Agilent CytoGenomics 2.0

Setup and Quality Review

User Guide

Research Use Only. Not for Diagnostic Procedures

Agilent Technologies

Notices

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In This Guide...

This guide describes how to use the Agilent CytoGenomics 2.0 software to set up report templates, analysis methods, workflows, standard notes, queries, and tracks. It also describes how to organize samples and use Quality Tools to examine trends and sample quality. The functions described in this guide are available to users with an assigned role of "Scientist" or "Administrator."

1 Configuration and System Setup

This chapter gives an overview of the tasks typically performed by users with the role of Scientist. This includes creating report templates, analysis methods, and workflows, and reviewing and signing off results.

2 Creating and Editing Cyto Report Templates

This chapter describes how to create and edit report templates that are used to generate results reports during a workflow.

3 Configuring Analysis Methods and Workflows

This chapter describes how to set up analysis methods and workflows for performing Feature Extraction and/or analysis of CGH and CGH+SNP microarrays.

4 Displaying and Organizing Samples

This chapter describes how to use the Content tab of Agilent CytoGenomics to manage samples and their attributes.

5 Quality Tools

This chapter provides instructions on how to use the tools in the Quality module of Agilent CytoGenomics to query, evaluate, and chart quality trends.

Contents

1 Configuration and System Setup

In Agilent CytoGenomics, the role of the Scientist is to create and configure report templates, analysis methods, and workflows. Technicians or Scientists then use them to extract and/or analyze the data, generate reports, and review results. The Scientist or Administrator also reviews and signs off the results after the workflows are completed. This user guide gives instructions on how to perform configuration tasks (Scientist or Administrator), review results (all roles), and sign off results (Scientist or Administrator.)

This chapter gives an overview of the set-up and data review tasks. The following chapters contain step-by-step instructions for performing those tasks.

For information on the tasks performed only by users with the Administrator role, see the *Installation and Administration Guide*. For information on running workflows and reviewing results (typical tasks for

the Technician role), see the *Running CytoGenomics Analyses User Guide*. For information on windows, command ribbons, dialog boxes, and reports you see in the Agilent CytoGenomics program, see the *CytoGenomics Reference Guide*.

Getting Help

To get help within Agilent CytoGenomics

Agilent CytoGenomics has several help guides. To open a help guide, on the right side of the Agilent CytoGenomics tab ribbon, click the **Manuals** arrow. Then select the help guide you want to display. Help guides are opened in Adobe® Reader® software.

- To run manual or auto-processing workflows, and review and sign-off results, follow the instructions in the *Running CytoGenomics Analyses User Guide*.
- To learn how to configure system defaults and use Quality Tools to review and chart QC data, see the *Setup and Quality Review User Guide*.
- To learn how to add users and assign roles and change database parameters for client computers, see the *Installation and Administration Guide*.
- Descriptions of windows and dialog boxes for all roles are in the *CytoGenomics Reference Guide*.

Help videos are also available from within the Agilent CytoGenomics program. These short videos give you instructions for doing basic tasks within Agilent CytoGenomics. To start a help video, on the right side of the Agilent CytoGenomics tab ribbon, click **Help Videos.** Then select the video you want to watch.

To contact Agilent Technical Support

Technical support is available by phone and/or e- mail message. Various useful information is also available on the Agilent Technical Support website.

Resource	To find technical support contact information
Agilent Technical Support website	**1** Go to http://genomics.agilent.com. **2** Click **Support**. **3** Select from the available links to display support information.
Contact Agilent Technical Support by telephone or e-mail message (United States and Canada)	Telephone: (800-227-9770) E-mail message: informatics_support@agilent.com
Contact Agilent Technical Support by telephone or e-mail message (for your country)	**1** Go to http://genomics.agilent.com. **2** At the top of the page, select **Contact Us**. **3** Under Worldwide Sales and Support, click to select a country. Complete e-mail message and telephone contact information for your country is displayed.

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User Roles and Capabilities

The following table lists the capabilities allowed for the three different user roles within Agilent CytoGenomics. Tasks highlighted in **bold** are included in this guide.

Table 1 User roles and capabilities

Role	Tasks and capabilities

Technician	• Run manual or auto-processing workflows
	• Add sample information
	• Monitor workflow jobs
	• Display reports and aberration results
	• Triage samples
	  • Check in/out samples
	  • Add notes
	  • Add or suppress calls
	  • Add/change interval classifications
	  • Review and change SNP peak assignment
	• Transfer results to Cartagenia BENCH
	• Display auto-processing logs
	(Technicians cannot sign off samples.)

Scientist	Technician tasks, plus:
	• **Configure system preferences, including**
	  • **Default input/output folders**
	  • **Auto-processing workflow settings**
	  • **Default view preferences**
	  • **Default genome build**
	  • **Standard notes, classifications, and queries**
	  • **Dynamic and static tracks**
	  • **Cartagenia BENCH data transfer settings**
	• **Set analysis and workflow settings**
	• **Create standard notes, classifications, and queries**
	• **Create customized cyto report templates**
	• **Manage samples**
	• **Evaluate and chart QC trends**
	• Sign off results

Administrator	Complete system access, including all Technician and Scientist tasks, plus • Add users and roles • Change database connection settings for client systems

Overview of CytoGenomics Setup Tasks

The following diagram shows the main configuration tasks performed by users in Agilent CytoGenomics with a role of Scientist or Administrator (shown in red). For information on how all users run workflows (shown in blue), see the *Running CytoGenomics Analyses User Guide*.

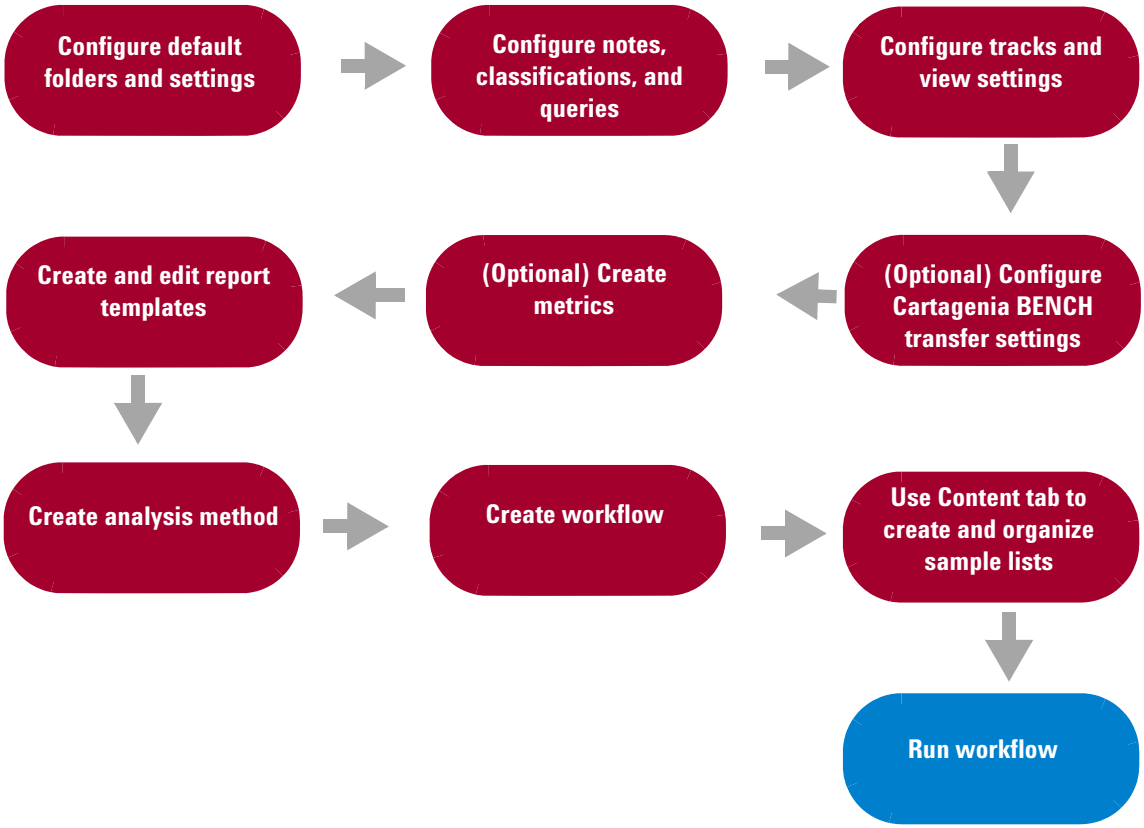


Figure 1 Overview of CytoGenomics tasks for configuring the program

Configuring Default Settings

Before you start using Agilent CytoGenomics, set up the default data folder locations, and preferences for display and analysis of data within the program. These tasks are performed in the Config tab.

Tasks for setting program defaults

Use the Settings command in the Config tab to open tabs where you can set the following default preferences:

Default Settings tab	Configure these settings
Global Settings (applies to all clients connected to a single server)	• Default genome build • Allow use of nondefault genome builds • Allow analysis using designs other than the designs used to extract the data • Default output settings (for naming report and sample output folder)
Local Settings (applies to the local client only)	• Default locations for output, array input, design input, and sample attributes
Auto-Processing Settings (applies to all clients connected to a single server)	• Assign default workflows to array designs • Locations for tiff images, sample attribute files, and auto-processing output • Number of retries in case of workflow failure • Tiff image archive settings

NOTE

By default, reports and results from *manual workflows* are saved in the **\Agilent CytoGenomics<version>\CytoGenomics_Client\Workflow Output** folder.

By default, reports and results from *auto-processing* are saved in the **\Agilent CytoGenomics <version>\CytoGenomics_Server\CommonStorage_<version>\Automation Workflow** folder.

To configure global settings

The following tasks describe how to configure settings common to all CytoGenomics clients.

Table 2 Tasks for setting global defaults

To do this task	Follow these instructions	Comments
Set up default genome build	**1** In the Config tab click **Settings**. **2** Click the **Global Settings** tab. **3** Under Genome Build Settings, click the arrow next to **Default Genome Build**, and select the genome build to set as default. **4** Select **Allow using genome build other than default genome build** to run workflows using genome builds other than the selected default. (This option is selected by default.) **5** Select **Allow running of analysis with different design files than were used for Feature Extraction.** This option lets you perform analysis using a later design version than the original design used to extract the data. (This option is selected by default.) **6** Click **Apply Settings**.	• Global settings are common to all CytoGenomics clients connected to the same CytoGenomics server.

Table 2 Tasks for setting global defaults (continued)

To do this task	Follow these instructions	Comments
Set up default folder and report names	**1** In the Config tab, click **Settings**. **2** Click the **Global Settings** tab. **3** Under Output Settings, select **Use Global Display Name** to use the global display name for the sign-off report and sample output folder names. (This setting is the default.) OR **1** Select **Use Sample Attribute Fields** to select one or more attributes to use for the sign-off report and sample output folder names. **2** Click **Select Sample Attribute**. The Select Sample Attributes dialog box appears. **3** In the Select Sample Attributes dialog box, select one or more attributes to use for the sign-off report and sample output folder names. **4** Click **OK.** **5** Click **Apply Settings**.	• Global settings are common to all CytoGenomics clients connected to the same CytoGenomics server. • Up to three sample attributes can be selected for use in output names. • When a workflow starts, the program checks to make sure that the folder names are unique. If a duplicate folder name exists, the workflow will fail. • Auto-processing uses the defaults set in the Auto-Processing Settings tab. See "To configure auto-processing settings" on page 14.
Display global settings audit trail	**1** In the Config tab, click **Settings**. **2** Click the **Global Settings** tab. **3** Click **Settings Audit**. The Global Setting History Summary dialog box appears. **4** Click **Close**.	

To configure local settings

The following tasks describe how to configure local default settings.

Table 3 Tasks for setting local defaults

To do this task	Follow these instructions	Comments
Set up default output folder	**1** In the Config tab, click **Settings**. **2** Click the **Local Settings** tab. **3** Next to **Select Output Directory**, type the path for the location where you want to save reports and results files. OR **a** Click **Browse** and search for the folder location where you want to save reports and results. **b** Double-click the folder location, then click **Open**. **4** Click **Apply Settings**.	• Local settings are applied only to the client where you are currently logged on. When more than one client uses the same server, each client uses its own local settings. • Manual workflows save reports and results by job name, in the default Output Directory. Within the job folder, results for each sample are saved in subfolders named by the sample global display name, or a combination of selected sample attributes you select in the Global Settings tab. • Auto-processing uses the defaults set in the Auto-Processing Settings tab. See "To configure auto-processing settings" on page 14.
Set up default array input folder	**1** In the Config tab, click **Settings**. **2** Click the **Local Settings** tab. **3** Next to **Select Array Input Directory**, type the path for the location where you want to find extracted arrays (FE files) for import. OR **a** Click **Browse** and search for the folder location. **b** Double-click the folder location, then click **Open**. **4** Click **Apply Settings**.	• Local settings are applied only to the client where you are currently logged on. When more than one client uses the same server, each client uses its own local settings. • The default array input folder is used by default as the location when a manual workflow requires FE Files or .tif images as the input. • Auto-processing uses the defaults set in the Auto-Processing Settings tab. See "To configure auto-processing settings" on page 14.

Table 3 Tasks for setting local defaults (continued)

To do this task	Follow these instructions	Comments
Set up default design input folder	1 In the Config tab, click Settings. 2 Click the **Local Settings** tab. 3 Next to **Select Design Input Directory**, type the path for the location where your designs are located. OR **a** Click **Browse** and search for the folder location. **b** Double-click the folder location, then click **Open**. 4 Click **Apply Settings**.	• Local settings are applied only to the client where you are currently logged on. When more than one client uses the same server, each client uses its own local settings. • By default, the program opens this location when you add designs in a workflow (for design files that are not currently in the database).
Set up default sample attribute file input folder	1 In the Config tab, click **Settings**. 2 Click the **Local Settings** tab. 3 Next to **Sample Attribute File Input Directory**, type the path for the location where the sample attribute files (SAF) are located. OR **a** Click **Browse** and search for the folder location. **b** Double-click the folder location, then click **Open**. 4 Click **Apply Settings**.	• Local settings are applied only to the client where you are currently logged on. When more than one client uses the same server, each client uses its own local settings. • By default, the program opens this location when you add sample attribute files. • Auto-processing uses the defaults set in the Auto-Processing Settings tab. See "To configure auto-processing settings" on page 14.

To configure auto-processing settings

The auto-processing default settings are used to determine what workflows are used for each design, and the folder locations for saving data and results.

Table 4 Tasks for setting auto-processing defaults

To do this task	Follow these instructions	Comments
Set up default workflows for array designs	**1** In the Config tab, click **Settings**. **2** Click the **Auto-Processing Settings** tab. **3** Under AMADID-Workflow Configuration, next to Default Workflow or Default Workflow (CGH+SNP), click the arrow next to **Default Workflow.** Then, select a workflow to use as the default workflow. The selected workflow is assigned when you import a design of this type. **4** Under Workflow, click the arrow next to an array design and select a workflow to use for samples with the designated AMADID.	• AMADID stands for **A**gilent **M**icro**A**rray **D**esign **ID**entifier. The first six digits of a design file is its AMADID. • When you start auto-processing, the program uses the default workflow assigned to the array design for each microarray image. • To use a customized workflow as the default for a design, make sure that the workflow was published (marked as "public").

Table 4 Tasks for setting auto-processing defaults (continued)

To do this task	Follow these instructions	Comments
Set up default input, output, and archive folders	**1** In the Config tab, click **Settings**. **2** Click the **Auto-Processing Settings** tab. **3** Click **Browse** next to **Tiff Image Input Directory,** and find the path for the folder where auto-processing looks for tiff image files. **4** Click **OK.** **5** Repeat steps 2 through 4 to set locations for all of the designated folders. **6** Under Tiff Image Archive Directory, select how you want to archive Tiff images. If you select Archive Tiff to Different Location, click **Browse** and browse to a location where you want to save the Tiff images after the workflow is complete. Or, type the path for the folder.	• The Tiff Image Input Directory is the folder where Agilent CytoGenomics auto-processing looks for scanned image files. This folder is typically the location where your scanner deposits the scanned .tif files. • The SAF File Input Directory contains sample attribute files for samples you are auto-processing. Auto-processing picks up these files and imports them into the Agilent CytoGenomics database. To auto process microarray images, sample information for each array must be present in the database of the program. Sample information is displayed in the Content tab. • The SAF Archive Directory is the location where the SAF files are archived after they are imported to the database during auto processing. • The Auto-Processing Output Directory is where the program saves reports and files generated by workflows run in auto processing mode.
Display auto-processing settings audit trail	**1** In the Config tab, click **Settings**. **2** Click the **Auto-Processing Settings** tab. **3** Click **Settings Audit**. The Automated Workflow Setting History Summary dialog box appears. **4** Click **Close**.	

Configuring Tracks

Agilent CytoGenomics contains a set of default tracks, and you can create custom tracks from Queries or BED files. You can display tracks next to genes and microarray data in the Gene View and/or in reports.

To configure display of tracks

You can choose to show tracks in the Gene View of the Triage View or the View Aberrations windows, and/or in reports.

Table 5 Tasks for displaying tracks

To do this task	Follow these instructions	Comments
Show tracks in Gene View	**1** In the Config tab, click **Tracks**. The tracks settings window appears. **2** In the tracks table, under Show in UI, select the tracks you want to display in the Gene View. **3** In the tracks table, under Show in Report, select the tracks you want to show in reports. OR **1** In Gene View of the Triage View or View Aberrations window, right-click anywhere within the scatter plot, then click **Track Settings**. The Track Settings dialog box appears. See the *CytoGenomics Reference Guide* for more information. **2** Click **Tracks**. **3** Select the **Show In UI** check box of each desired track. **4** Click **OK**. The program displays the selected tracks in Gene View.	• Tracks are only displayed in reports that are generated *after* you select to show tracks in reports. • In the report, a column for the selected track is added to the results. The column contains the hits from the track for each aberrant interval. • In Triage View, only tracks of the same genome builds are displayed. For example, if an analyzed sample has a genome build of hg19, in Triage View, only tracks of hg19 genome build are displayed; tracks of hg18 and other genome builds are not displayed. • For more information on how tracks appear in the Gene View, see the *CytoGenomics Reference Guide*.

Table 5 Tasks for displaying tracks (continued)

To do this task	Follow these instructions	Comments
Display all tracks as a single track	**1** In the Config tab, click **Tracks**. The tracks settings window appears. **2** Under Tracks, select **Show Overlaid**. The program combines the annotations of all selected tracks into a single track named Overlaid Track. **3** To show tracks individually again, clear **Show Overlaid**.	

To create new tracks

There are three ways to create custom tracks for CytoGenomics:

- Create a custom track from a BED file
- Create a dynamic custom track from a query
- Create a custom track graphically

Table 6 Tasks for creating new tracks

To do this task	Follow these instructions	Comments
Search for tracks of interest	**1** In the Config tab, click **Tracks**. The tracks parameters appear, with track search settings at the top. **2** Next to Search in column, select the column you want to use for the search. **3** Next to Operator, select a logical operator to use for comparison of the selected column to the value. **4** Next to Value, select or type the desired value to use for the search. **5** Click **Search**. The tracks that meet the selected search criteria are displayed in the table.	• You can search for tracks by Genome Build, Type, or by User. • To restore the complete list of Tracks, click **Reset**.

Table 6 Tasks for creating new tracks (continued)

To do this task	Follow these instructions	Comments
Create a custom track from a BED file	**1** In the Config tab, click **Tracks**. **2** At the bottom of the window, click **Create Custom Track from BED File**. The Import Track dialog box appears. **3** Next to **Species**, select the species to which the track applies. **4** Next to **Build Name**, select the specific genome build of the species to which the track applies. **5** (Optional) Under Color, click **Change**. **a** In the Swatches tab of the Choose Track Color dialog box, select a color for the track. **b** Click **OK**. **6** Next to **Track Name**, type a name for the track. This name identifies the track within the program, including the name that appears if you include the track in Gene View. **7** Next to **Track File,** click **Browse**. The Open dialog box appears. **8** Select the name of the track (*.bed) file to import. **9** Click **Open**. The location of the file appears in Track File. **10** Click **OK**.	• When you create a track, it appears in the list of tracks, and can be displayed in the UI and/or Reports.

Table 6 Tasks for creating new tracks (continued)

To do this task	Follow these instructions	Comments
Create a custom track from a query	**1** In the Config tab, click **Tracks**. **2** At the bottom of the window, click **Create Custom Track From Query**. The Create Custom Track dialog box appears. **3** Next to **Track Name**, type a name for the track. This name identifies the track within the program, including the name that appears if you include the track in Gene View. **4** Next to **Build Name**, select the specific genome build of the species to which the track applies. **5** (Optional) Under Color, click **Change**. **a** In the Swatches tab of the Choose Track Color dialog box, select a color for the track. **b** Click **OK**. **6** Under Track Query, next to **Select Interval Classification**, click the arrow and select a custom classification to use for the query. **7** (Optional) Select the box next to **And**. Under Track Query, next to **Select Sample Attribute**, click the arrow, and select a sample attribute to use for the query. **8** Under Track Query, next to **Value**, select or type a value for the attribute to use in the query. **9** Click **Create**.	• When you create a track, it appears in the list of tracks, and can be displayed in the UI and/or Reports.

Table 6 Tasks for creating new tracks (continued)

To do this task	Follow these instructions	Comments
Create a custom track graphically	**1** Follow these steps to select a chromosomal region for your track. If you know the exact start and end locations of the chromosomal region, skip to step 2. **a** Open a sample in Triage View or View Aberrations. **b** In Genome View, select the chromosome. The selected chromosome appears in Chromosome View. **c** In Chromosome View, in the plot area to the right of the chromosome, drag the pointer over the approximate chromosomal region of interest. **d** The program draws a blue box around the region, and displays the region in greater detail in Gene View. **e** In Gene View, adjust the view so only the genes of interest appear. For a description of the adjustment commands available in Gene View, see the *CytoGenomics Reference Guide*. **2** Right-click anywhere within the log ratio plot area in Gene View, then click **Create Track**. The Create Track dialog box appears. **3** Under Set Chromosome Start-Stop, click **User Defined**. **4** Type the Name for the track. **5** (Optional) Type a description. **6** Under Set Chromosome Start-Stop, select the chromosome, and type the chromosomal region for the new track. (If you selected the region graphically as described in Step 1, the defined region appears in the boxes.	• For more information on the Triage View and View Aberrations windows, and the Create Track dialog box, see the *CytoGenomics Reference Guide*. • To display a track after you create it, select it in the Config > Tracks window or in the Track Settings dialog box. For information, see the *CytoGenomics Reference Guide*.

Table 6 Tasks for creating new tracks (continued)

To do this task	Follow these instructions	Comments
	7 Under Select Track Source, select **Aberration Results**. **8** (Optional) Under Color, click **Change**. The Change Track Color dialog box appears. **a** In the Swatches tab of the Choose Track Color dialog box, select a color for the track. **b** Click **OK** to accept the color and close the dialog box. **9** Click **OK**.	

Configuring Display Preferences

Agilent CytoGenomics uses a "Genomic Viewer" to display graphical depictions of aberrations and results. You can customize how data are displayed within the Genomic Viewer. For more information on the Genomic Viewer, see the *CytoGenomics Reference Guide*.

To set default display preferences

Use the tasks in the following table to set default preferences for how data and tracks are displayed in the program. These settings can be changed later from within the data views.

Table 7 Tasks for setting default display preferences

To do this task	Follow these instructions	Comments
Set up default preferences for showing tracks	**1** In the Config tab, click **Tracks**. The tracks parameters appear. **2** Select the default preferences for how tracks are displayed in the program. For more information, see the Track Settings dialog box in the *CytoGenomics Reference Guide*. **3** Click **Apply** to save the settings and apply them to the program.	• Tracks contain information for specific genomic locations. A multitude of tracks from diverse sources is available for many species. You can display tracks next to genes and microarray data in Gene View. • To change these settings from within the View Aberrations or Triage View windows, right-click in the Gene View, and select **Track Settings.**
Set up default preferences for display of data in the Genomic Viewer	**1** In the Config tab, click **Views**. The View Preferences parameters appear. For more information, see the View Preferences dialog box in the *CytoGenomics Reference Guide.* **2** Select the default preferences for how data are displayed in the program. **3** Click **Apply** to save the settings and apply them to the program.	• To change these settings from within the View Aberrations or Triage View windows, right-click in the Gene View, and select **View Preferences.**

Creating Standard Notes

Standard notes are predefined notes that you can add to a sample during Triage. You can also select to add standard notes to sign off reports. They are server-based, and any client connected to the same server can access them.

Table 8 Tasks for creating and editing Standard Notes

To do this task	Follow these instructions	Comments
Create a standard note	**1** In the Config tab, click **Notes**. **2** At the bottom of the window, click **Create Standard Note.** The Standard Note dialog box opens. **3** Next to **Name**, type a name to identify the note. **4** Next to **Text**, type the note. **5** Click **OK**. The note appears as a line in the notes list.	• Standard notes are available to all users and can be added to a sample during triage, or to the sign-off report. • You can create a maximum of 20 standard notes.
Edit a standard note	**1** In the Config tab, click **Notes**. **2** Locate the note you want to change in the notes list. Under Action, click **Edit/View**. The Standard Note dialog box opens. **3** Type the changes. **4** Click **OK**. Notification that the note was edited successfully appears. **5** Click **OK**.	

Creating Custom Classifications

A custom classification is a user-defined term used to annotate or classify an aberrant interval. In Triage View, users can search the database for signed-off samples that contain aberrations with selected classifications. For example, search for all "unknown" aberrations over location of chr1:5,000,000-5,100,000. In addition, you can create a track that maps to all intervals that contain a common classification. The track is updated automatically every time a sample is signed off.

Table 9 Tasks for creating and changing custom classifications

To do this task	Follow these instructions	Comments
Create a custom classification	**1** In the Config tab, click Classification. **2** At the bottom of the window, click **Create Classification**. The Classifications dialog box appears. **3** Next to **Name**, type the name of the custom classification. **4** Click **OK**. The classification is added to the custom classifications table, and a message shows that the classification was added successfully. **5** Click **OK** to close the message.	• Custom classifications are assigned to aberration intervals of a sample in the Triage View. For information on Triage View, see the *Running CytoGenomics Analyses User Guide*.
Remove a custom classification	**1** In the Config tab, click **Classification**. **2** In the classification list, locate the classification you wish to remove. **3** Under Action, click **Delete**. A Confirm message appears. **4** Click **OK** to confirm that you want to delete the classification.	• When you delete a classification, it is not removed from any sample intervals where it was previously assigned. • The classification no longer appears in the list of classifications that you can assign in Triage View.

Creating and Editing Queries

In Agilent CytoGenomics, queries are used to search for samples in the database that match criteria defined by the query. A set of default queries is supplied with the program. You can also create your own custom queries. The instructions in the following table describe how to create and edit custom queries.

Table 10 Tasks for creating and editing queries

To do this task	Follow these instructions	Comments
Create a custom query	**1** In the Config tab command ribbon, click **Query**. The Query window appears. **2** At the bottom of the window, click **Create Query**. **3** In the Create Query dialog box, type the name for the new query, and click **Ok**. **4** Under Sample Attribute, click the arrow and select one of the available attributes to use for the search. **5** Select **Equals to** or **Contains**. **6** Under Value, select the attribute value, or (if required) type the value. **7** Click **Add**. The criteria is added to the Conditions for the query. **8** (Optional) Under Logical Operation, click the arrow and select the logic to apply for the next condition. • Perform step 4 through step 7 until all conditions are set up. **9** Click **Save Query** to save the query. The query appears in the query table.	• Logical Operation AND matches samples only if both conditions are true. Logical Operation OR matches samples if any of the conditions are true. • To remove all conditions for a query and start again, click **Clear Conditions**. • Other users cannot see or edit custom queries that you create.

Table 10 Tasks for creating and editing queries (continued)

To do this task	Follow these instructions	Comments
Change a custom query	**1** In the Config tab command ribbon, click **Query**. The Query window appears. **2** In the query table, next to the query you want to change, click **View/Edit**. **3** In the Conditions table, select a condition to change. **4** Click **Edit Condition**. **5** Under Sample Attribute, click the arrow and select one of the available attributes to use for the search. **6** Select **Equals to** or **Contains**. **7** Under Value, select the attribute value, or (if required) type the value. **8** (Optional) To delete a condition, select the condition in the conditions table, and click **Delete Condition**. **9** Click **Update Query** to save the changes to the query. **10** (Optional) Under Logical Operation, click the arrow and select the logic to apply for the next condition. • Perform step 5 through step 7 and click **Add** to add a condition for the query. **11** Click **Update Query** to save the changes to the query. **12** When you are finished, click **Close**.	• You cannot change the default Last 10 Records query. • Logical Operation of AND matches samples only if both conditions are true. Logical Operation of OR matches samples if any of the conditions are true. • Click **Clear Conditions** to remove all conditions for a query and start again.
Delete a custom query	**1** In the Config tab command ribbon, click **Query**. The Query window appears. **2** In the query table, next to the query you want to change, click **Delete**.	• You cannot delete the default queries.

Setting Up Data Transfer for Cartagenia BENCH

You can configure Agilent CytoGenomics to transfer selected reports and images automatically to Cartagenia BENCH. This data transfer happens as part of a workflow run, or manually after a sample is analyzed. Workflows that are configured to send data to Cartagenia BENCH automatically use these settings when they run.

NOTE You must have a valid Cartagenia BENCH account, and know your user name and password to use this feature. You can only transfer data from genome builds supported by Cartagenia BENCH. If a sample is analyzed with a genome build not supported by Cartagenia BENCH, data transfer will fail.

Table 11 Tasks for configuring Cartagenia BENCH data transfer

To do this task	Follow these instructions	Comments
Set Cartagenia BENCH login credentials	**1** In the Config tab, click **Partners**. The Cartagenia BENCH tab appears. **2** Under Cartagenia BENCH Login Credentials, type the URL for your Cartagenia BENCH login website location. **3** Next to **Username**, type your Cartagenia BENCH username. **4** Next to **Password**, type your Cartagenia BENCH password. **5** Click **Test Connection**. A message appears that your connection was successful. If a failed connection message appears, check the information you typed in steps 2 to 4. **6** Click **Apply**.	

Table 11 Tasks for configuring Cartagenia BENCH data transfer (continued)

To do this task	Follow these instructions	Comments
Select data to transfer to Cartagenia BENCH	**1** In the Config tab, click **Partners**. The Cartagenia BENCH tab appears. **2** Under Data to be sent to Cartagenia BENCH, select the reports to send to Cartagenia BENCH. **3** Click **Apply**.	• Only workflows that create the same reports as configured for Cartagenia BENCH transfer data to Cartagenia BENCH.
Select proxy settings for Cartagenia BENCH	**1** In the Config tab, click **Partners**. **2** Under Set Proxy settings, select **Use Proxy settings**. **3** Next to **Proxy Host**, type the name of your proxy server host. **4** Next to **Proxy Domain**, type the name of your proxy domain. **5** Next to **Proxy Port**, type the proxy port number to use. **6** Next to **Proxy User Name**, type your proxy user name. **7** Next to **Proxy Password**, type your proxy password. **8** Click **Apply**.	• If your company uses a firewall, provide valid proxy server settings. • Contact your local network administrator if you do not know the required information for your proxy server settings.

2 Creating and Editing Cyto Report Templates

Agilent CytoGenomics uses report templates to generate Cyto Reports at the end of a workflow. A Cyto Report template is a design for how the report looks and what information it contains. This chapter describes how to create and edit customized Cyto Report templates for use in workflows.

Creating and Editing Cyto Report Templates – Overview

Within CytoGenomics, you can choose to generate result reports automatically as part of the workflow. Two default Cyto Report templates are provided. You can use these reports as-is, or you can edit them to create your own custom report templates. This section describes how to create or edit Cyto Report templates.

NOTE Create or edit the Cyto Report templates *before* you set up workflows.

The following diagram shows the steps for creating or editing a Cyto Report template.

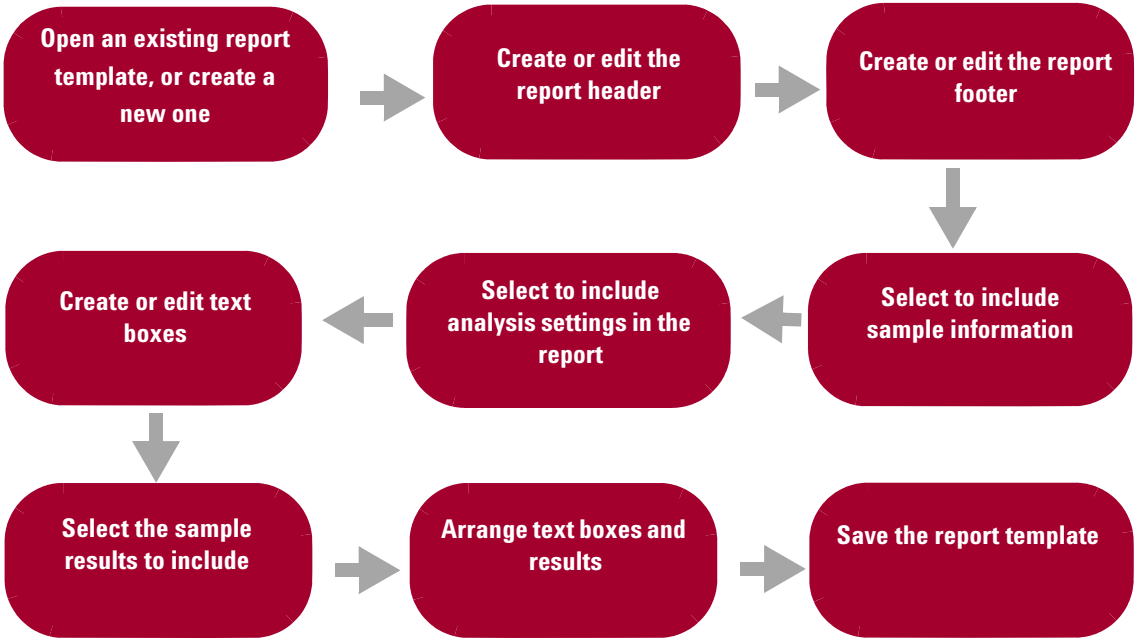


Figure 2 Steps for creating or editing a Cyto Report template

Tasks for Creating or Editing Cyto Report Templates

Table 12 Tasks for creating or editing Cyto Report templates

To do this task	Follow these instructions	Comments
Create a Cyto Report template	1 On the Config ribbon, click **Report**. 2 Click **Create Template.** 3 Type a name for the new template. The Create Cyto Report Template wizard appears.	• You use the Create Cyto Report Template wizard to create a report template from a blank template. Except for the report contents when you start the wizard, the Edit Cyto Report Template wizard and the Create Cyto Report Template wizard are identical. • To go to the next step of the wizard, click **Next**. To return to the previous step, click **Back**.
Create a report template from an existing Cyto Report template	1 On the Config ribbon, click **Report**. 2 Click **Create Template.** 3 In the report template list, in a report template row, click **View/Edit**. The Edit Cyto Report Template wizard appears. 4 Complete the wizard, and then click **Save As**. 5 In the Input dialog box, type the new name for the template and click **OK**.	• Except for the report contents when you start the wizard, the Edit Cyto Report Template wizard and the Create Cyto Report Template wizard are identical. You use the Create Cyto Report Template wizard to create a report template from a blank template. • To go to the next step of the wizard, click **Next**. To return to the previous step, click **Back**. • The program includes two default Cyto Report templates; one template for CGH analyses and one for CGH+SNP analyses. You can change these templates and save them using a different name, but you cannot save changes to the default templates.

Table 12 Tasks for creating or editing Cyto Report templates (continued)

To do this task	Follow these instructions	Comments
Create or edit the report header	**1** Open an existing report template, or create a new one. See Create a Cyto Report template. **2** In Step 1 of the Create Cyto Report Template wizard, under Header, select **Include** to include a header in the report. **3** In the Text box, type the information you want to show in the header of all reports. **4** Optional: Select **Image** to include a graphic image in the header of the report. **a** Click **Browse**. **b** In the Open dialog box, browse to and select the image file you want to include in the report header. **c** Click **Open**. The path to the image file is shown in the wizard.	• A report header can include text, or text and an image.

Table 12 Tasks for creating or editing Cyto Report templates (continued)

To do this task	Follow these instructions	Comments
Create or edit the report footer	**1** Open an existing report template, or create a new one. See Create a Cyto Report template. **2** In Step 1 of the Create Cyto Report Template wizard, under Footer, select **Include** to include a header in the report. **3** In the **Text** box, type the information you want to show in the footer of all reports. **4** Optional: Select **Image** to include a graphic image in the footer of the report. **a** Click **Browse**. **b** In the Open dialog box, browse to and select the image file you want to include in the report footer. **c** Click **Open**. **d** The path to the image file is shown in the wizard. **5** Optional: Select **Date** to include the date in the footer. • Next to **Align**, select where you want to place the date. **6** Optional: Select **Page Number** to include the page number in the footer. • Next to **Align**, select where you want to place the page number.	• A report footer can include one or any combination of the following: text, an image, the date, page number.

Table 12 Tasks for creating or editing Cyto Report templates (continued)

To do this task	Follow these instructions	Comments
Select to include sample information	**1** Open an existing report template, or create a new one. See Create a Cyto Report template. **2** In Step 1 of the Create Cyto Report Template wizard, under Sample Information, select **Sample Information**. **3** In the box next to **Title**, type a title that is shown in the report above the sample information **4** In the box next to **Number of fields**, select the number of sample information fields you want to include. **5** Click **Add**. **6** For each sample information field, select the sample attribute from the list. **7** Optional: Type a name to display for the sample information item.	• Choices for the sample information fields are the sample attributes set in the Sample Manager.
Select to include analysis settings	**1** Open an existing report template, or create a new one. See Create a Cyto Report template. **2** In Step 1 of the Create Cyto Report Template wizard, under Sample Information, select **Show Analysis Settings in the end of report**.	
Select to include QC Metrics	**1** Open an existing report template, or create a new one. See Create a Cyto Report template. **2** In Step 1 of the Create Cyto Report Template wizard, select **Show QC Metrics**.	

Table 12 Tasks for creating or editing Cyto Report templates (continued)

To do this task	Follow these instructions	Comments
Create or edit text boxes	**1** Open an existing report template, or create a new one. See Create a Cyto Report template. **2** In Step 2 of the Create Cyto Report Template wizard, under Text Boxes, select **Text Boxes**. **3** Next to **Number of fields**, select the number of text fields to add to the report, and then click **Add**. **4** Under Name, type a name for the text field (this name is not shown on the report.) **5** Under Value, type the information you want to show in this text box on the report. **6** If you want the text to be editable after the report is generated, select the **Editable** box. **7** To delete a text field from the report, under Delete, click **X**.	• To type information in an editable text box, open the Cyto Report and click **Edit** in the Manage Cyto Report dialog box.

Table 12 Tasks for creating or editing Cyto Report templates (continued)

To do this task	Follow these instructions	Comments
Select the sample results to include	**1** Open an existing report template, or create a new one. See Create a Cyto Report template. **2** In Step 3 of the Create Cyto Report Template wizard, select the boxes for the data you want to show in the report. **3** To load an external track file and show it in the Cyto Report, select **Gene View**, and select **Load Tracks.** • Browse to a tracks file, select it, and click **Open**.	• The choices for how results are displayed in the report are similar to data displayed in the Triage View. For details on the choices in this step of the wizard, see the *CytoGenomics Reference Guide*. • To load a track and show it in a report, an external track file must be available. The track file must be of the same genome build as the sample.
Arrange text boxes and results	**1** Open an existing report template, or create a new one. See Create a Cyto Report template. **2** In Step 4 of the Create Cyto Report Template wizard, a diagram of the report layout is displayed. Drag and drop the boxes so that they appear in the order you want displayed in the report. **3** Click **Save** to save the report using the current report name. OR Click **Save As** and type a new name for the report. **4** Click **OK**.	• For more information on the Edit Cyto Report Template wizard, see the *CytoGenomics Reference Guide*. • You cannot change the default report templates. If you open one of these templates and change it, save the template using another name.

3 Configuring Analysis Methods and Workflows

In Agilent CytoGenomics, *workflows* are used to automate feature extraction and/or analysis of CGH and CGH+SNP microarrays. Each workflow specifies an *analysis method*, that contains a set of actions and parameters for analyzing the data during the workflow. This chapter gives instructions on how to create analysis methods and workflows to automate feature extraction and data analysis.

For information on how to run a workflow, see the *Running CytoGenomics Analyses User Guide*. For information on the dialog boxes and parameter panels that appear when you set up analysis methods and workflows, see the *CytoGenomics Reference Guide*.

Creating an Analysis Method – Overview

An *analysis method* is a set of instructions that tells the program how to analyze the data. When you run the workflow, the program automatically uses the analysis method that is selected in the workflow.

The procedure described in this section shows you how to create and configure an analysis method to analyze CGH and CGH+SNP microarray data.

NOTE To configure analysis methods, you must have the role of Scientist or Administrator.

The following flowchart describes how you create and configure an analysis method.

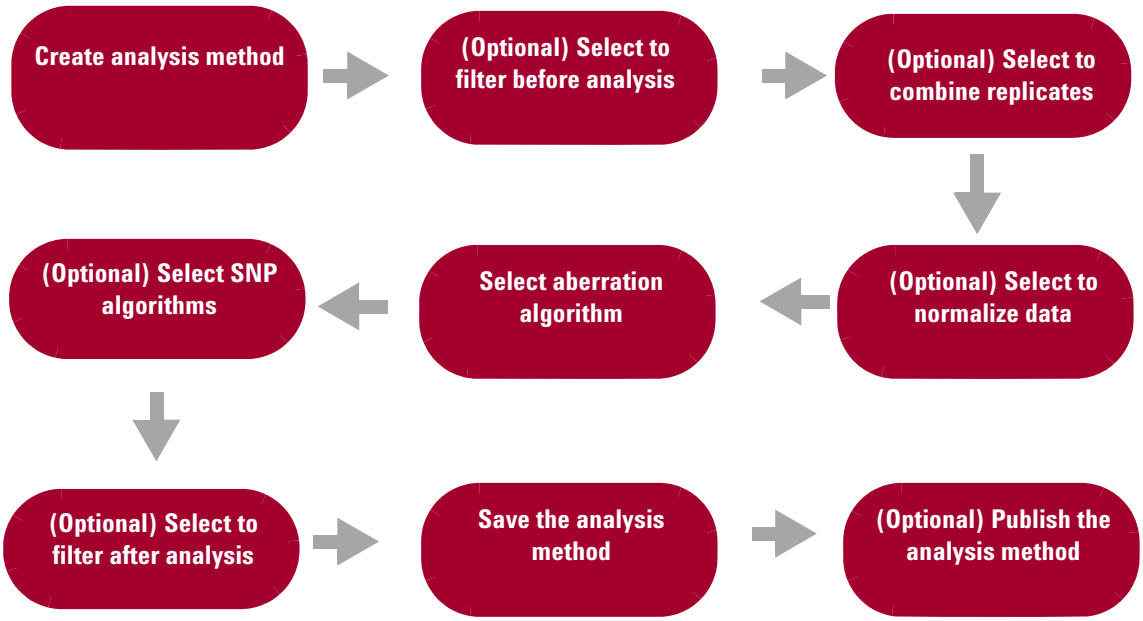


Figure 3 Configuring an analysis method

Tasks for Creating and Configuring an Analysis Method

The tasks in this section show you how to create and configure an analysis method. The program includes two default analysis methods. These default analysis methods cannot be changed.

Table 13 Tasks for creating and configuring an analysis method

To do this task	Follow these instructions	Comments
Create or change an analysis method	**1** In the Config tab, click **Analysis**. **2** Click **Create Analysis Method** to create an analysis method. The Create Analysis Method dialog box appears. **3** Type a name for the analysis method, and then click **OK**. The Analysis method window opens, **4** Under Description, type information to describe the analysis method, if desired. OR Click **View/Edit** for an analysis method you want to change. You cannot change a default analysis method or an analysis method with a status of *Public*. **5** Use the tasks shown in the next steps to set the analysis method parameters.	• The name you type for the analysis method appears in the selection list under Method Name. • You can create more than one analysis method for use in multiple workflows. • To use a default or published analysis method as a "template" for a new analysis method, click **View** to open the analysis method. Then click **Save As** to save the analysis method with a different name. You can then change the parameters and publish the new analysis method. • If you upgraded to Agilent CytoGenomics 2.0 from a previous version, make sure to use the new default analysis method parameters for analysis of constitutional and mosaic samples. Recommended settings are: ◦ Default Design level filter ◦ Centralization (legacy) OFF ◦ GC correction ON ◦ Adjust Diploid peak ON

Table 13 Tasks for creating and configuring an analysis method (continued)

To do this task	Follow these instructions	Comments
(Optional) Select to use filters before analysis	**1** In the Analysis method dialog box, under Filter Before Analysis, select one of the check boxes, **Design Level Filter,** Feature Level Filter, or **Array Level Filter**. A parameter panel appears for the selected filter type. **2** In the parameter panel, • Select the default filter OR • Select a defined filter from the list OR • Click **New** and create a filter	• You can mark more than one filter to use in the analysis method. • When you apply a Design Level probe filter, you include or exclude probes, based on design filter conditions. • When you apply an Array filter, microarrays that fail the attribute filter criteria are not included in the evaluation. • When you apply a Feature Level filter, features from the array that fail the criteria are not included in the evaluation. • For details on the parameter panels, see the *CytoGenomics Reference Guide*.
(Optional) Select to combine replicates	• To combine replicates, in the Analysis method dialog box, under Combining Replicates, select **Intra-Array Replicates**.	• If your arrays contain probes that are replicated in the array, this option combines them to increase the confidence of your analysis. When the program combines replicates, it selects probes with common probe names, and calculates a weighted average of their values to create a single point.
(Optional) Normalize the data	• To normalize the data so that zero represents the most common ploidy, select the **Centralization (legacy)** check box. • To correct for baseline waviness by performing a regression fit to GC content in the genomic region flanking the probes, select **GC Correction**. • To normalize the data so that the log ratios of the diploid regions are centered around zero, select **Diploid Peak Centralization**.	• Diploid Peak Centralization and GC Correction are recommended for CGH analysis, SNP analysis, and LOH (Lack/Loss of heterozygosity) analysis. • In order for the program to display copy number peak assignments for CGH arrays in the CGH&SNP Fit window, make sure Diploid Peak Centralization normalization is selected.

Table 13 Tasks for creating and configuring an analysis method (continued)

To do this task	Follow these instructions	Comments
Set up the aberration algorithm	**1** In the Analysis method dialog box, under Aberration, select one of the aberration algorithms. **2** Type or change parameters.	• ADM1 and 2 can use Fuzzy Zero to make the results more reliable by taking into account the global error across the chromosomes. • If the Nesting Filter value is set to zero, it filters out all nested aberrations. By default, no filter is applied. • For SNP and LOH analyses, Z-score is not allowed. • See the "Statistical Algorithms" chapter of the *CytoGenomics Reference Guide* for information on the aberration algorithm calculations and suggested thresholds.
(For CGH+SNP) Setup the SNP algorithms	**1** Select **SNP Copy Number** and (optionally) **LOH** for analysis of CGH+SNP data. **2** Type or change the parameters. • **SNP Copy Number** – detects allele-specific copy numbers for SNP probes. • **LOH** – Detects regions that show loss or lack of heterozygosity.	• To select **SNP Copy Number,** select an aberration algorithm (other than Z-score). • To select **LOH**, first select **SNP Copy Number**. • For SNP Copy Number and LOH analyses, GC Correction and Diploid Peak Centralization are recommended.
(Optional) Select to filter after the analysis	**1** In the Analysis method dialog box, under Filter After Analysis, select **Aberration Filter**. **2** Select the **DefaultAberrationFilter** or another filter from the list, or create a new one.	• Default aberration filter: 3 probes, 0.25 minimum log ratio. • Suggested filter for mosaic sample analysis: 5 probes, 0.15 minimum average log ratio. • For more information on this parameter panel, see the *CytoGenomics Reference Guide.*

Table 13 Tasks for creating and configuring an analysis method (continued)

To do this task	Follow these instructions	Comments
(Optional) Select to include or exclude calls within a genomic boundary	**1** In the Analysis method dialog box, under Genomic Boundary, select **Genomic Boundary.** **2** In the Genomic Boundary Parameter Panel, under Select Track, click the arrow and select a track to use. **3** Select **Include** or **Exclude**.	• When you select **Include**, detection of aberration calls is limited to the genomic boundaries of the selected track. • When you select **Exclude**, detection of aberration calls within the selected track are excluded from the analysis.
Save the analysis method	• In the Analysis method dialog box, click **Save** or **Save And Close**. OR **1** To save the analysis method with a new name, click **Save As.** The Create Analysis Method dialog box appears. **2** In the Create Analysis Method dialog box, type the name to use for the analysis method. **3** Click **OK**.	• After you *save* an analysis method, you can go back later and change it. After you *publish* an analysis method, it cannot be changed.
(Optional) Delete an analysis method	**1** In the Config tab, under Workflow, click **Analysis**. **2** Find the row with the analysis method you want to delete. Under Action, click **Delete**.	• You cannot delete an analysis method with a Status of *Public*. • You cannot delete an analysis method that is part of a workflow.
(Optional) Publish an analysis method	**1** In the Config tab, under Workflow, click **Analysis**. **2** Click **Publish**. **3** Click **Close** to close the dialog box.	• When you publish an analysis method, its Status becomes *Public* and it becomes available for anyone to use in a workflow. • After you publish an analysis method, it cannot be changed.

Table 13 Tasks for creating and configuring an analysis method (continued)

To do this task	Follow these instructions	Comments
(Optional) Export an analysis method	**1** In the Config tab, click **Analysis**. **2** Find the row with the analysis method you want to delete. Under Action, click **Export**. The Export dialog box appears. **3** In the Export dialog box, browse to the location where you want to save the file. **4** Next to **File name**, type a name for the file. **5** Click **Export**.	
(Optional) Import an analysis method	**1** In the Config tab, click **Analysis**. **2** At the bottom of the window, click **Import Analysis Method**. The Import Analysis Method(s) dialog box appears. **3** Browse to and select the analysis method file (or more than one analysis method files), and click **Import**. The Import dialog box appears. **4** Under Import, select the box next to the analysis methods you want to import. **5** Click **Ok.** The Import Status dialog box appears. **6** Click **Ok**.	

Setting Up Workflows – Overview

Agilent CytoGenomics lets you set up a workflow that performs analysis for a set of samples.

A workflow starts with one of the following input types:

- Image files that are extracted with Agilent Feature Extraction for CytoGenomics software (FE) during the workflow run to produce a Feature Extraction output file and a QC Report that contains sample QC information
- Extracted Feature Extraction output files that are imported during the workflow process
- Files that were previously imported into the database

The workflow then runs data analysis with CGH and/or CGH+SNP algorithms and generates one or more selected reports.

You can also run workflows in fully automated, on-time mode, where samples are extracted and analyzed as soon as they are scanned. For information on running workflows in manual or auto-processing mode, see the *Running CytoGenomics Analyses User Guide*.

The following flow chart describes the general steps for creating or editing a workflow.

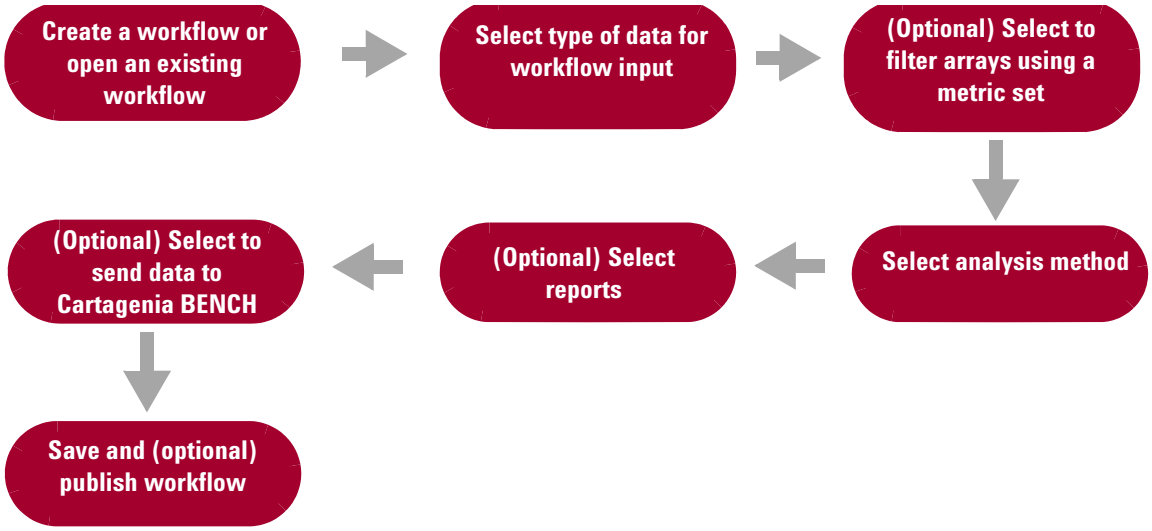


Figure 4 Steps for creating or changing a workflow

Creating or Changing a Workflow

The following table gives instructions on how to create or change a workflow. The program provides two default workflows. These workflows cannot be changed.

Table 14 Tasks for creating or changing a workflow

To do this task	Follow these instructions	Comments
Create a workflow or change an existing workflow	1 In the Config tab, click **Workflow**. 2 Click **Create Workflow**. The Create Workflow dialog box appears. 3 Next to **Enter Workflow Name**, type a name for the new workflow, and then click **OK**. 4 The Workflow dialog box appears. OR **To change an existing workflow** • Click **View/Edit** for the workflow you want to change. You can only change a workflow with a Status of *Private.* Use the following tasks to configure the workflow.	• Workflows that have been published have a Status of *Public* and cannot be changed. • To use a published workflow as a "template" for a new workflow, click **View** to open the workflow. Then click **Save As** to save the workflow with a different name. You can then change the parameters and publish the new workflow.
Select type of data for workflow input	• In the Workflow dialog box, under Input, select the type of data to use as the input for your workflow. Choices include: ◦ Import FE Files – files for images that were previously feature extracted (.txt format) ◦ Select Imported Data – files that are already in the database ◦ Image Files – TIFF image files created by a scanner	• For manual workflows, you select the actual files to use when you run the workflow. • For auto-processing workflows, the input file type must be Image Files. • For auto-processing workflows, the program uses input files from the Tiff Image Input Directory configured in the Auto-Processing Settings. For information on how to configure auto-processing workflow settings, see "To configure auto-processing settings" on page 14.

Table 14 Tasks for creating or changing a workflow (continued)

To do this task	Follow these instructions	Comments
(Optional for image files) Review and change FE parameters	**1** In the Workflow dialog box, under Input, select Image Files to use as the input for your workflow. **2** Click **FE default parameters being used**. The Feature Extraction Properties dialog box appears. **3** In the Default Value column for the selected property, click the arrow and select a new property. **1** Click **Save**.	• **Important**: Changes to the FE default parameters are applied to *all* workflows where image files are the input.
(Optional) Select to filter arrays using a metric set filter	**1** In the Workflow dialog box, under Metric Evaluation, select **Metric Set Array Filter** to filter out arrays based on a filter created from a metric set. **2** In the Metric Set Filter Parameter Panel, under Edit Metric Set Filter, click the arrow next to **Name** and select a metric set filter to use for filtering the arrays before analysis. OR **1** In the **Metric Set Filter Parameter Panel**, click **New**. The Input dialog box appears. **2** Type a name for the new metric set filter, and then click **OK**. **3** Add and configure conditions for the metric set filter. **4** Click **Save**.	

Table 14 Tasks for creating or changing a workflow (continued)

To do this task	Follow these instructions	Comments
Select analysis method	**1** In the Workflow dialog box, under Analysis, select **Run Analysis.** **2** In the Run Analysis Application parameter panel, next to **Analysis Method**, select the analysis method to use for the workflow. OR Click **New** to create an analysis method for the workflow or **View/Edit** to change an existing analysis method. For instructions on how to create or change an analysis method, see "Creating an Analysis Method – Overview" on page 40**.**	
Select Reports	• Select any of the following Reports: ◦ **CGH Aberration Report** – Gives overall deletion and amplification tabular results, along with *p*-values (log 10). Select Probe Based or Interval Based report type, or both. ◦ **Cyto Report** – Gives deletion and amplification tabular and graphical results with all the parameter settings, according to the selected cyto report template. You can select to use the default Cyto Report template, or you can select a custom report template. ◦ **SNP Genotype Report** – Reports genotype and *p*-values for each SNP position that the SNP probes on the microarray cover. ◦ **Aberration & LOH Report** – Reports amplification, deletion, and LOH aberrations on a per interval basis.	• For information on the options in the parameter panel for each type of report, see the *CytoGenomics Reference Guide*. • Cyto reports are generated when you run the workflow, and are saved in the job folder in the Output Directory specified in the default settings. See "Configuring Default Settings" on page 9. • Cyto reports are saved in .PDF and .XML formats. • Select SNP Genotype Report or Aberration & LOH Report only for workflows with analysis methods that include SNP Copy Number and LOH analysis. • For information on how to create or change a report template, see Chapter 2, "Creating and Editing Cyto Report Templates".

Table 14 Tasks for creating or changing a workflow (continued)

To do this task	Follow these instructions	Comments
(Optional) Select to send data to	• Under Send Data, select **Cartagenia BENCH**.	• To send data to Cartagenia BENCH, set your Cartagenia BENCH user name and password in Agilent CytoGenomics. See "Setting Up Data Transfer for Cartagenia BENCH" on page 28. • Only the reports selected in the Cartagenia BENCH configuration are sent to Cartagenia BENCH. These reports must match the reports selected in the workflow. • As of release date, Cartagenia BENCH supports data transfer for hg18 genome build only. Check with Cartagenia for updated information on what builds are supported.
Save the workflow	• Click **Save** or **Save And Close** to save the workflow using the current workflow name. OR **1** Click **Save As**. The Create Workflow dialog box appears. **2** Type a new name for the workflow, and then click **OK**. **3** Click **Close** to close the workflow and keep it private for later changes.	• Only the owner of a workflow with a status of *Private* can change or run it.
(Optional) Publish the workflow	• In the Workflow dialog box, click **Publish** to publish the workflow.	• When you publish a workflow, its Status is set to *Public* and it can no longer be changed. Any user can run a *Public* workflow. Only the owner of a workflow with a status of *Private* can change or run it.

Table 14 Tasks for creating or changing a workflow (continued)

To do this task	Follow these instructions	Comments
(Optional) Import a workflow	**1** In the Config tab, click **Workflow** and then click **Import Workflow.** The Import Workflow(s) dialog box appears. **2** Browse to and select the workflow file (or more than one workflow files), and click **Import**. The Import dialog box appears. **3** Under Import, select the box next to the workflows you want to import. **4** Click **Ok.** The Import Status dialog box appears. **5** Click **Ok**.	• You can only import workflow files that were exported using Agilent CytoGenomics.
(Optional) Export a workflow	**1** In the Config tab, click **Workflow**. Under Action, click **Export** for the workflow you want to export. **2** In the Export dialog box, browse to a location where you want to save your workflow file. Type a name to use for the exported file. **3** Click **Export**. The selected workflow is saved in the specified location.	• Exported workflows can be imported to Agilent CytoGenomics at a later time, or imported to a different Agilent CytoGenomics server.

4
Displaying and Organizing Samples

In Agilent CytoGenomics, the Content tab is used to manage samples and display Feature Extraction content. This chapter gives instructions on how to manage samples and their attributes, and display Feature Extraction protocols and information about extracted samples.

Sample Management – Overview

The Sample Manager area in the Content tab is used to display and manage samples and their attributes.

Sample Attributes

Sample attributes are used in various ways throughout Agilent CytoGenomics. They are a powerful way to organize, annotate, and search your samples. Some uses for sample attributes include:

- Use Green Sample (or Red, if polarity is flipped) to associate a genotype reference with a CGH+SNP (Comparative Genomic Hybridization with Single Nucleotide Polymorphism) microarray.
- Use a combination of up to three sample attributes to name folders for workflow output and sample cyto reports.
- Create queries to search for samples with common sample attributes. Or, create dynamic tracks for aberrations that occur in samples with common attributes.
- In the Quality tab, create queries and QC Charts based on selected attributes.

When you process a set of samples in interactive mode, define sample attributes for each sample. The workflow adds one row to the Sample Manager table in the Content tab for each sample in the workflow. You can later use sample attributes to find common samples.

NOTE After the workflow starts, you cannot change the *required* sample attributes: Array ID, Global Display Name, Green Sample, Red Sample, Polarity.

Array IDs

To run a workflow in auto-processing mode, first add an Array ID for each sample to the Sample Manager table. Then define the sample attributes for each. Otherwise, save a valid sample attribute file (SAF) in the configured SAF Input Directory for auto-processing workflows. A valid SAF file contains one row for each sample in the workflow. In order for

the auto-processing workflow to complete successfully, make sure that the required sample attributes for each array are present in the Sample Manager table or in the SAF file.

NOTE

When you run a *manual* workflow, a row for each sample is automatically added to the Sample Manager table. Sample attributes are attributes you set when you run the workflow. Alternatively, you can create a row in the Sample Manager table before you run a workflow, either manually, or by importing a SAF. This row is used by the next workflow that contains that sample Array ID.

For a description of each part of this module, including all of the dialog boxes that can appear when you click buttons and other elements, see the *CytoGenomics Reference Guide*.

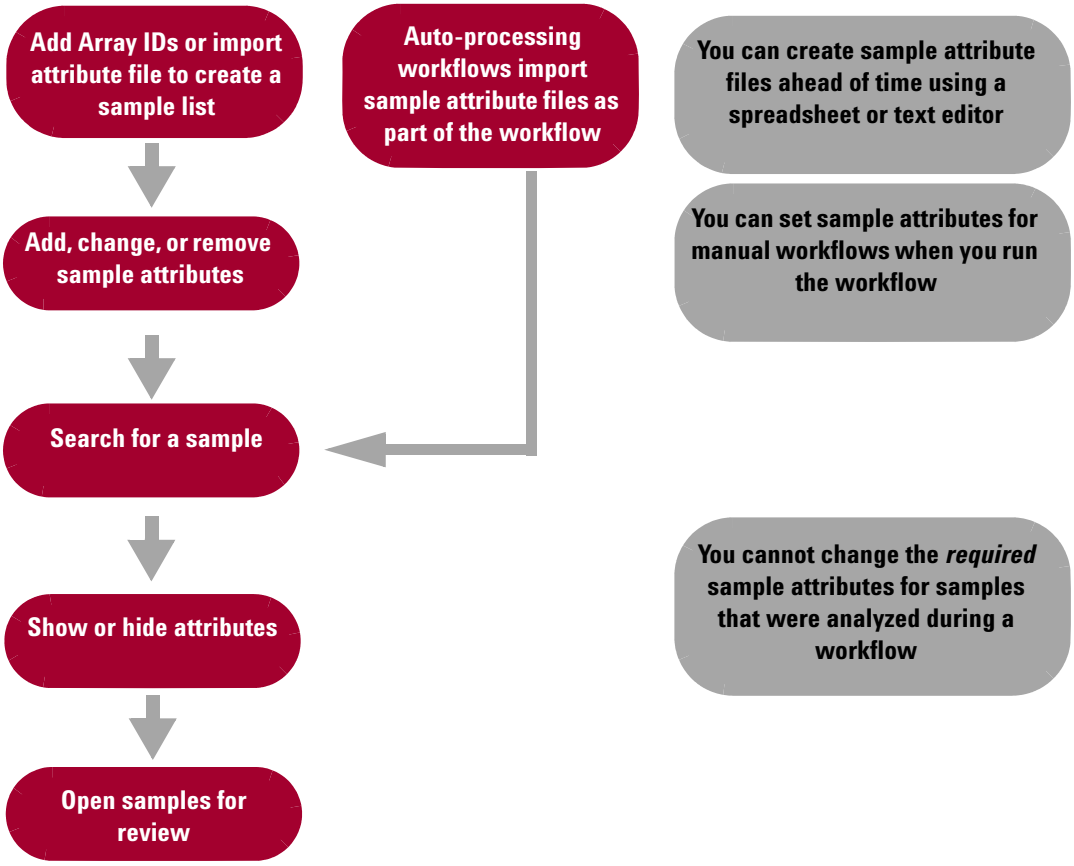


Figure 5 Managing samples overview

Sample management terminology

The terms in the following table are used in the Content tab, as well as in other areas of Agilent CytoGenomics.

Term	Definition
Array ID	The unique identifier for each array on a microarray slide. You cannot change the Array ID.
Global Display Name	By default, the name of the extraction. This name is used to identify the microarray sample throughout the program. You can change the Global Display Name before running a workflow.
FE File	A .txt file that contains the Feature Extraction data for the microarray.
Attribute	A piece of information about the specific microarray; for example, hybridization temperature or polarity.
Attribute file	A file that contains attributes for one or more microarrays. Attribute files are created and saved as .xls or .txt files by the user, or they are created within Agilent CytoGenomics and then exported.

About the Sample Manager table

When you extract a microarray or import an extracted data file (using an Agilent CytoGenomics workflow), the Array ID appears as a row in the Sample Manager table. The array also appears in the Sample Manager Array Data Navigator, under the associated design in the Imported FE Data folder. These rows appear gray in the table. If you add an Array ID manually or import a Sample Attribute File (SAF), the row appears white. After the sample is analyzed in a workflow, the row turns gray. Arrays that are imported as FE Stats and Parameters from the Quality tab are displayed as white rows in the Array Data Navigator, in the Only Stats and Params folder.

You can also add an array ID to the Sample Manager table and assign attributes *before* you run a workflow. Array IDs that you add manually appear as white rows in the Sample Manager table. These arrays are also displayed in the Array Data Navigator, under the folder name you created when you added the Array ID.

After you organize your samples and define their attributes, you run a workflow. The workflow converts the scanned images of your arrays automatically into feature extracted data, or imports the already-extracted text file, and then analyzes the data. The attributes that are assigned for each Array ID are carried with the Array ID during the analysis using Agilent CytoGenomics.

NOTE After a sample is analyzed with a workflow, you cannot change its *required* sample attributes: Array ID, Global Display Name, Green Sample, Red Sample, Polarity.

About sample attribute files

A sample attribute file (SAF) contains identification information (Array ID and Global Display Name) and attributes for one or more microarray samples. A sample attribute file is a convenient way to add a list of samples to the program.

Array attributes can vary, but sample attribute files must follow these guidelines:

- The first line of the file contains the names of array attributes, separated by tabs.
- The rest of the lines of the file contain the values of each attribute, one line per array. The values must be in the same order as the attributes in the first line of the file.
- The file must contain the following mandatory attributes:
  - Array ID
  - Global Display Name
  - Green Sample
  - Red Sample (for 2-color arrays)
  - Polarity (for 2-color arrays)

Here is an example:

Array ID	Global Display Name	Green Sample	Red Sample	Polarity
252192413168_1_2	252192413168_1_2	European Male (NA12891_v1)	NA04592	1
252192413168_1_1	252192413168_1_1	European Male (NA12891_v1)	NA04592	1
252192413168_1_3	252192413168_1_3	European Male (NA12891_v1)	NA04592	1
252192413168_1_4	252192413168_1_4	European Male (NA12891_v1)	NA04592	1
252192413168_2_1	252192413168_2_1	European Male (NA12891_v1)	NA04592	1
252192413168_2_2	252192413168_2_2	European Male (NA12891_v1)	NA04592	1
252192413168_2_3	252192413168_2_3	European Male (NA12891_v1)	NA04592	1
252192413168_2_4	252192413168_2_4	European Male (NA12891_v1)	NA04592	1

Figure 6 Example of a sample attribute file

You can create sample attribute files with a text editor or spreadsheet program. Save the sample attribute files as tab-delimited text files or .xls files.

NOTE If you create an attribute file using Microsoft® Excel, save it as an Excel 97-2003 workbook .xls file or a tab-delimited .txt file only.

About Global Display Names

The Global Display Name for a microarray is, by default, the name of the extraction. This name is displayed throughout Agilent CytoGenomics for the microarray; for example, in the Triage view, and in reports. To make it easier to identify and organize your samples, you can change the global display names of samples before you run a manual workflow. Or, change the global display names in the sample attribute file (SAF) before running an auto-processing workflow. Each Global Display Name must be unique.

NOTE After a sample is analyzed with a workflow, you cannot change its *required* sample attributes: Array ID, Global Display Name, Green Sample, Red Sample, Polarity.

Tasks for Managing Samples and Attributes

The tasks in this section show you how to manage samples and their attributes. It also explains how to import design files and genotype reference files for CGH+SNP analysis.

To manage samples and their attributes

Table 15 Tasks for managing samples and their attributes

To do this task	Follow these instructions	Comments
Add an array ID	**1** In the Content tab, in the Navigator, click **Sample Manager**. The Sample Search pane and Sample Manager table are displayed. **2** Click **Add Array ID**. The Add Array ID to Data Folder dialog box appears. **3** Select the folder, or create a folder, where you want to add an Array ID. **4** Click **OK**. A row is added to the array list. **5** In the blank fields, type the attributes for the microarray and then click **Save Changes**.	• You can add Array IDs one at a time, or you can import a sample attribute file to add multiple Array IDs. See Import a sample attribute file. • Required attributes include Array ID, Global Display Name (must be unique and not blank), Green Sample, Red Sample (for 2-color array), Polarity (for 2-color array). Array ID rows that do not contain all required attributes are not added. • Arrays that are imported or extracted using a workflow are automatically added to the Sample Manager table.

Table 15 Tasks for managing samples and their attributes (continued)

To do this task	Follow these instructions	Comments
Import a sample attribute file	**1** In the Content tab, select **Sample Manager**. **2** On the command ribbon, under Sample Attribute, click the green arrow.  The Import Attribute Files dialog box opens. **3** Browse to a folder location and click to select a file. OR Type the name of the file to import. **4** Click **Open**. The Attribute File Importer - Map Column Headers dialog box opens. **5** For each header field, select a sample attribute to which it corresponds. See "About sample attribute files" on page 58. **6** (Optional) To set the mapping of this sample attribute file as the default for sample attribute files imported as part of an auto-processing workflow, select **Set to be Automated Import format**. **7** Click **Import**. The data in the file is imported and displayed in the Sample Manager table.	• Sample attribute files must be either tab-delimited .txt files or .xls files, and cannot be formulas (only simple text). If you want to import an .xlsx spreadsheet file, first save it as a tab-delimited .txt or .xls file (Excel 97-2003 Workbook format.) • Sample attribute files must contain the following required attributes for each Array: Array ID, Global Display Name (must be unique and not blank), Green Sample, Red Sample (for 2-color array), Polarity (for 2-color array). Array ID rows that do not contain all required attributes are skipped and not imported. • Sample attribute files contain only Array IDs and the associated sample attributes. They do not contain feature data from the scanned images of microarrays. • Sample attribute files for auto-processing workflows are imported as part of the workflow process.

Table 15 Tasks for managing samples and their attributes (continued)

To do this task	Follow these instructions	Comments
Export a sample attribute file	**1** In the Content tab, select **Sample Manager**. **2** On the command ribbon, under Sample Attribute, click the Export Attribute File yellow arrow.  The Export Attribute Files dialog box appears, that contains an Array tab and an Attribute tab. **3** In the Array tab, select the sample containing the microarrays whose attributes you want to export. **4** In the Array list, select the sample arrays whose attributes you want to export. Click one or more of the arrow buttons to move the arrays to the Selected Array List. **5** Click **Next**. The Attribute tab is displayed. **6** Select the Attributes to export and then click **OK**. The Export dialog box opens. **7** In the Export dialog box, find the folder to save the exported attribute file and type a name for the file. **8** Click **Export** to save it.	

Table 15 Tasks for managing samples and their attributes (continued)

To do this task	Follow these instructions	Comments
Add, change, or remove sample attributes	**To add a microarray attribute** **1** In the Content tab, in the Sample Manager window, click **Add/Edit Attribute.** The Attributes dialog box appears. **2** At the bottom of the dialog box, click **New** (*not* New Value). The Input dialog box appears. **3** Type a name for the attribute of up to 50 characters in length, and then click **OK.** The name appears in the Attributes dialog box. **4** Edit the details of the attribute. See the *CytoGenomics Reference Guide* for a description of the available options and commands. **5** Click **Update.** **6** Click **Close.** The attribute is now available for any sample in the program. **To change an existing attribute** **1** In the Content tab, in the Sample Manager window, click **Add/Edit Attribute.** The Attributes dialog box appears. **2** Click the **Name** arrow and select the attribute you want to change. **3** Edit the details of the attribute. **4** Click **Update.** For details about editing options, see the *CytoGenomics Reference Guide.* **5** Click **Close.**	• Deleting an attribute permanently removes the attribute and its associated values from Agilent CytoGenomics. To restore the attribute, create it again.

Table 15 Tasks for managing samples and their attributes (continued)

To do this task	Follow these instructions	Comments
	To remove an attribute **1** In the Content tab, in the Sample Manager window, click **Add/Edit Attribute.** The Attributes dialog box appears. **2** Click the **Name** arrow and select the attribute you want to delete. **3** Click **Delete** (*Not* Delete Value). You are asked to confirm that you want to delete the attribute. **4** Click **Yes** to delete the attribute permanently. **5** Click **Close.**	• When you delete an attribute, it is no longer available, even if samples exist in the database with values for that attribute. To restore the attribute, create it again. The values saved for samples in the database are then restored.
Show or hide attributes	**1** In the Content tab, at the bottom of the Sample Manager table, click **Show/Hide Attributes**. The Show/Hide Columns dialog box appears. **2** Under Show in Table or Show in Triage View, select the check box next to each attribute you want to display in the Sample Manager table or in Triage View. **3** Click again to clear a box. **4** When all desired attributes are selected, click **Save** to update the Sample Manager table and save the selections. **5** Click **Close** to close the Show/Hide Columns dialog box.	• Columns that show by default are grayed-out. • You can select to display sample attributes in the Sample Manager table, in the Triage View, or both. • Select the box next to **Show in Table** or **Show in Triage View** to select all attributes. Clear the box to clear all selections.

Table 15 Tasks for managing samples and their attributes (continued)

To do this task	Follow these instructions	Comments
Show or hide FE parameters	**1** In the Content tab, at the bottom of the Sample Manager table, click **Show/Hide Attributes**. The Show/Hide Columns dialog box appears. **2** Click the **FE Params** tab. **3** Under Show in Table, select the box next to each Feature Extraction parameter you want to display in the Sample Manager Table. **4** Click **Save** to update the table and save the selections. **5** Click **Close** to close the dialog box.	• For information on Feature Extraction parameters, see the *FE for CytoGenomics Reference Guide,* located in the CytoGenomics_Client\Feature Extraction\Docs folder of your Agilent CytoGenomics installation folder.
Show or hide FE statistics	**1** In the Content tab, at the bottom of the Sample Manager table, click **Show/Hide Attributes**. The Show/Hide Columns dialog box appears. **2** Click the **FE Stats** tab. **3** Under Show in Table, select the box next to each Feature Extraction statistic you want to display in the Sample Manager Table. **4** Click **Save** to update the table and save the selections. **5** Click **Close** to close the dialog box.	• For information on Feature Extraction statistics, see the *FE for CytoGenomics Reference Guide,* located in the \ **CytoGenomics_Client\Feature Extraction\Docs** folder of your Agilent CytoGenomics installation folder.

Table 15 Tasks for managing samples and their attributes (continued)

To do this task	Follow these instructions	Comments
Show or hide the FE QC Report, CGH&SNP Fit, and CGH&SNP QC Flag columns	**1** In the Content tab, at the bottom of the Sample Manager table, click **Show/Hide Attributes**. The Show/Hide Columns dialog box appears. **2** Click to select one or more of the columns: • Show FE QC report column • Show CGH&SNP Fit column • Show CGH&SNP QC Flag column **3** Click **Save**. **4** Click **Close** to close the dialog box.	• When the FE QC Report column is displayed, "FE QC Report" appears whenever an FE QC Report is available for a sample. Click **FE QC Report** to display the FE QC Report for a sample. • For information on the contents of FE QC Reports, see the *FE for CytoGenomics Reference Guide,* located in the \ **CytoGenomics_Client\Feature Extraction\Docs** folder of your Agilent CytoGenomics installation folder. • Selections you make in the Show/Hide Attributes dialog box apply to all sample tables that display sample attributes (Sample Manager, Review, and Describe Samples).
Edit attribute values	**1** In the Content tab, click **Sample Manager.** **2** In the Sample Manager table, click a microarray to select it. **3** Double-click the sample attribute you want to change for this microarray. The selected attribute field becomes active. **4** Type the attribute value or click the arrow to select the value from a list. **5** When finished, click **Save Changes**.	• Before you run the workflow, you can change all attributes for Array IDs within the Content tab. • After you run the workflow, you cannot change *required* sample attributes for microarrays. (Array ID, Global Display Name, Green Sample, Red Sample, Polarity)

Table 15 Tasks for managing samples and their attributes (continued)

To do this task	Follow these instructions	Comments
Associate a genotype reference to a microarray	**1** In the Content tab, click **Sample Manager.** **2** Click on **Green Sample** (or **Red Sample**, if polarity is flipped) in the array row. The box becomes active and a selection arrow appears. **3** Click the selection arrow, and select a genotype reference sample from the list of genotype reference samples in the database. OR Type the name of the genotype reference sample. **4** At the bottom of the window, click **Save Changes**.	• Before you run the workflow, you can change all attributes for Array IDs within the Content tab. • After you run the workflow, you cannot change *required* sample attributes for microarrays. (Array ID, Global Display Name, Green Sample, Red Sample, Polarity) • To perform CGH+SNP analysis in a workflow, first select a reference genotype sample for the CGH+SNP array. • The program comes with a set of preloaded genotype references. You can also import your own genotype reference file. See "Import a genotype reference file" on page 72.
Remove an Array Data folder	**1** In the Content tab, click **Sample Manager.** **2** In the Sample Manager Array Data Navigator, right-click the data folder you want to delete. **3** Select **Delete**. The selected data folder is deleted from the Sample Manager.	• A data folder is created when a user adds an Array ID manually within the Sample Manager. • After data is imported into a data folder, you cannot remove the data folder. • You cannot remove a data folder created by another user.

Table 15 Tasks for managing samples and their attributes (continued)

To do this task	Follow these instructions	Comments
Search for a sample	1 In the Content tab, select **Sample Manager**. 2 Under Sample Search, click the arrow next to **Search in column**, and select an array attribute to search for. 3 Click the arrow next to **Value**, and select the value for the selected attribute to search for. 4 Click **Search**. The Sample Manager table displays all of the arrays that meet the selected criterion. 5 To reset the table contents to the previously displayed values, click **Reset**.	

To examine sample results

From the Sample Manager, you search for and review sample results in various ways. These tasks are described in the following table.

Table 16 Tasks for examining sample results

To do this task	Follow these instructions	Comments
Open the FE QC Report	• In a sample row of the Sample Manager table, click **FE QC Report**. The FE QC Report .pdf file opens.	• The FE QC Report is the report generated by the Feature Extraction program when the sample image was feature extracted. For more information, see the *Feature Extraction for CytoGenomics Reference Guide.* • Only samples that were feature extracted using Agilent CytoGenomics have a FE QC Report available.
Display CGH&SNP Fit Plot and metrics	1 In a sample row of the Sample Manager table, under CGH&SNP Fit, click **View**. 2 If the sample has more than one analysis result, the Analyzed Sample dialog box appears. Select the sample you want to view, and then click **Ok**. The CGH&SNP Fit plots window opens.	• Although you can open the CGH&SNP Fit Plot for any sample, only samples that were analyzed using the SNP Copy Number algorithm have valid SNP data. • For more information on the CGH&SNP Fit Plot window, see the *CytoGenomics Reference Guide*.
Open a sample in Triage View	1 For a sample in the Sample Manager table, click its **Status**. 2 If the sample has more than one analysis result, the Analyzed Sample dialog box appears. Select the sample you want to view, and then click **Ok**. The sample opens in Triage View.	• Only samples that were successfully analyzed within Agilent CytoGenomics open when you click their status. • For more information on the Triage View window, see the *Running CytoGenomics Analyses User Guide* and the *CytoGenomics Reference Guide*.

To import designs

To run a workflow Feature Extract and/or analyze extracted data, the design that matches the microarray must be present in the Agilent CytoGenomics database. You can import design files automatically during manual workflows that use extracted Feature Extraction files. Or, you can import designs manually from the Content tab, as described in the following table.

NOTE If a matching design is not already present in the database, auto-processing workflows and manual workflows that extract image files fail.

Table 17 Tasks for importing designs

To do this task	Follow these instructions	Comments
Import a Design file	**1** In the Content tab, click **Sample Manager**. **2** On the Sample Manager ribbon, under Design File, click the green import arrow.  The Import Design Files dialog box appears. **3** Browse to the design file to import, select it, and then click **Open**. The Import GEML design files dialog box opens. The program validates the selected files, and the Import GEML Design Files dialog box appears. • If a design file passes validation, the Status column shows **Valid** in green. • If the design is a newer version of an existing design, the Status shows Update in green. • If a design file exists in the database, the Status shows **Overwrite** in yellow. (Except for different builds of the same design, which are added as a separate design.) If you continue, the existing design is replaced with the imported design. • If a design file fails validation, **Corrupt** appears in the Status column beside it, and the program does not import the file. To remove the corrupt design from the list, click its **Remove** button  . **4** For CGH+SNP designs, mark the **Import SNP Probes** box. **5** Click **Start Import.**	• The program comes with a preloaded suite of designs. Import design files if the designs you use are not already in the database. You cannot run a workflow unless the design is already in the database. For a list of the preloaded designs, see the Content Tab Window topic in the *CytoGenomics Reference Guide*.

To import genotype reference files

To perform CGH+SNP analysis, first associate a reference genotype sample in the database to the CGH+SNP array. To associate a genotype reference, select it in the Green Sample or Red Sample (if flipped) sample attribute. For more information on selecting genotype reference files, see the *Running CytoGenomics Analyses User Guide*. A genotype reference file contains reference genotypes for one or more genotype reference samples. If the genotype reference is not in the database, you can import the genotype reference file from the Content tab. For information on the format requirements for a genotype reference file, see the *CytoGenomics Reference Guide*.

Table 18 Task for importing and displaying genotype reference files

To do this task	Follow these instructions	Comments
Import a genotype reference file	**1** In the Content tab command ribbon, under Ref. Sample, click the green import arrow.  The Import Genotype Reference Files dialog box appears. **2** Browse to a location and select the genotype reference file to import. **3** Click **Open**. The Genotype Reference Importer dialog box appears. **4** Click **OK**.	• After you import the genotype reference file, the reference samples it contains are available in the database and you can associate them with CGH+SNP samples. • For information on how to create a custom genotype reference file, see the *Running CytoGenomics Analyses User Guide*.
Display details of a genotype reference file	**1** In the Content tab, locate the genotype reference file in the Genotypes pane. **2** Right-click the genotype reference file, and select **View Details**. The Genotype Reference Details dialog box opens. **3** To close the dialog box, click **Close**.	

Displaying Feature Extraction Content

The Content tab of Agilent CytoGenomics lets you display Feature Extraction content such as protocols and information about extractions in the database.

The following table shows the capabilities that you have within the Content tab to display Feature Extraction information.

Table 19 Tasks available in the Feature Extraction module of the Content tab

To do this task	Follow these instructions	Comments
Display a Feature Extraction protocol	1 At the bottom of the Navigator of the Content tab, click **Feature Extraction**. 2 In the FE Protocols pane of the Navigator, double-click the protocol you want to display. The protocol is shown in the FE tab.	• Protocols are sets of instructions used during a feature extraction. CGH protocols include the following steps: ◦ Place grid ◦ Optimize grid fit ◦ Find Spots ◦ Flag outliers ◦ Compute background, bias, and error ◦ Correct dye biases ◦ Compute ratios ◦ Calculate metrics ◦ Generate results • You cannot change the parameters shown for a protocol. To create protocols, use the Feature Extraction program that was installed with Agilent CytoGenomics.
Use quick-display links to display extractions	1 At the bottom of the Navigator of the Content tab, click **Feature Extraction**. 2 In the FE Data pane of the Navigator, double-click any of the quick-display links (Extraction by barcode, or Extraction by date, for example.) The extractions that match the selection are displayed in the table.	• Information about the extracted sample, such as extraction date and FE QC Report location, are shown in the table – not the results.

Table 19 Tasks available in the Feature Extraction module of the Content tab (continued)

To do this task	Follow these instructions	Comments
Open a Feature Extraction QC Report	**1** At the bottom of the Navigator of the Content tab, click **Feature Extraction**. **2** In the FE Data pane of the Navigator, search for the extraction you want to display. **3** In the FE QC Report Name column, double-click the name of the FE QC Report. The Feature Extraction QC Report for the selected sample is displayed.	• For information on what the Feature Extraction QC Report contains, see the *Feature Extraction for CytoGenomics Reference Guide*.
Start the Feature Extraction program	**1** In the Navigator of the Content tab, click **Feature Extraction**. The Feature Extraction navigator appears. **2** In the command ribbon, under Launch, click **Launch Feature Extraction**.  The Feature Extraction program starts.	• The Feature Extraction software is a separate program. You can use it to extract images, and display and create protocols, metric sets, and grid files. For information about the capabilities of Feature Extraction, see the User Guides provided in the Help menu of the Feature Extraction program.

5 Quality Tools

This chapter provides instructions on how to use the tools in the Quality module of Agilent CytoGenomics to query, evaluate, and chart microarray extraction data. For a description of each part of the module window, including all of the dialog boxes that can appear when you click buttons and other elements, see the *CytoGenomics Reference Guide*. For more information, see the Agilent Technologies application note, "Enhanced Quality Assessment Using Agilent Feature Extraction QC Metric Sets, Thresholds, and Charting Tools" by G. Delenstarr and C. Troup. To download this application note from the Agilent Technologies literature library, go to http://chem.agilent.com and then click **Literature Library**.

What are the Quality Tools?

Quality is a module consisting of a set of tools that analyze summary statistics from Feature Extraction output files and optional annotation files. The results from quality analysis are used to monitor microarray processing performance. The set of Quality tools consists of tools for:

- Building and running queries
- Creating metrics
- Creating metric sets and setting thresholds
- Creating and displaying charts

Feature Extraction data are kept in the CytoGenomics relational database. Queries enable you to search this data. You can save the queries to allow the display of specific subsets of the data. For instance, queries can select the data found in specific designs, workflows, batches, or dates of processing. Using the data, you can create metrics that monitor aspects of the microarray processing workflow.

NOTE The Quality tab is only available if you have a user role of Scientist or Administrator.

Additionally, you can create metric sets that combine metrics, and you can set thresholds for metrics within a metric set. A default metric set is included in the Agilent CytoGenomics database. Agilent optimized the default metric set for the workflow using the Agilent microarray scanner, Feature Extraction default protocol, and Agilent laboratory protocol. You can change and save it using a new name, to create one or more custom metric sets; for example, if you need to optimize the thresholds for your custom protocols.

With the Quality tools, you select a query to define which extractions to display and select a metric set to define which metrics to display. You then graphically plot the results from current or historical microarray data and create thresholds for the metrics that are appropriate for your experimental conditions and processing environment.

You can also use metric sets and thresholds to create Metric Set filters, which are then used in workflow extractions or preprocessing filtering of data.

These Quality tools were designed for use in a production environment where:

- Microarray processing protocols are standardized and personnel want to know the effect of specific variables on performance, such as:
  - Operators
  - Wet-lab protocols
  - FE parameter protocols
- Monitoring run-to-run consistency is an important goal to:
  - Identify extractions that fall outside the established normal range
  - Identify systematic data trends

For the most recent information and to download QC metric sets, go to the website: **http://www.agilent.com/chem/feqcmetrics**.

Example use cases for quality tools

Agilent Feature Extraction for CytoGenomics (FE) generates output to assist in quality assessment. A table of array-wide, or global statistics (the "Stats" table in FE), is useful in data quality determination for each extraction. These global statistics capture information from every independent FE step - for example, the numbers of outliers, the averages of negative control signal statistics, and spike-in regression values. In addition, the table includes key fields from the FE Parameters table, which shows user, software version, scanner date, grid, and protocol date. Sample attributes added by the user are also available in the table.

The number of output fields are sometimes cumbersome without a tool for quality assessment of each extraction. The Quality tools capture key global statistics to use as metrics and create graphs for easy visual assessment of metrics. Additionally, you can use the metrics that are used for analysis for specific monitoring requirements.

This section provides several common use cases for the Quality tools.

Use Case 1 Analysis of Feature Extraction output

Feature Extraction analysis is a common way to use the Quality tools with everyday extraction monitoring. An example of this type of analysis was performed using a collection of microarrays from several workflows.

Some of these microarrays were previously annotated by the operator as having issues in the labeling, hybridization, and/or washing steps. These microarrays had poor correlation with their replicate microarray sets. They were chosen because they each had at least one metric flagged as having values outside of the normal range.

This analysis of Feature Extraction consisted of:

1 A comparison to all other extractions within the extraction set.

2 A comparison to thresholds associated with the default metric set. For information on creating metric sets, see "Creating Metric Sets and Setting Thresholds" on page 92.

By default, the chart generated by the Quality charting tool shows extractions in the order they were performed. Because the Quality charting tool has customized sorting, color-by, and shape-by attributes, it is a powerful tool for visualizing and highlighting trends in patterns. You can see these trends in the results of this analysis.

Figure 7 on page 79 confirms the presence of processing artifacts and replicate microarray outliers. The chart shows that several microarrays have more than one metric out of normal range (represented by red circles). Values in range are also displayed (blue triangles). The inset window zooms in on the "rNegCtrlAveGBSubSig" metric, which is the average of the red-channel negative-control background-subtracted signals. For more information about Feature Extraction statistics used for metrics, see the *Feature Extraction for CytoGenomics Reference Guide* located at **<program folder>\FeatureExtraction\Docs,** where **<program folder>** is the location where your Agilent CytoGenomics client software is installed.

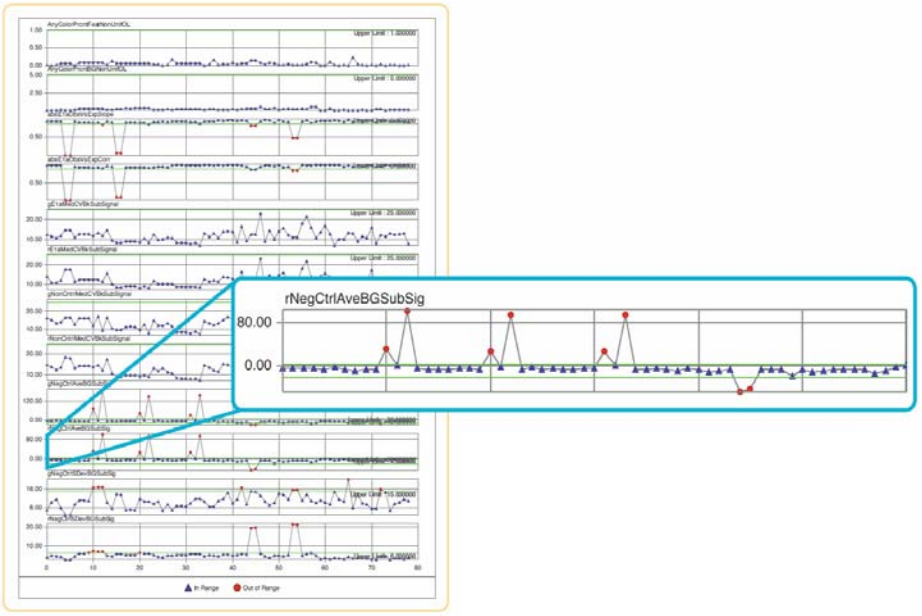


Figure 7 Assessment of extraction statistics used for metrics with the Quality tools

Use Case 2 Analysis of the user effect on extraction quality

Often the effects of specific variables on extraction quality are needed in a production setting. One such variable is the effect of the user. A retrospective analysis of different users was generated using the following steps:

1. A query was created to select only those extractions of interest and applied to a chart as the x-axis. For information on creating queries, see "Building and Running Queries" on page 86.
2. A two-color gene expression metric set was chosen and applied to the chart as the Y-axis. For information on creating metric sets, see "Creating Metric Sets and Setting Thresholds" on page 92.
3. The extractions were then color-coded to reflect the three different operators who had processed the arrays in those extractions.

Figure 8 on page 80 shows data from microarrays processed by users A (represented by blue squares), B (red circles), and C (green triangles). Threshold limits appear in upper right corners and as green lines within each plot. The inset window zooms in on the "rNegCtrlAveGBSubSig" metric, which is the average of the red-channel negative-control background-subtracted signals. For more information about Extraction statistics used for metrics, see the *Feature Extraction for CytoGenomics Reference Guide*.

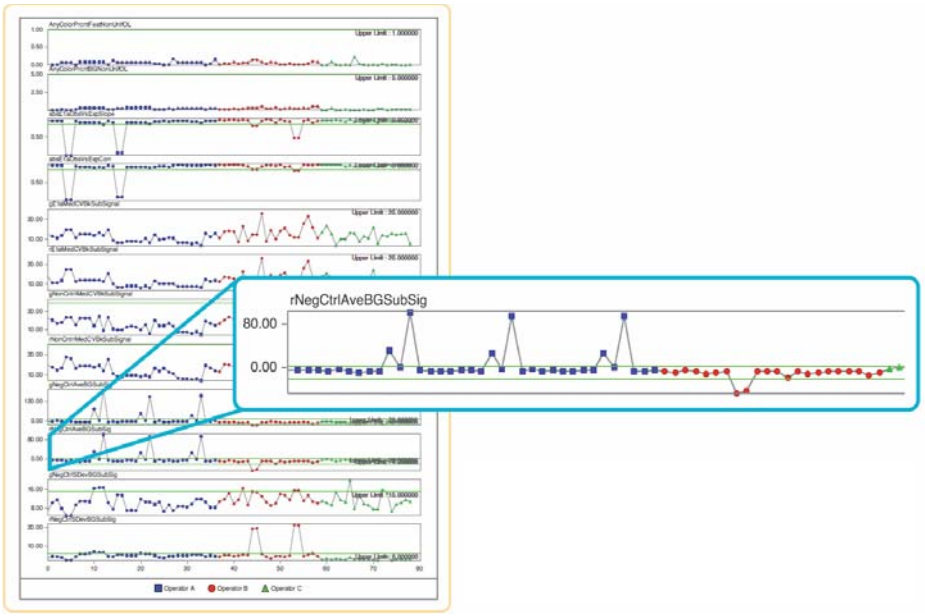


Figure 8 Comparison of the effect of different users on microarray performance using the Quality tools

Use Case 3 Analysis of the effect of changing the FE parameter protocols

Analysis of Extraction statistics used for metrics with the Quality tools gives an intuitive evaluation of competing protocol methods, such as background processing algorithms. An 18-array set was extracted with either the default FE parameter protocol "Spatial Detrend" background method or an alternative "Minimum Signal" background method and processed using the standard metric set in Quality tools.

Figure 9 on page 81 shows extractions with either the default Spatial Detrend background (represented by blue squares), or the alternate Minimum Signal background data (red circles). For this data set, more favorable metric values were clearly seen with the Spatial Detrend method. This improvement is seen especially with the average negative control background-subtracted signal. This value is closer to the expected value of zero for extractions using the Agilent default background algorithm, and therefore a more accurate estimate of background.

Additionally, undersubtraction of background, as seen with the Minimum Signal method, results in compression of log ratios. The inset window shows a plot of observed versus expected spike-in ratios (the "absE1aObsVsExpSlope" metric), where Spatial Detrend background yields a slope closer to 1.0. For more information about Feature Extraction statistics used for metrics, see the *Feature Extraction for CytoGenomics Reference Guide*.

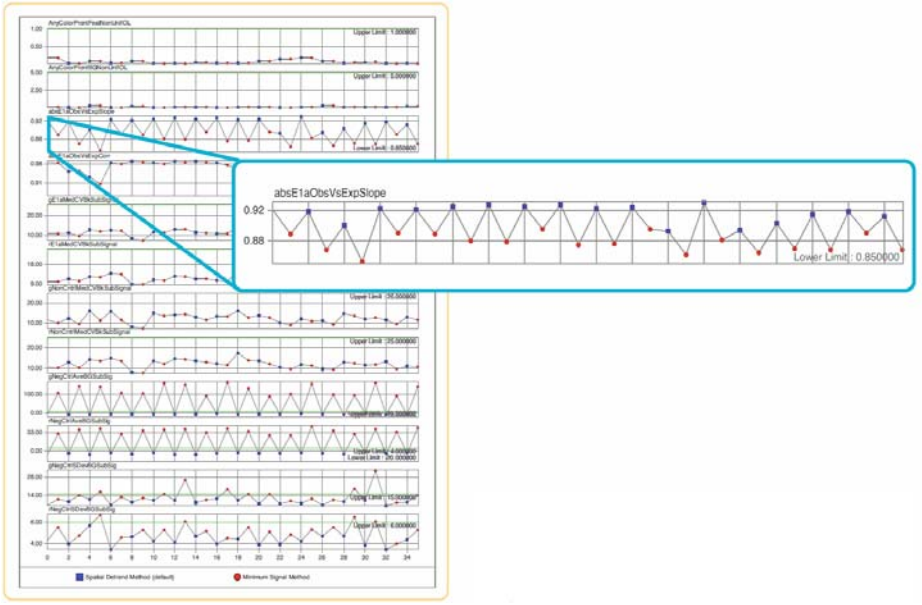


Figure 9 Comparison of the effect of FE parameter protocols on extraction quality

5 Quality Tools

Example use cases for quality tools

Creating Queries, Metric Sets, and Charts to Evaluate Extractions – Overview

This section provides an overview of using the tools in the Quality module. For details on the commands and dialog boxes that appear, see the *CytoGenomics Reference Guide*.

The following diagram shows how you typically use the Quality tools.

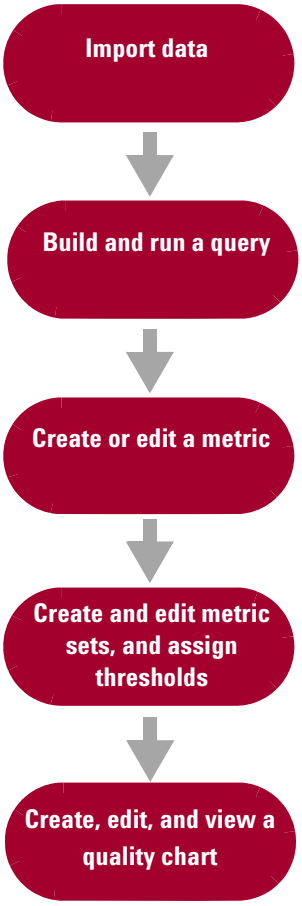


Figure 10 Using the quality tools

Importing Data

The Quality module can easily access the database of feature extractions (global statistics and FE parameters) that are imported or extracted, making it easy to monitor microarray quality control, and analyze historical quality control trends.

When you set up a Workflow to extract an image file, or import an extracted FE file, the extraction is imported into the Agilent CytoGenomics database. The appropriate FE statistics and parameters are also imported into the Quality database.

The Quality module also includes a way to import only the quality information (Stats and Parameters) from Feature Extraction output files that are not in the CytoGenomics database.

The following table gives instructions on how to manually perform the import tasks shown in Figure 10 on page 83. Usually, it is not necessarry to perform import tasks manually; data is imported as part of the workflow.

Table 20 Tasks for importing data in the Quality module

To do this task	Follow these instructions	Comments
Import FE statistics and parameter information	**1** On the Quality ribbon, click the **Import File** button, and then click **FE Stats and Parameters**. The Import FE Files dialog box appears. **2** Browse to the Feature Extraction output files, and highlight to select them. **3** Click **Open**. The Agilent Feature Extraction Importer dialog box appears. **4** Click **OK**. The Feature Extraction statistics and parameters information is added to the list of extractions in the Quality table. The names of the arrays are also added to the Array Data pane of the Sample Manager Navigator, under Stats and Parameters.	• On the Quality ribbon, move the cursor over the button to see the name of the button. • When you click the **Import File** button, a menu of Import commands appears. • **Important:** In order to associate sample attributes with an array before running the workflow, add the array in the Content > Sample Manager table, or import an SAF.
Import a saved query	**1** On the Quality ribbon, click the **Import File** button, and then click **Query Result**. The Import Query Result dialog box appears. **2** Move to the previously exported query result file, select it, and click **Open**.	• You can only import query result files that were previously exported from the Quality tab.

Building and Running Queries

The Quality module has a query builder that lets you select a subset of the extractions from the Agilent CytoGenomics database, based on criteria that you select.

A query is used to define a subset of extractions for a representative data set, for use in metric and threshold development, and in producing Charts. For example, you can build a query that contains data from similar biological samples processed under identical conditions. Or, you can query for different types of samples, or for different processing methods. With the latter example, you can then use the different processing attributes to color-code a chart. See "Example use cases for quality tools" on page 77 for examples of color-coding a chart based on the processing attributes.

A subset of extractions is defined in a query by specific FE Parameter fields, or by user-added attribute fields.

The following table gives instructions on how to perform the tasks shown in Figure 10 on page 83 for building and running queries.

Table 21 Tasks for building and running a query

To do this task	Follow these instructions	Comments
Create a query	**1** To start the query builder, on the Quality ribbon, click the **Queries** button, and then click **New**. **2** In the Column Name drop-down list, select the parameter to set. **3** In the Operator drop-down list, select the appropriate operator. **4** In the text box on the right, select the value with which to compare the value of the Column Name parameter. **5** Click **Add**. **6** In the Query Name area, type a name for the query. **7** Click **Save**.	• When you click the **Queries** button, a menu of commands appears. • A Query can be used in developing thresholds for metrics or used to define a Quality chart. The query can be run, and the results can be exported and saved. Queries can also be renamed and deleted • Once you create a query, you can run, delete, rename, edit, or export that query using the menu that appears when you right-click the query name in the Extractions pane of the Navigator.
Create a composite query	**1** Create a basic query as described in the previous step. **2** Click **AND** or **OR**. Use AND to find extractions that meet all criteria. Use OR to find extractions that meet at least one criterion. **3** Create the next basic query. **4** To group composite queries, click a query, and then click **"("** or **")"**. Repeat for the query at the other end of the group. **5** In the **Query Name** area, type a name for the query. **6** Click **Save.** The Quality main user interface is displayed.	• This example is a query that finds all 1-color gene expression extractions: ColorMode = 1-Color AND QCReportType=GeneExpression
Edit a query	**1** In the Quality Navigator, in the Extractions pane, under Queries, select the query to edit. **2** On the Quality ribbon, click the **Queries** button, and then click **Edit.** Alternatively, right click on the query of interest and select **Edit Query.** **3** In the Query Builder dialog box, make any necessary changes. **4** Click **Save.**	

Table 21 Tasks for building and running a query

To do this task	Follow these instructions	Comments
Run a query	**1** In the Navigator, in the Extractions pane, under Queries, double-click the query to run. OR Select the query to run. Then, do one of the following: • On the Quality ribbon, click the **Queries** button, and then click **Run.** • Right click on the query of interest and select **Run Query.**	
Delete a query	**1** In the Navigator, in the Extractions pane, click the **Queries** button, and select the query to delete. **2** On the Quality ribbon, click the **Queries** button, and then click **Delete.** Alternatively, right click on the query of interest and select **Delete Query.** **3** In the Delete Query dialog box, click **Yes** to confirm the deletion.	

Table 21 Tasks for building and running a query

To do this task	Follow these instructions	Comments
Rename a query	**1** In the Navigator, in the Extractions pane, select the query to rename. **2** On the Quality ribbon, click the **Queries** button, and then click **Rename.** Alternatively, right click on the query of interest and select **Rename Query**. **3** In the Enter New Name dialog box, enter the name for the query. **4** Click **OK**.	
Export a query	**1** In the Navigator, in the Extractions pane, select the query to export. **2** On the Quality ribbon, click the **Queries** button, and then click **Export.** Alternatively, right click on the query of interest and select **Export Query.** **3** In the Export Query Result dialog box, type the path and name of the export query result file. **4** Click **OK**.	

Creating and Editing Metrics

Metrics are defined in order for you to track desired statistical values within a set of extractions. These metrics can be associated in a metric set. The following table gives instructions on how to perform the tasks shown in Figure 10 on page 83 for creating and editing metrics.

Table 22 Creating and editing metrics

To do this task	Follow these instructions	Comments
Create a new metric	1 On the Quality ribbon, click the **Metrics** button, and then select **New**. The Create a new Metric dialog box opens. 2 Select a metric from the Choose Metric Column list and then click **Add**. 3 Define a new metric as an expression. Use the operator buttons and type numbers, if needed, in the Numerical Constant text field to create a formula. 4 In the **Save Metric** text box, type a name to save the new metric. 5 Click **Save**. 6 The new metric now appears in the Metric list of the Configure Metrics and Thresholds dialog box.	**Example** Feature Extraction calculates a slope for the eQC spike-ins (observed versus expected Log Ratio). Depending upon the hybridization, this spike-in mixture can be present as "+1" or "–1" polarity. If it is "–1", then any threshold that is set (for example, Slope > 0.85), will not pass. Therefore you can make a derivative metric by taking the absolute of the slope, using the following steps. 1 On the Quality ribbon, click the **Metrics** button, and then click **New** to open the Create a new Metric dialog box. 2 Click **Abs**. The term **Abs (**appears in the Metric Calculation box. 3 From the Choose Metric Column list, select the statistic **eQCObsVsExpLRSlope**, click **Add**, and then select "**)**" to finish the expression. 4 Validate and save the metric as Abs_eQCSlope. The new metric now appears in the Metric list of the Create a new Metric dialog box.

Table 22 Creating and editing metrics

To do this task	Follow these instructions	Comments
Delete a metric	**1** On the Quality ribbon, click the **Metrics** button and then click **Delete**. **2** Select a metric or hold down the control button and select several metrics previously defined shown in the Delete Metrics dialog box. **3** Click **Delete.**	• If a metric is part of a metric set, it does not appear in the list, and cannot be deleted.

Creating Metric Sets and Setting Thresholds

Metric Sets are combinations of existing metrics applied with optional user-defined thresholds. Metric sets can be saved and exported for future use.

The following table gives instructions on how to perform the tasks shown in Figure 10 on page 83 for creating metric sets and setting thresholds.

Table 23 Steps to define metric sets and set thresholds

To do this task	Follow these instructions	Comments
Create a new metric set	**1** On the Quality ribbon, click the **Metric Sets** button and then click **New**. **2** When the Metric Set Configuration dialog box appears, select the **Add Metrics to Metric Set** tab. **3** Mark the metrics from the Existing Metrics checklist to associate with the selected metric set. **4** Set the appropriate thresholds for the Metric to add to the Metric Set. See "Edit a metric set". **5** Optional: assign an Extraction Query to the metric set. **6** In the Metric Set Name area, type the name for the Metric Set. **7** Click **Save**.	• You can also create a metric set by editing an existing metric set, then saving it with a new name. • The Extraction Query option is available in Standard, Robust, and Percentage Threshold Calculation modes. This query filters the appropriate extractions from the database. The data from these queried extractions is used to calculate the statistical summary values for setting the thresholds. • You can display, edit, delete, rename, import, or export a metric set using the menu that appears when you right-click the metric set name in the Metric Sets pane of the Navigator.
Edit a metric set	**1** Select the Metric Set of interest in the Metric Sets pane of the Navigator. **2** On the Quality ribbon, click the **Metric Sets** button, and click **Edit**. **3** Follow the steps outlined in "Create a new metric set".	

Table 23 Steps to define metric sets and set thresholds

To do this task	Follow these instructions	Comments
Set thresholds in a metric set	**1** Open the Metric Set Configuration dialog box. See "Create a new metric set" on page 92 or "Edit a metric set" on page 92. **2** Select the **Add Metrics to Metric Set** tab. **3** Select the metrics from the Existing Metrics checklist that is currently associated or will be associated with the selected metric set. **4** In the Threshold Type area, select the type of threshold to associate with the metric: • Upper Limit • Upper Warning Limit • Lower Warning Limit • Lower Limit **5** In the Threshold Calculation area, select the limit calculation type: • Manual • Standard • Robust • Percentage The relevant calculations and their limits are displayed. **6** Optional: assign an Extraction Query to the metric set. **7** Edit the limits as appropriate. The text boxes accept floating point constants. **8** If necessary, enter or change the name for the metric set in the Metric Set Name area. **9** In the Edit Thresholds dialog box, click **Save** to add the chosen metric along with its limits to the selected metric-set. If the metric is already present, it is updated with the new limits.	• The Extraction Query option is available in Standard, Robust, and Percentage Threshold Calculation modes. This query filters the appropriate extractions from the database. The data from these queried extractions is used to calculate the statistical summary values for setting the thresholds.

Table 23 Steps to define metric sets and set thresholds

To do this task	Follow these instructions	Comments
Export a metric set	**1** In the Metric Sets pane of the Navigator, select the metric set you want to export. **2** On the Quality ribbon, click the **Metric Sets** button and then click **Export**. The Export Metric Set dialog box appears. **3** Browse to the location where you want to save the file, type a name for the file, and then click **Export**.	• A Metric Set can be exported to an XML file and reimported in Quality Tools or Feature Extraction.
Import a metric set	**1** On the Quality ribbon, click the **Metric Sets** button and then select **Import**. The Import Metric Set dialog box appears. **2** Browse to a location, select the metric set file, and then click **Open**. **3** If a warning appears that asks if you want to overwrite Threshold settings for the matching metrics, click **Yes**.	• You can import metric sets that have been exported, or that have been downloaded to your computer from the Agilent website at http://www.agilent.com/chem/feqcmetrics.
Duplicate a metric set	**1** In the Navigator, in the Metric Sets panel, right-click the metric set to duplicate. **2** Click **Duplicate Metric Set**. **3** Type a new name for the metric set, and click **Ok**.	• The default metric sets that come with the software are read-only. To change them, first duplicate them and save them using a different name.
Delete a metric set	**1** In the Navigator, in the Metric Sets panel, select the metric set to remove. **2** On the Quality ribbon, click the **Metric Sets** button and then click **Delete**. Alternatively, right-click the metric set in the Navigator and select **Delete Metric** set.	• You cannot delete the standard metric sets. • You cannot delete a metric set if it is used in a Chart.

Table 23 Steps to define metric sets and set thresholds

To do this task	Follow these instructions	Comments
Remove a metric from a metric set	**1** In the Metric Sets pane of the Navigator, right-click the metric set you want to rename, and then click **Edit Metric set**. The Metric Set Configuration dialog box appears. **2** Clear the metrics in the Existing Metrics checklist you want to remove from the selected metric set. **3** Click **Save**.	• You cannot remove metrics from the standard metric sets. Click **Save As** to save the updated metric set with a different name.
Rename a metric set	**1** In the Metric Sets pane of the Navigator, right-click the metric set you want to rename, and then click **Rename**. The Enter new name dialog box appears. **2** Type a new name for the metric set. **3** Click **Ok**.	• You cannot rename a standard metric set.

Creating and Displaying Charts

A chart is drawn on a metric set for a chosen query. The chart is defined in the Chart Configuration dialog box. The following table gives instructions on how to perform the tasks shown in Figure 10 on page 83 for creating and displaying quality charts.

Table 24 Tasks for creating and displaying quality charts

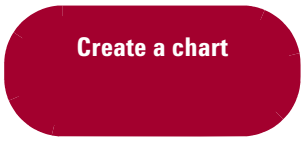
To do this task	Follow these instructions	Comments
	**1** On the Quality ribbon, click the **Chart** button and then select **New**. The Chart Configuration dialog box appears. **2** Select the metric set to use for the chart from the Metric Set drop-down list. **3** Optional: Select the Extraction Query from the drop-down list for the metric set. **4** Optional: In the Sort by Columns area, select any ordering of the metrics for display in the chart. **5** Optional: Mark the **Color and shape by in/out of the threshold range** check box to set the color and shape of all points by whether their values are in or out of range. Otherwise, *clear* the **Color and shape by in/out of the threshold range** check box and set the color and shape for the chart as follows:	

Table 24 Tasks for creating and displaying quality charts

To do this task	Follow these instructions	Comments
	• In the Color By area, select any color for one of the metrics to display in the chart. • In the Shape By area, select any shape for one of the metrics to display in the chart. **6** In the Chart Name area, type a name for the chart. **7** Click **Save**. You can now display the chart. You can also edit, rename, and delete charts.	
Display a chart	**1** In the Navigator, in the Charts panel, select the chart to view. **2** On the Quality ribbon, click the **Chart** button and then click **View**. Alternatively, double-click the chart in the Navigator. **3** The metric set is displayed in the Tab View.	• To return the Tab View to another metric set, chart, or query result, click the appropriate entity in the Navigator.
Edit a chart	**1** In the Navigator, in the Charts panel, select the chart to edit. **2** On the Quality ribbon, click the **Chart** button and then click **Edit**. Alternatively, right-click the chart in the Navigator and select **Edit Chart**. The Chart Configuration dialog box appears. **3** Configure the dialog box according to your needs. See "Create a chart" on page 96. **4** If necessary, change the name of the chart in the Chart Name area. **5** Click **Save**.	

Table 24 Tasks for creating and displaying quality charts

To do this task	Follow these instructions	Comments
Delete a chart	**1** In the Navigator, in the Charts panel, select the chart to delete. **2** On the Quality ribbon, click the **Chart** button and then click **Delete**. Alternatively, right-click the chart in the Navigator and select **Delete Chart**. **3** When the Delete Chart dialog box appears, click **Yes**.	
Rename a chart	**1** In the Navigator, in the Charts panel, select the chart to rename. **2** On the Quality ribbon, click the **Chart** button and then click **Rename**. Alternatively, right-click the chart in the Navigator and select **Rename Chart**. The Enter New Name dialog box appears. **3** Type the new name for the chart, and then click **Ok**.	

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In this book

This guide describes how to use the Agilent CytoGenomics 2.0 software to set up report templates, analysis methods, workflows, standard notes, queries, and tracks. It also describes how to organize samples and use Quality Tools to examine trends and sample quality. The functions described in this guide are available to users with an assigned role of “Scientist” or “Administrator.”

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