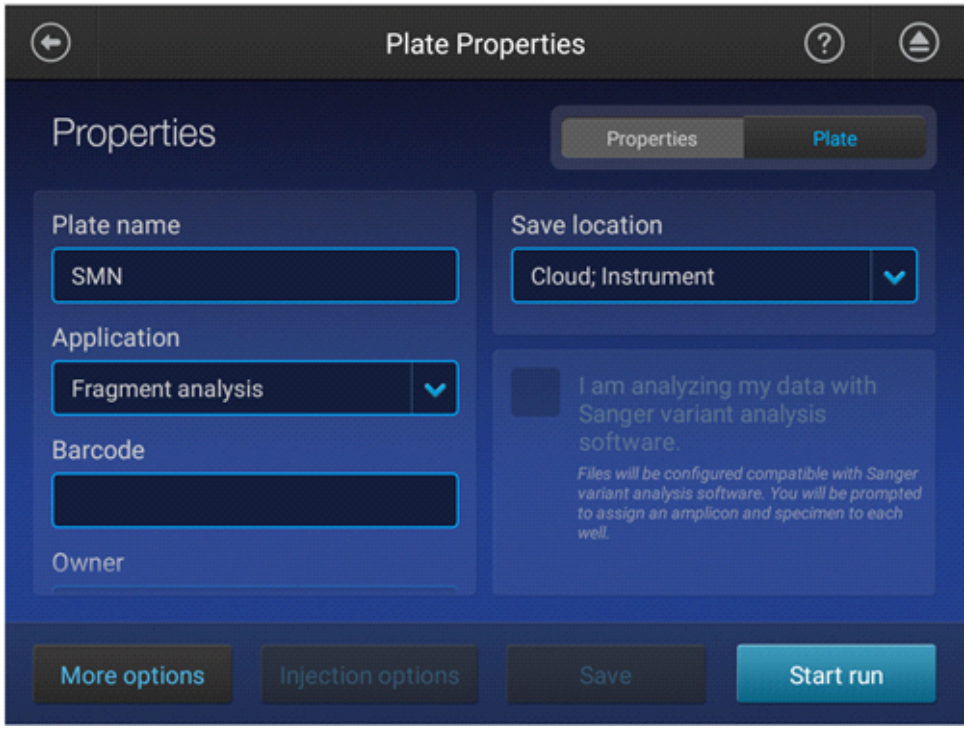
4. Tap **Done**.

Create a new plate setup

1. On the SeqStudio™ Genetic Analyzer, tap **Setup run** ▸ **Create new plate setup**.

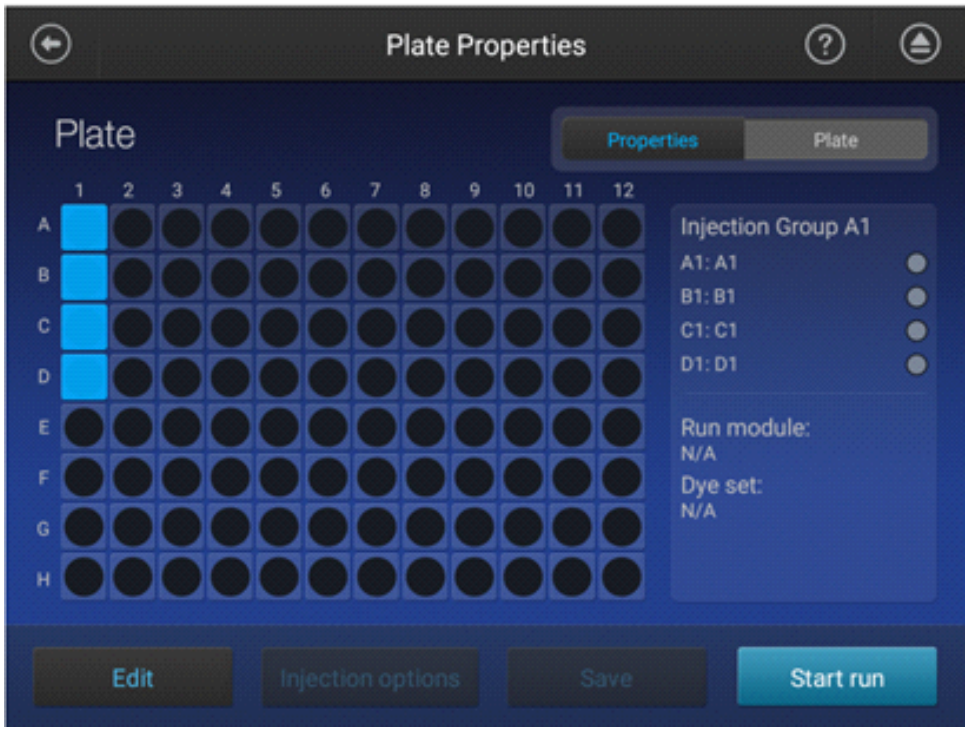


2. In the **Properties** tab of the **Plate Properties** screen, enter the following settings.
   - **Plate Name—SMN**
   - **Application—Fragment analysis**
   - **Save location—Cloud; Instrument**

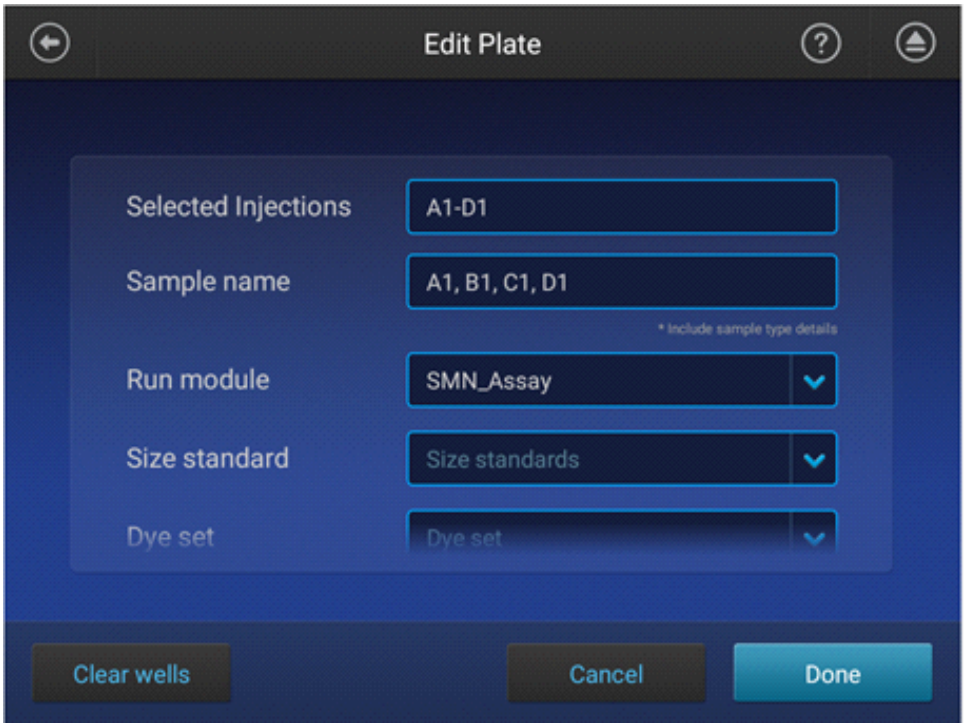


3. Tap the **Plate** tab.

4. Highlight the **Injection Groups**, then tap **Edit**.

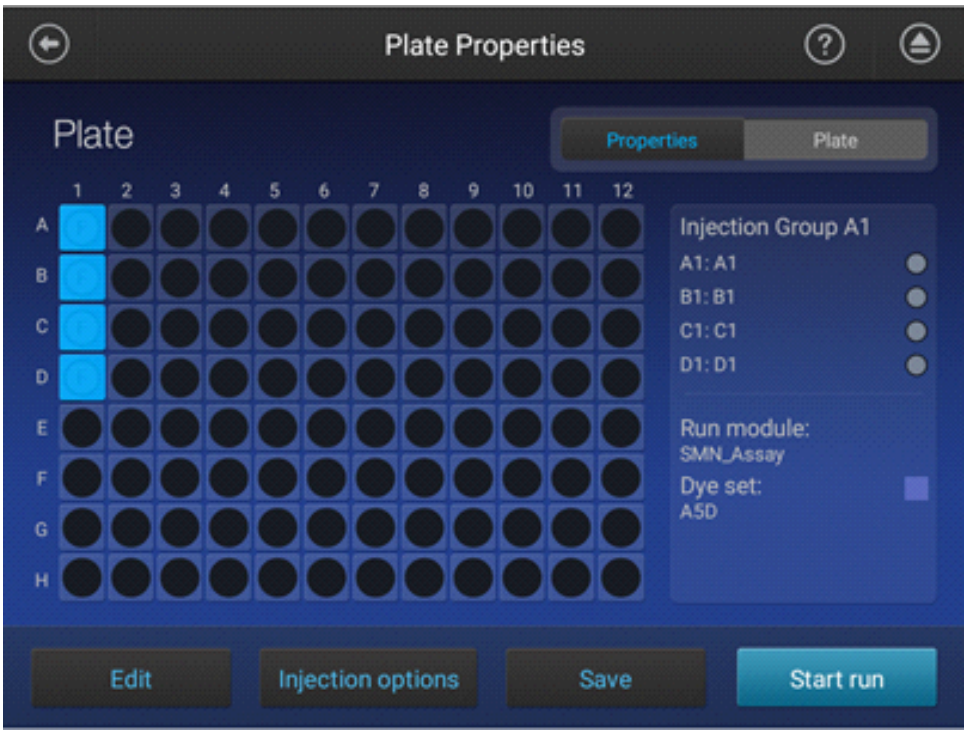


5. In the **Edit Plate** screen, make the following selections.
   - **Run module—SMN_Assay**
   - **Size standard—CM500**
   - **Dye set—A5D**

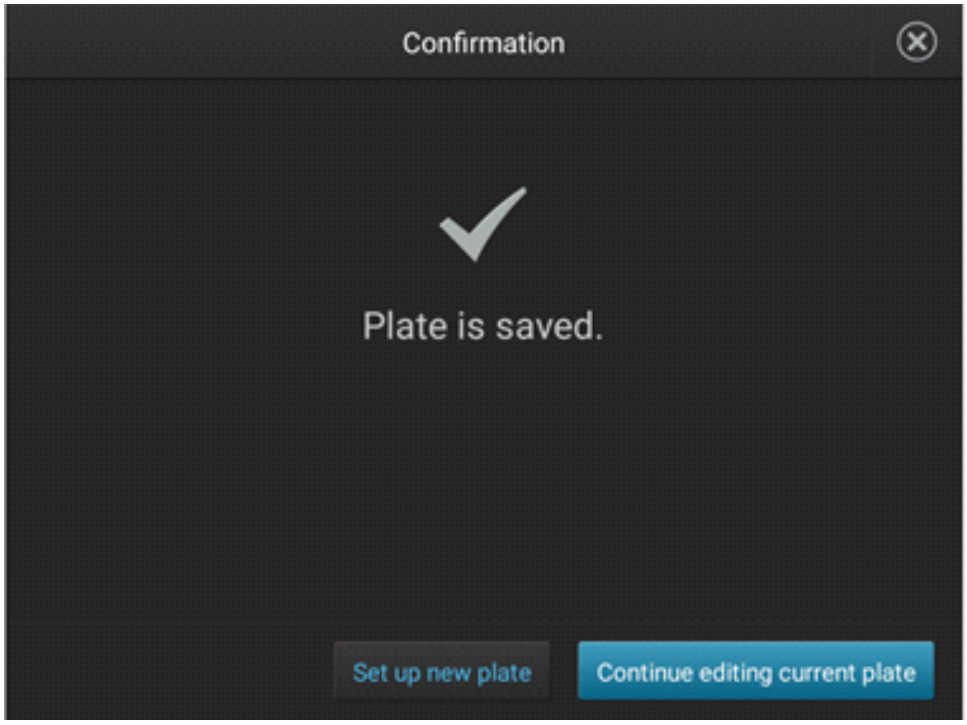


6. Tap **Done**.

7. Highlight the injection groups to confirm the correct **Run module** and **Dye set** have been selected, then tap **Save**.



The plate is saved on the instrument.


Confirmation
Plate is saved.
Set up new plate
Continue editing current plate

3 Prepare samples and run the PCR

DNA sample preparation

This kit is optimized for use with genomic DNA. We recommend extraction with the PureLink™ Genomic DNA Mini Kit (Cat. No. K1820-00) and fluorometric methods for DNA quantification such as the Qubit™ Fluorometer and the Qubit™ dsDNA HS (High Sensitivity) Assay Kit (Cat No. Q32851). Alternatively, dsDNA can also be quantitated using Quant-iT™ PicoGreen™ dsDNA Assay Kit (Cat. No. P7589) on a microplate reader. The optimal and recommended DNA concentration is 10 ng/ µL. Store at 2–8°C for up to one week, or at –20°C for up to 6 months.

Note: We do not recommend methods such as densitometry for this assay (for example, a NanoDrop™ Spectrophotometer). These methods do not discriminate between DNA and RNA and are sensitive to small RNA fragments leading to over-estimation of dsDNA yield.

Optimize all protocols with your standard laboratory procedures.

Set up the PCR reactions

- Thaw all components (except the CarrierMax™ SMN1/SMN2 Reagent Taq DNA Polymerase) to room temperature, then vortex and briefly centrifuge to mix. Place the tubes on ice.
- Remove the tube of CarrierMax™ SMN1/SMN2 Reagent Taq DNA Polymerase from the freezer immediately before use, gently flick the tube 3 times to mix, then centrifuge briefly. Place the tube on ice.

IMPORTANT! Perform all steps on ice.

1. In a 1.5 mL microfuge tube (Nonstick, RNase-Free Microfuge Tubes, 1.5 mL), prepare the **PCR Reaction Mix**.

Component	Volume per reaction[1]	
	CNV	SNP
CarrierMax™SMN1/SMN2 Reagent 2xPCR Reaction buffer	10 µL	10 µL
CarrierMax™ SMN1/SMN2 Reagent CNV Primers Mix	0.5 µL	—
CarrierMax™ SMN1/SMN2 Reagent SNP Primers Mix	—	0.5 µL
CarrierMax™ SMN1/SMN2 Reagent Taq DNA Polymerase	0.4 µL	0.4 µL

(continued)

Component	Volume per reaction[1]	
	CNV	SNP
CarrierMax™ SMN1/SMN2 Reagent Nuclease-free Water	8.1 µL	8.1 µL
Total volume	**19 µL**	**19 µL**

[1] Scale reaction volume according to the number of reactions, including overage for pipetting.

2. Vortex the **PCR Reaction Mix**, then centrifuge briefly before use.
3. To the labeled PCR reaction plate, add the following components.
   a. Add 19 µL of PCR reaction mix to each sample and no template control (NTC) well.
   b. Add one of the following to sample wells:
      - 1 µL of sample DNA (10 ng recommended)
      - 1 µL of CarrierMax™ SMN1/SMN2 Reagent Nuclease-free Water (NTC)
      - (*Optional*) 10 ng of control from the CarrierMax™ SMN Control DNA Kit. See “Required materials not supplied” on page 6.
4. Seal the plate tightly with adhesive film. Vortex briefly to mix, then centrifuge briefly to bring the mixture to the bottom of the tube and eliminate air bubbles.
5. Immediately proceed to “Run the PCR” on page 29.

Run the PCR

1. Program the thermal cycling conditions.

Step	Temperature	Time	Cycles
Hot start	95°C	5 minutes	1
Denature	95°C	30 seconds	35
Anneal	57°C	30 seconds	
Extend	72°C	30 seconds	
Final extension	72°C	10 minutes	1
Hold	4°C	∞	

2. Set the appropriate reaction volume, then load the plate into the thermal cycler.
3. Close the heated cover, then start the run.
4. When the run is complete, remove the plate from the thermal cycler.

IMPORTANT! Protect the amplified DNA from light.

Amplified DNA can be stored at 2°C to 8°C for up to 24 hours, or at –25°C to –15°C for up to 72 hours. Avoid multiple freeze-thaw steps.

Dilute the PCR products

Dilute the PCR products before capillary electrophoresis.

Instrument	Dilution
3500xL Genetic Analyzer	Dilute PCR products 1:10. For example, 1 µL PCR product + 9 µL of nuclease-free water.
SeqStudio™ Genetic Analyzer	Dilute PCR products 1:20. For example, 1 µL PCR product + 19 µL of nuclease-free water.

Note: If signal saturation is observed after capillary electrophoresis, further dilute, then re-test the sample.

Prepare samples for capillary electrophoresis

Prepare the samples for capillary electrophoresis immediately before loading. This protocol is for 96-well plates, but MicroAmp™ 8-Tube Strips (0.2-mL) with MicroAmp™ 8-Cap Strips can also be used.

1. In a 1.5 mL microfuge tube, prepare the mix of Hi-Di™ Formamide and CarrierMax™ SMN1/SMN2 Reagent CM500 Size Standard for the required number of reactions.

Component	Volume per reaction
CarrierMax™ SMN1/SMN2 Reagent CM500 Size Standard	0.3 µL
Hi-Di™ Formamide	8.7 µL
Total volume	9 µL

IMPORTANT! The volume of size standard is a suggested amount. Determine the appropriate amount based on your experiments and results.

2. Vortex the mix of Hi-Di™ Formamide and CarrierMax™ SMN1/SMN2 Reagent CM500 Size Standard well, then centrifuge briefly before use.

3. Prepare the fragment analysis reactions. To a MicroAmp™ Optical 96-Well Reaction Plate, add the following components.
   - 9 µL of the Hi-Di™ Formamide and CarrierMax™ SMN1/SMN2 Reagent CM500 Size Standard mix
   - 1 µL of PCR product (see “Run the PCR” on page 29)

   Note: For blank wells, add 10 µL of Hi-Di™ Formamide.

4. Seal the reaction plate with MicroAmp™ Clear Adhesive Film.

5. Vortex the reaction plate well, then centrifuge briefly to bring the contents to the bottom of the wells and eliminate air bubbles.

6. Denature the DNA fragments:
   a. Incubate the mixture at 95°C for 5 minutes.
   b. Incubate the mixture on ice for 3 minutes.

7. Centrifuge the plate for 1 minute to bring the contents to the bottom of the wells and eliminate air bubbles.

8. Remove the adhesive film, then cover the plate with a septa.
   a. Align the holes on the septa with the wells of the plate.
   b. Press firmly until the septa snaps into position.

9. Assemble the plate with the retainer and base, then load on the instrument. Reactions can be run on the 3500/3500xL Genetic Analyzer or the SeqStudio™ Genetic Analyzer.

See the instrument user guide for specifics on setting up the run.

Naming conventions

IMPORTANT! Sample names must match exactly in the GeneMapper™ Software and the CSV export for the CarrierMax™ Software to recognize CNV and SNP paired reactions.

CNV or SNP must be added to the file name to be correctly imported.

Example sample names:

- CNV-XXX
- SNP-XXX

The XXX part of the name must match exactly in the CarrierMax™ Software in order for the CNV and SNP files to be analyzed as a paired result from the sample.

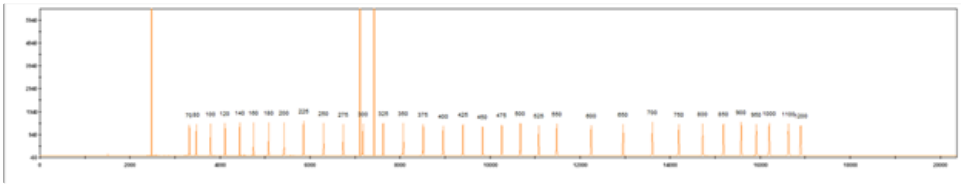
Analyze the results

Data analysis workflow

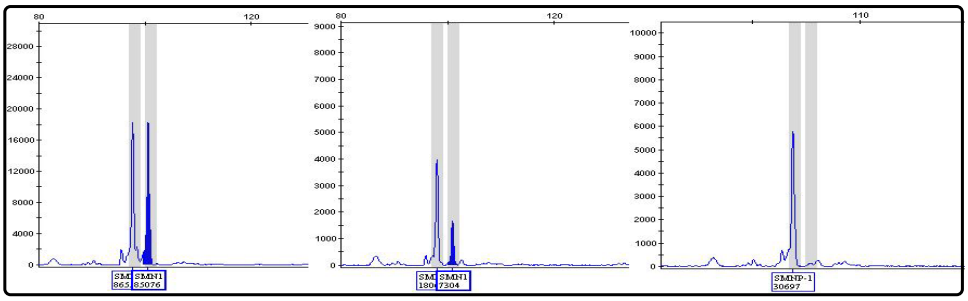
Create a project and analyze sample files with GeneMapper™ Software

Note: Before first use, the latest instrument-specific settings files must be imported. See "About importing files into the GeneMapper™ Software database" on page 34.

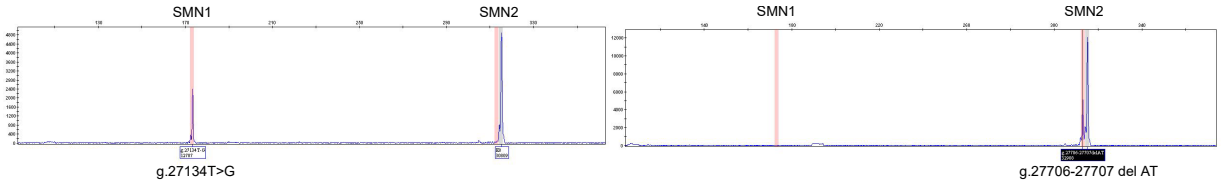
Determine sizing quality (SQ), then correct as required



Review the CNV peak results



Review the SNP peak results



Export the genotypes table

Ensure that the sample naming conventions have been followed. See "Naming conventions" on page 31.

Analyze the data with CarrierMax™ Software

If there are **No call** results, see the "Results troubleshooting" on page 55.

Analyze the data with GeneMapper™ Software

Overview of the GeneMapper™ Software

GeneMapper™ Software is a flexible genotyping software package that provides DNA sizing and quality allele calls for all Applied Biosystems™ electrophoresis-based genotyping systems. GeneMapper™ Software helps increase data processing efficiency with a multiuser, client-server deployment. The software uses Process Quality Values (PQVs) for automated identification that reduces data review time for high throughput genotyping. In addition, the security and audit features help users meet 21 CFR 11 requirements.

For information on obtaining the GeneMapper™ Software, contact your local field application specialist.

About importing files into the GeneMapper™ Software database

Before you use GeneMapper™ Software to analyze assay data for the first time, you must import the latest settings files.

IMPORTANT! Settings files are instrument-specific. Ensure that you import files for the instrument that corresponds to the data you will analyze.

Note: The names and version numbers of the files that are shown in this section may differ from the file names and version numbers that you see when you download or import files.
If you need help to determine the correct files to use, contact your local field applications scientist or technical support.

File to import	File type	Description	See
Panel	TXT	Defines the markers (loci) that are being interrogated. **IMPORTANT!** Use only panel and bin TXT files that are provided by Thermo Fisher Scientific. If you use panel and bin files that you create in the GeneMapper™ Software, you may see off-ladder alleles.	"Import panel and bins" on page 35
Bins	TXT	Defines bins (location of expected alleles) for each marker.	
Analysis Method	XML	Defines the settings used for peak detection, allele calling, and peak quality flags.	"Import analysis method" on page 37
Size Standard	XML	Defines the sizes of the fragments present in the size standard.	"Import the size standard" on page 36

(continued)

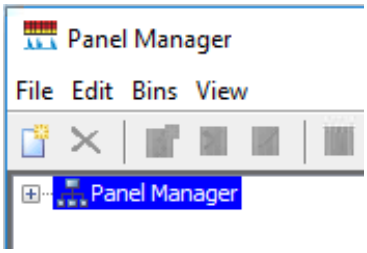
File to import	File type	Description	See
Table Settings	XML	Hide or show columns, filter results, sort order.	"Import table settings and plot settings" on page 38
Plot Settings	XML	Defines settings for electropherogram plot display including Sample Header Settings, Genotypes Header settings, Sizing Table settings, peak labels settings, and display settings for Sample and Genotype plots.	

Import panel and bins

IMPORTANT! Use only panel and bin TXT files that are provided by Thermo Fisher Scientific. If you use panel and bin files that you create in the GeneMapper™ Software, you may see off-ladder alleles.

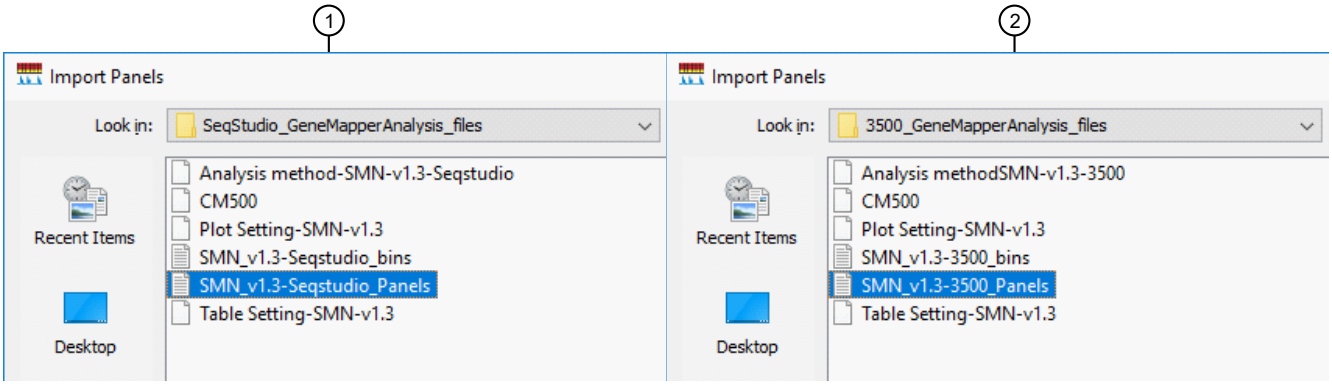
1. Start the GeneMapper™ Software, then log in with the appropriate user name and password.
2. Select **Tools** ▸ **Panel Manager**.
3. In the left pane, click **Panel Manager**.

   Note: If you do not click on **Panel Manager**, the import commands are not available.



4. Import the panel file:
   a. Select **File** ▸ **Import Panels** to open the **Import Panels** dialog box.
   b. Navigate to, then select, the appropriate panel file for your instrument.

      Note: The files can be downloaded from **thermofisher.com/carriermax-smn1**.

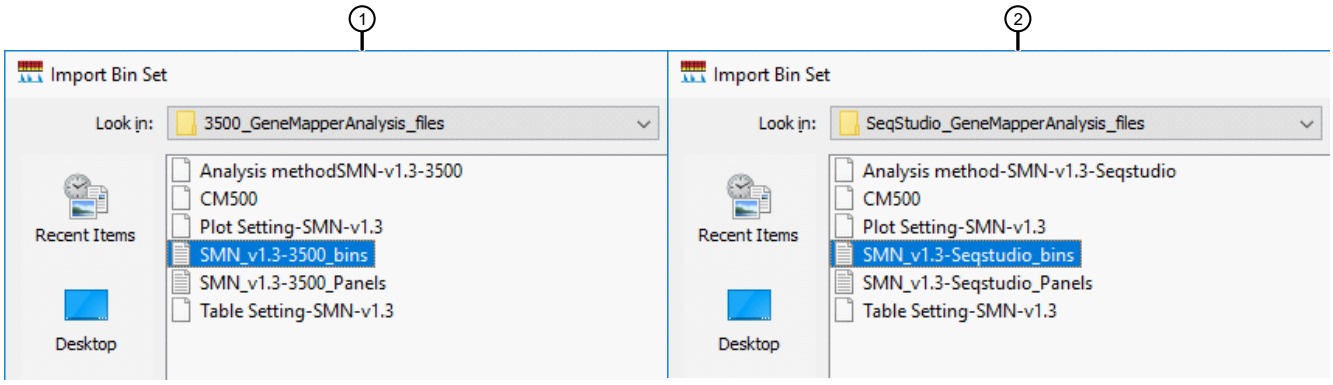


① 3500/3500xL Genetic Analyzer
② SeqStudio™ Genetic Analyzer

c. Click **Import**.

Note: Importing this file creates a new folder in the navigation pane of the **Panel Manager**. This folder contains the panel and associated markers.

5. Import the bins file:
   a. Select the SMN panel.
   b. Select **File ▸ Import Bin Set** to open the **Import Bin Set** dialog box.
   c. Navigate to, then select, the appropriate bin file for your instrument.



① 3500/3500xL Genetic Analyzer
② SeqStudio™ Genetic Analyzer

6. *(Optional)* View the imported panels and bins in the navigation pane: Double-click the SMN folders in the left pane.

   The panel information is displayed in the right pane and the markers are displayed below it.

7. Click **Apply**, then click **OK** to add the panel and bins to the GeneMapper™ Software database.

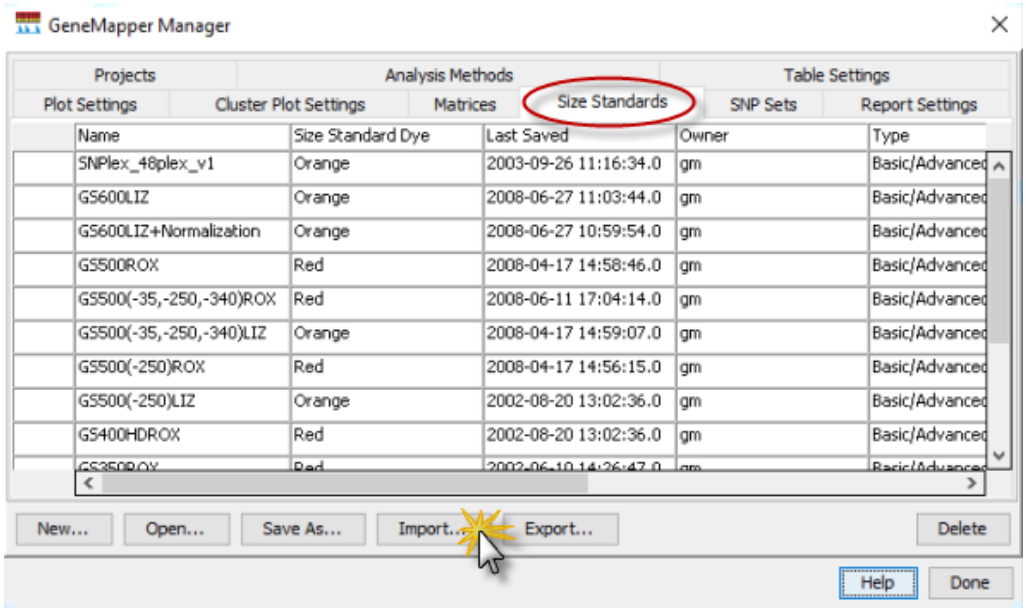
IMPORTANT! If you close the **Panel Manager** without clicking **Apply**, the panels, bin sets, and marker stutter are not imported into the software database.

Import the size standard

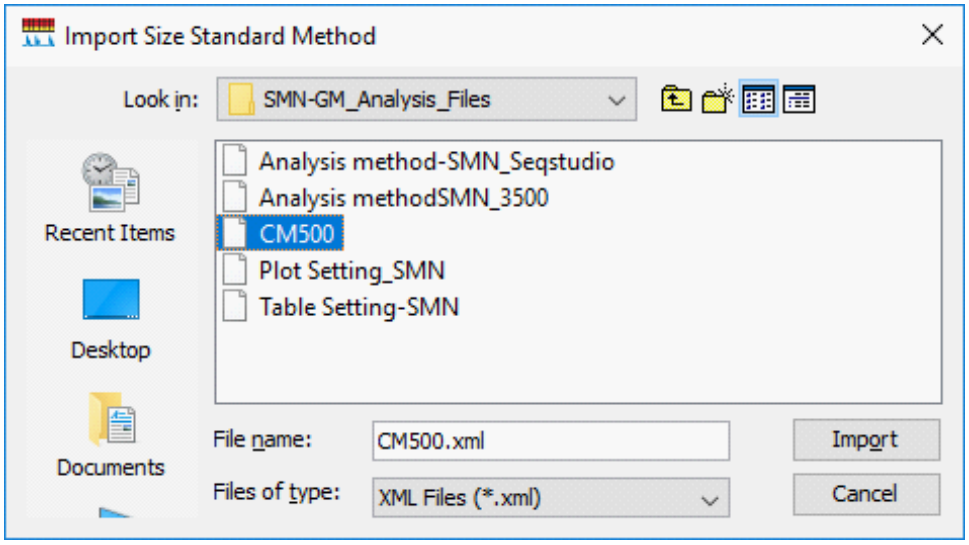
Import the latest size standard from the website into the GeneMapper™ Software database:

1. In the GeneMapper™ Software, select **Tools ▸ GeneMapper Manager**.
2. In the **GeneMapper Manager** window, select the **Size Standards** tab.

3. In the **Size Standards** tab, click **Import.**



4. Navigate to, then select, the appropriate size standards file.



5. Click **Import**.

6. Click **Done**.

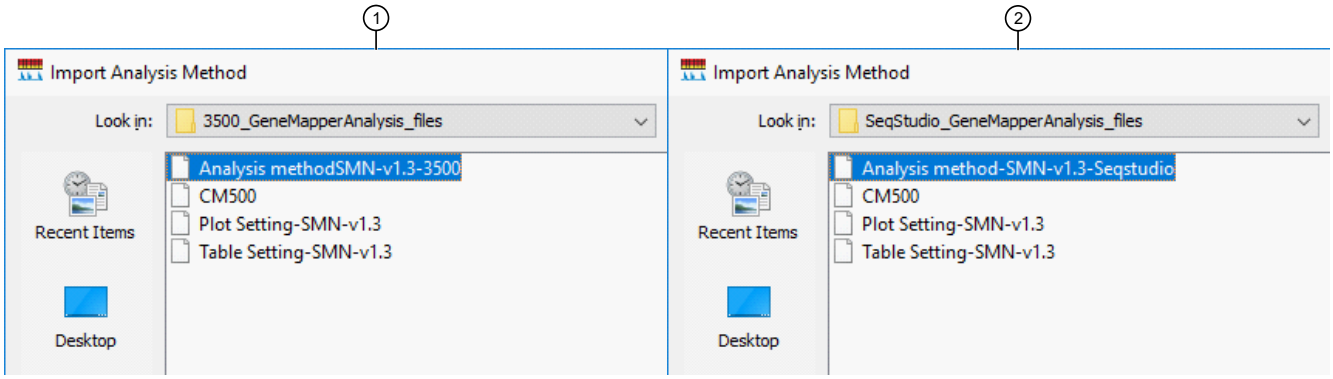
Import analysis method

1. In the GeneMapper™ Software, select **Tools ▸ GeneMapper Manager**.

2. In the **GeneMapper Manager** window, select the **Analysis Methods** tab.

3. In the **Analysis Methods** tab, click **Import**.

4. Navigate to, then select, the SMN file for your instrument type.



① 3500/3500xL Genetic Analyzer
② SeqStudio™ Genetic Analyzer

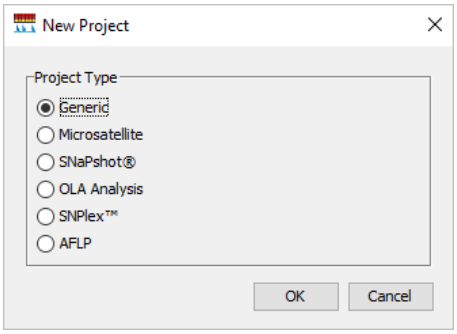
5. Click **Done**.

Import table settings and plot settings

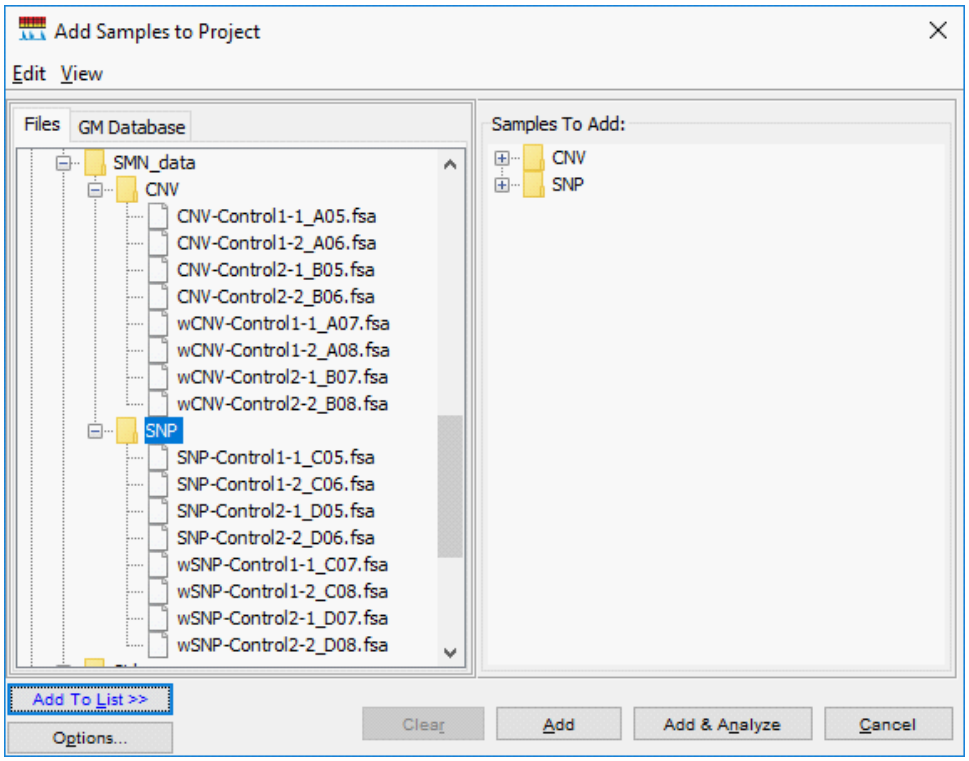
1. In the GeneMapper™ Software, select **Tools** ▸ **GeneMapper Manager**.
2. In the **Table Settings** tab, click **Import**.
3. Navigate to, then select, the appropriate table setting file.
4. Click **Import**.
5. In the **Plot Settings** tab, click **Import**.
6. Navigate to, then select, the appropriate plot setting file.
7. Click **Import**.
8. Click **Done**.

Create a project and analyze sample files with GeneMapper™ Software

1. If a project is currently open, select **File** ▸ **New Project**, then select **Generic**.

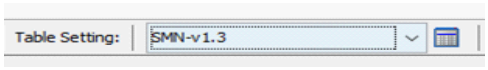


2. In the **Project** window, select **File ▸ Add Samples to Project**, select the folder that contains the sample files, click **Add to List**, then click **Add**.



IMPORTANT! Duplicate or triplicate samples must have unique **Sample Names** to be analyzed separately.

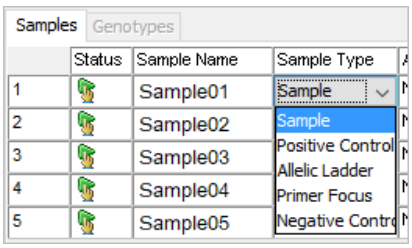
3. Select the table setting that determines the columns that are displayed in samples table and genotypes table: Click the arrow next to the **Table Setting** list, then select the setting.



4. Apply analysis settings to the samples in the project:

   a. Click the **Sample Type** field in the first row, then select the appropriate sample type.

   Repeat for each sample.



   b. Repeat for the **Analysis Method**, **Panel**, and **Size Standard** fields.

   IMPORTANT! Analysis method is instrument-specific. Ensure that you select the appropriate analysis method for the sample files in the project.

   c. *If this is the first time the analysis method is used*: Double-click the analysis method in the first row, then click the **Allele** tab. Select the bin set you imported.

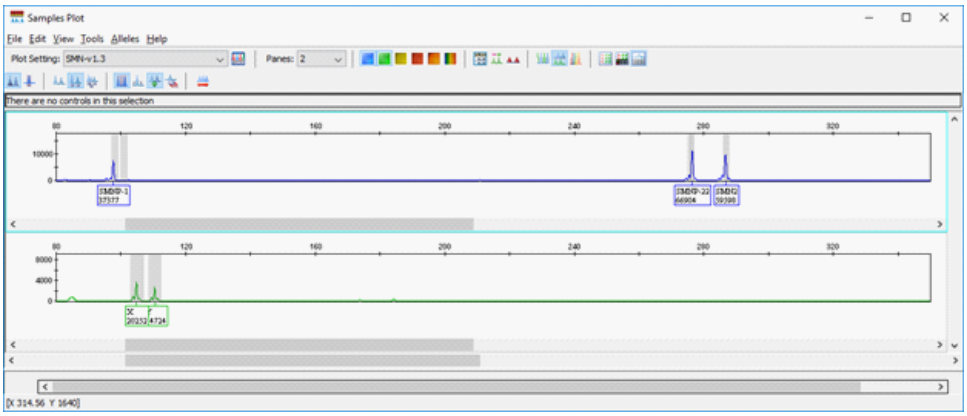
d. Fill in remaining rows.

Note: You can fill in the first row, click the column header to select all rows, then use **Ctrl+D** to fill down remaining rows.

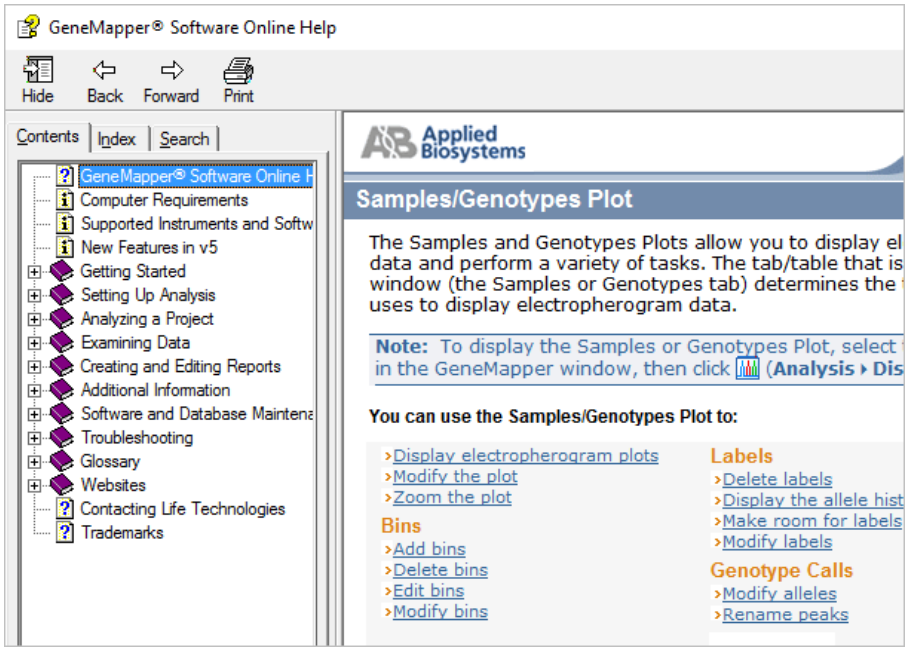
5. Click **(Analyze)**, enter a name for the project (in the **Save Project** dialog box), then click **OK** to start analysis.
   - The status bar displays the progress of analysis as a completion bar.
   - The table displays the row of the sample currently being analyzed in green (or red if analysis failed for the sample).
   - When analysis is complete, the **SQ** field displays a PQV result, and the **Genotypes** tab is available.

Display and zoom on sample plots

1. To view the electropherogram for a sample, click the sample in the sample table, then click **(Display Plots)**.

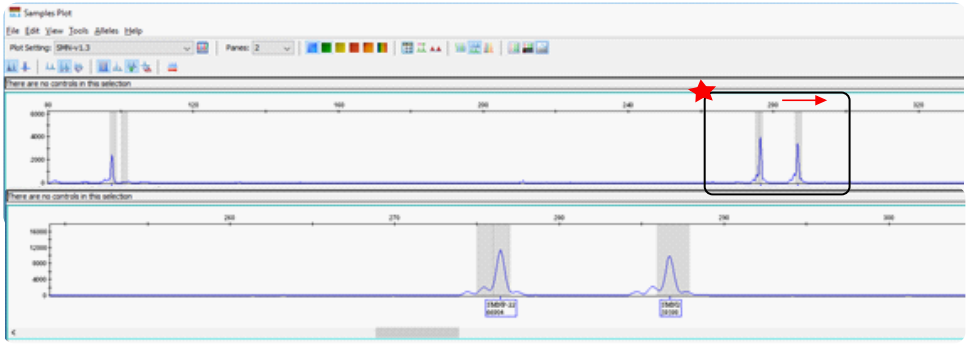


For more information on the samples plot, select **Help ▸ Contents and Index**.



2. Select the plot setting that determines the headers that are displayed in sample and genotype plot: Click the arrow next to the **Plot Setting** list at the top of the screen, then select the setting.

3. To zoom on the plot, select **View ▸ Zoom**, or:
   - To zoom on the x- axis, place the cursor on the x-axis *at the top of the plot*, then click-drag to the right.



   - To zoom on the y- axis, place the cursor on the 0 y-axis, then click-drag up.
   - To unzoom double-click the x-axis.

4. To close the sample plot, click the X in top-right corner of the screen.

Results guidelines

IMPORTANT! Negative and positive controls must be run with every sample run.

Expected results:

- No template controls—Negative controls are normal if they do not show amplification products within the detection range of the SMN1 gene (100-103 bp) or the SMN2 gene (283-293 bp). Negative controls are called homozygote affected in the CarrierMax™ Software.
- Positive controls—(*Optional*) Use Corielle controls. See "Required materials not supplied" on page 6 for controls and their expected results. Control DNA is available to show 0, 1, or 2+ copies of the SMN1 gene.
- Samples—peaks of each amplicon should be similar. Peaks should not be included if they display an abnormal peak shape.

For more information see Appendix A, "Troubleshooting".

Determine sizing quality (SQ), then correct as required

1. In the sample table, select a sample with a **SQ** label, then click **(Size Match Editor)**.
2. In the Size Match Editor, select **Add**, **Delete**, or **Change** for individual peaks as needed.
3. Repeat for each sample with a **SQ** label.

Review the CNV peak results

1. Perform a quality control check by following the "Results guidelines" on page 42.
2. In the **Samples** tab, select **Analysis ▸ Display Plots**. Click **Binning Mode**, then zoom in on the target CNV peak areas as needed.

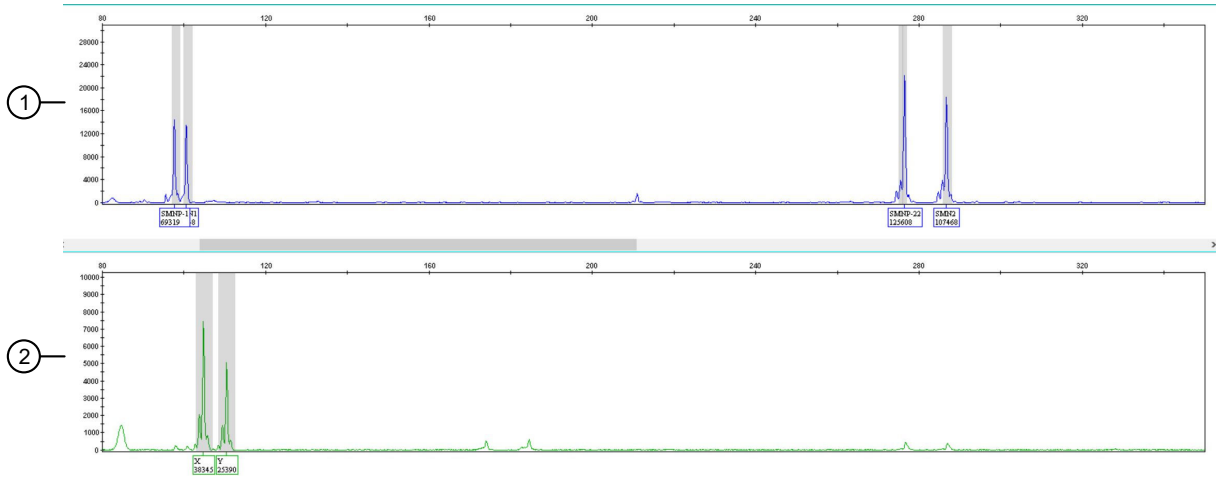


Figure 1 Example bins and peaks using the CarrierMax™ SMN1/SMN2 Reagent CNV Primers Mix

① FAM™ dye
② HEX™ dye

3. Ensure that all the copy number variant peaks (FAM™ dye) are labeled. For the SMN1 gene, target peaks are between 100-103 bp. SMN2 peaks are between 283-293 bp.

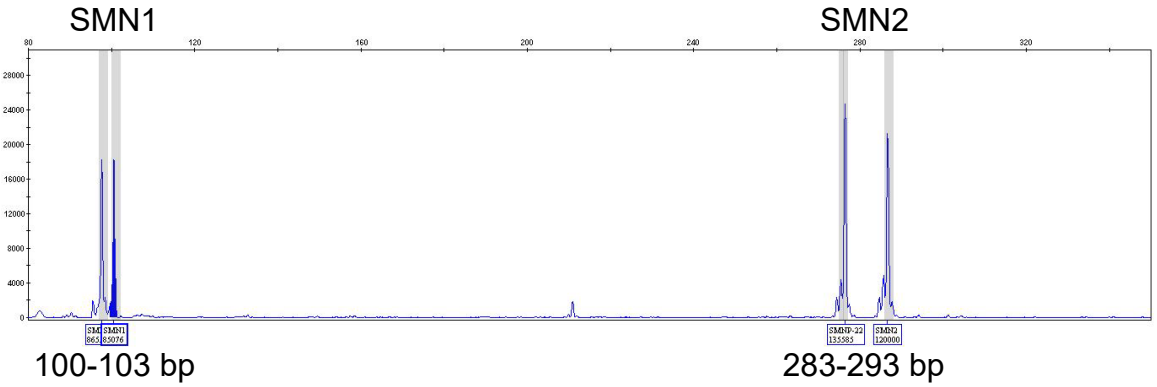


Figure 2 Example of SMN1 and SMN2 peaks using the CarrierMax™ SMN1/SMN2 Reagent CNV Primers Mix

---

Note: If any peaks are not labeled, highlight the peak, left-click, then add the **Allele Name**. Not labeling peaks can result in miscounts of allele copies.

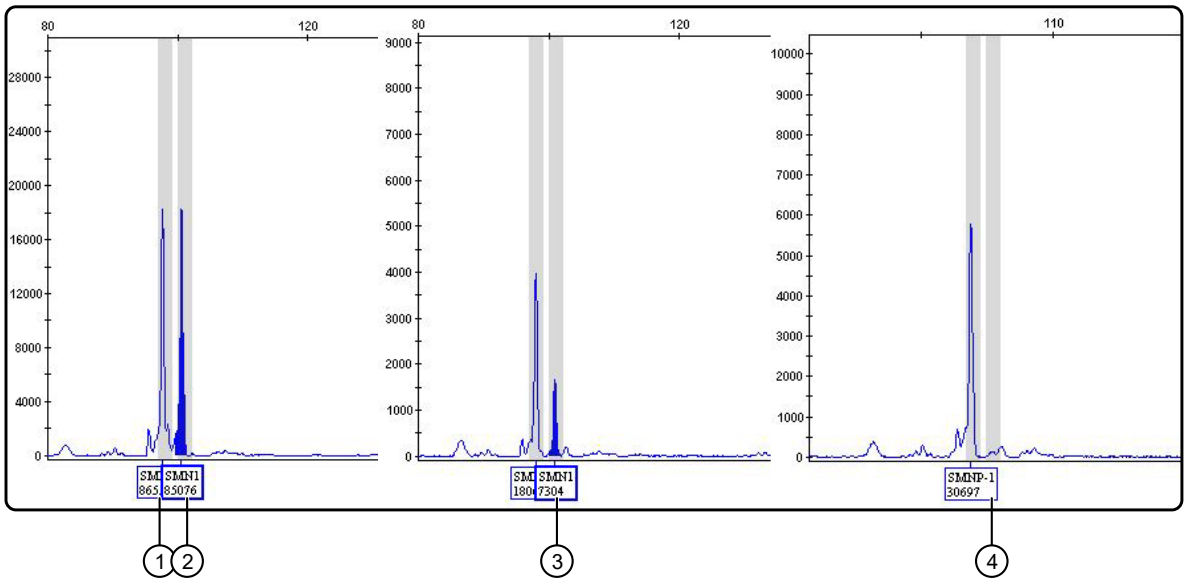


Figure 3 Example of SMN1 peaks using the CNV primers. Representative peaks for 0, 1, and 2 copies of SMN1

① Control SMN pseudogene
② SMN1—2 copies
③ SMN1—1 copy
④ SMN1—0 copies

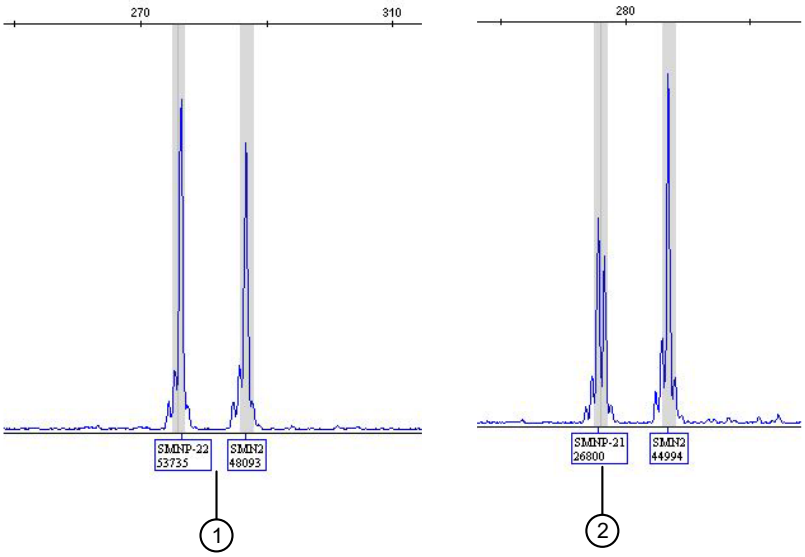


Figure 4 Example of SMN2 peaks using the CarrierMax™ SMN1/SMN2 Reagent CNV Primers Mix

① Standard peaks for SMN2—2 copies
② Example of split peaks for SMN2. One peak is not labeled.

Note: Bifurcation, or splitting, of the SMN2 control peak may be observed in the CNV reaction of some samples. Two peaks are generated in the presence of a SNP (commonly a G deletion) in the region amplified by the SMN2 control primers. These peaks correspond to the two alleles (one with SNP and one without). When this phenomenon occurs, the area of the control peak is calculated by adding the areas from both peaks.

4. Ensure that the X and Y peaks (HEX™ dye) are labeled. If labels aren't present, the result is **No Call**.

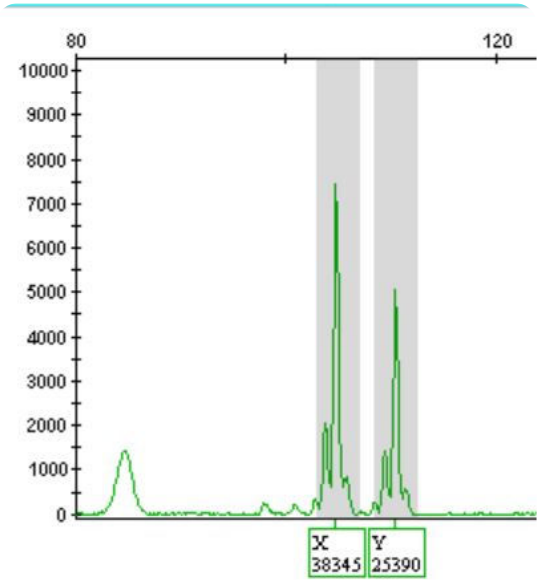


Figure 5 Example of X and Y peaks using the CarrierMax™ SMN1/SMN2 Reagent CNV Primers Mix

Review the SNP peak results

1. Perform a quality control check by following the “Results guidelines” on page 42.

2. In the **Samples** tab, select **Analysis ▸ Display Plots**. Click **Binning Mode**, then zoom in on the target SNP peak areas as needed.

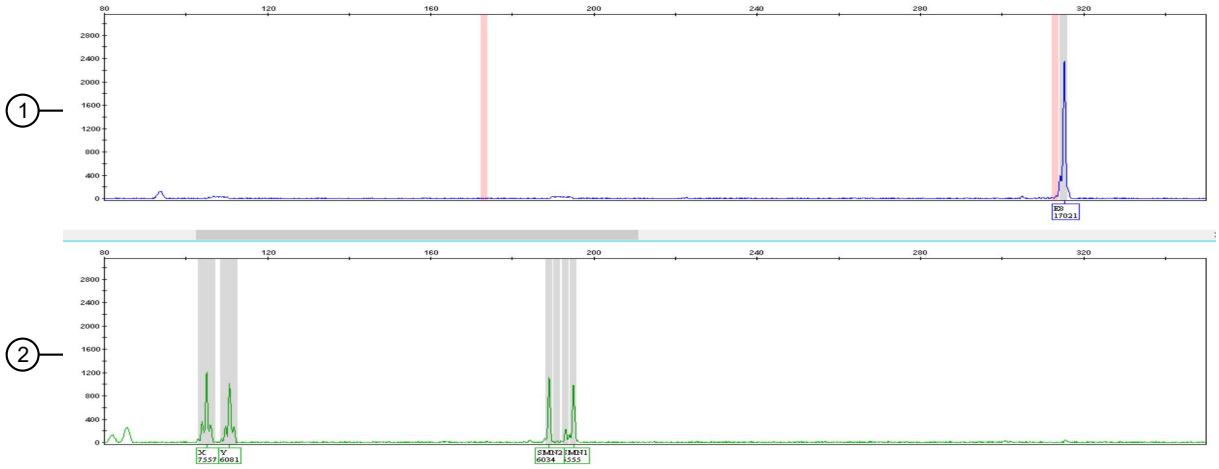


Figure 6 Example bins and peaks using the CarrierMax™ SMN1/SMN2 Reagent SNP Primers Mix

① FAM™ dye
② HEX™ dye

3. Ensure that all the SNP peaks (FAM™ dye) are labeled. The CarrierMax™ SMN1/SMN2 Reagent SNP Primers Mix results in target peaks between 172-174 bp and between 316-318 bp.

   The mutations shown in the pink bins are indicators of 2 alleles on the same chromosome (2+0 haplotype), and are counted as silent carriers during screening.

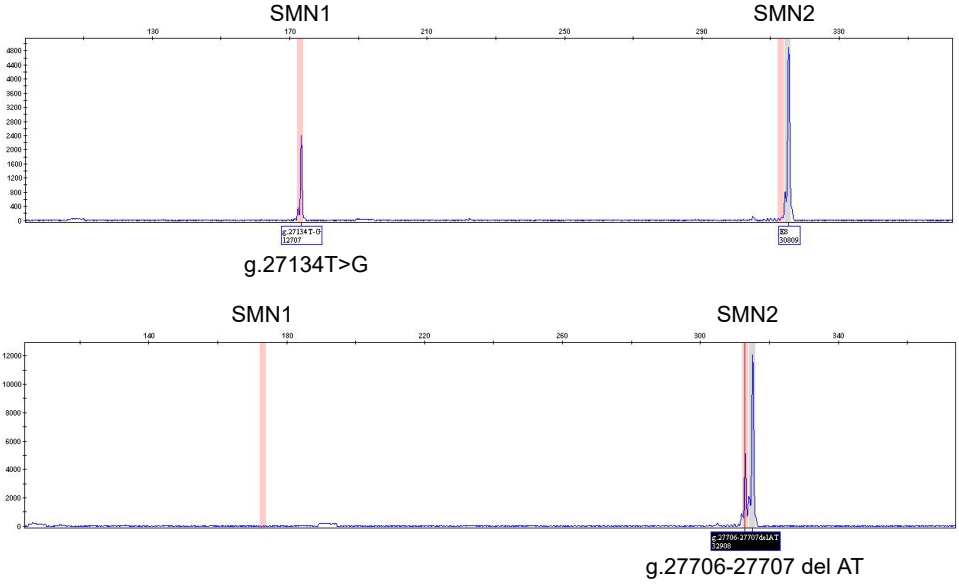


Figure 7 Examples of two common mutations (FAM™ dye). Top: T>G SNP; Bottom: AT deletion

Note: If any peaks are not labeled, highlight the peak, left-click, then add the **Allele Name**. Not labeling peaks can result in miscounts of allele copies.

4. Ensure that all the SNP peaks (HEX™ dye) are labeled.

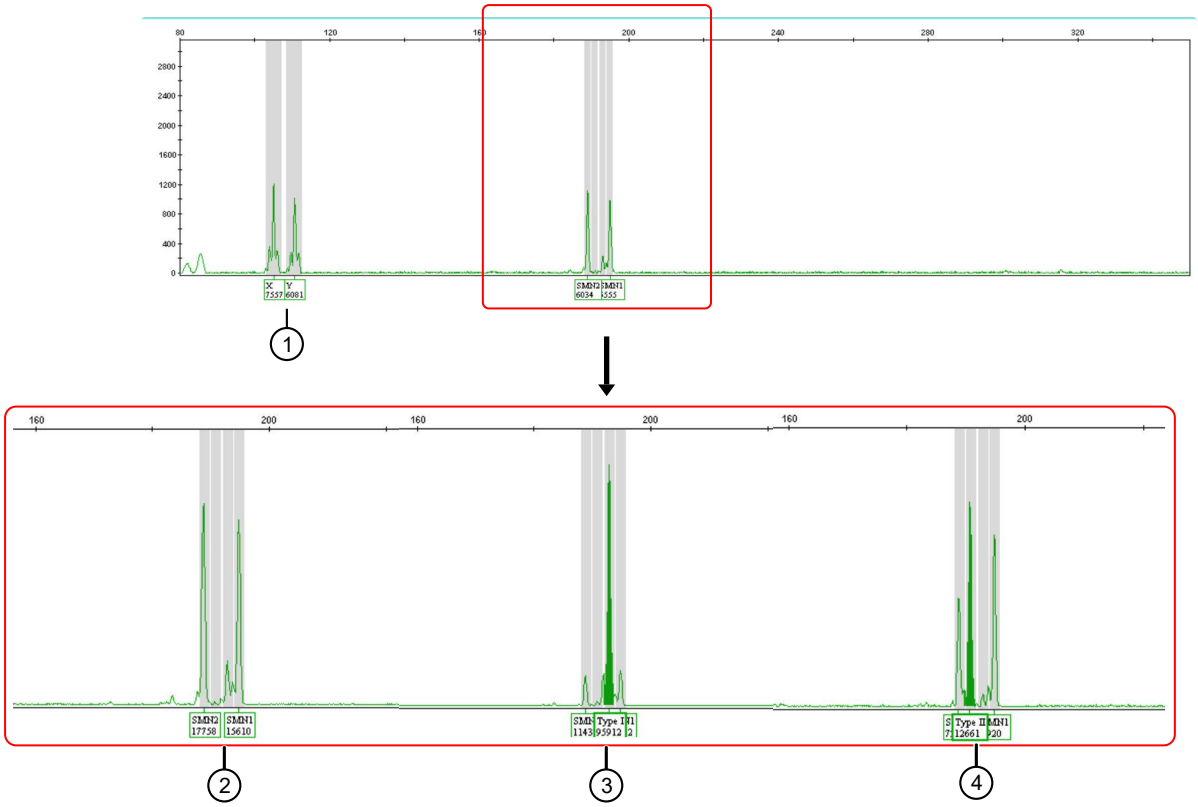


Figure 8 Examples of gene conversion events (HEX™ dye)

Gene conversion events are rare, but can be identified with the CarrierMax™ SMN1/SMN2 Reagent SNP Primers Mix.

① X and Y peaks
② No gene conversion event
③ Type 1 gene conversion (classified as **Normal**)
④ Type 2 gene conversion (classified as **Carrier**)

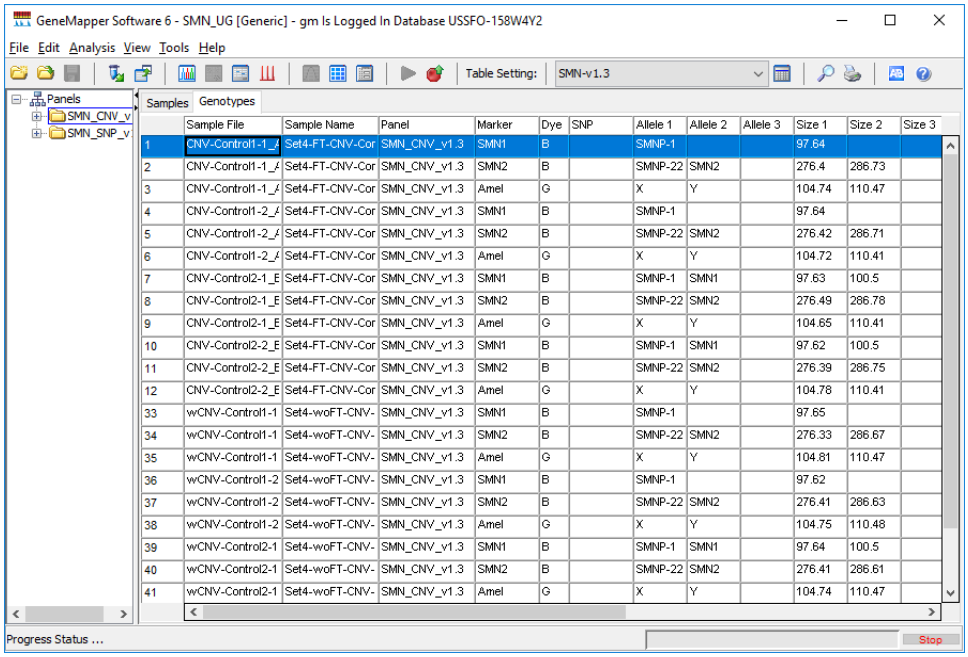
Note: True gene conversion events are characterized by large peak heights. Disregard small shoulder peaks.

IMPORTANT! Ensure that the X and Y peaks are also labeled correctly. If labels aren't present, the result is **No Call**.

After reviewing all peaks, and labeling peaks that were missed by the software, go to “Export the genotypes table” on page 47.

Export the genotypes table

1. Open the project that contains the samples of interest.
2. Click the **Genotypes** tab.



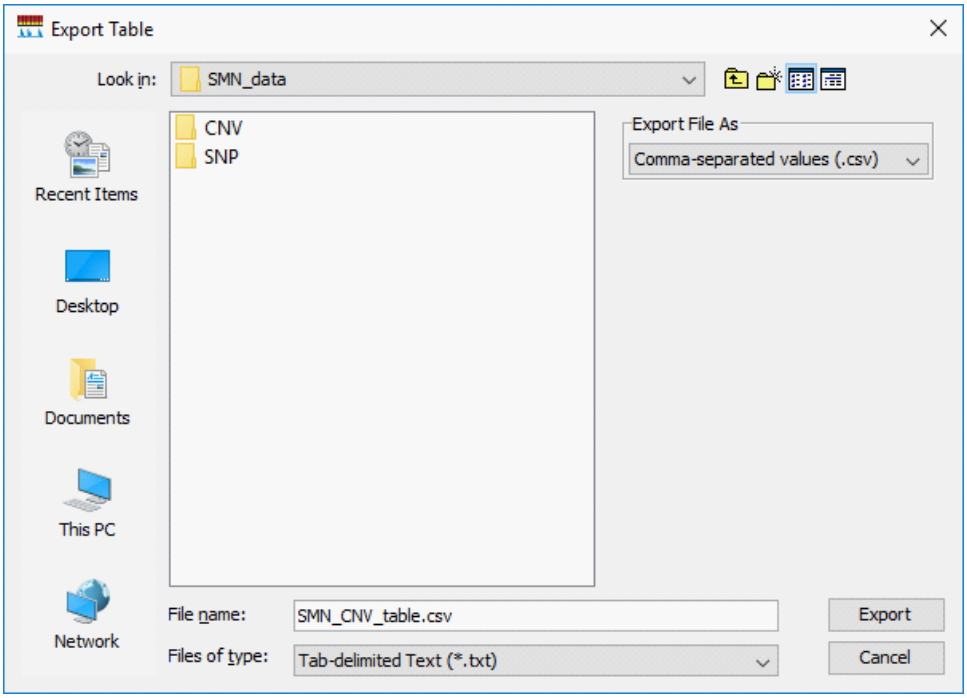
	Sample File	Sample Name	Panel	Marker	Dye	SNP	Allele 1	Allele 2	Allele 3	Size 1	Size 2	Size 3
1	CNV-Control1-1_A	Set4-FT-CNV-Cor	SMN_CNV_v1.3	SMN1	B		SMNP-1			97.64		
2	CNV-Control1-1_A	Set4-FT-CNV-Cor	SMN_CNV_v1.3	SMN2	B		SMNP-22	SMN2		276.4	286.73	
3	CNV-Control1-1_A	Set4-FT-CNV-Cor	SMN_CNV_v1.3	Amel	G		X	Y		104.74	110.47	
4	CNV-Control1-2_A	Set4-FT-CNV-Cor	SMN_CNV_v1.3	SMN1	B		SMNP-1			97.64		
5	CNV-Control1-2_A	Set4-FT-CNV-Cor	SMN_CNV_v1.3	SMN2	B		SMNP-22	SMN2		276.42	286.71	
6	CNV-Control1-2_A	Set4-FT-CNV-Cor	SMN_CNV_v1.3	Amel	G		X	Y		104.72	110.41	
7	CNV-Control2-1_E	Set4-FT-CNV-Cor	SMN_CNV_v1.3	SMN1	B		SMNP-1	SMN1		97.63	100.5	
8	CNV-Control2-1_E	Set4-FT-CNV-Cor	SMN_CNV_v1.3	SMN2	B		SMNP-22	SMN2		276.49	286.78	
9	CNV-Control2-1_E	Set4-FT-CNV-Cor	SMN_CNV_v1.3	Amel	G		X	Y		104.65	110.41	
10	CNV-Control2-2_E	Set4-FT-CNV-Cor	SMN_CNV_v1.3	SMN1	B		SMNP-1	SMN1		97.62	100.5	
11	CNV-Control2-2_E	Set4-FT-CNV-Cor	SMN_CNV_v1.3	SMN2	B		SMNP-22	SMN2		276.39	286.75	
12	CNV-Control2-2_E	Set4-FT-CNV-Cor	SMN_CNV_v1.3	Amel	G		X	Y		104.78	110.41	
33	wCNV-Control1-1	Set4-woFT-CNV-	SMN_CNV_v1.3	SMN1	B		SMNP-1			97.65		
34	wCNV-Control1-1	Set4-woFT-CNV-	SMN_CNV_v1.3	SMN2	B		SMNP-22	SMN2		276.33	286.67	
35	wCNV-Control1-1	Set4-woFT-CNV-	SMN_CNV_v1.3	Amel	G		X	Y		104.81	110.47	
36	wCNV-Control1-2	Set4-woFT-CNV-	SMN_CNV_v1.3	SMN1	B		SMNP-1			97.62		
37	wCNV-Control1-2	Set4-woFT-CNV-	SMN_CNV_v1.3	SMN2	B		SMNP-22	SMN2		276.41	286.63	
38	wCNV-Control1-2	Set4-woFT-CNV-	SMN_CNV_v1.3	Amel	G		X	Y		104.75	110.48	
39	wCNV-Control2-1	Set4-woFT-CNV-	SMN_CNV_v1.3	SMN1	B		SMNP-1	SMN1		97.64	100.5	
40	wCNV-Control2-1	Set4-woFT-CNV-	SMN_CNV_v1.3	SMN2	B		SMNP-22	SMN2		276.41	286.61	
41	wCNV-Control2-1	Set4-woFT-CNV-	SMN_CNV_v1.3	Amel	G		X	Y		104.74	110.47	

3. Display the columns that you want to export.

   IMPORTANT! CNV and SNP genotype results must be exported as separate tables.

4. Highlight the CNV panel, then select **File ▸ Export Table**.
5. Select **CSV** as the export file type (the default is TXT).

6. Click **Export**.



7. Highlight the SNP panel, then select **File ▸ Export Table**.
8. Select **CSV** as the export file type (the default is TXT).
9. Click **Export**.

Analyze the data with CarrierMax™ Software

Download and install the CarrierMax™ Software

1. Navigate to **thermofisher.com/carriermax-smn1**.
2. Download the **CMAXSetup.exe** file.

   Note: The CarrierMax™ Software is compatible with Windows™ 7 and Windows™ 10 operating systems.

3. Double-click the **CMAXSetup.exe** file to launch the InstallShield Wizard, then follow the prompts.

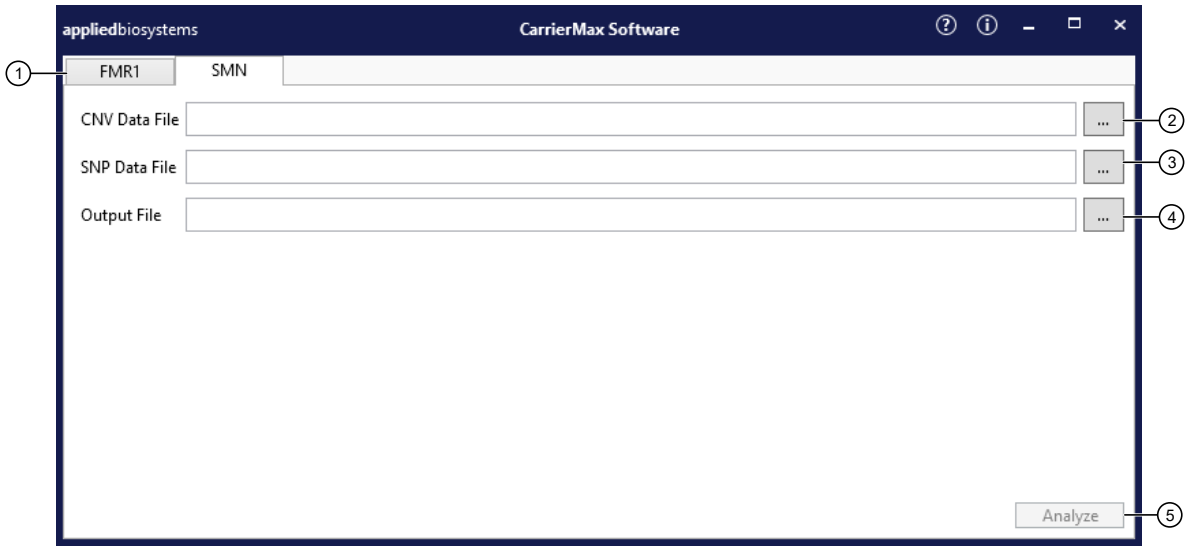
   Note: If a security warning is displayed, click **Run**.

4. Upon installation completion, click **Finish**.

Import sample files

1. In the **Home** screen, click the **SMN** tab.
2. In the **Home** screen, click **... (Ellipses)** for the **CNV Data File**.
3. Navigate to, then select the sample files (CSV) to import. Click **Open**.
4. In the **Home** screen, click **... (Ellipses)** for the **SNP Data File**.
5. Navigate to, then select the sample files (CSV) to import. Click **Open**.
6. In the **Home** screen, click **... (Ellipses)** for the **Output File**. This is the file where your results will be stored. The file must also be in CSV format.
7. Navigate to, then select the output file (CSV) for export. Click **Open**.

8. Click **Analyze**.

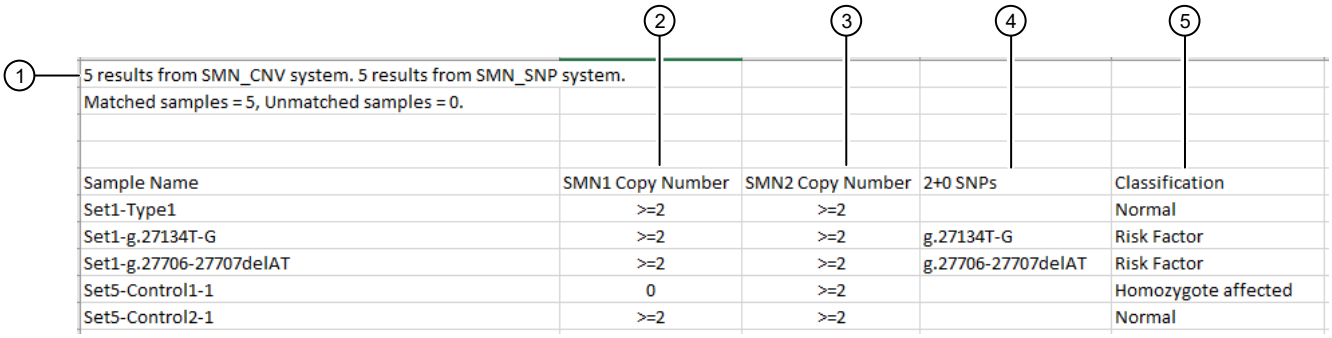


① Assay tab
② Ellipses. Use to select the **CNV Data File**.
③ Ellipses. Use to select the **SNP Data File**.
④ Ellipses. Use to select the **Output File**.
⑤ **Analyze** button

A popup window appears when the analysis is complete. Click **OK** and navigate to the output file to view the results.

View the results

Navigate to the **Output File** selected in "Import sample files" on page 49. Open the file.



5 results from SMN_CNV system. 5 results from SMN_SNP system.				
Matched samples = 5, Unmatched samples = 0.				
Sample Name	SMN1 Copy Number	SMN2 Copy Number	2+0 SNPs	Classification
Set1-Type1	>=2	>=2		Normal
Set1-g.27134T-G	>=2	>=2	g.27134T-G	Risk Factor
Set1-g.27706-27707delAT	>=2	>=2	g.27706-27707delAT	Risk Factor
Set5-Control1-1	0	>=2		Homozygote affected
Set5-Control2-1	>=2	>=2		Normal

① Rows 1–2 display the number of matched files imported.

Note: If the numbers of files imported doesn't match the number of files matched, there was a sample naming discrepancy.

② **SMN1 Copy Number**—**0**, **1**, **≥2**, **ND (Not Determined)**

③ **SMN2 Copy Number**—**0**, **1**, **≥2**, **ND (Not Determined)**

④ **2+0 SNPs**—lists the identity of any 2+0 SNP that is found. Two SNPs are identified with this kit: SMN1 g.27134T>G and SMN1 g.27706-27707delAT.

⑤ **Classification**—lists the SMN classifications called by the software.

Table 5 SMN classifications

Classification	Description
Homozygote affected	No copies of SMN1 detected.
Carrier	1 copy of SMN1 detected.
Normal	≥2 copies of SMN1 detected. No 2+0 SNP mutations detected.
Risk factor	≥2 copies of SMN1 detected. ≥1 2+0 SNP mutations detected.
No Call	SMN1 copy number could not be determined.

A

Troubleshooting

PCR and CE troubleshooting

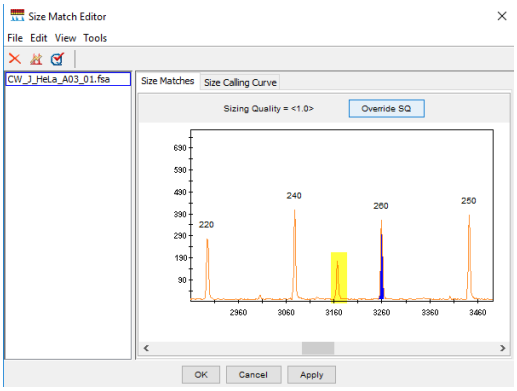
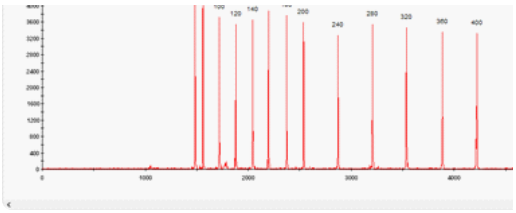
Observation	Possible cause	Recommended action
Faint or no signal from the controls and test samples at all loci	The **PCR Reaction Mix** reagents were not vortexed thoroughly before aliquoting	All mixes should be thawed at room temperature, then vortexed thoroughly before aliquoting.
	Incorrect volume of reagents added.	Repeat amplification ensuring that all components have been added correctly.
	Insufficient sample quantity or quality	Ensure that both sample quantity and quality are at recommended levels.
	Incorrect thermal cycler parameters	Use the recommended thermal cycler parameters.
	Insufficient PCR product injected（and the peaks of size standards are also low or absent)	Check the sample volume to ensure capillaries are going into the sample. Check for bubbles at the bottom of the sample, if present briefly centrifuge samples to remove the bubbles in the well.
	Degraded Hi-Di™ Formamide	Use the recommended storage conditions for Hi-Di™ Formamide . Do not thaw and refreeze multiple times.
Signal obtained for the positive control, but not the test samples	Quantity of the test DNA sample is too low.	Quantitate DNA, then add 10 ng of DNA. Repeat the test to ensure that each target peak is above 50 rfu.
	The test sample contains a high concentration of PCR inhibitor (for example, heme compounds or certain dyes)	Quantitate the DNA, then add the minimum necessary volume. Repeat the test.
		Purify the sample, then repeat the test.
	The sample DNA is degraded.	Evaluate the sample DNA quality by running an agarose gel. If the DNA is degraded, reamplify with an increased amount of DNA.
Poor peak height balance	Too much DNA. Excess DNA causes low-size peaks to appear much higher.	Use the recommended amount of DNA.

Observation	Possible cause	Recommended action
Poor peak height balance *(continued)*	The test sample contains a high concentration of PCR inhibitor (for example, heme compounds or certain dyes)	Quantitate the DNA, then add the minimum necessary volume. Repeat the assay.
		Purify the sample. Repeat the assay.
	The sample DNA is degraded.	Re-extract the sample DNA.
	The PCR Reaction Mix reagents were not vortexed thoroughly before aliquoting.	All mixes should be thawed at room temperature, then vortexed thoroughly before aliquoting.
	The PCR reaction mix was not vortexed thoroughly before aliquoting.	The PCR reaction mix should be vortexed for 10 seconds before aliquoting it into the reaction plate.

GeneMapper™ Software troubleshooting

Troubleshoot a failing SQ in GeneMapper™ Software

1. In the sample table, select a sample with a **SQ** label, then click **(Size Match Editor)**.
2. Follow the steps to **Add**, **Delete**, or **Change** as described in "Determine sizing quality (SQ), then correct as required" on page 42.

Observation	Possible cause	Recommended action
When you click **(Analyze)**, error message is displayed "There are samples that do not meet analysis requirements. Please see Error Message in the info view of each sample."	Multiple possible causes.	Select a sample, then select **View** ▸ **Sample Info**. Review the information in the **Error Message** section of the screen. Correct the issue indicated in the **Error Message** section of the screen.
Cannot select peak in **Size Match Editor** 	The peak signal is below the **Peak Amplitude Threshold** in the analysis method.	Dilute the PCR product further. Increase the amount of size standard.
No Sizing Data error	The size standards did not pass the quality check.	Go to the **Size Match Editor**, then edit each size peak manually. Save, then re-analyze the sample.  **Figure 9 Example: Expected size standard**

Results troubleshooting

Observation	Possible cause	Recommended action
Peak height is too high	Sample DNA concentration is too high.	Use the recommended amount of DNA and dilute PCR products as instructed.

B

Safety

WARNING! GENERAL SAFETY. Using this product in a manner not specified in the user documentation may result in personal injury or damage to the instrument or device. Ensure that anyone using this product has received instructions in general safety practices for laboratories and the safety information provided in this document.

- Before using an instrument or device, read and understand the safety information provided in the user documentation provided by the manufacturer of the instrument or device.
- Before handling chemicals, read and understand all applicable Safety Data Sheets (SDSs) and use appropriate personal protective equipment (gloves, gowns, eye protection, and so on). To obtain SDSs, visit **thermofisher.com/support**.

Chemical safety

WARNING! GENERAL CHEMICAL HANDLING. To minimize hazards, ensure laboratory personnel read and practice the general safety guidelines for chemical usage, storage, and waste provided below. Consult the relevant SDS for specific precautions and instructions:

- Read and understand the Safety Data Sheets (SDSs) provided by the chemical manufacturer before you store, handle, or work with any chemicals or hazardous materials. To obtain SDSs, see the "Documentation and Support" section in this document.
- Minimize contact with chemicals. Wear appropriate personal protective equipment when handling chemicals (for example, safety glasses, gloves, or protective clothing).
- Minimize the inhalation of chemicals. Do not leave chemical containers open. Use only with sufficient ventilation (for example, fume hood).
- Check regularly for chemical leaks or spills. If a leak or spill occurs, follow the manufacturer cleanup procedures as recommended in the SDS.
- Handle chemical wastes in a fume hood.
- Ensure use of primary and secondary waste containers. (A primary waste container holds the immediate waste. A secondary container contains spills or leaks from the primary container. Both containers must be compatible with the waste material and meet federal, state, and local requirements for container storage.)
- After emptying a waste container, seal it with the cap provided.
- Characterize (by analysis if needed) the waste generated by the particular applications, reagents, and substrates used in your laboratory.
- Ensure that the waste is stored, transferred, transported, and disposed of according to all local, state/provincial, and/or national regulations.
- **IMPORTANT!** Radioactive or biohazardous materials may require special handling, and disposal limitations may apply.

Biological hazard safety

WARNING! Potential Biohazard. Depending on the samples used on this instrument, the surface may be considered a biohazard. Use appropriate decontamination methods when working with biohazards.

WARNING! BIOHAZARD. Biological samples such as tissues, body fluids, infectious agents, and blood of humans and other animals have the potential to transmit infectious diseases. Conduct all work in properly equipped facilities with the appropriate safety equipment (for example, physical containment devices). Safety equipment can also include items for personal protection, such as gloves, coats, gowns, shoe covers, boots, respirators, face shields, safety glasses, or goggles. Individuals should be trained according to applicable regulatory and company/ institution requirements before working with potentially biohazardous materials. Follow all applicable local, state/provincial, and/or national regulations. The following references provide general guidelines when handling biological samples in laboratory environment.

- U.S. Department of Health and Human Services, *Biosafety in Microbiological and Biomedical Laboratories (BMBL)*, 5th Edition, HHS Publication No. (CDC) 21-1112, Revised December 2009; found at:
  https://www.cdc.gov/labs/pdf/CDC-BiosafetymicrobiologicalBiomedicalLaboratories-2020-P.pdf
- World Health Organization, *Laboratory Biosafety Manual*, 3rd Edition, WHO/CDS/CSR/LYO/2004.11; found at:
  www.who.int/publications/i/item/9789240011311

C Documentation and support

Related documentation

Document	Publication number
CarrierMax™ A5D Matrix Standard Kit Product Information Sheet	MAN0018893
3500/3500xL Genetic Analyzer with 3500 Series Data Collection Software v3.3 User Guide	100079380
SeqStudio™ Genetic Analyzer Instrument and Software User Guide	MAN0018646
GeneMapper™ Software 5 Installation and Administration User Guide	4476603
GeneMapper™ Software v4.1 Quick Reference Guide	4403615

Customer and technical support

Visit **thermofisher.com/support** for the latest service and support information.

- Worldwide contact telephone numbers
- Product support information
  - Product FAQs
  - Software, patches, and updates
  - Training for many applications and instruments
- Order and web support
- Product documentation
  - User guides, manuals, and protocols
  - Certificates of Analysis
  - Safety Data Sheets (SDSs; also known as MSDSs)

Note: For SDSs for reagents and chemicals from other manufacturers, contact the manufacturer.

Limited product warranty

Life Technologies Corporation and/or its affiliate(s) warrant their products as set forth in the Life Technologies' General Terms and Conditions of Sale at **www.thermofisher.com/us/en/home/global/terms-and-conditions.html**. If you have any questions, please contact Life Technologies at **www.thermofisher.com/support**.