

Agilent Genomic Workbench Lite Edition 6.5

Product Overview Guide

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Agilent Technologies

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Software Revision

This guide is valid for 6.5 and later revisions of the Agilent Genomic Workbench Lite Edition software, until superseded.

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In This Guide...

This guide gives high-level descriptions of the programs within Agilent Genomic Workbench Lite Edition, a suite of tools that allows you to:

- Manage information about samples
- Interactively investigate aberration patterns, protein-DNA binding events, and DNA methylation
- Simultaneously detect and investigate high resolution copy number and copy neutral variations for CGH+SNP (Comparative Genomic Hybridization with Single Nucleotide Polymorphism) microarrays.
- Run a workflow to automate feature extraction, the assessment of data quality, and data analysis.

1 Overview

This chapter gives a high-level overview of the major features of the Lite Edition of Agilent Genomic Workbench.

2 Getting Started

This chapter describes how to start the programs in Agilent Genomic Workbench Lite Edition and find Help, and how to enter your license information.

3 Detailed Descriptions

This chapter gives more detailed descriptions of the major features of the Lite Edition of Agilent Genomic Workbench.

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1 Overview

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Agilent Genomic Workbench 6.5 is a comprehensive design and analysis tool that you use to design microarrays and SureSelect Target Enrichment Kits, and to analyze your microarray experiments. Its capabilities are included in two software packages: a Standard Edition that gives full functionality, and a Lite Edition that provides a subset of the features. This guide gives an overview of the Lite Edition of Agilent Genomic Workbench.

This chapter gives you a high-level overview of the Lite Edition of Agilent Genomic Workbench. Chapter 2 tells you how to start and find Help for each module, and Chapter 3 gives you more details about each application.



Features of the Agilent Genomic Workbench Lite Edition

The Agilent Genomic Workbench Lite Edition software provides a robust data management and integrated data analysis environment for Agilent genomics applications, including comparative genomic hybridization (CGH), chromatin immunoprecipitation (ChIP), and methylation (CH3).

When used with Agilent Feature Extraction software (installed separately), the Agilent Genomic Workbench Lite Edition lets you manage, extract, analyze, and produce reports for your microarrays with little or no intervention. You can also use the program interactively to analyze and examine data in more detail.

Using the Agilent SureSelect Target Enrichment system and the Agilent eArray Web site, you can design bait libraries that retrieve specific DNA fragments for sequencing. You can then use the Agilent Genomic Workbench Lite Edition SureSelect Target Enrichment application to run read depth and enrichment analyses on your next-generation sequencing data.

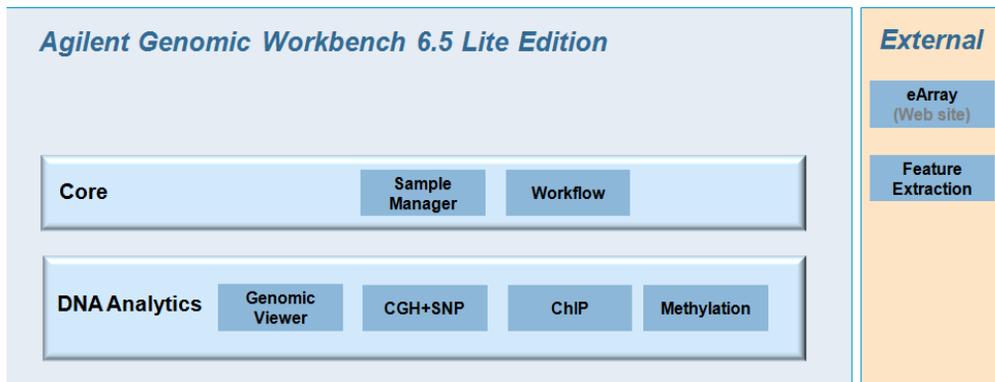


Figure 1 Programs available in Agilent Genomic Workbench Lite Edition

The software helps you:

- Manage sample information through the analysis process
- Start a program to extract features from microarray *.tif files
- Analyze data interactively

Features of the Agilent Genomic Workbench Lite Edition

- Run a workflow to automate feature extraction and data analysis
- Produce QC reports and do enrichment analysis for target genomic regions, based on sequence read data

Automated Feature Extraction and Analysis

The Agilent Genomic Workbench Workflow is uniquely designed to meet the needs of users who are extracting and analyzing microarray data from a large number of samples.

With Agilent Feature Extraction installed (license required) and a DNA Analytics program license (CGH or ChIP), you can use Sample Manager to associate a list of image files with Array IDs and sample attributes, and then set up and run a workflow that extracts selected images. The Feature Extraction results are then automatically analyzed with a DNA Analytics analysis method.

Feature Extraction works together with the Agilent Genomic Workbench Lite Edition core utilities to create the final analytical results (Figure 3).

Sample Manager is used with Workflow and Feature Extraction to keep sample IDs and their results together during extraction; then Workflow calls a specified analysis method to run a selected algorithm on the extracted results. You can save the workflow and the analysis method for later use.

The Agilent Genomic Workbench Lite Edition user interface has Sample Manager and Workflow tabs (Figure 2). You can set up samples and run the workflow with the commands under these tabs, if you have a CGH or ChIP license.

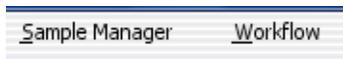


Figure 2 Sample Manager and Workflow tabs

When you run an extraction workflow, Feature Extraction runs in the background and uses a grid template that matches the AMADID in the image barcode and a default Feature Extraction protocol associated with the grid template. If you want to check images before a run, or run a sample interactively, you use the Feature Extraction software, which is separate from the Agilent Genomic Workbench Lite Edition. Refer to the *Agilent Feature Extraction User Guide* for more information on using the Feature Extraction software.

The entire Agilent microarray research pathway with the Lite Edition of Agilent Genomic Workbench looks like this:

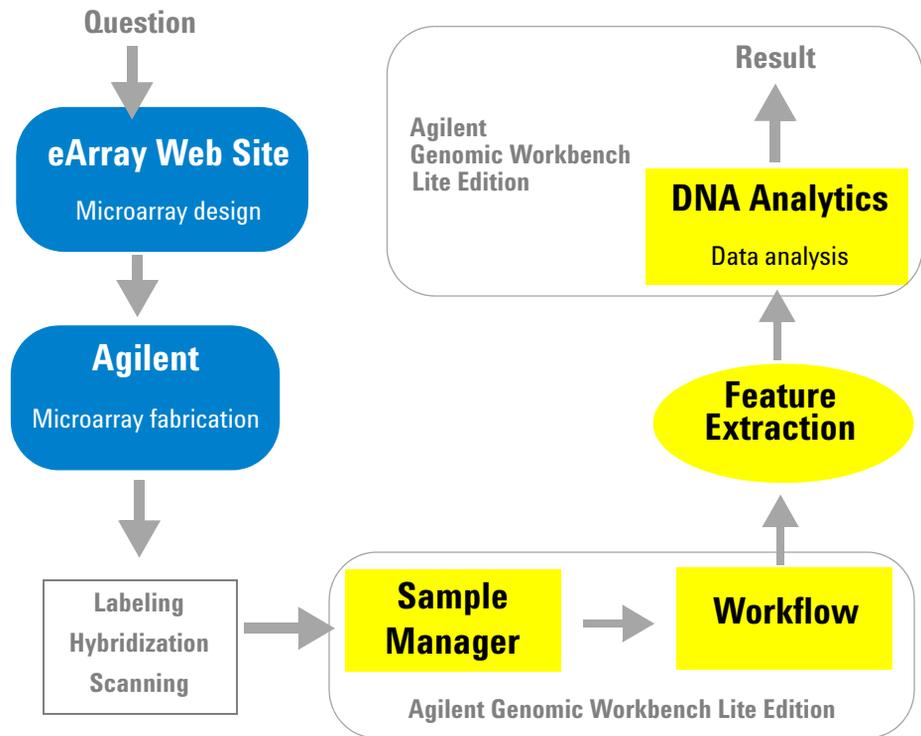


Figure 3 Microarray research pathway with Agilent Genomic Workbench Lite Edition and Feature Extraction

Interactive Data Analysis for CGH, ChIP, or Methylation (CH3)

With one or more DNA Analytics licenses – CGH, ChIP, Methylation (CH3) – you can set up preprocessing, analysis, and reporting parameters interactively. The CGH program also has many postprocessing capabilities, called Discovery.

Table 1 Comparison of DNA Interactive Analysis capabilities with Agilent Genomic Workbench Lite Edition

Tabs	CGH capabilities	ChIP capabilities	Methylation (CH3) capabilities
Preprocessing	<ul style="list-style-type: none"> • Add feature, array, and design filters • Apply GC content correction • Turn on Centralization • Add design filters • Combine array designs and replicates • Display QC metrics 	<ul style="list-style-type: none"> • Select/Edit normalization calculation • Select/Edit error model • Combine designs and replicates • Display QC metrics 	<ul style="list-style-type: none"> • Combine array designs
Analysis	<ul style="list-style-type: none"> • Calculate a moving average on log ratio data • Select an aberration detection algorithm and set up its parameters • Perform SNP analysis, including SNP Copy Number (with or without manual reassignment of peaks) and LOH (Loss/Lack of Heterozygosity). 	<ul style="list-style-type: none"> • Select/Edit/Apply event detection model 	<ul style="list-style-type: none"> • Calculate a moving average on log ratio data • Apply probe methylation • Calculate a moving average on Z-scores generated by probe methylation algorithm • Apply Batman algorithm

Table 1 Comparison of DNA Interactive Analysis capabilities with Agilent Genomic Workbench Lite Edition

Tabs	CGH capabilities	ChIP capabilities	Methylation (CH3) capabilities
Discovery	<ul style="list-style-type: none"> • Add aberration filters • Compare arrays with common aberrations • Make graphical penetrance diagrams • Set up to view CNVRs (copy number variant regions) • Compare CGH data with expression data • Compare arrays with different aberrations • Do a cluster analysis • Do a heatmap analysis • Generate a genotype reference 	<ul style="list-style-type: none"> • Not applicable 	<ul style="list-style-type: none"> • Not applicable
Reports	<ul style="list-style-type: none"> • Manage reports • Create Aberration report • Create SNP Genotype report • Create Aberration & LOH report • Create Penetrance report • Create Cyto Report 	<ul style="list-style-type: none"> • Probe report • Gene report • QC report 	<ul style="list-style-type: none"> • Probe report • Batman report

Summary of capabilities

The modules in the Lite Edition of Agilent Genomic Workbench give you the capabilities shown in [Table 2](#):

Table 2 What you can do with Agilent Genomic Workbench Lite Edition and associated programs

If you want to do this:	Start this program or click this tab: (See Figure 4 on page 18 and Figure 7 on page 25)	Read this guide:
Design a microarray or SureSelect Target Enrichment library	eArray Web site	<ul style="list-style-type: none"> Go to https://earray.chem.agilent.com/earray/ and click Help SureSelect Target Enrichment User Guide
Manage samples – Associate array IDs and attributes (sample information) with image files and/or imported extracted data	Sample Manager (Agilent Feature Extraction 10.7 or higher license and one or more DNA Analytics (CGH, ChIP, CH3) licenses required. Note: Feature Extraction has no tab in the Agilent Genomic Workbench user interface.)	Sample Manager User Guide
Run Feature Extraction Workflow – Set up and run a workflow where scanned image files are automatically extracted	<ul style="list-style-type: none"> Workflow (Agilent Feature Extraction 10.7 or higher license and one or more DNA Analytics (CGH, ChIP) licenses required. Note: Feature Extraction has no tab in the Agilent Genomic Workbench user interface.) In order to run feature extraction on CGH+SNP microarrays, you must have Feature Extraction version 10.10 or higher installed. 	Workflow User Guide
Run Analysis Workflow – Set up and run a workflow for automated, unattended CGH or ChIP analyses	Workflow (CGH or ChIP license required)	Workflow User Guide

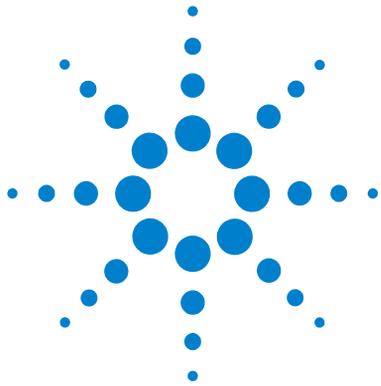
Table 2 What you can do with Agilent Genomic Workbench Lite Edition (continued) and associated programs

If you want to do this:	Start this program or click this tab: (See Figure 4 on page 18 and Figure 7 on page 25)	Read this guide:
<p>Run Feature Extraction and Analysis Workflow – Set up and run a workflow where image files are automatically extracted and the results are automatically analyzed</p>	<ul style="list-style-type: none"> Workflow (Agilent Feature Extraction 10.7 or higher required and either a CGH or ChIP license required) In order to run feature extraction on CGH+SNP microarrays, you must have Feature Extraction version 10.10 or higher installed. 	Workflow User Guide
<p>Display Feature Extraction data – Import Feature Extraction data and view it next to chromosomes and genes</p>	Genomic Viewer	Data Viewing User Guide (if you do not have a CGH, ChIP, or Methylation license), OR CGH Interactive Analysis User Guide ChIP Interactive Analysis User Guide Methylation (CH3) Analysis User Guide
<p>Create and apply array, feature, and design filters in the CGH Interactive interface</p>	DNA Analytics (CGH license required)	CGH Interactive Analysis User Guide
<p>Analyze extracted data interactively for CGH, ChIP or Methylation (CH3) application types</p>	DNA Analytics (CGH, ChIP or Methylation license required)	CGH Interactive Analysis User Guide ChIP Interactive Analysis User Guide Methylation (CH3) Analysis User Guide
<p>Create and view reports</p>	DNA Analytics (CGH, ChIP, and/or CH3 license required)	CGH Interactive Analysis User Guide ChIP Interactive Analysis User Guide Methylation (CH3) Analysis User Guide
<p>Assess quality of pull-downs of targeted genomic fragments when you use the Agilent SureSelect Target Enrichment System</p>	SureSelect Quality Analyzer	SureSelect Quality Analyzer User Guide

Capabilities Without Licenses

You do not need to purchase licenses to get some component capability with Agilent Genomic Workbench Lite Edition. Without an Agilent Feature Extraction or DNA Analytics program license you can still:

- Use the Home commands and Navigator (see [Figure 7](#) on page 25) to import, manage and display extracted log ratio data and other content in Genomic Viewer
- Use SureSelect Quality Analyzer to assess the quality of results from the Agilent SureSelect Target Enrichment system



2 Getting Started

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This chapter helps you get started with the Lite Edition of Agilent Genomic Workbench.

Before you read this chapter, install the Agilent Genomic Workbench Lite Edition 6.5. If you need to reinstall the program, see the *Agilent Genomic Workbench Lite Edition 6.5 Installation Guide*. If you do not have the guide, download it from the Agilent Web site.



Starting Your Application and Finding Help

After you install the software and start the program, you should see the Open Application tab of the main window.

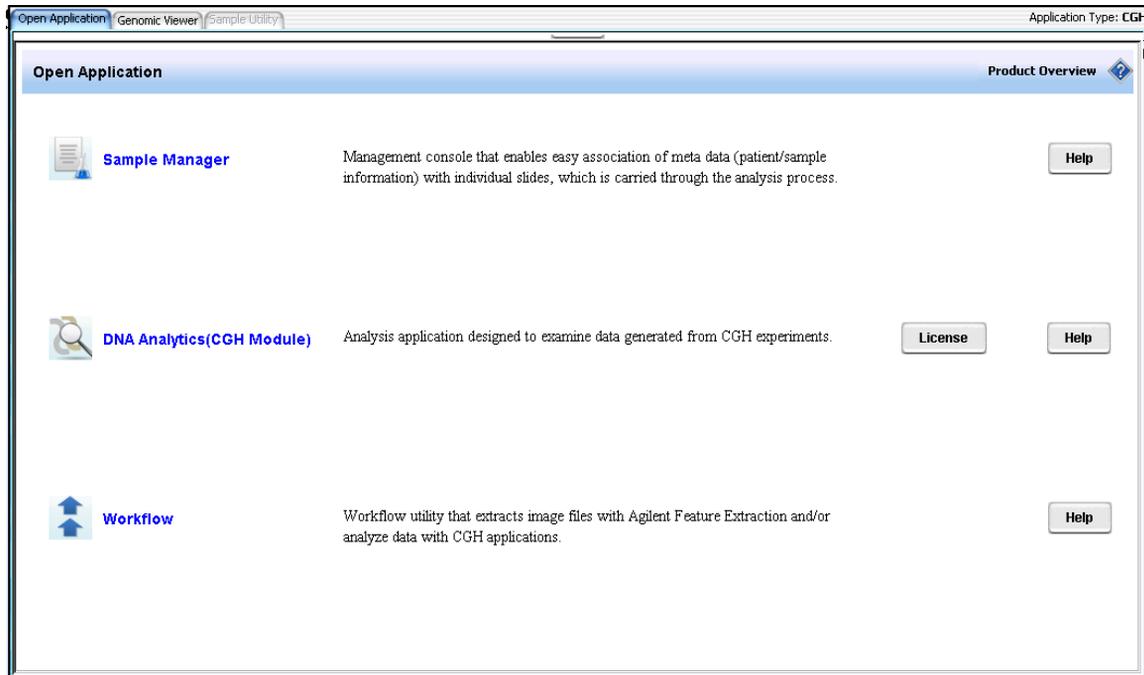


Figure 4 Open Application tab in Agilent Genomic Workbench Lite Edition, shown when CGH is selected under Switch Application

This section describes how to start each application and find help, and how to enter license(s) for individual modules.

To use eArray to update design files

Agilent regularly makes updates to probe annotations on its eArray Web portal. If you have imported Agilent array designs into Agilent Genomic Workbench, and you are a registered eArray user, you can download the updated design files from within Agilent Genomic Workbench. For more information about eArray, go to <https://earray.chem.agilent.com> and click **Help**.

- 1 In the Home tab, click **User Preferences**.

The User Preferences dialog box appears.

- 2 In the Miscellaneous tab, under eArray User Details, type your **eArray Username and Password**. For more information on User Preferences, see the *User Guide* for the application you want to use.
- 3 Click OK.

To start and find help for Sample Manager

This application requires a Feature Extraction license and one or more DNA Analytics license (CGH, ChIP, or CH3).

1 (Optional) To display the *Sample Manager User Guide*, in the Open Application tab, click **Help** next to the description of Sample Manager.

2 In the Open Application tab, click the icon by **Sample Manager**  .
OR

Click the **Sample Manager** tab.

The Sample Manager window appears. Two folders with demo data appear in the Navigator. See [Figure 5](#). For more information about the window components and how to use them, see the “Getting Started” chapter in the *Sample Manager User Guide*.

You do not have to change applications to work with Sample Manager for a different program – CGH, ChIP, or Methylation (CH3).

3 (Optional) To display the *Sample Manager User Guide*, click the **Help** tab, then click **Sample Manager**.

See also “[Organizing and Assigning Array Attributes with Sample Manager](#)” on page 38.

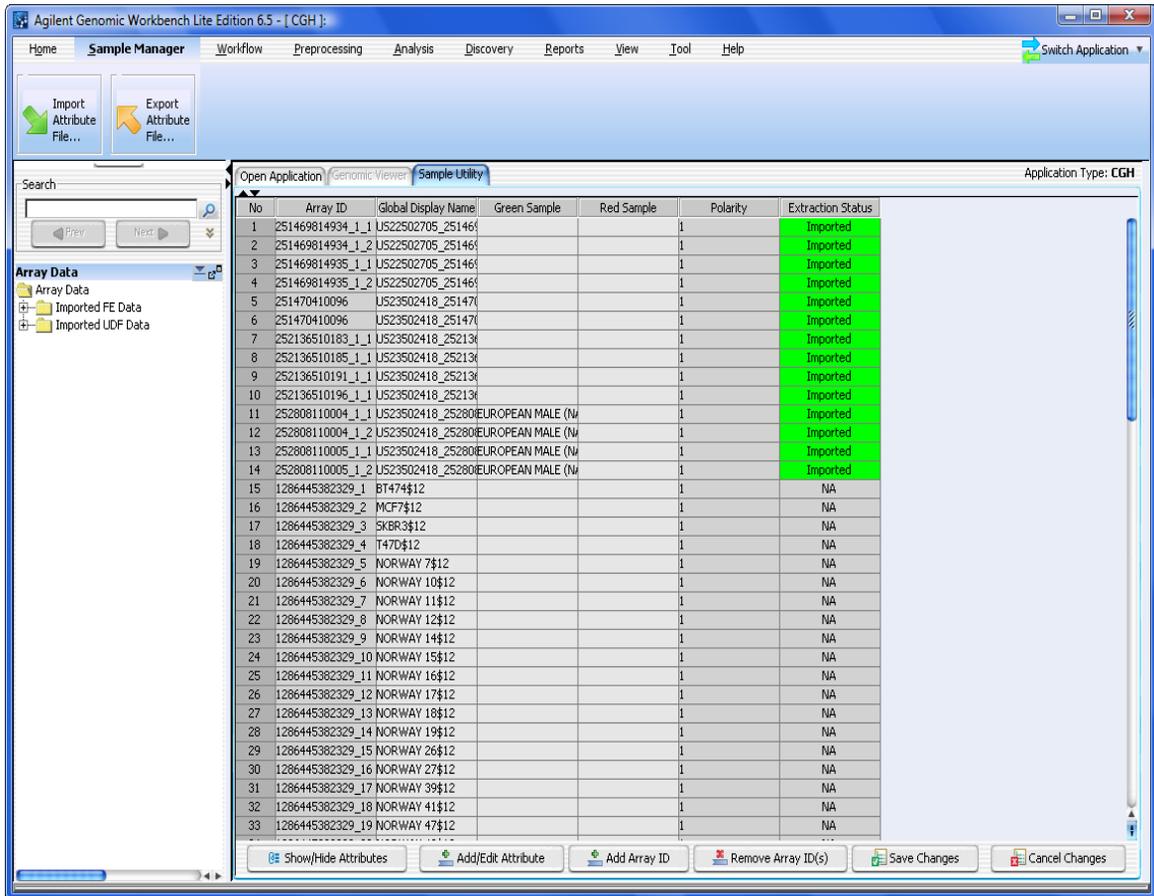


Figure 5 Agilent Genomic Workbench Lite Edition – Sample Manager tab

To start and find help to run workflows

This application requires one or more licenses.

- You must have an Agilent Feature Extraction 10.10 (or higher) license and one or more DNA Analytics licenses (CGH or ChIP) to run an *extraction* workflow. You can run an *extraction* workflow for many types of arrays, including CGH, ChIP, and gene expression.
 - You must have a CGH and/or ChIP license to run an *analysis* workflow. You can run an *analysis* workflow only for CGH and ChIP arrays.
- 1 (Optional) To display the *Workflow User Guide* in the Open Application tab, click **Help** next to the description of Workflow.
 - 2 In the Open Application tab, click the icon next to **Workflow** .

OR

Click the **Workflow** tab.

The Workflow window appears and the content of the Navigator changes. You initially see the Workflow Navigator. See [Figure 6](#). For more information about the window components and how to use them, see the “Getting Started” chapter in the *Workflow User Guide*.

You must change applications to set up a workflow for a different DNA Analytics program (CGH or ChIP). If you change applications, the Workflow tab is not available unless you have a valid license installed for that application. See [“To use Server Location to enter the license\(s\)”](#) on page 27 or [“To use Text License to enter the license\(s\)”](#) on page 27.

- 3 (Optional) To display the *Workflow User Guide*, click the **Help** tab, then click **Workflow**.

See also [“Setting Up and Running Workflows for Extraction and/or Analysis”](#) on page 39.

To start and find help to run workflows

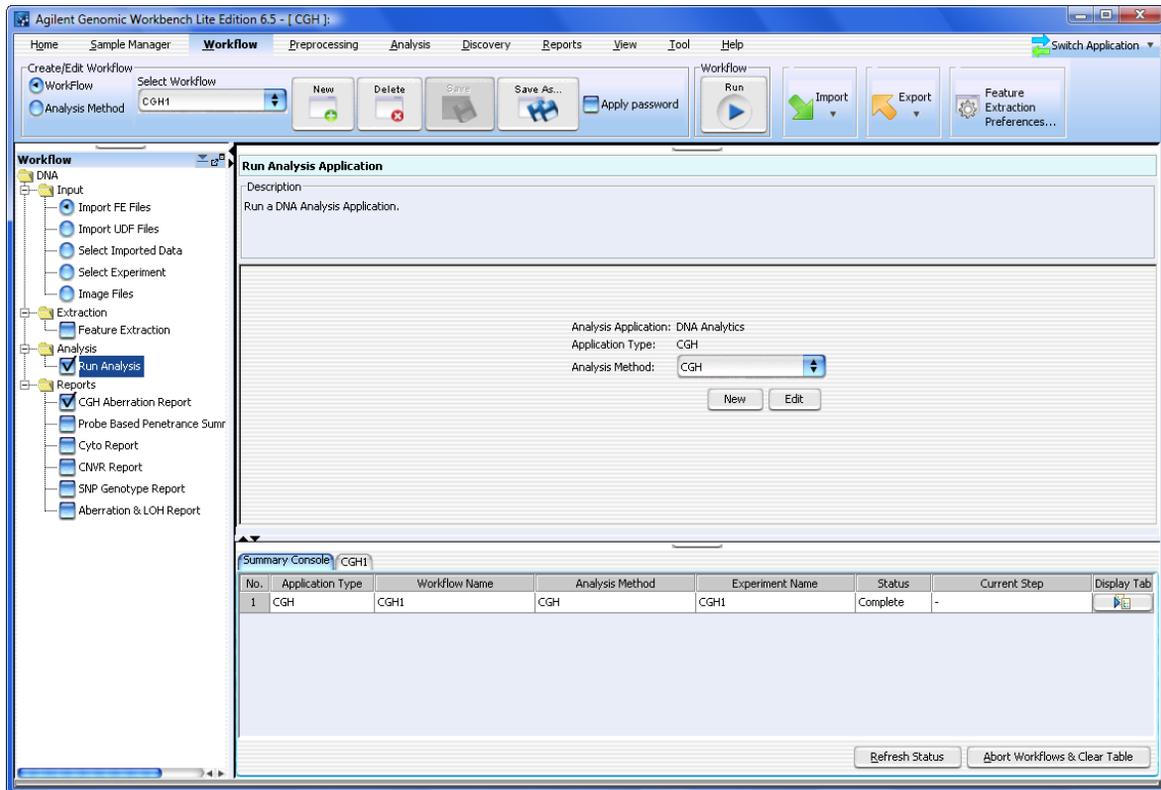


Figure 6 Agilent Genomic Workbench Lite Edition – Workflow tab

To start and find help to display CGH, ChIP, or Methylation (CH3) data

This application requires no license to display data only.

- 1 Click the **Genomic Viewer** tab at the top of the Agilent Genomic Workbench main window to display the Genomic Viewer pane. See [Figure 7](#). For more information about the window components and how to use them, see the “Getting Started” chapter in the *Data Viewing User Guide*.

You can now display the data for the application you selected when you started the program.

- 2 If you want to display another type of data, click **Switch Application** in the upper right corner of the application window, and click the appropriate application type.

You must change applications to view other data types because some of the options are different for each data type.

- 3 To display the *Data Viewing User Guide*, click the **Help** tab, then click **Data Viewing**.

See also “[Displaying Data/Results in Genomic Viewer](#)” on page 42.

NOTE

In the Switch Application menu, you actually select the “application type.” Your selection turns on the viewing capabilities that are available with or without a license for the selected data type.

To start and find help to display CGH, ChIP, or Methylation (CH3) data

The screenshot displays the Agilent Genomic Workbench Lite Edition 6.5 - [CGH]-Unlicensed Version interface. The ribbon at the top includes options like 'User Preferences...', 'Import', 'Export', 'Create Experiment', 'Save Experiment Result', 'GoTo Gene/Genomic location', and 'Print'. The 'Switch Application' button is highlighted in the top right. The main workspace is divided into several panes:

- Genomic Viewer:** Shows a chromosome ideogram with a selected region on chromosome 16.
- Chromosome View:** Displays a detailed view of the selected region on chromosome 16, showing bands and a red signal track.
- Gene View:** Shows a scatter plot of data points for the selected region, with a 'ScatterPlot' dropdown menu.
- Genome View:** Shows a genome-wide view of the data.
- Tab View:** A table at the bottom displays a list of arrays with columns for ProbeName, ChrName, Start, Stop, FeatureNum, and log2 ratios for two arrays (US2250270 and US22502705).

Other panes include 'Data' (showing a file tree), 'Experiment' (showing a folder structure), and 'My Entity List' (showing entities like Gene List and Tracks). The status bar at the bottom shows 'hg19 log2 ratio Selected Row = 1186 4605 x 7'.

Figure 7 Agilent Genomic Workbench Lite Edition – Genomic Viewer tab for unlicensed CGH application type

2 Getting Started

To start and find help to analyze CGH, ChIP or Methylation (CH3) data interactively

To start and find help to analyze CGH, ChIP or Methylation (CH3) data interactively

This application requires a DNA Analytics license (CGH, ChIP, or Methylation) for analysis of data.

- 1 (Optional) To view the *CGH* or *ChIP Interactive Analysis User Guide* or the *Methylation (CH3) Analysis User Guide*, click **Help** next to the description of DNA Analytics in the Open Application tab.

The appropriate User Guide is displayed.

- 2 In the Open Application tab, click **License** next to the description of DNA Analytics.

The License tab of the User Preferences dialog box appears.

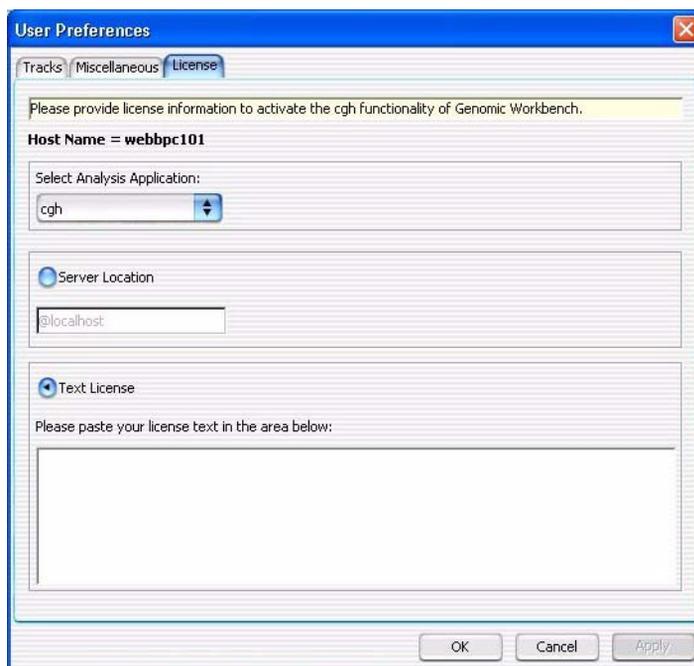


Figure 8 License tab of the User Preferences dialog box

To start and find help to analyze CGH, ChIP or Methylation (CH3) data interactively

You can enter your license information in one of two ways:

- Let Agilent Genomic Workbench automatically read the license(s) – uses Server Location shown in [Figure 8](#)
- Copy the text for the license(s) into the box – uses Text License shown in [Figure 8](#)

To use Server Location to enter the license(s)

- 1 Under Select Analysis Application, select the application for the license you wish to enter. (See [Figure 8](#) on page 26.)
- 2 Unzip all your DNA Analytics text license file(s) into a folder on your computer or server. Make sure this is a folder to which the program has access. Copy this path to the Clipboard.
- 3 In the License tab of the User Preferences dialog box, click **Server Location**. Paste the license folder path into the box below Server Location. (To paste the license for both Windows and Mac computers, hold down the **Ctrl** key and press **V**.)
- 4 Click **Apply**. Agilent Genomic Workbench automatically reads the license for the selected application.
- 5 If you have no other license, click **OK**.
If you have another license, repeat [step 1](#) and [step 4](#).

To use Text License to enter the license(s)

- 1 Under Select Analysis Application, select the application for the license you wish to enter. (See [Figure 8](#) on page 26.)
- 2 Find the folder that contains the DNA Analytics program license.
- 3 Double-click the license name to open the file in Notepad (or open the file using a text editor).
- 4 Copy the text to the Clipboard.
- 5 In the User Preferences License tab, click **Text License**.
- 6 Paste the license information into the License text box. (To paste the license for both Windows and Mac computers, hold down the **Ctrl** key and press **V**.)
- 7 Click **Apply**.
- 8 If you have no other license, click **OK**.
If you have another license, repeat [step 1](#) through [step 7](#).

2 Getting Started

To start and find help to analyze CGH, ChIP or Methylation (CH3) data interactively

To open the program

- 1 In the Open Application tab, click the icon next to **DNA Analytics**.
The Genomic Viewer for the selected application – CGH, ChIP, or Methylation (CH3) – appears and the Preprocessing tab is selected. See [Figure 9](#). For more information about the window components and how to use them, see the “Getting Started” chapter in a DNA Analytics *User Guide* (CGH or ChIP Interactive Analysis or Methylation (CH3) Analysis).
- 2 (Optional) To change application type, click **Switch Application** in the upper right corner of the application window, and click the program for which you entered another license.

NOTE

In the Switch Application menu, you actually select the “application type.” Your selection then makes available all of the various programs for the application type. For example, when you select **CGH** as the application type, and you have a CGH license installed, you gain access to the CGH functionality of the Sample Manager program, the Workflow for CGH, and the CGH Interactive Analysis programs.

- 3 (Optional) To view a DNA Analytics *User Guide* (CGH or ChIP Interactive Analysis or Methylation (CH3) Analysis), click the **Help** tab, then click **Application Guide** in the ribbon.

To start and find help to analyze CGH, ChIP or Methylation (CH3) data interactively

The screenshot displays the Agilent Genomic Workbench Lite Edition 6.5 - [CGH]: SNP interface. The window title is "Agilent Genomic Workbench Lite Edition 6.5 - [CGH]: SNP". The ribbon menu includes tabs for Home, Sample Manager, Workflow, Preprocessing, Analysis, Discovery, Reports, View, Tool, and Help. A "Switch Application" button is highlighted in green. The interface is divided into several panes:

- Search:** A search bar with "Play" and "Next" buttons.
- Data:** A tree view showing folders for Data, Experiment, My Entities, and Genotypes.
- Genome View:** A grid of chromosomes 1-22, X, Y.
- Chromosome View:** A detailed view of chromosome 8 with cytobands p22, p21.2, q12.1, q13.2, q21.11, q21.13, q21.3, q22.2, q23.1, q23.3, q24.12, q24.21, q24.23.
- Gene View:** A scatter plot of gene expression data.
- Tab View:** A table of arrays with columns: ProbeName, ChrName, Start, Stop, FeatureNum, and log2 ratio.

ProbeName	ChrName	Start	Stop	FeatureNum	log2 ratio
A_16_P384...	chr8	78981633	78981692	88366	-0.071
A_18_P165...	chr8	79014545	79014604	15531	-0.223
A_16_P183...	chr8	79034685	79034944	384443	-0.137
A_16_P384...	chr8	79058540	79058599	245445	-0.040
A_16_P183...	chr8	79088572	79088631	254846	-0.129
A_16_P183...	chr8	79123782	79123841	278252	0.059
A_16_P183...	chr8	79123782	79123841	73353	0.056
A_16_P183...	chr8	79123782	79123841	401005	-0.098
A_16_P183...	chr8	79123782	79123841	299941	0.017

The status bar at the bottom shows "hg18 log2 ratio Selected Row = 7482 13856 x 6".

Figure 9 Agilent Genomic Workbench Lite Edition – Genomic Viewer tab for CGH application type

To start and find help to assess the effectiveness of the SureSelect Target Enrichment System

This application requires no license.

- 1 Click **Switch Application** in the upper right corner of the application window, and click **SureSelect Target Enrichment**.
- 2 (Optional) To display the *SureSelect Quality Analyzer User Guide*, in the Open Application tab, click **Help** next to the description of the SureSelect Quality Analyzer.
- 3 Click the **Quality Analyzer** tab.

The Quality Analyzer window appears. See [Figure 10](#). For more information about the window components and how to use them, see the “Getting Started” chapter in the *SureSelect Quality Analyzer User Guide*.

See also “[Using SureSelect Quality Analyzer](#)” on page 47.

To start and find help to assess the effectiveness of the SureSelect Target Enrichment System

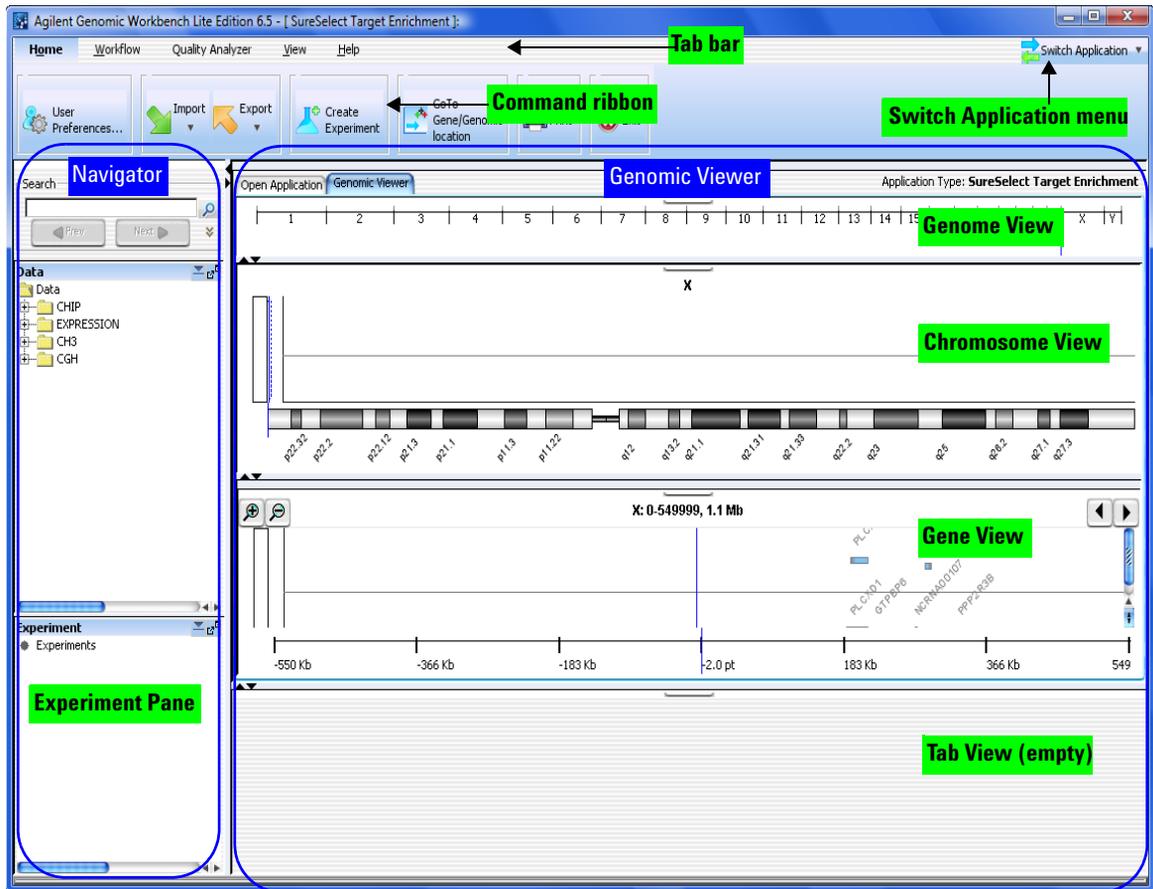


Figure 10 Agilent Genomic Workbench Lite Edition main window – Quality Analyzer tab for SureSelect Target Enrichment application

Getting Help Within the Applications

To get help within Agilent Genomic Workbench

The program has several built-in help resources, described in the following table. Help guides open in Adobe® Reader®.

Help resource	Description/instructions
Data Viewing User Guide	<p>This guide supplies comprehensive help on all available Data Viewing tasks. You can access it easily from anywhere within the program.</p> <ol style="list-style-type: none">1 In any tab of Agilent Genomic Workbench Lite Edition, click the Help tab.2 On the Help Ribbon, click Data Viewing. The Agilent Genomic Workbench Lite Edition Data Viewing Guide opens.
User Guides	<p>The Help tab in the program lets you view any of the available user guides that apply to the currently selected application type.</p> <ol style="list-style-type: none">1 Set the desired application type.2 In the Agilent Genomic Workbench tab bar, click Help. The names of the available user guides appear in the command ribbon. See Figure 11 for an example.3 Click the desired user guide. The selected user guide opens.
Product Overview Guide	<p>An additional guide, which you are currently reading, gives an overview of the capabilities within Agilent Genomic Workbench and describes how to start and find help for all of the programs. In addition, it helps you with system administration and troubleshooting.</p> <ol style="list-style-type: none">1 In any tab of Agilent Genomic Workbench, click the Open Application tab.2 At the upper right corner of the Open Application tab, click Product Overview.

Help tab

The Help tab contains commands that open the user guides that are available for Agilent Genomic Workbench Lite Edition. It also contains additional commands that let you display version and license information for the program, view information about the software updates that have been installed, and check the Agilent Web site for software updates that are available.

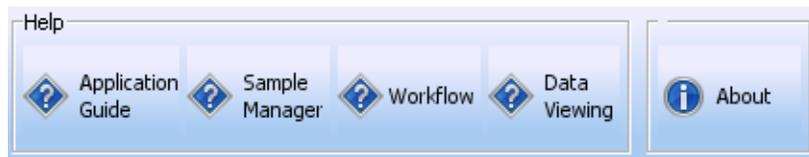


Figure 11 Help ribbon for CGH and CHIP applications

Help buttons

These buttons let you view the user guides that are available for Agilent Genomic Workbench Lite Edition. The guides that are available vary by application type. Each opens in Adobe® Reader®. These buttons can appear:

Button	Description
Application Guide	<p>For each of these application types, this button opens the indicated user guide:</p> <ul style="list-style-type: none"> • CGH – Opens the <i>CGH Interactive Analysis User Guide</i>. This guide describes how to use the CGH application of Agilent Genomic Workbench to analyze comparative genomic hybridization data and create reports. • ChIP-on-chip – Opens the <i>ChIP Interactive Analysis User Guide</i>. This guide describes how to use the ChIP application of Agilent Genomic Workbench to analyze chromatin immunoprecipitation data and create reports. • CH3 – Opens the <i>Methylation (CH3) Analysis User Guide</i>. This guide describes how the use the Methylation (CH3) application of Agilent Genomic Workbench to apply algorithms that help identify methylated regions. • SureSelect Target Enrichment – Opens the <i>SureSelect Quality Analyzer User Guide</i>. This guide describes how to use the SureSelect Quality Analyzer application of Agilent Genomic Workbench to assess the effectiveness of fragment pull-down for target enrichment experiments
Sample Manager	<p>(Available for all application types except SureSelect Target Enrichment) Opens the <i>Sample Manager User Guide</i>. This guide describes how to use the Sample Manager module of Agilent Genomic Workbench to organize microarrays and edit their attributes.</p>
Workflow	<p>(Available for the CGH and ChIP-on-chip application types) Opens the <i>Workflow User Guide</i>. This guide describes how to use the workflow module of Agilent Genomic Workbench to extract image files with Agilent Feature Extraction software and/or analyze data using the CGH and ChIP analysis applications.</p>

Button	Description
Data Viewing	(Available for all application types) Opens the <i>Data Viewing User Guide</i> . This guide describes how to import, organize, manage, export, and display data and other content (experiments, gene lists, tracks) within Agilent Genomic Workbench. It is targeted for users who have no DNA Analytics application license(s).
About	Opens a dialog box that displays version and copyright information for your installation of Agilent Genomic Workbench Lite Edition. You can also use this dialog box to display the License Agreement for the Agilent Genomic Workbench software.

To get help with the eArray Web site

The eArray Web site contains a comprehensive online Help system that describes how to use the Web site and all of its available tools. You do not need to log in to the site, or be a registered user on the site to view the online Help.

- 1 In your Web browser, go to <https://earray.chem.agilent.com>.

The login page of the eArray Web site appears.

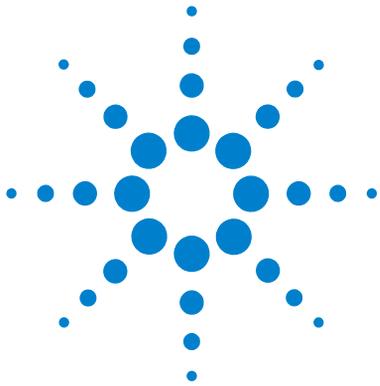
- 2 At the top of the page, click **Help**.

The online Help system for the eArray Web site opens in a new window.

In addition, Info links appear throughout the site that give additional details and instructions about selected parameters, criteria, and commands. Click [Info](#) where it appears.

2 Getting Started

To get help with the eArray Web site



3

Detailed Descriptions

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This chapter gives you more details about each of the modules within the Lite Edition of Agilent Genomic Workbench 6.5.



Organizing and Assigning Array Attributes with Sample Manager

When you scan a hybridized microarray slide, the Agilent Microarray Scanner creates a *.tif image file that includes the Array ID from the slide. This Array ID, contained in the header of the *.tif file, uniquely identifies the slide. A microarray slide can contain one array, or for multi-pack arrays, multiple replicate arrays on the same slide. The scanned data from each microarray represents a particular sample, and each array on a slide has a unique Array ID; thus each sample has a specific Array ID.

In Sample Manager, you define and edit the sample attributes for each Array ID, such as the biological sample used, the amount of label used, or the hybridization date. See the *Sample Manager User Guide* for more information.

The attributes that you assign for each Array ID stay with the Array ID for the rest of the analysis in Agilent Genomic Workbench. This sample information can appear in reports. By default, the Global Display Name is the same as the extraction name. However, you can change the Global Display Name and your new name will appear throughout the rest of the user interface for that array (for example, in experiments, reports, etc.).

You can export attributes to an Attribute File. Later, you can import the file into Agilent Genomic Workbench to use the saved attributes.

An Attribute File contains identification information (Array ID) and attributes for one or more microarray samples. An Attribute File for a two-color experiment must at a minimum include Array ID, Global Display Name, Red Sample, Green Sample, and Polarity values. An Attribute File for a one-color experiment must include the Array ID, Global Display Name, and Green Sample. Other information may be included as well. You can create Attribute Files with a spreadsheet program, and save them as a tab-delimited text file or *.xls file.

Setting Up and Running Workflows for Extraction and/or Analysis

You can set up and run a workflow that runs Feature Extraction to automatically create log ratio data from the scanned images of your arrays. You can use this capability for many types of arrays, including CGH, ChIP, and Methylation (CH3). If you have a CGH or ChIP license, you can also set up the workflow to analyze the CGH or ChIP log ratio data right after the extraction.

NOTE

You must have Feature Extraction version 10.7 or higher installed in order to run a Feature Extraction workflow using Agilent Genomic Workbench Lite Edition 6.5. To run a Feature Extraction workflow for CGH+SNP microarrays, you must have Feature Extraction version 10.10 or higher installed.

Optionally, you can use Sample Manager to organize your microarrays and define their attributes, before or after running a workflow. See [“Organizing and Assigning Array Attributes with Sample Manager”](#) on page 38 for more information.

Agilent Genomic Workbench lets you set up and run a workflow with no need for intervention ([Figure 12](#)).

3 Detailed Descriptions

Setting Up and Running Workflows for Extraction and/or Analysis

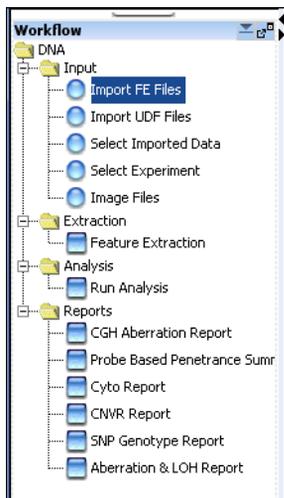


Figure 12 Workflow Navigator for CGH

You can:

- Select from a variety of input files for the workflow: import extracted data, select imported data already in the database, use files from a selected experiment, or select image files to extract
- Extract the data (from image files) using Feature Extraction
- Analyze extracted data with a CGH or ChIP analysis method
- Apply a design, feature, or array filter to the extracted data to include or exclude arrays based on user-selected criteria (CGH application only)
- Save Workflows and Analysis methods for later use
- Do one of the following:
 - Run the configured workflow to extract image files with Agilent Feature Extraction and create a QC report that contains statistics for the extraction
 - Run the configured workflow to analyze CGH, CGH+SNP, or ChIP (not Methylation) Feature Extraction data with DNA Analytics and create DNA Analytics reports
 - Run the configured workflow to extract image files and then analyze the extracted results to create both sets of reports.

Setting Up and Running Workflows for Extraction and/or Analysis

With Workflow analysis, you configure an analysis method ahead of time and then run it in a workflow. This method is useful for unattended operation and consistent analyses of multiple data sets. To review workflow results, you use the Genomic Viewer. For more details and quick-start instructions to set up and run a workflow, see the *Workflow User Guide*.

Displaying Data/Results in Genomic Viewer

What is Genomic Viewer?

Genomic Viewer is the graphics and tabular display section of the Agilent Genomic Workbench main window. You can use this data *viewing* capability in Agilent Genomic Workbench without a license to view data for many types of arrays, including CGH, CGH+SNP, ChIP, and Methylation (CH3). You can use the data *analysis* capability in Agilent Genomic Workbench only if you have a license for one or more of the DNA Analytics programs (CGH, ChIP, or Methylation).

Figure 13 shows the main window of Agilent Genomic Workbench Lite Edition, and identifies the names of its components. In the Genomic Viewer, extracted data and analysis results are tabulated and displayed next to depictions of the genome, selected chromosome, and selected genes of the species whose array data you are analyzing.

Genomic Viewer contains four main views:

- **Genome View** – A graphical representation of the entire genome for the selected species. Use this view to select the chromosome to show in the other views.
- **Chromosome View** – A graphical representation of the selected chromosome, displayed with cytobands and a plot area. Click or drag the mouse to select a region to display in the Gene View.
- **Gene View** – A more detailed view of the chromosomal region selected in the Chromosome View.
- **Tab View** – A view that displays design annotation, log ratio data, and optional signal data related to the chromosome you select in Chromosome View

To learn how to display log ratio data and content without a license (experiments, gene lists and tracks), see the *Data Viewing User Guide*.

To learn how to display log ratio data, content, and results, see the *User Guide* for which you have a DNA Analytics program license.

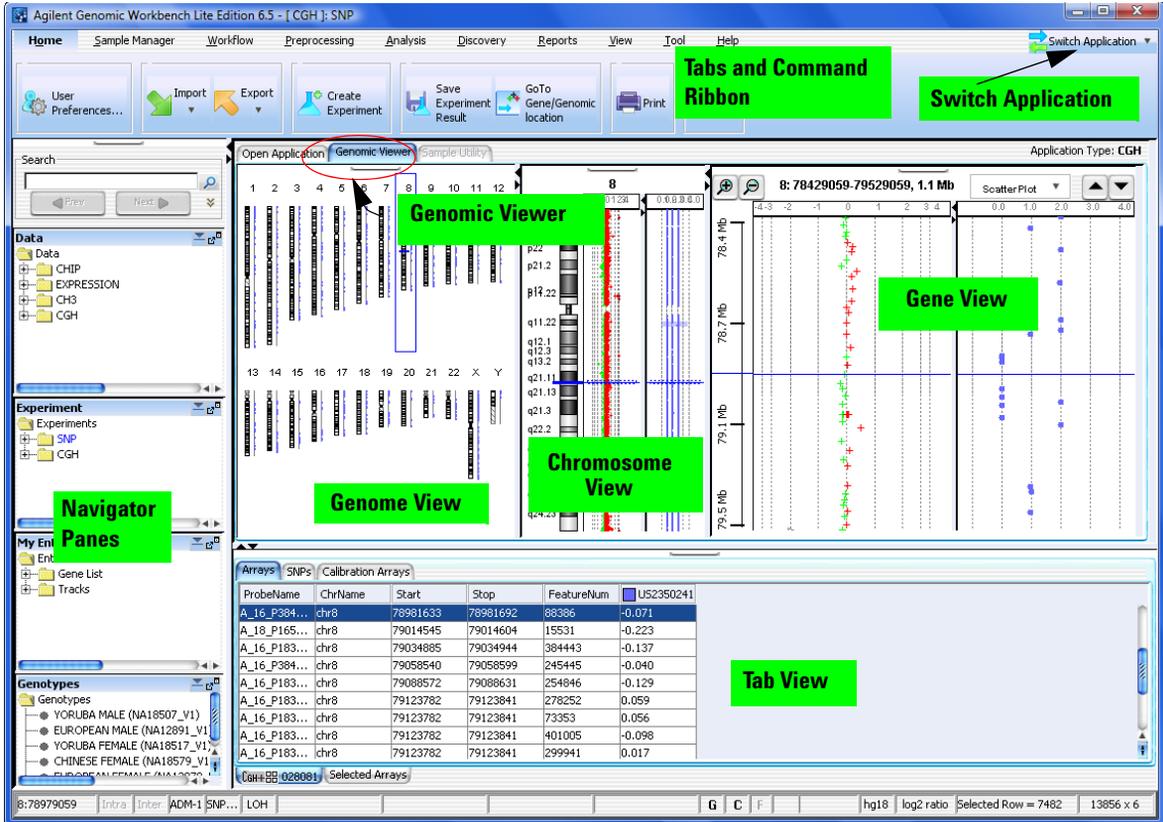


Figure 13 Agilent Genomic Workbench Lite Edition main window with major components – CGH

Analyzing CGH Data Interactively

The Agilent Genomic Workbench lets you visually explore, detect, and analyze aberration patterns from multiple Comparative Genomic Hybridization (CGH) microarray profiles, either interactively or by setting up an analysis method to use with a workflow. With CGH Interactive Analysis, you set up an experiment and apply analysis algorithms to your data one experiment at a time. You also set parameters for some report templates for use in Workflow analysis.

You do most of your operations with the Preprocessing, Analysis, Discovery and Reports tabs. See [Table 1](#) on page 12. Also see the *CGH Interactive Analysis User Guide*.

With the interactive portion of the Agilent Genomic Workbench, you can:

- Import Agilent Feature Extraction data, Axon data, or UDF files and use the Genomic Viewer to visualize this data along the chromosome
- Preprocess the data:
 - Apply feature, array, and design filters
 - Combine designs and/or inter- or intra-array replicates
 - Centralize the data, including nonunique probes
 - Display QC metrics on the original data
- Use five different robust statistical aberration detection algorithms to detect and map aberration regions with high confidence
- Display chromosomal deletions and amplifications at multiple zoom levels simultaneously
- Use Discovery options to display the analysis results in many different ways
 - Apply aberration filters on the results
 - Use the CGH program for CNV (Copy Number Variation) applications
 - Find common aberrations between several CGH samples, as well as do a differential aberration analysis
 - Do a correlation analysis of gene expression and CGH data, do a cluster analysis, and display data with a “heatmap”
- Save aberration results
- Make customizable Cytogenetic Reports for individual CGH samples

Analyzing ChIP Data Interactively

The ChIP program is a complete environment that lets you analyze chromatin immunoprecipitation (ChIP) microarray data. ChIP microarray analysis can identify the genomic loci that contain proteins that bind to DNA, including individual transcription factors, chromatin modifiers, and components of the general transcription machinery.

With ChIP interactive analysis, you set up an experiment and apply analysis algorithms to your data one experiment at a time. You also set parameters for some report templates for use in Workflow analysis.

You do most of your operations with the Preprocessing, Analysis, and Reports tabs. See [Table 1](#) on page 12. Also see the *ChIP Interactive Analysis User Guide*.

With the interactive portion of the ChIP program, you can:

- Import data from the Agilent Feature Extraction and Axon programs
- Use an intuitive graphical interface to display data and annotations in the context of an organism's genome, at several simultaneous levels of detail
- Normalize your data with several statistical methods
- Use error modeling and event detection algorithms to identify probes, genes, and genomic loci that have significant binding
- Display significant binding events graphically within the program, and export report files that you can analyze further with other programs

Analyzing Methylation (CH3) Data

The Methylation (CH3) program is a framework used to identify methylation events in your samples. CH3 microarray analysis can help identify the CpG islands where methylation has occurred. The software helps you:

- Identify molecular events associated with DNA methylation
- Find and validate gene regulation and regulatory networks by creating high-resolution, genome-wide methylation profiles
- Show modes of action and potential therapeutic activities of compounds and target genes by understanding the relation of DNA methylation to transcriptional control

With the Methylation (CH3) program, you can:

- Import data from the Agilent Feature Extraction and Axon programs, and import UDF files
- Use an intuitive graphical interface to display data and annotations in the context of an organism's genome, at several simultaneous levels of detail
- Use a *Z*-score or Batman algorithm to calculate the probability that the regions covered by specific probes are methylated or unmethylated
- Compare the moving average of your log ratio and *Z*-score data within the boundaries of CpG Island tracks

You cannot run methylation event detection within a Workflow.

Using SureSelect Quality Analyzer

SureSelect Quality Analyzer is a program that lets you assess the effectiveness of the pull-down of targeted genomic fragments when you use the Agilent SureSelect Target Enrichment system. This system uses libraries of biotinylated RNA oligonucleotide “baits” to harvest genomic DNA fragments of interest for sequencing. This forms the basis of a powerful selection method that lets you focus your sequencing efforts.

The selective nature of the process makes it ideal for targeted resequencing using next-generation sequencing technology. After you sequence the harvested fragments, you can use SureSelect Quality Analyzer to calculate statistical metrics and perform read depth analysis. You can also perform enrichment analysis, and view the results in the UCSC Genome Browser next to the annotation tracks of your choice.

For more information, see the *Agilent Genomic Workbench Lite Edition SureSelect Quality Analyzer User Guide*.

3 Detailed Descriptions

Using SureSelect Quality Analyzer

www.agilent.com

In this book

This book gives an overview of the capabilities within the Agilent Genomic Workbench Lite Edition 6.5. It also describes how to start each of the component programs and find Help, and how to enter your license information.

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