

# **GeneMapper® *ID* Software** **Versions 3.1 and 3.2** **Human Identification Analysis**

Tutorial

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Part Number 4357520 Rev. A

11/2004

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# Preface

## How to Use This Tutorial

**Purpose of This Tutorial** The Applied Biosystems GeneMapper® ID Software Versions 3.1 and 3.2 Human Identification Analysis Tutorial provides example workflows for using GeneMapper ID Software versions 3.1 and 3.2.

The tutorial consists of casework analysis and database analysis using data provided with the software from AmpFℓSTR® PCR Amplification Kits. The settings included in this tutorial should serve as a guideline for subsequent analysis. For detailed information on the features and capabilities of GeneMapper ID Software version 3.2, including support of the AmpFℓSTR® Yfiler™ PCR Amplification Kit, please refer to the GeneMapper ID Software Version 3.2 User Bulletin (PN 4352543).

**Audience** This tutorial is intended for novice GeneMapper *ID* software users who use the software to analyze forensic casework and single-source or parentage samples amplified with the AmpFℓSTR PCR Amplification Kits.

**Text Conventions** This guide uses the following conventions:

- **Bold** indicates user action. For example:  
Type **0**, then press **Enter** for each of the remaining fields.
- *Italic* text indicates new or important words and is also used for emphasis. For example:  
Before analyzing, *always* prepare fresh matrix.
- A right arrow bracket (>) separates successive commands you select from a drop-down or shortcut menu. For example:  
Select **File > Open > Project**.  
Right-click the sample row, then select **View Filter > View All Runs**.

## User Attention Words

Two user attention words appear in Applied Biosystems user documentation. Each word implies a particular level of observation or action as described below:

**Note:** Provides information that may be of interest or help but is not critical to the use of the product.

**IMPORTANT!** Provides information that is necessary for proper instrument operation, accurate chemistry kit use, or safe use of a chemical.

Examples of the user attention words appear below:

**Note:** The size of the column affects the run time.

**Note:** The Calibrate function is also available in the Control Console.

**IMPORTANT!** To verify your client connection to the database, you need a valid Oracle user ID and password.

**IMPORTANT!** You must create a separate Sample Entry Spreadsheet for each 96-well microtiter plate.

## How to Obtain More Information

### Related Documentation

The following related documents are available:

- *GeneMapper® ID Software Version 3.1 User Guide*  
(PN 4338775)
- *GeneMapper® ID Software Version 3.2 User Bulletin*  
(PN 4352543)
- *AmpFSTR® Yfiler™ PCR Amplification Kit Users Manual*  
(PN 4358101)

Portable document format (PDF) versions of this tutorial and the user guide are also available on the GeneMapper ID software installation CD. PDF versions of the other tutorials are available on the Applied Biosystems Web site. See “How to Obtain Support” on page v.

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At the Support page, you can:

- Search through frequently asked questions (FAQs)
- Submit a question directly to Technical Support
- Order Applied Biosystems user documents, MSDSs, certificates of analysis, and other related documents
- Download PDF documents
- Obtain information about customer training
- Download software updates and patches

In addition, the Support page provides access to worldwide telephone and fax numbers to contact Applied Biosystems Technical Support and Sales facilities.





# Software Setup

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# 1

This chapter covers:

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## Before You Start

### Considerations for HID Analysis

When using GeneMapper® ID Software version 3.1 to perform Human Identification (HID) analysis with AmpFℓSTR® kits, consider the following:

- HID analysis requires the presence of at least one allelic ladder sample per project. Your laboratory can use multiple ladder samples in an analysis, provided individual laboratories conduct the appropriate validation studies.

Samples are genotyped according to the allelic bins calculated from ladder(s) within the same run folder. When multiple run folders are imported into a project, each sample is genotyped according to the ladders with the respective run.

- Allelic ladder samples in a single run folder are considered to be from a single run.

When the software imports multiple run folders into a project, only ladders within a single run folder are used for calculating allelic bin offsets and subsequent genotyping. Multiple ladders within a single run folder are averaged by the software to create bin offsets.

- Allelic ladder samples need to be identified as “Allelic Ladder” in the Sample Type column in a project. Failure to make this setting for ladder samples results in failed analysis.
- Allelic bin definitions are stored in the AmpFℓSTR panels in the Panel Manager.
- Lanes or injections containing the allelic ladder should be analyzed with the same analysis method and parameters used for samples.
- Alleles not found in the AmpFℓSTR Allelic Ladders do exist. These off-ladder alleles may contain full and/or partial repeat units. An off-ladder allele is defined as an allele falling outside of the  $\pm 0.5$ -bp bin window of any known allelic ladder allele or virtual bin.

**Note:** If a sample allele peak is called as an off-ladder allele, then run the sample again to verify the result.

- The marker-specific stutter ratios included in the kit panels and cutoff value used in the tutorial serve as a tool and a guideline. Base final conclusions on careful examination of the STR profiles.

**Note:** GeneMapper® *ID* Software version 3.1 has undergone a verification process defined by Applied Biosystems. However, human identification laboratories that choose to use GeneMapper *ID* Software to analyze forensic, paternity, databasing, and single-source samples, should perform their own appropriate validation studies.

## Software Setup Tutorial Overview

Perform the following tasks before you analyze fragment sample (.fsa) files for HID the first time.

1. Import panels and bins into the Panel Manager (page 1-5).
2. Create an analysis method with the appropriate bin set option (page 1-8).
3. Define custom views of analysis tables (page 1-16).
4. Define custom views of plots (page 1-19).
5. View and set HID analysis options (page 1-27).
6. If necessary, convert any GeneScan® software sample files generated on the Macintosh® platform to the .fsa format using the Mac-to-Win AppleScript® software provided with GeneMapper *ID* software. Conversion is described in the *GeneMapper® ID Software Version 3.1 User Guide*.

## Panels and Bins

**Overview** In this tutorial, you import panels and bin sets in the form of text files (.txt extension) provided for use with the AmpF $\mathcal{L}$ STR kits and included with the GeneMapper<sup>®</sup> ID software.

For information on creating new panels, see the Panel Manager Window Commands section in the *GeneMapper<sup>®</sup> ID Software Version 3.1 User Guide*.


**Definitions** The following table lists some terms and definitions that are used often in this tutorial.

Term	Definition
Kit	Collection of panels. For example, an AmpF $\mathcal{L}$ STR kit is a panel.
Panel	Collection of markers (A primer set specific to an AmpF $\mathcal{L}$ STR kit). For example, Identifiler has 16 markers.
Marker	A loci or primer pair. For example, THO1 or vWA. The marker information contains the: <ul style="list-style-type: none"><li>• Name</li><li>• Dye color</li><li>• 9947A control alleles</li><li>• Min. and max. size</li><li>• Marker specific stutter ratio</li><li>• Allelic ladder alleles</li></ul>
Bin	An expected location for a particular allele within a marker.
Binset	Collection of expected locations for markers contained within a panel or kit.

## Importing Panels and Bin Sets

Use this procedure to import panels and bin sets into the GeneMapper database for subsequent analysis and to view imported panels, markers, and bins. Import the panels and bin sets the first time you use the software and when updated versions of panels and bin sets are provided.

### To import panels and bin sets:

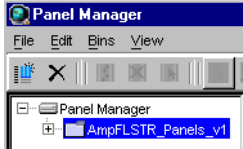
1.	<p>Start the GeneMapper <i>ID</i> software:</p> <ol style="list-style-type: none"> <li>Select <b>Start &gt; Programs &gt; Applied Biosystems &gt; GeneMapper &gt; GeneMapper ID</b>.</li> <li>In the login box, leave the Password box empty if this is the first time you are launching the software. If not, enter your password.</li> <li>The first time you start the software, you are prompted to change the password. When the password dialog box opens: <ul style="list-style-type: none"> <li>– Leave the Old Password box blank.</li> <li>– Type a the New Password.</li> <li>– Type the new password again to verify it.</li> </ul> <p>The GeneMapper Project window opens with a blank, untitled project.</p> </li> </ol>
2.	<p>Select <b>Tools &gt; Panel Manager</b> to open the Panel Manager.</p>
3.	<p>Locate and open the folder containing the panels and bins:</p> <ol style="list-style-type: none"> <li>Select <b>Panel Manager</b> in the navigation pane.</li> </ol> <div data-bbox="548 1119 1085 1275" style="border: 1px solid black; padding: 5px;">  </div> <ol style="list-style-type: none"> <li>Select <b>File &gt; Import Panels</b> to open the Import Panels dialog box.</li> </ol>

To import panels and bin sets: *(continued)*

4. Import **AmpFLSTR\_Panels\_v1**:
  - a. Select **AmpFLSTR\_Panels\_v1.txt**.
  - b. Click **Import**.

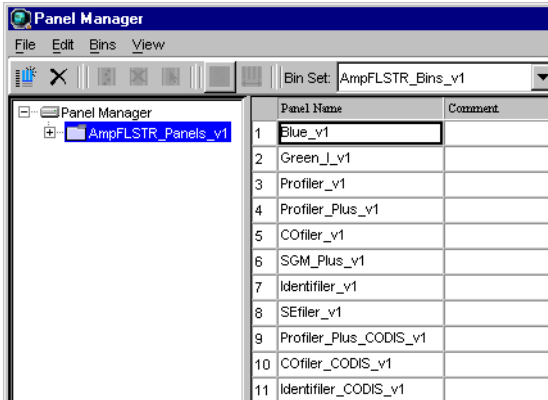
**Note:** Importing this file creates a new folder in the navigation pane of the Panel Manager, AmpFISTR\_Panels\_v1, containing the panels and associated markers.

5. Import **AmpFLSTR\_Bins\_v1.txt**:
  - a. Select the **AmpFLSTR\_Panels\_v1** folder in the navigation pane.



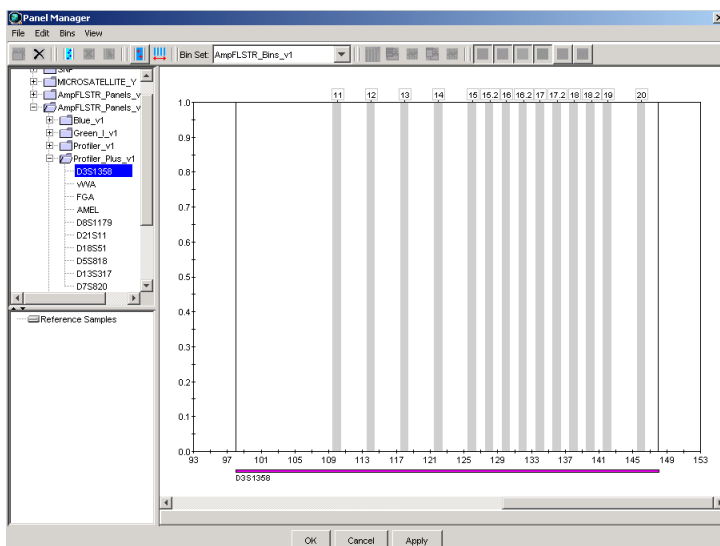
- b. Select **File > Import Bin Set** to open the Import Bin Set dialog box.
  - c. Select **AmpFLSTR\_Bins\_v1.txt**.
  - d. Click **Import**.

**Note:** Click **Apply** or **OK** to make this file associate the bin set with the panels in the AmpFISTR\_Panels\_v1 folder.



To import panels and bin sets: *(continued)*

6. View the markers and display the Bin view:
  - a. Select the **Profiler\_Plus\_v1** folder in the navigation pane to display the list of markers it contains in the right pane.
  - b. Double-click the **Profiler\_Plus\_v1** folder in the navigation pane to display the list of markers below it.
  - c. Select **D3S1358** in the navigation pane to display the Bin view for the marker in the right pane.



7. Click **Apply** to add the Profiler\_Plus\_v1 panel to the project window.
8. Click **OK** to close the Panel Manager.  
**IMPORTANT!** If you close the Panel Manager without clicking OK, the panels and bins will not be available for analysis.

**What's Next?**

You have now imported the necessary panel, marker, and bin information that you need for this tutorial and subsequent analysis. You are ready to continue setting up the software.

# HID Analysis Methods

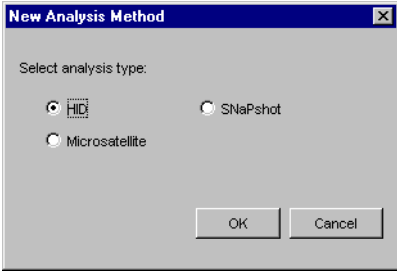
**Overview** In this tutorial, you create two analysis methods suitable for Human Identification (HID) analysis:

- **HID\_Classic** – Used in Chapter 2, “Casework Analysis,” for processing casework. This training analysis method includes GeneScan® analysis parameters for Macintosh computers, and maker-specific stutter percentages.
- **HID\_Advanced** – Used in Chapter 3, “Database Analysis,” for processing database samples. This training analysis method includes GeneScan analysis parameters for the Microsoft Windows NT operating system, and a 20% stutter filter (similar to Kazam 20%).

**Note:** These analysis methods are examples for training. When analyzing your own data, adjust the analysis range and peak amplitude thresholds for your samples and validation.

## Creating Analysis Methods for HID Classic

To create analysis methods for HID Classic:

1.	Select <b>Tools &gt; GeneMapper Manager</b> to open the GeneMapper Manager.
2.	Create an analysis method called HID_Classic: a. Select the <b>Analysis Methods</b> tab and click <b>New</b> to open the New Analysis Method dialog box.  b. Select <b>HID</b> and click <b>OK</b> to open the Analysis Method Editor with the General tab selected.



To create analysis methods for HID Classic: *(continued)*

- |    |                                                                                                                                                                                                                 |
|----|-----------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------|
| 3. | Select the settings shown in Table 1-1 on page 1-10<br><b>IMPORTANT!</b> You must select your settings on all the tabs before you Click <b>OK</b> to save the analysis method and return to GeneMapper Manager! |
|----|-----------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------|

# HID\_Classic Settings

Table 1-1 HID\_Classic analysis method settings

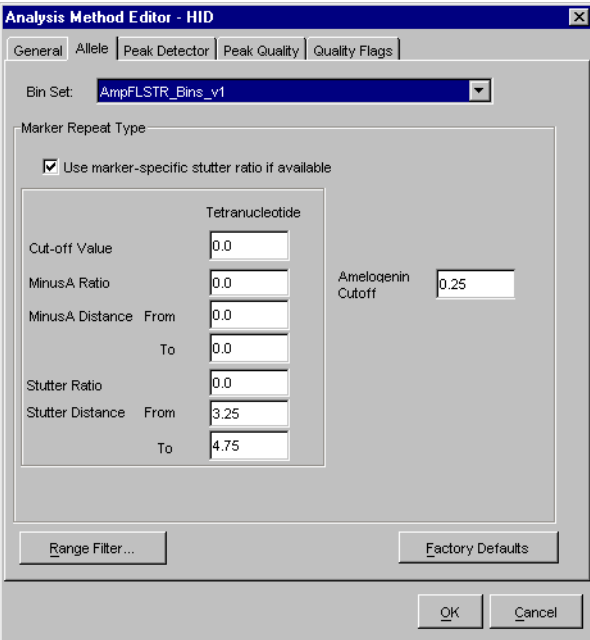
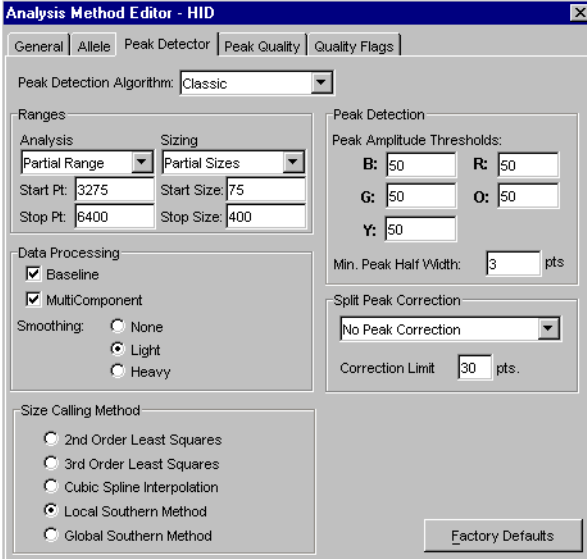
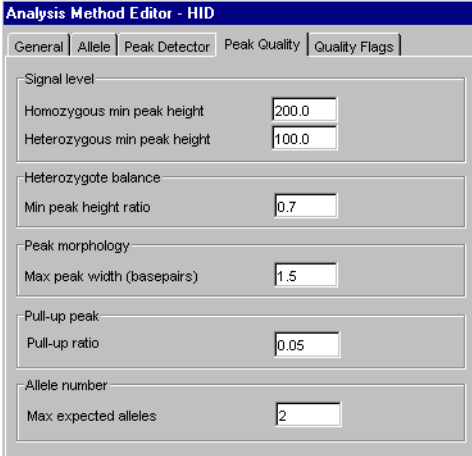
Tab	Settings
General	Name: HID_Classic
Allele	 <p><b>Note:</b> Stutter values included with kit panels are the same as those included in Genotyper template version 7. Values listed are simple ratios, rather than percent differences. For specific information on these values, see the appropriate user guide for your AmpFLSTR PCR Amplification kit.</p> <p><b>Note:</b> The values in the current macros do not match the values in the user manual. Genotyper template version 7 applies only to Macintosh computers.</p>

Table 1-1 HID\_Classic analysis method settings (continued)

Tab	Settings
Peak Detector	 <p><b>Note:</b> The Analysis Partial Range is defined for tutorial casework sample files.</p> <p><b>Note:</b> For more information, see Peak Detection Algorithms on page A-3.</p>
Peak Quality	

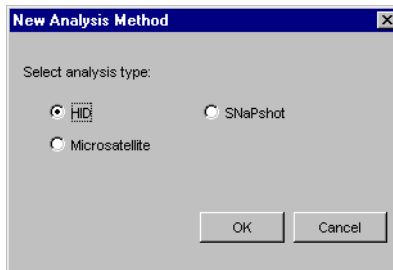
**Table 1-1** HID\_Classic analysis method settings (*continued*)

Tab	Settings																																												
Quality Flags	<p>Quality flag settings:</p> <div style="border: 1px solid gray; padding: 5px; background-color: #f0f0f0;"> <p>Quality weights are between 0 and 1.</p> <p>Quality Flag Settings</p> <table style="width: 100%; border-collapse: collapse;"> <tr> <td style="width: 50%;">Spectral Pull-up</td> <td style="width: 10%; text-align: center;">0.8</td> <td style="width: 20%;">Control Concordance</td> <td style="width: 10%; text-align: center;">1.0</td> <td style="width: 5%;"></td> </tr> <tr> <td>Broad Peak</td> <td style="text-align: center;">0.8</td> <td>Low Peak Height</td> <td style="text-align: center;">0.3</td> <td></td> </tr> <tr> <td>Out of Bin Allele</td> <td style="text-align: center;">0.8</td> <td>Off-scale</td> <td style="text-align: center;">0.8</td> <td></td> </tr> <tr> <td>Overlap</td> <td style="text-align: center;">0.8</td> <td>Peak Height Ratio</td> <td style="text-align: center;">0.3</td> <td></td> </tr> </table> </div> <p>PQV thresholds:</p> <div style="border: 1px solid gray; padding: 5px; background-color: #f0f0f0;"> <p>PQV Thresholds</p> <table style="width: 100%; border-collapse: collapse;"> <tr> <td colspan="2"></td> <td style="background-color: #90EE90; text-align: center;">Pass Range:</td> <td colspan="2"></td> <td style="background-color: #FFB6C1; text-align: center;">Low Quality Range:</td> </tr> <tr> <td>Sizing Quality:</td> <td>From</td> <td style="text-align: center;">0.75</td> <td>to</td> <td>1.0</td> <td>From 0.0 to</td> </tr> <tr> <td>Genotype Quality:</td> <td>From</td> <td style="text-align: center;">0.75</td> <td>to</td> <td>1.0</td> <td>From 0.0 to</td> </tr> <tr> <td></td> <td></td> <td></td> <td></td> <td></td> <td style="text-align: center;">0.25</td> </tr> </table> <p style="text-align: right; margin-top: 5px;">Factory Defaults</p> </div>	Spectral Pull-up	0.8	Control Concordance	1.0		Broad Peak	0.8	Low Peak Height	0.3		Out of Bin Allele	0.8	Off-scale	0.8		Overlap	0.8	Peak Height Ratio	0.3				Pass Range:			Low Quality Range:	Sizing Quality:	From	0.75	to	1.0	From 0.0 to	Genotype Quality:	From	0.75	to	1.0	From 0.0 to						0.25
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Broad Peak	0.8	Low Peak Height	0.3																																										
Out of Bin Allele	0.8	Off-scale	0.8																																										
Overlap	0.8	Peak Height Ratio	0.3																																										
		Pass Range:			Low Quality Range:																																								
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Genotype Quality:	From	0.75	to	1.0	From 0.0 to																																								
					0.25																																								

### Creating Analysis Methods for HID Advanced

To create analysis methods for HID Advanced:

1. In the GeneMapper Manager, create an analysis method called HID\_Advanced:
  - a. Select the **Analysis Methods** tab and click **New** to open the New Analysis Method dialog box.



- b. Select **HID** and click **OK** to open the Analysis Method Editor with the General tab selected.

To create analysis methods for HID Advanced: *(continued)*

2. Select the settings shown in Table 1-2, “HID\_Advanced analysis method settings.”  
**IMPORTANT!** You must select your settings on all the tabs before you Click **OK** to save the analysis method and return to GeneMapper Manager!

**HID\_Advanced Settings**

Table 1-2 HID\_Advanced analysis method settings

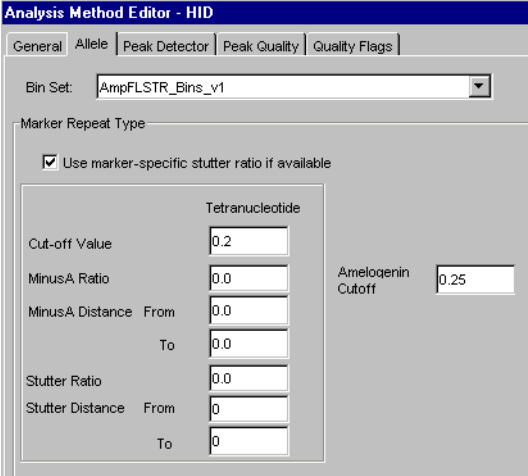
Tab	Settings
General	Name: HID_Advanced
Allele	 <p><b>Note:</b> For more information about the Cutoff Value setting, see page 3-2.</p>

Table 1-2 HID\_Advanced analysis method settings (continued)

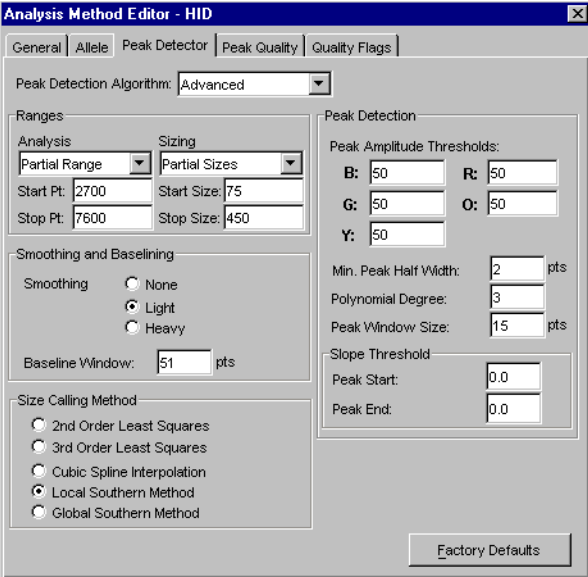
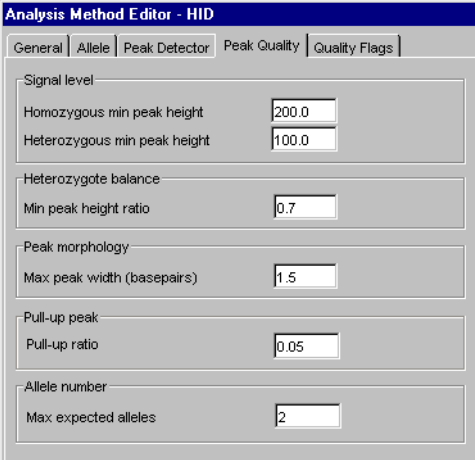
Tab	Settings
Peak Detector	 <p><b>Note:</b> The Analysis Partial Range is defined for tutorial database sample files.</p> <p><b>Note:</b> For more information, see “Peak Detection Algorithms” on page A-3.</p>
Peak Quality	

Table 1-2 HID\_Advanced analysis method settings (continued)

Tab	Settings																																												
Quality Flags	<p>Quality flag settings:</p> <div style="border: 1px solid gray; padding: 5px; background-color: #f0f0f0;"> <p>Quality weights are between 0 and 1.</p> <p>Quality Flag Settings:</p> <table style="width: 100%; border-collapse: collapse;"> <tr> <td style="width: 50%;">Spectral Pull-up</td> <td style="width: 10%; text-align: center;"><input type="text" value="0.8"/></td> <td style="width: 20%;">Control Concordance</td> <td style="width: 10%; text-align: center;"><input type="text" value="1.0"/></td> <td style="width: 5%;"></td> </tr> <tr> <td>Broad Peak</td> <td style="text-align: center;"><input type="text" value="0.8"/></td> <td>Low Peak Height</td> <td style="text-align: center;"><input type="text" value="0.3"/></td> <td></td> </tr> <tr> <td>Out of Bin Allele</td> <td style="text-align: center;"><input type="text" value="0.8"/></td> <td>Off-scale</td> <td style="text-align: center;"><input type="text" value="0.8"/></td> <td></td> </tr> <tr> <td>Overlap</td> <td style="text-align: center;"><input type="text" value="0.8"/></td> <td>Peak Height Ratio</td> <td style="text-align: center;"><input type="text" value="0.3"/></td> <td></td> </tr> </table> </div> <p>PQV thresholds:</p> <div style="border: 1px solid gray; padding: 5px; background-color: #f0f0f0;"> <p>PQV Thresholds</p> <table style="width: 100%; border-collapse: collapse;"> <tr> <td colspan="2"></td> <td style="background-color: #90EE90; text-align: center; color: white;">Pass Range:</td> <td colspan="2"></td> <td style="background-color: #FF6347; text-align: center; color: white;">Low Quality Range:</td> </tr> <tr> <td>Sizing Quality:</td> <td>From</td> <td style="text-align: center;"><input type="text" value="0.75"/></td> <td>to</td> <td>1.0</td> <td>From 0.0 to</td> </tr> <tr> <td>Genotype Quality:</td> <td>From</td> <td style="text-align: center;"><input type="text" value="0.75"/></td> <td>to</td> <td>1.0</td> <td>From 0.0 to</td> </tr> <tr> <td></td> <td></td> <td></td> <td></td> <td></td> <td style="text-align: center;"><input type="text" value="0.25"/></td> </tr> </table> <p style="text-align: right; margin-top: 5px;"><input type="button" value="Factory Defaults"/></p> </div>	Spectral Pull-up	<input type="text" value="0.8"/>	Control Concordance	<input type="text" value="1.0"/>		Broad Peak	<input type="text" value="0.8"/>	Low Peak Height	<input type="text" value="0.3"/>		Out of Bin Allele	<input type="text" value="0.8"/>	Off-scale	<input type="text" value="0.8"/>		Overlap	<input type="text" value="0.8"/>	Peak Height Ratio	<input type="text" value="0.3"/>				Pass Range:			Low Quality Range:	Sizing Quality:	From	<input type="text" value="0.75"/>	to	1.0	From 0.0 to	Genotype Quality:	From	<input type="text" value="0.75"/>	to	1.0	From 0.0 to						<input type="text" value="0.25"/>
Spectral Pull-up	<input type="text" value="0.8"/>	Control Concordance	<input type="text" value="1.0"/>																																										
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					<input type="text" value="0.25"/>																																								

# Analysis Tables

**Overview** In this tutorial, you define a custom view of Samples and Genotypes tables for viewing HID tutorial data. For subsequent analysis with your own samples, edit this table setting or create a new table setting.

**Creating a Table Setting** To create a table setting:

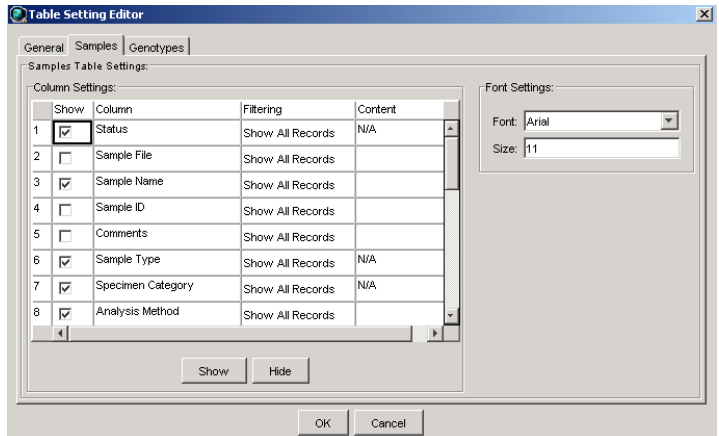
1.	From the <b>GeneMapper Manager</b> , select the <b>Table Settings</b> tab.
2.	Click <b>New</b> to open the Table Setting Editor with the General tab selected.
3.	Enter a name in the text box. For this tutorial, type <b>HID Table</b> .



To create a table setting: *(continued)*

4. Select the **Samples** tab and make the following selections:
- Column settings:
    - Show: 1, 3, 6–13, 17, 18, 22, 23
    - Hide: 2, 4, 5, 14–16, 19–21, 24, 25

**Note:** The box in the Show column indicates whether columns are shown or hidden in the table. Check the box to show the column and deselect the box to hide the column.
  - Font settings:
    - Font: **Arial**
    - Size: **11**



To create a table setting: *(continued)*

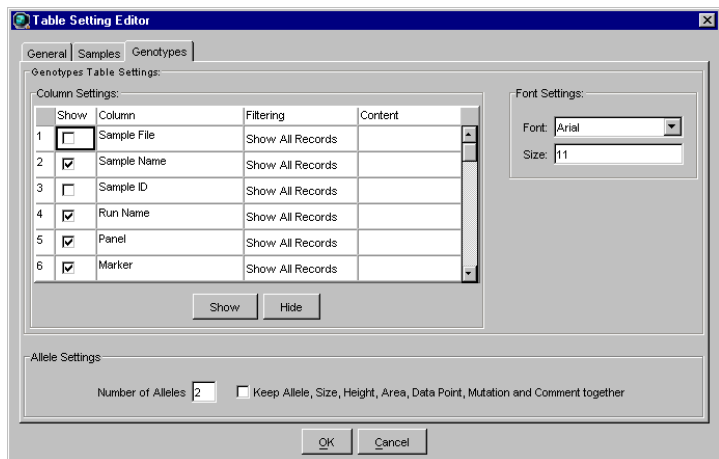
5. Select the **Genotypes** tab (graphic below) and make the following selections:

- Column settings:
  - Show: 2, 4–8, 14, 16, 17, 19, 24–29, 32, 33, 35
  - Hide: 1, 3, 9–13, 15, 18, 20–23, 30, 31, 34, 36–38

- Number of Alleles: 2

**Note:** To display columns for more alleles, this value can be increased.

- Font settings:
  - Font: **Arial**
  - Size: **11**



6. Click **OK** to save the table setting, close the Table Setting Editor, and return to the GeneMapper Manager.

# Plots

**Overview** In this tutorial, you create five custom views of electropherogram plots:

- HID Genotyping
- HID Sizing
- Overlay GS500 LIZ Dye
- Overlay GS500 ROX Dye
- Last Used

To create these custom views, follow the table steps and the software settings for each view.

## Creating HID Genotyping Plot Settings

To create HID Genotyping plot settings:

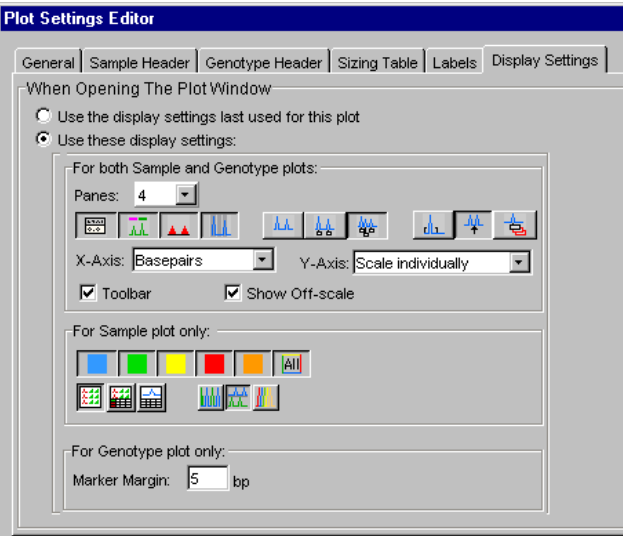
1.	From the <b>GeneMapper Manager</b> , select the <b>Plot Settings</b> tab.
2.	Click <b>New</b> to open the Plot Settings Editor with the General tab selected.
3.	Select the settings shown in Table 1-3, “HID Genotyping plot settings.”
4.	Click <b>OK</b> to save the plot settings and to close the Plot Settings Editor.  <b>Note:</b> Do not click OK until <i>after</i> you select settings on <i>all</i> tabs.
5.	Click <b>Done</b> to close the GeneMapper Manager if you have finished creating all plot settings.

## HID Genotyping Plot Settings

Table 1-3 HID Genotyping plot settings

Tab	Settings
General	Name: HID Genotyping
Sample Header	Hide: 1 and 5 Show: 2, 3, 4, and 6

Table 1-3 HID Genotyping plot settings (continued)

Tab	Settings
Genotype Header	Show: 2–5, 10–15, 18, 19, and 20 Hide: 1, 6–9, 16, and 17
Sizing Table	Show: 1–8
Labels	Label 1: Allele call Label 2, Label 3, and Label 4: None Show data type prefixes: deselected Show type of edit: selected Invert mutant labels: deselected Label Color: Dye Color-Border Label Font: Arial, size 11
Display Settings	

## Creating HID Sizing Plot Settings

To create HID Sizing plot settings:

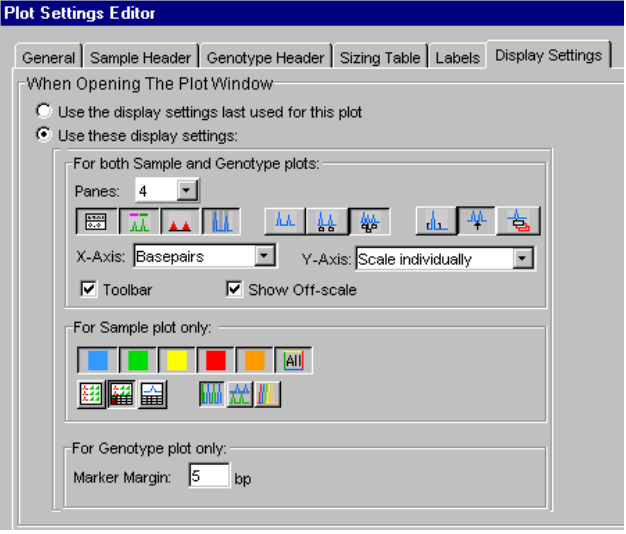
1.	From the <b>GeneMapper Manager</b> , select the <b>Plot Settings</b> tab.
2.	Click <b>New</b> to open the Plot Settings Editor with the General tab selected.
3.	Select the settings shown in Table 1-4, “HID Sizing plot settings.”
4.	Click <b>OK</b> to save the plot settings and to close the Plot Settings Editor.  <b>Note:</b> Do not click OK until <i>after</i> you select settings on <i>all</i> tabs.
5.	Click <b>Done</b> to close the GeneMapper Manager if you have finished creating all plot settings.

## HID Sizing Plot Settings

Table 1-4 HID Sizing plot settings

Tab	Settings
General	Name: HID Sizing
Sample Header	Show: 2, 3, 4, and 6 Hide: 1 and 5
Genotype Header	Show: 2–5, 10–15, 18, 19, and 20 Hide: 1, 6–9, 16, and 17
Sizing Table	Show: 1–8 Font: Arial, size 11
Labels	Label 1: Allele call Label 2, Label 3, Label 4: None Show data type prefixes: deselected Show type of edit: deselected Invert mutant labels: deselected Label Color: Dye Color-Border Label Font: Arial, size 11

Table 1-4 HID Sizing plot settings (*continued*)

Tab	Settings
Display Settings	

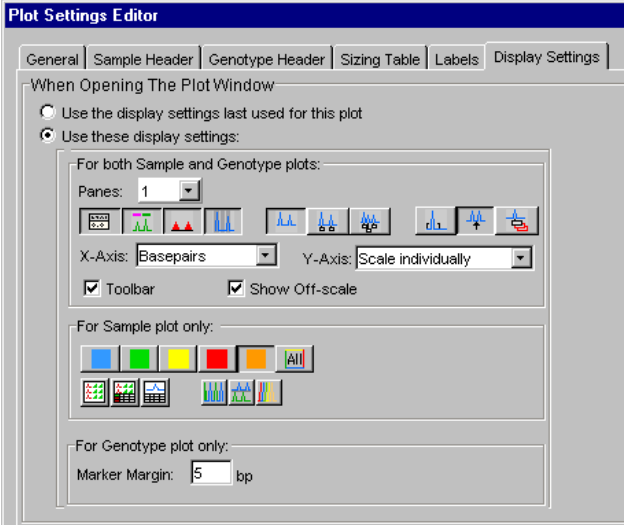
### Creating Overlay GS500 LIZ Dye Plot Settings

To create the Overlay GS500 Liz Dye plot settings:

1.	From the <b>GeneMapper Manager</b> , select the <b>Plot Settings</b> tab.
2.	Click <b>New</b> to open the Plot Settings Editor with the General tab selected.
3.	Select the settings shown in Table 1-5, “Overlay GS500 LIZ® Dye plot settings.”
4.	Click <b>OK</b> to save the plot settings and to close the Plot Settings Editor.  <b>Note:</b> Do not click OK until <i>after</i> you select settings on <i>all</i> tabs.
5.	Click <b>Done</b> to close the GeneMapper Manager if you have finished creating all plot settings.

## Overlay GS500 LIZ Dye Plot Settings

Table 1-5 Overlay GS500 LIZ® Dye plot settings

Tab	Settings
General	Name: Overlay GS500 LIZ Dye
Sample Header	Show: 2, 3, 4, and 6 Hide: 1 and 5
Genotype Header	Show: 2–5, 10–15, 18, 19, and 20 Hide: 1, 6–9, 16, and 17
Sizing Table	Show: 1–8 Font: Arial, size 11
Labels	Label 1: Allele call Label 2, Label 3, and Label 4: None Show data type prefixes: deselected Show type of edit: deselected Invert mutant labels: deselected Label Color: Dye Color-Border Label Font: Arial, size 11
Display Settings	 <p><b>Plot Settings Editor</b></p> <p>General   Sample Header   Genotype Header   Sizing Table   Labels   Display Settings</p> <p>When Opening The Plot Window</p> <p><input type="radio"/> Use the display settings last used for this plot</p> <p><input checked="" type="radio"/> Use these display settings:</p> <p>For both Sample and Genotype plots:</p> <p>Panels: 1</p> <p>X-Axis: Basepairs Y-Axis: Scale individually</p> <p><input checked="" type="checkbox"/> Toolbar <input checked="" type="checkbox"/> Show Off-scale</p> <p>For Sample plot only:</p> <p>For Genotype plot only:</p> <p>Marker Margin: 5 bp</p>

## Creating Overlay GS500 ROX Dye Plot Settings

To create the Overlay GS500 ROX Dye plot settings:

1.	From the <b>GeneMapper Manager</b> , select the <b>Plot Settings</b> tab.
2.	Click <b>New</b> to open the Plot Settings Editor with the General tab selected.
3.	Select the settings shown in Table 1-6, “Overlay GS500 ROX Dye plot settings.”
4.	Click <b>OK</b> to save the plot settings and to close the Plot Settings Editor.  <b>Note:</b> Do not click OK until <i>after</i> you select settings on <i>all</i> tabs.
5.	Click <b>Done</b> to close the GeneMapper Manager if you have finished creating all plot settings.


## Overlay GS500 Rox Dye Plot Settings

Table 1-6 Overlay GS500 ROX Dye plot settings

Tab	Settings
General	Name: Overlay GS500 ROX Dye
Sample Header	Show: 2, 3, 4, and 6 Hide: 1 and 5
Genotype Header	Show: 2–5, 10–15, 18, 19, and 20 Hide: 1, 6–9, 16, and 17
Sizing Table	Show: 1–8 Font: Arial, size 11
Labels	Label 1: Allele call Label 2, Label 3, and Label 4: None Show data type prefixes: deselected Show type of edit: deselected Invert mutant labels: deselected Label Color: Dye Color-Border Label Font: Arial, size 11



Table 1-6 Overlay GS500 ROX Dye plot settings (*continued*)

Tab	Settings
Display Settings	

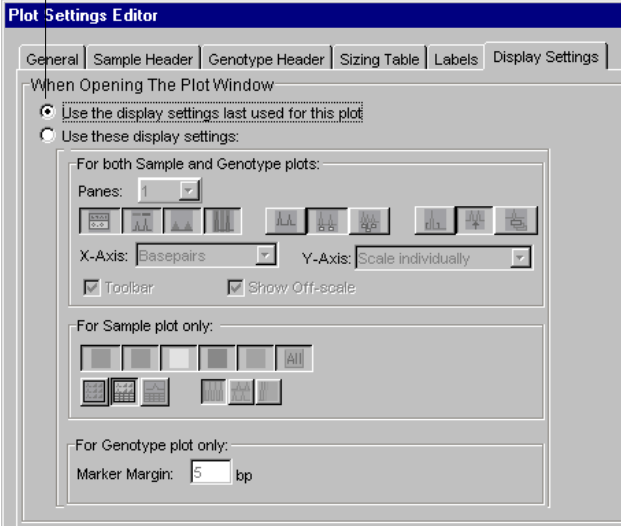
### Creating Last Used Plot Settings

To create the Last Used plot settings:

1.	From the <b>GeneMapper Manager</b> , select the <b>Plot Settings</b> tab.
2.	Click <b>New</b> to open the Plot Settings Editor with the General tab selected.
3.	Select the settings shown in Table 1-7, “Last Used plot settings.”
4.	Click <b>OK</b> to save the plot settings and to close the Plot Settings Editor.  <b>Note:</b> Do not click OK until <i>after</i> you select settings on <i>all</i> tabs.
5.	Click <b>Done</b> to close the GeneMapper Manager if you have finished creating all plot settings.

## Last Used Plot Settings

Table 1-7 Last Used plot settings

Tab	Settings
General	Name: Last Used
Sample Header	Show: 2, 3, 4, and 6 Hide: 1 and 5
Genotype Header	Show: 2–5, 10–15, 18, 19, and 20 Hide: 1, 6–9, 16, and 17
Sizing Table	Show: 1–8 Font: Arial, size 11
Labels	Label 1: Allele call Label 2, Label 3, and Label 4: None Show data type prefixes: deselected Show type of edit: deselected Invert mutant labels: deselected Label Color: Dye Color-Border Label Font: Arial, size 11
Display Settings	<p>Select this radio button</p> 

# HID Analysis Options

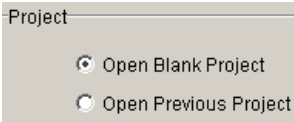
**Overview** In preparing GeneMapper® *ID* software to analyze tutorial data from AmpF $\mathcal{L}$ STR kits, view and set options for:

- Startup of GeneMapper *ID* software
- Analysis settings when adding samples to a project
- Analysis displays
- Users of GeneMapper *ID* software

## Viewing and Setting Options

**Note:** These options are active only for the user currently logged into the software.

To view and set options:

1.	Select <b>Tools &gt; Options</b> to open the Options dialog box.
2.	<p>From the <b>Startup</b> tab, view the default startup options:</p>  <p><b>Note:</b> Later, you may select <b>Open Previous Project</b> to open the last project you analyzed using GeneMapper <i>ID</i> software.</p>

To view and set options: *(continued)*

3. Select the **Add Samples** tab to view the default Add Samples options:

**Options**

Startup | **Add Samples** | Analysis | Users

When adding new samples, automatically...

Set Analysis Method to:

Default for all samples.

Read from the Sample.

Set Size Standard to:

377\_F\_HID\_GS500 for all samples.

Read from the Sample.

Set 310/377 Matrix to:

for all samples.

Read from the Sample.

Set Panel to:

Select a Panel for all samples.

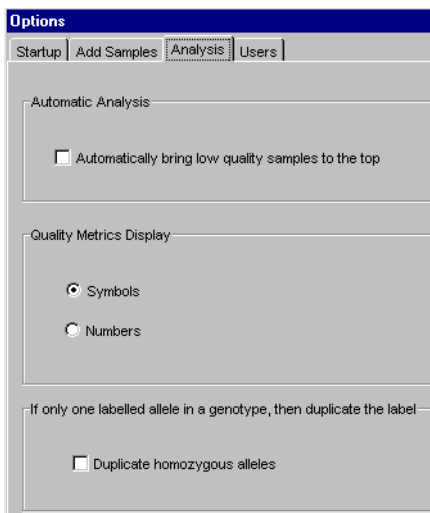
Read from Data collection 'Comment/Panel field'.

Set Sample Type to:

**Note:** For subsequent analyses, you may change these settings to set the same analysis method, size standard, 310/377 matrix, panel, and/or sample type for all samples. Select the alternative radio button and then select the setting from the drop-down list.

## To view and set options: (continued)

4. Select the **Analysis** tab to set the Analysis options:



The screenshot shows the 'Options' dialog box with the 'Analysis' tab selected. The dialog has four tabs: 'Startup', 'Add Samples', 'Analysis', and 'Users'. The 'Analysis' tab is active and contains three sections:

- Automatic Analysis:** A checkbox labeled 'Automatically bring low quality samples to the top' is currently unchecked.
- Quality Metrics Display:** Two radio buttons are present: 'Symbols' (which is selected) and 'Numbers'.
- If only one labelled allele in a genotype, then duplicate the label:** A checkbox labeled 'Duplicate homozygous alleles' is currently unchecked.

- a. For the Automatic Analysis option, deselect the **Automatically bring errors to the top of the table** check box.

**Note:** Later, you can select the check box to display samples with analysis errors at the top of the Project window automatically.

- b. Set the Quality Metrics Display option to **Symbols**.

**Note:** Changing this option to Numbers affects only the Sizing Quality (SQ) column in the Samples view and the Genotype Quality (GQ) column in the Genotypes view.

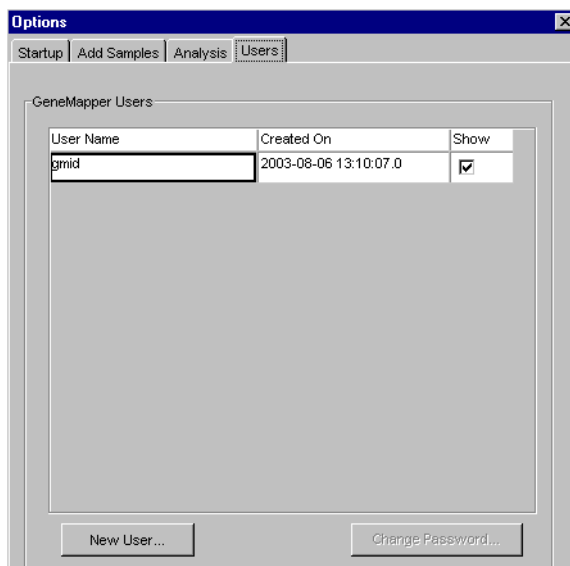
The Quality Metrics Display option is part of the Process Component-Based Quality Value (PQV) system (for more information, see “PQV System Description” on page A-3 and the *GeneMapper® ID Software Version 3.1 User Guide*).

- c. Deselect the **Duplicate homozygous alleles** check box.

To view and set options: *(continued)*

5. **IMPORTANT!** We do not recommend creating new users since the software license is limited to 5 users.

Select the **Users** tab to view the Users options:



Column	Contents
User Name	The user name that you used to log into GeneMapper <i>ID</i> software is shown (“gmid” in the example above).
Created On	The date a particular user either registered or chose a name on this tab.
Show	indicates whether the user name is presented at GeneMapper <i>ID</i> software login. Deselect the box in the Show column to hide the name at GeneMapper <i>ID</i> software login.

**To view and set options: (continued)**

6.	<p>To add a user name, click <b>New User</b> to open the New User dialog box and then:</p> <ol style="list-style-type: none"><li>a. Type a user name of your choice into the New User Name text box.</li><li>b. Type a password and confirm the password in the corresponding text boxes.</li><li>c. Click <b>OK</b> to add the user name and close the dialog box.</li></ol>
7.	<p>Click <b>OK</b> to exit Options and implement the changes you made on all Options tabs.</p> <p><b>IMPORTANT!</b> If you click <b>Cancel</b> to exit, you discard any changes made on the Options tabs!</p>





# Casework Analysis

---

# 2

This chapter covers:

Casework Workflow .....	2-2
Setting Up a Casework Project .....	2-3
Examining and Editing Results .....	2-14

## Casework Workflow

**Description** In this chapter, you perform analysis of tutorial casework samples using the software settings from Chapter 1, Software Setup.

### **Casework Analysis Tutorial Overview**

Overview of the tutorial for analyzing a casework project:

1. Add samples to be analyzed to a new project (page 2-5).
2. Apply analysis settings to the samples in the project (page 2-7).
  - a. Select the analysis method.
  - b. Create a custom size standard definition for your data using the Classic peak detection algorithm.
  - c. Select the size standards for the samples.
3. Analyze the project (page 2-14).
4. Examine the size standard (page 2-15).
  - a. Examine the size standard peak assignments.
  - b. Examine the 250-bp size standard peak.
5. Examine data (page 2-20).
  - a. Examine allelic ladder calls.
  - b. Examine allele calls.
6. Edit labels (page 2-25).
7. View allele history and comments (page 2-25).
8. Complete analysis (page 2-25).
9. Check concordance for shared markers (page 2-28).
10. Export the table (optional) (page 2-30).

# Setting Up a Casework Project

## Adjusting the Project Window

As you examine the Project window, you may need to adjust the window to see as many of the table columns as possible. The amount of resizing needed depends on the number of columns displayed and on the size and screen resolution of your monitor. In general, perform the following steps to view all columns in the Project window.

Adjust column width by placing cursor on lines between columns, and then dragging.

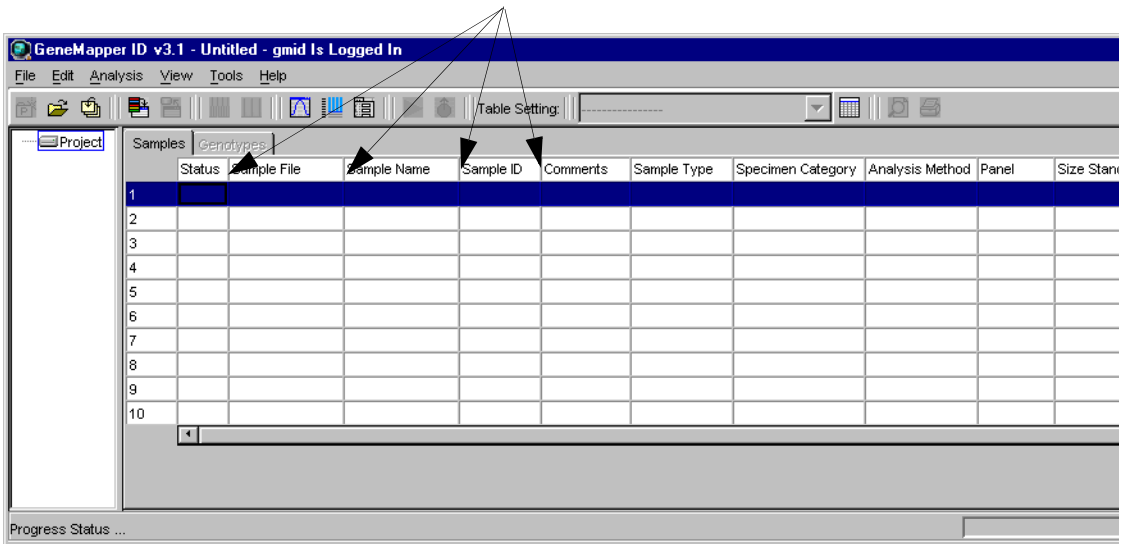



Figure 2-1 Adjusting the column width

**Note:** Altered column widths are not saved when you close the window.

To adjust the Project window:

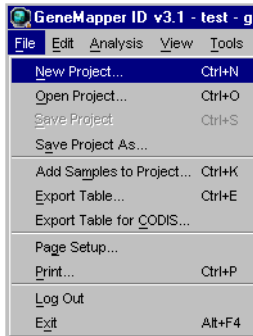
1. Click the square in the upper right corner of the window to maximize the window. 

**To adjust the Project window: (continued)**

2.	<p>Deselect <b>Show Navigator</b> from the View menu to hide the navigation pane.</p> <p>This action expands the Samples and Genotypes tabs to the width of the Project window. Select <b>Show Navigator</b> from the View menu to restore the navigation pane.</p>
3.	<p>Resize columns by dragging the separating lines:</p> <ol style="list-style-type: none"> <li>a. Position the cursor over the line separating two columns until the cursor changes to sizing arrows.</li> <li>b. Click and drag the sizing arrows. Dragging to the left narrows the column to the left.</li> </ol>

**Adding Samples**

You should have a blank, untitled Project window open. To create a blank project if a blank project window is not open, or if an existing project is already open, select **File > New Project**.

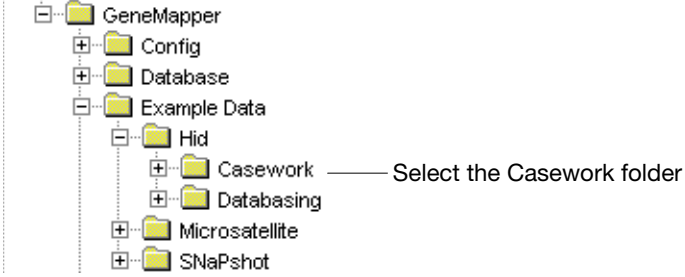


**Figure 2-2 New Project**

**To add samples to the project:**

1.	<p>From the Project window, select <b>File &gt; Add Samples to Project</b> to navigate to the disk or directory containing the tutorial sample files.</p> <p>The initial view of the dialog box is for local disk access. For more information on the GeneMapper ID software database, see the <i>GeneMapper® ID Software Version 3.1 User Guide</i>.</p>
----	-----------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------

## To add samples to the project: (continued)


2.	<p>Navigate to the Casework folder: X:\Applied Biosystems\GeneMapper\Example Data\HID\Casework</p> <p><b>Note:</b> X is the drive where you installed GeneMapper ID software. For subsequent analysis using your data, navigate to the disk/directory containing your files.</p>
3.	<p>Select the <b>Casework</b> folder and click <b>Add To List</b> at the bottom of the screen.</p>  <p><b>Note:</b> The casework folder appears in the <b>Samples To Add</b> box on the right.</p> <p><b>Note:</b> If you make an error in moving a file to the list, select the files to remove from the Samples to Add list and click <b>Clear</b>.</p>
4.	<p>Click <b>Add</b> to import the files into the project and close the dialog box.</p> <p>The samples are displayed in the Project window.</p>

## Applying Analysis Settings

In this procedure, you select the analysis method for the samples, create a new size standard custom definition, and set the size standard for the samples.

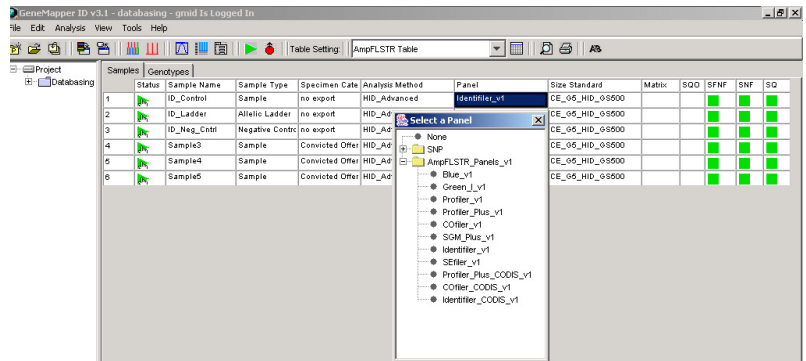
**Note:** The definitions for the 377\_F\_HID\_GS500, 377\_G5\_HID\_GS500, CE\_F\_HID\_GS500, and CE\_G5\_HID\_GS500 standards are provided for use with the Advanced peak detection algorithm. See “Size Standard Definitions” on page A-2 for more information.

**To apply analysis settings:**

1.	Select <b>HID Table</b> from the Table Setting drop-down list at the top of the project window.
2.	<p>Select the analysis method for the samples:</p> <ol style="list-style-type: none"> <li>a. Click the first empty (<b>None</b>) cell in the Analysis Method column in the Samples view.</li> <li>b. Select <b>HID_Classic</b> from the drop-down list.</li> <li>c. Click the <b>Analysis Method</b> column header to select the column.</li> </ol>  <p>d. Select <b>Edit &gt; Fill Down</b> to apply the HID_Classic analysis method to all samples.</p>

## To apply analysis settings: (continued)

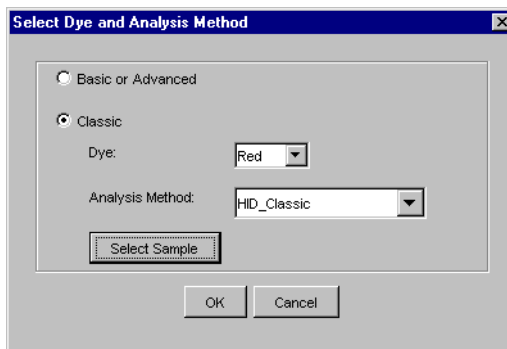
3. Select the appropriate panel for the sample. (Samples with a CO prefix use the COfiler\_v1 panel and samples with the PP prefix use the Profiler Plus\_Plus\_v1 panel.)



**Note:** In the example above, the first four samples are Cofiler® and the last four samples are Profiler Plus™.

To apply analysis settings: (*continued*)

3. Create a new size standard custom definition.
  - a. Click the first empty (**None**) cell in the Size Standard column.
  - b. Select **New Size Standard** from the drop-down list.
  - c. Specify the parameters below.



- Sizing method: **Classic**
- Dye: **Red**
- Analysis Method: **HID\_Classic**
- Select Sample: **CO\_Control.fsa**

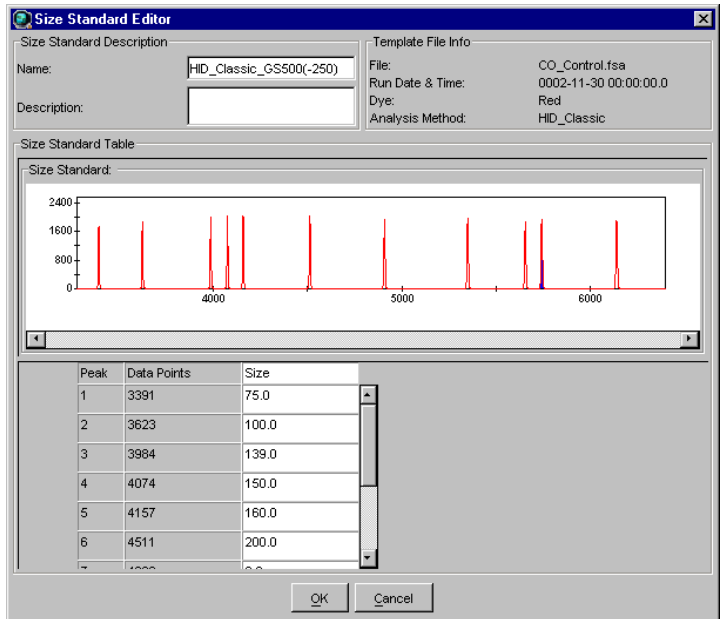
**Note:** If you want to use another sample to make the size standard definition, click **Select Sample** and then browse to the location of the sample file. *For this tutorial, use the CO\_Control.fsa sample already displayed.*

- d. Click **OK** to access the Size Standard Editor.



## To apply analysis settings: (continued)

The Size Standard Editor shows the electropherogram and a table of peaks for the dye color and sample selected.



For more information about setting size standard parameters, see the *GeneMapper® ID Software Version 3.1 User Guide*.

4. Enter a name for the size standard:  
**HID\_Classic\_GS500(-250)**
5. If needed, adjust the size of the Size Standard Editor window:
  - a. Position the cursor over a border or corner of the Size Standard Editor until the cursor changes to sizing arrows.
  - b. Click and drag the sizing arrows to achieve the desired size.

**To apply analysis settings: (continued)**

6.	<p>Zoom in and out of the Size Standard Editor electropherogram for easy viewing:</p> <ul style="list-style-type: none"><li>a. Place the cursor along the x-axis until the cursor changes to a magnifying glass.</li><li>b. Click and drag to create a box and release when the box contains the desired viewing range. Always drag along the x-axis that contains the numbers. The x-axis changes depending on the type of plot.</li><li>c. Move the cursor toward the x-axis until the cursor changes to a magnifying glass, and then double-click to restore the plot to full view.</li></ul>
7.	<p>Explore the Size Standard Editor:</p> <ul style="list-style-type: none"><li>a. Select a peak in the electropherogram, and notice that the corresponding row in the table is highlighted.</li><li>b. Select a peak in the table, and notice that the corresponding peak in the electropherogram is highlighted.</li></ul>

To apply analysis settings: *(continued)*

8. Assign sizes to the peaks of the size standard: 75, 100, 139, 150, 160, 200, 0 (skip the 250 peak), 300, 340, 350, 400.

**Note:** Do not assign a size for the 250-bp peak. This peak can be used as an indicator for precision within a run.

- a. Click the first peak in the electropherogram to select it.
- b. Press **Enter**.
- c. Type the fragment size for the selected peak in the corresponding cell in the table.
- d. Press the down arrow key to move to the next peak/cell.
- e. Press **Enter**.
- f. Repeat steps c through e for all fragments sizes.

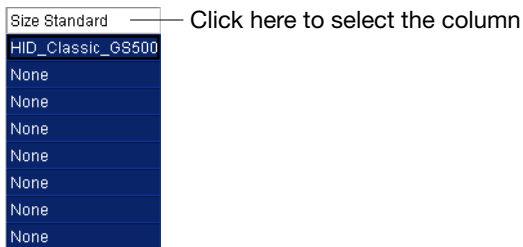
Peak	Data Points	Size
1	3391	75.0
2	3623	100.0
3	3984	139.0
4	4074	150.0
5	4157	160.0
6	4511	200.0
7	4908	0.0
8	5350	300.0
9	5655	340.0
10	5743	350.0
11	6140	400.0

—— Skip the 250-bp peak

- g. Click **OK** to save the size standard, close the Size Standard Editor, and return to the Project window.

To apply analysis settings: *(continued)*


9. Set the size standard for the samples:
  - a. Click the **Size Standard** column header to select the column.



- b. Select **Edit > Fill Down** to apply the size standard to all samples.

## Analyzing the Project

To analyze the project:

1. Click  (Analyze), and the Save Project dialog box opens.
  2. Type **Casework Project** and click **OK** to initiate analysis and save each analyzed sample to the project.
    - The status bar displays progress of analysis:
      - As a completion bar extending to the right with the percentage indicated
      - With text messages on the left
    - The table displays the row of the sample currently being analyzed in green (or red if analysis failed for the sample). See Figure 2-3.
- Note:** Auto-saving takes place after analysis of every 10 sample files is completed.
- The Genotypes tab becomes available after analysis.

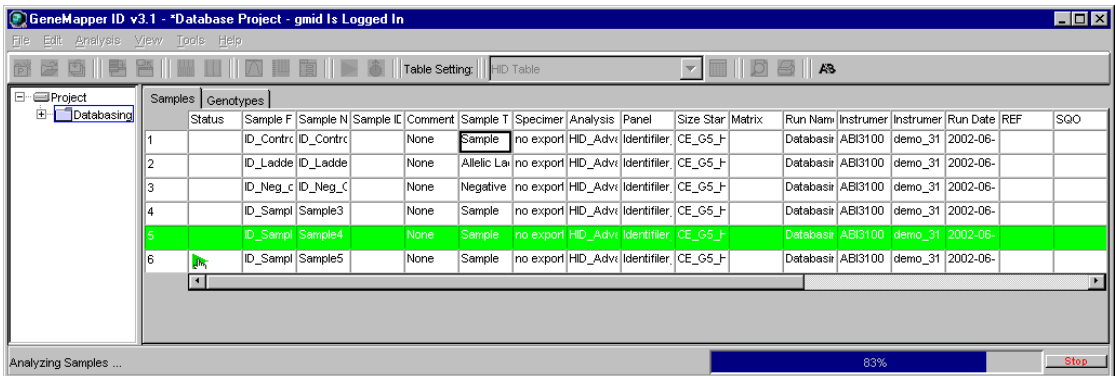



Figure 2-3 The Database Project window

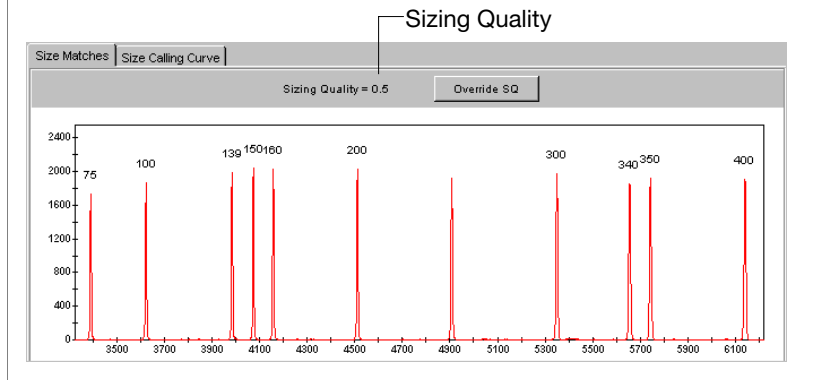
# Examining and Editing Results

**Overview** You can display electropherogram plots from the Samples and Genotypes tabs of the Project window to examine the data. These procedures start with the Samples tab of the Project window (assuming that analysis is complete).

**Examining the Size Standard** When using the Classic peak detection algorithm and at least three size standard peaks match, the software assigns a Sizing Quality (SQ) value of 0.5, which corresponds to a yellow triangle (Check). In this procedure, you verify that the size standard peak assignments are correct and override the SQ value to display a green square (Pass).

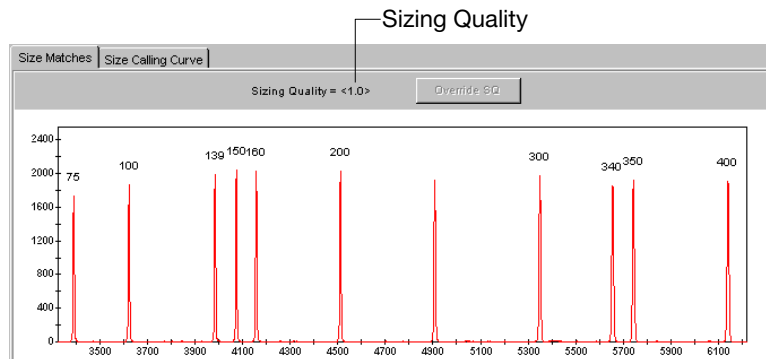
**To examine the size standard:**

1. Scroll to the right side of the table to see the yellow sizing qualities.
2. Select **Edit > Select All** and click  (Size Match Editor) to view the size standard for all samples.
3. Notice that the Sizing Quality value equals 0.5.



To examine the size standard: *(continued)*

4. View the peak assignments for each sample:
  - a. Press the down arrow key to scroll through the samples on the left side of the Size Match Editor screen.
  - b. Confirm that the size for the peaks in the **HID\_Classic\_GS500(-250)** size standard (GeneScan® 500 ROX™ Size Standard) is correctly assigned for each sample.
5. Select **Edit > Override All SQ** to override the SQ value for all samples.
6. Notice that the Sizing Quality is changed to <1.0>, which indicates that user verified the size standard.



7. Click **Apply**, then click **OK** to close the Size Match Editor and return to the Project window.

**To examine the size standard: (continued)**

8. Notice that after overriding the SQ values:
- The SQ flags are changed to green squares (Pass).
  - The Sizing Quality Override (SQO) column is checked.


SQO	SFNF	SQ
X	■	■
X	■	■
X	■	■
X	■	■
X	■	■
X	■	■
X	■	■
X	■	■

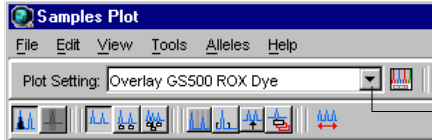
9. Define a new size standard for the affected samples, and then reanalyze the sample if one of the following occurs:
- Size standard peak assignments are incorrect for one or more of the samples in a subsequent analysis.
  - Fewer than three peaks are matched and a red flag is displayed.



## Examining the 250-bp Peak

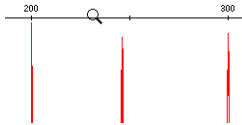
To examine the 250-bp peak:

1. Display the Overlay GS500 ROX Dye plot:
  - a. With all samples still selected, click  (Display Plots) to display the Samples Plot window.
  - b. Select the **Overlay GS500 ROX Dye** plot from the drop-down list below the menu bar.

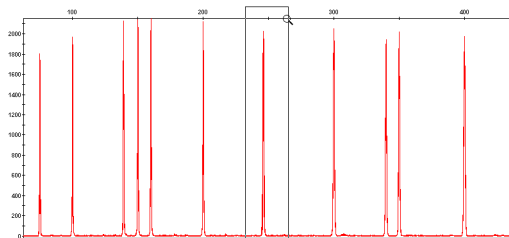


Click here to display the list of plots

2. Zoom in on the 250-bp peak:
  - a. Place the cursor to the left of the 250-bp peak, along the top x-axis until the cursor changes to a magnifying glass.



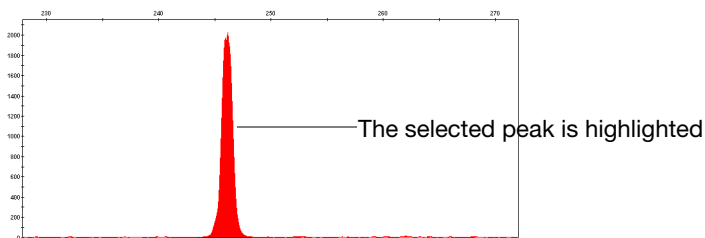
- b. Click and drag to create a box and release when the box includes the 250-bp peak.



**Note:** If you zoom in inaccurately, move the cursor toward the x-axis until the cursor changes to a magnifying glass, and then double-click to restore the plot to full view.

To examine the 250-bp peak: *(continued)*

3. Select the 250-bp peak by clicking it.



4. Select **View > Table Filter > Show Selected Rows**, and verify that the values in the Size column are approximately 246 bp (expected for this set of data). However, not all instruments will produce a 246 migration of the 250 peak.

	Dye/Sample Pe	Sample File Na	Marker	Allele	Size	Height	Area	Data Point
■	R,7	CO_Control.fsa			245.99	1924	14500	4908
■	R,7	CO_Ladder.fsa			245.95	1811	13741	4905
■	R,7	CO_Sample1.f			245.98	1977	14977	4906
■	R,7	CO_Sample2.f			245.98	1935	14989	4906
■	R,7	PP_Control.fsa			246.25	1923	14261	4860
■	R,7	PP_Ladder.fsa			246.17	2024	14976	4905
■	R,7	PP_Sample1.f			246.07	1950	14598	4882
■	R,7	PP_Sample2.f			245.99	1951	14660	4889



**Note:** This peak was not defined in the size standard. The 250-bp peaks should size consistently and overlap. In a typical run, the 250-bp peaks all fall within a size window of approximately 1 bp. Temperature fluctuations in the laboratory may cause variations > 1 bp.

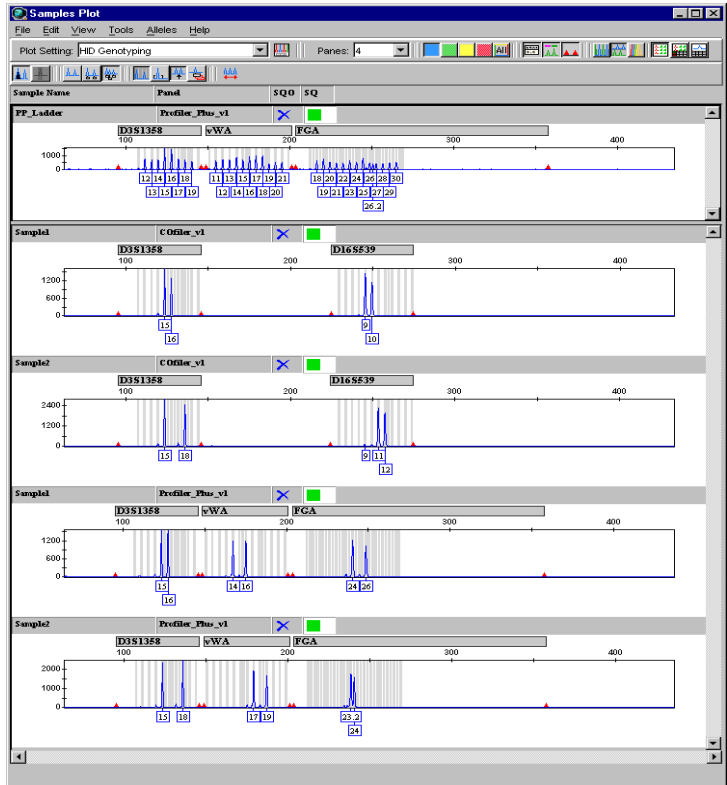
**Note:** Laboratory temperature fluctuations can cause size shifts. For the ABI PRISM® 310 Genetic Analyzer only: if the temperature of the laboratory fluctuates, inject the appropriate AmpF<sup>®</sup>STR allelic ladder approximately every 10 injections, or 5 h.

5. Close the sample plot.

**What's Next?** You have now confirmed the sizing precision, and you are ready to view and edit the plots and allele calls.

## Examining Data To examine data:

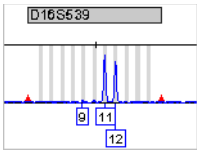
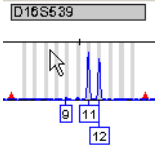
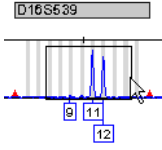



1. With all samples still selected in the Project window, select the **HID Genotyping** plot from the drop-down list below the menu bar of the Samples Plot window.
2. Click  (Hide All) and then click  (Show Blue Dye).



**Note:** If the electropherogram plots display “No room for labels,” then do one or more of the following to view the labels:

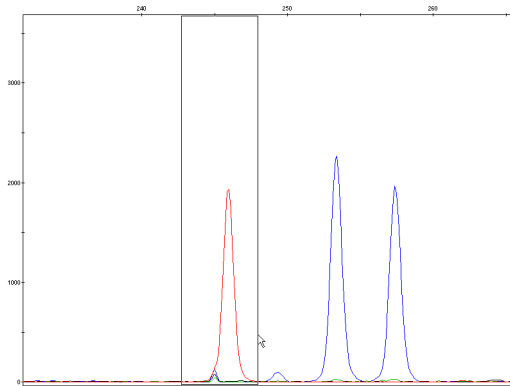
- Reduce the number of panes displayed.
- Remove the ladder from the view.
- Increase the screen resolution.
- Expand the top window by placing the cursor on the line, then click and drag.


To examine data: (continued)

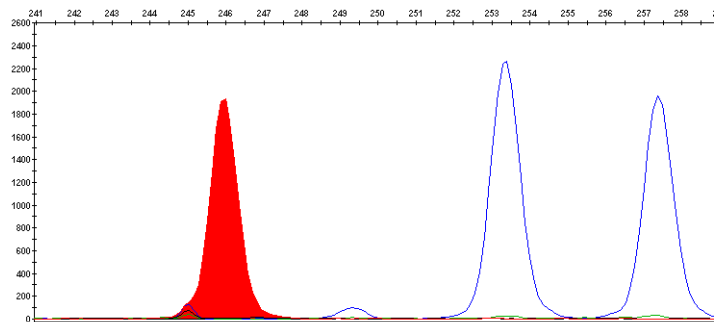
3.	Examine the allelic ladder calls and verify that the allelic ladder was called correctly.
4.	<p>View the electropherogram plots and notice that there are three allele calls for Sample2 (COfiler_v1 panel) at the D16S539 locus.</p>  <p>To zoom in on a particular marker: click and drag to create a rectangle around the marker of interest. Release the mouse to zoom in.</p>
5.	<p>View the D16S539 alleles for Sample2 (COfiler_v1 panel) more closely in a single pane:</p> <ol style="list-style-type: none"> <li>a. Select the D16S539 labels:             <ol style="list-style-type: none"> <li>1. Place the cursor to the left of the peaks within the plot.</li>  <li>2. Click and drag to the right to create a box.</li> <li>3. Release when the box includes the three peaks.</li>  </ol> </li> <li>b. Select <b>1</b> from the Panes drop-down list in the toolbar.             <p><b>Note:</b> Panes for the selected peaks are presented in the Project window.</p> </li> <li>c. Click  (Don't Bring Controls to Top).</li> </ol>
6.	<p>Examine the peak for allele 9:</p> <p>Click  (Combine Dyes) and click  (Show All).</p>

## To examine data: (continued)

7. Observe that the allele 9 peak is not caused by spectral pull-up:
- Select the 250-bp size standard peak and the allele 9 peak by clicking within the plot and dragging to create a box that includes these peaks.




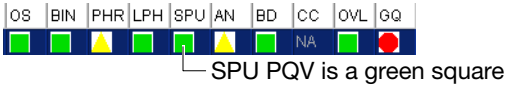
- Click  (Sizing Table) and select **View > Table Filter > Show Selected Rows**.
- View the peak location in the graph and note that the blue peak for allele 9 is shifted to the left of the red peak for the 250-bp size standard.



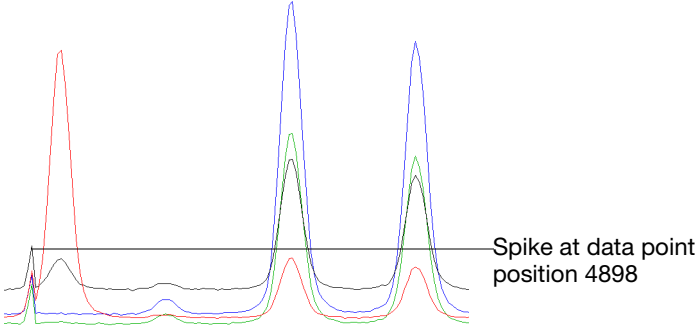
- View the data point values for the selected peaks and note that the data point for allele 9 (4898) differs from the data point for the 250-bp size standard (4906) by more than 1.

	Dye/Sample Pe	Sample File Na	Marker	Allele	Size	Height	Area	Data Point
	B,6	CO_Sample2.f	D16S53	9	245.03	127	434	4898
	Y,1	CO_Sample2.f			245.03	76	264	4898
	R,7	CO_Sample2.f			245.98	1935	14989	4906


## To examine data: (continued)

8.	<p>With the peaks still selected, click  (Genotypes Table) and observe that the spectral pull-up (SPU) PQV is a green square (Pass).</p>  <p><b>Note:</b> The SPU PQV denotes no peak above the analysis threshold and no peak below the 0.05 pull-up ratio (set on page 1-11) within <math>\pm 1</math> data point of allele 9.</p>
9.	<p>Observe the other PQVs and note that PHR (peak height ratio) and the AN (allele number) are flagged yellow.</p> <ul style="list-style-type: none"> <li>• <i>PHR</i>: Indicates that the peak height ratio between the 9, 11, 12 peaks are not as expected. In the Analysis Method Peak quality tab, we set a 70% ratio. Any ratio less than 70% flags the PQV <b>yellow</b>. In this case, the peak height of the 9 peak is 127 and the peak height fo the 11 peak is 2265. This is an unacceptable ratio of 5.6%.</li> <li>• <i>AN</i>: Indicates that there are more than 2 alleles. In theAnalysis Method Peak Quality tab, we set the maxium expected alleles at 2. A marker with more than 2 alleles flags the PQV <b>yellow</b>.</li> </ul> <p>When these two components (PHR and AN) are both flagged yellow, the overall genotype quality is flagged <b>red</b>.</p>

To examine data: *(continued)*

10.	<p>Observe the raw data:</p> <ol style="list-style-type: none"><li>Select <b>View &gt; Raw Data</b>.</li><li>Zoom in on the data point position 4898, and observe that the allele 9 peak corresponds to a spike.</li></ol>  <p>The chromatogram displays several peaks of varying heights and colors (red, blue, green). A horizontal line is drawn across the plot, with a label 'Spike at data point position 4898' pointing to a specific peak. The peaks are arranged from left to right, with the highest peak being blue.</p>
11.	<p>Select <b>View &gt; Samples</b> to view the Samples tab of the Project window.</p>

## Editing Labels **To edit labels:**

1.	Click: <ol style="list-style-type: none"> <li>On the Samples Plot on the lower taskbar of your computer screen to return to the previous plot window</li> <li>Click  to separate dyes</li> </ol>
2.	Click within the plot (but <i>not</i> on a peak) to deselect previously selected peaks.
3.	Select the label for allele 9 by clicking the label or the peak.
4.	Right-click the label and select <b>Delete</b> .
5.	Type <b>spike</b> in the Edit Allele Comment dialog box.
6.	Click <b>OK</b> .

## Viewing Allele History and Comments


### To view allele history and comments:

1.	In the Samples Plot window, right-click the label for the edited allele and select <b>History</b> to view changes.
2.	Click <b>OK</b> to close the history.
3.	View the row for the locus D16S539 for Sample2 (COfiler_v1 panel) in the Genotypes table and observe that: <ul style="list-style-type: none"> <li>The PQVs are displayed as gray triangles, indicating that there is an override of the values due to the deletion of the allele call.</li> </ul>

## Completing Analysis



Complete the analysis of all of the samples by viewing the dyes individually in the HID Genotyping plot.

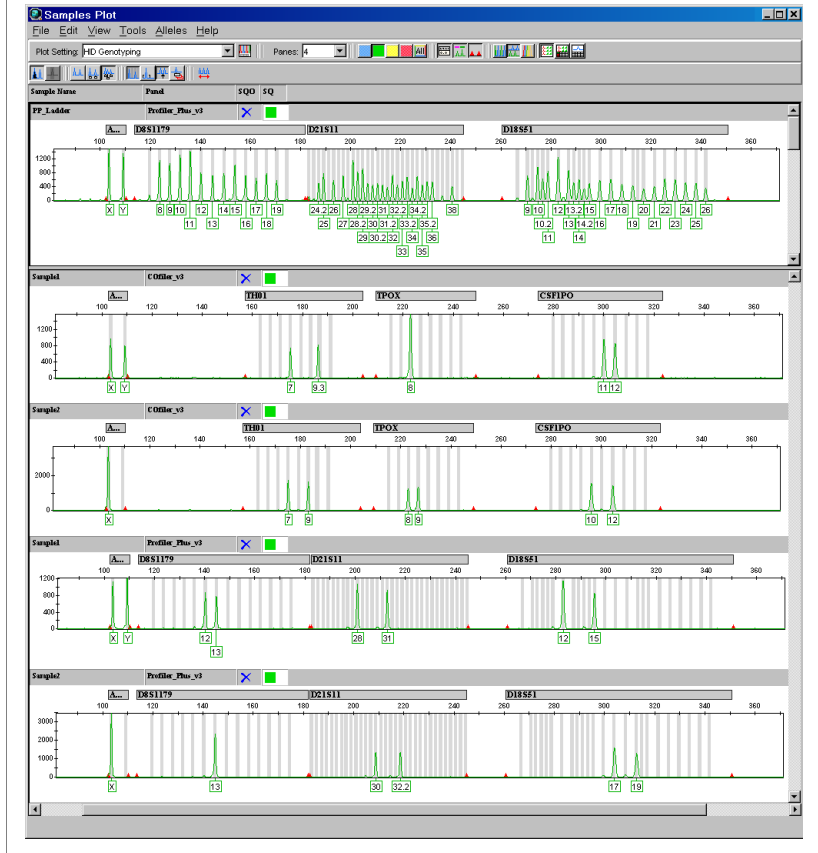
### To complete analysis:

1.	Return to the Project window, and select <b>Edit &gt; Select All</b> .
2.	Click  (Display Plots) to open the Samples Plot window.





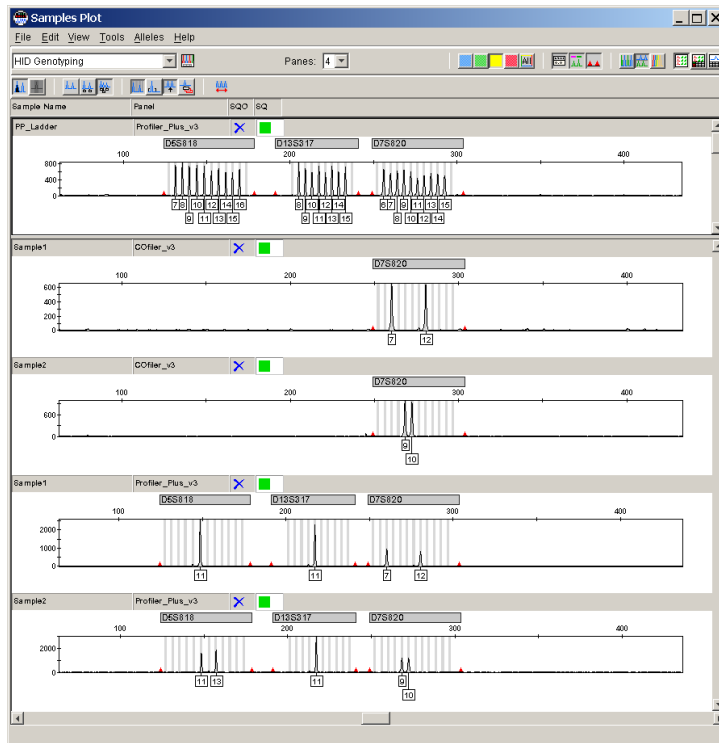
## To complete analysis: (continued)

3. With the samples plot still open, reselect the **HID Genotyping** plot from the drop list to refresh the window with the original settings. Note that it may appear as though it is already selected, but you need to open the box and select it again.
4. Click  (Hide All) and click  (Show Green Dye), and scroll through all samples



## To complete analysis: (continued)

5. Click  (Hide Green Dye), click  (Show Yellow Dye), and scroll through all samples.



6. Select **File > Close Plot Window** to close the Samples Plot window and return to the Project window.
7. Select **File > Save Project** to save all changes.

**Concordance Usefulness** Checking concordance can be useful for comparing a sample amplified using kits with shared markers.

**Concordance Requirements** Requirements for performing a concordance check:

- Add the sample to a project in the local GeneMapper database.
  - Note:** A sample added from outside of the GeneMapper database has no results associated with it for comparison.
- Samples to be compared must have the same sample name or user defined value.

### Checking Concordance for Shared Markers


**Note:** The concordance check in this tutorial compares markers shared between samples with the same sample name. Laboratories with automated processes for naming samples can still perform this procedure by typing in a shared name for two or more samples in the UD1 column. You can also use this option when setting up a sample sheet by typing | after the panel name in the comment field, for example, **Profiler\_Plus\_v1|sample 1**.

**To check concordance for shared markers:**

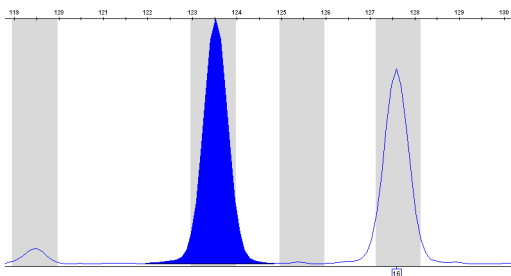
1.	In the Project window, select the <b>Genotypes</b> tab.
2.	Select <b>Analysis &gt; Non-concordant Samples to Top</b> .  <b>Note:</b> No samples are highlighted, indicating that there are no discrepancies in genotypes for shared markers of the same sample amplified with AmpF $\ell$ STR Profiler Plus and AmpF $\ell$ STR COfiler kits.

To check concordance for shared markers: *(continued)*

3. For this tutorial, introduce a discrepancy:
  - a. Select the row for Sample 1 (Profiler\_Plus\_v1 panel) containing the D3S1358 locus.
 

**Note:** The navigation pane contains a folder for each panel (AmpF<sup>®</sup>STR kit) analyzed. Selecting a marker in the navigation pane displays all samples containing that marker.
  - b. Click  (Display Plots) to open the Genotypes Plot window.
  - c. Select allele 15 by clicking the label or the peak and then right-click and select **Delete** from the pop-up menu to delete the allele.
 

**Note:** If you select multiple labels or peaks and then select Delete, the software prompts you with a warning that the corresponding alleles will be deleted.
  - d. Type **Concordance Test** in the Delete Allele Comment dialog box and click **OK**.
  - e. Notice that the label is deleted.




**Note:** Show deleted labels by selecting **Allele Changes** from the View menu.

4. Select **File > Close Plot Window** to return to the Project window.

To check concordance for shared markers: *(continued)*

5. Select **Analysis > Non-concordant Samples to Top**, and notice that rows 1 and 2 are highlighted and brought to the top. These samples contain the D3S1358 marker with nonconcordant genotypes.

	Sample Name	Run Name	Panel	Marker	Dye	Allele 1	Allele 2	AE Corri	AE Com	ADO	AE	OS	BIN	PHR	LPH	SPU	AN	BD	CC	OVL	GO
1	Sample1	Casework	COffler_v3	D3S1358	B	15	16														
2	Sample1	Casework	Profiler_Plus_v	D3S1358	B	16															

6. Click  (Display Plots) to open the Genotypes Plot window.

7. Restore the original allele call:
  - a. Select the peak without the label by clicking it.
  - b. Right-click the selected peak and select **Add Allele Call** from the pop-up menu.
  - c. Type **Concordance restored** in the Add Allele Comment dialog box and click **OK**.

8. Select **File > Close Plot Window** to return to the Project window and select **File > Save Project** to save the changes.

### Exporting the Table (Optional)

You may export the table data as a tab-delimited text file that can be used with spreadsheet software.

**Note:** To export a table for the CODIS database, go to Chapter 4, “CODIS Export.”

#### To export the table:

1. Select **File > Export Table** from the Project window.
2. Select a location for the file.
3. Enter **Casework Table** for the file name.
4. Click **Export Table**.
5. Using Microsoft<sup>®</sup> Excel<sup>®</sup> software or equivalent spreadsheet software, open the exported table file.



# Database Analysis

---

# 3

This chapter covers:

Database Workflow . . . . .	3-2
Setting Up a Database Project . . . . .	3-3
Examining and Editing Results . . . . .	3-9

## Database Workflow

**Description** In this chapter, you perform analysis of tutorial database samples using the software settings from Chapter 1, Software Setup.

**Cutoff Value Setting** The HID\_Advanced analysis method created in this tutorial for processing database samples removes labels from each peak with a height less than 20% of the highest peak in a marker's allele size range. A cutoff value selected from the Allele tab of the analysis method does not include any condition regarding the base pair size of the peak with a removed label relative to a higher peak. This option is provided for laboratories that wish to use one general value for removing labels from all loci. It can be used when a high level of filtering specificity is not required, as in the typing of single source samples, for example, database samples.

### Database Analysis Tutorial Overview

Overview of the tutorial for analyzing a database project:

1. Add samples to be analyzed to the project (page 3-4).
2. Apply analysis settings to the samples in the project (page 3-6).
  - a. Select the analysis method.
  - b. Select the size standards for the samples.
3. Analyze the project (page 3-8).
4. Examine the size standard (page 3-9).
  - a. Assess whether samples pass the sizing criteria.
  - b. Check the size standards for any samples that do not pass the sizing criteria.
5. Examine the allelic ladder calls (page 3-9).
6. Examine data and edit labels (page 3-10).
  - a. Assess whether sample markers pass the genotyping criteria.
  - b. Examine peaks for any sample markers that do not pass the genotyping criteria.
7. Export the table (optional) (page 3-13).




# Setting Up a Database Project

## Adjusting the Project Window

As you examine the Project window, you may need to adjust the window to see as many of the table columns as possible. The amount of resizing needed depends on the number of columns displayed and on the size and screen resolution of the monitor used to run the GeneMapper *ID* software. In general, perform the following steps to view all columns in the Project window.

To adjust the Project window:

1.	Click the square in the upper right corner of the window to maximize the window. 
2.	Deselect <b>Show Navigator</b> from the View menu to hide the navigation pane.  This action expands the Samples and Genotypes tabs to the width of the Project window. Select <b>Show Navigator</b> from the View menu to restore the navigation pane.
3.	Resize columns by dragging the separating lines: a. Position the cursor over the line separating two columns until the cursor changes to sizing arrows. b. Click and drag the sizing arrows. Dragging to the left narrows the column to the left.  <b>Note:</b> Altered column widths are not saved when you close the window.

**Adding Samples** You should have a blank, untitled Project window open. To create a project, select **File > New Project**.

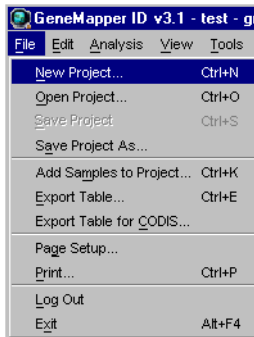


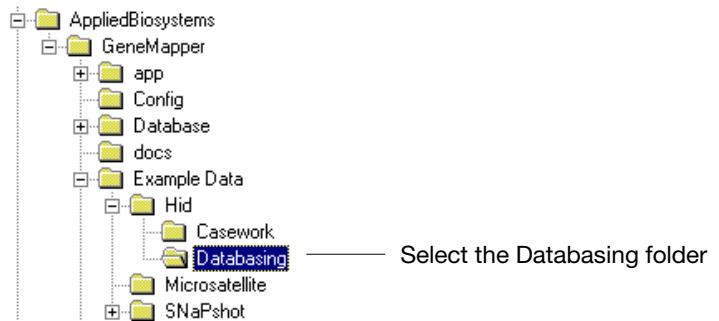
Figure 3-1 New Project

To add samples to the project:

- |    |                                                                                                                                                                                                                                                                                                                                                                 |
|----|-----------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------|
| 1. | <p>From the Project window, select <b>File &gt; Add Samples to Project</b> to navigate to the disk or directory containing the tutorial sample files.</p> <p>The initial view of the dialog box is for local disk access. For more information on the GeneMapper <i>ID</i> software database see the <i>GeneMapper® ID Software Version 3.1 User Guide</i>.</p> |
| 2. | <p>Navigate to the Databasing folder:</p> <p style="padding-left: 40px;"><i>X:\AppliedBiosystems\GeneMapper\Example Data\HID\Databasing\</i></p> <p><b>Note:</b> <i>X</i> is the drive where you installed GeneMapper <i>ID</i> software. For subsequent analysis using your data, navigate to the disk/directory containing your files.</p>                    |

## To add samples to the project: (continued)

3. Select the **Databasing** folder and click **Add To List**.

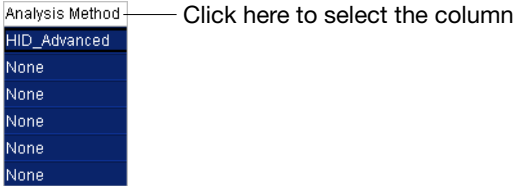


**Note:** If you make an error in moving a file to the list, select the files to remove from the Samples to Add list and click **Clear**.

4. Click **Add** to import the files into the project and close the dialog box.  
The samples are displayed in the Project window.

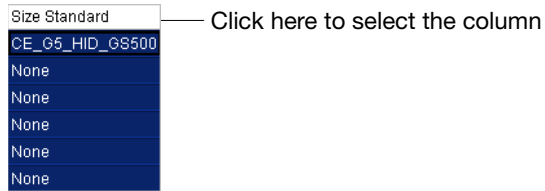
## Applying Analysis Settings

To apply analysis settings:

1.	Make sure that <b>HID Table</b> is selected from the Table Setting drop-down list.
2.	<p>Select the analysis method for the samples:</p> <ol style="list-style-type: none"> <li>a. Click the first empty (<b>None</b>) cell in the Analysis Method column in the Samples tab.</li> <li>b. Select <b>HID_Advanced</b> from the drop-down list.</li> <li>c. Click the <b>Analysis Method</b> column header to select the column.</li> </ol>  <p style="margin-left: 20px;">Analysis Method — Click here to select the column</p> <ul style="list-style-type: none"> <li>HID_Advanced</li> <li>None</li> <li>None</li> <li>None</li> <li>None</li> <li>None</li> </ul> <ol style="list-style-type: none"> <li>d. Select <b>Edit &gt; Fill Down</b> to apply the analysis method to the selected samples.</li> </ol>
3.	<p>Select the appropriate panel for the samples:</p> <ol style="list-style-type: none"> <li>a. Click on the first empty (<b>None</b>) cell in the Panel column and open the AmpFSTR_Panels_v1 folder.</li> <li>b. Double-click on the <b>Identifiler_v1</b> panel from the drop-down list. This places the Identifiler_v1 panel in the first sample row.</li> <li>c. Use the <b>Edit &gt; Fill Down</b> feature to place Identifiler_v1 in each sample row.</li> </ol>

**To apply analysis settings: (continued)**


4. Select the size standard for the samples:
  - a. Click the first empty (**None**) cell in the Size Standard column, and select **CE\_G5\_HID\_GS500** from the drop-down list.
  - b. Click the **Size Standard** column header to select the column.



- c. Select **Edit > Fill Down** to apply the size standard to the selected samples.

## Analyzing the Project

To analyze the project:

1. Click  (Analyze) to open the Save Project dialog box.
2. Type **Database Project** and click **OK** to initiate analysis and save each analyzed sample to the project.
  - The status bar displays progress of analysis:
    - As a completion bar extending to the right with the percentage indicated
    - With text messages
  - The table displays the row of the sample currently being analyzed in green (or red if analysis failed for the sample). See Figure 3-2.

**Note:** Auto-saving takes place after analysis of every 10 sample files and after analysis is completed.

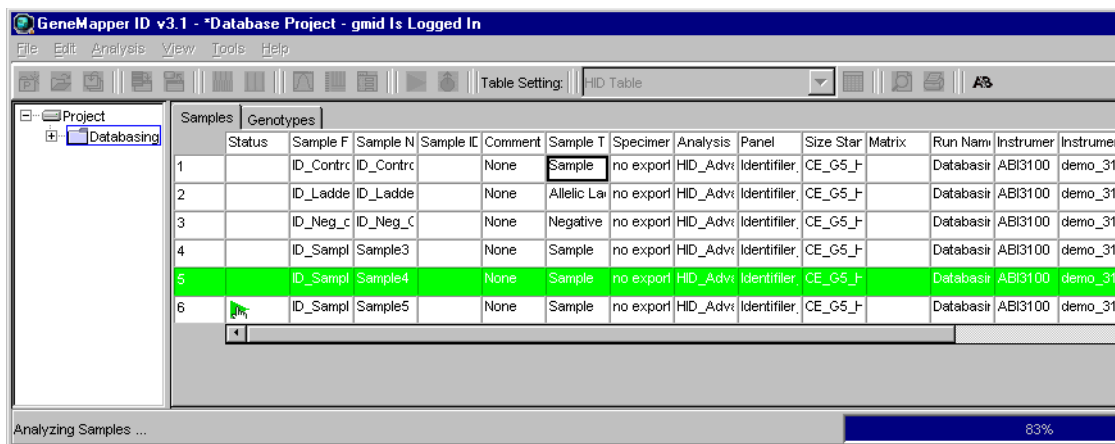


Figure 3-2 Database Project window

# Examining and Editing Results

**Overview** In this section, you examine the size standard and data, then edit labels.

**Sizing Quality** The advanced peak detection algorithm includes a sizing quality (SQ) value to assess the sizing of a sample.

You can override the SQ value assigned by the software and/or reassign incorrect size standard peaks using the Size Match Editor. Overriding the SQ value sets the value to 1.0; the override is indicated by a check mark in the SQO column.


## Examining the Size Standard

To examine the size standard:

1.	Examine the flags in the SQ column to assess sizing quality.
2.	Observe that all flags in the SQ column are green squares, indicating that all samples passed the sizing criteria.

## Examining the Allelic Ladder Calls

To examine the allelic ladder calls:


1.	Select the <b>Genotypes</b> tab.
2.	Find plots for all allelic ladders: <ol style="list-style-type: none"> <li>Select <b>Edit &gt;Find</b>.</li> <li>In the Find what field, type <b>ladder</b>.</li> <li>From the In column drop-down list, select <b>Sample Name</b>.</li> <li>Click <b>Find All</b>.</li> <li>Close the dialog box.</li> </ol>
3.	Display the HID Genotyping plot: <ol style="list-style-type: none"> <li>Click  (Display Plots) to display the Genotypes Plot window.</li> <li>Select the <b>HID Genotyping</b> plot from the drop-down list below the menu bar.</li> </ol>


To examine the allelic ladder calls: *(continued)*

4. Verify that the allelic ladder is called correctly for each marker.  
**Note:** Deselecting Controls to Top will display all panes chosen from Genotypes plots.
5. Close the Genotypes Plot window.

Examining Data and Editing Labels

To examine data and edit labels:

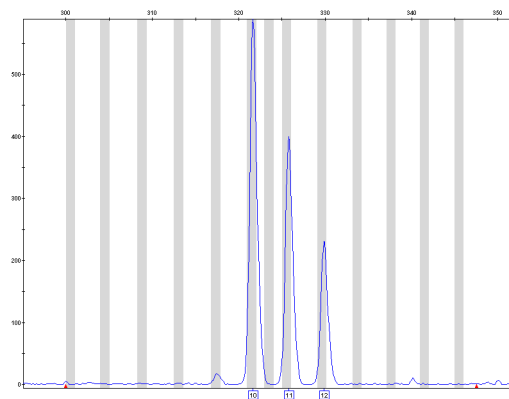
1. Click  (Low Quality to Top).  
**Note:** This option can be set as a default in Analysis options (see page 1-29).
2. Select **Sample5, Marker CSFIPO**, which displays yellow triangles (Check) in the Peak Height Ratio (PHR) and Allele Number (AN) columns and a red octagon (Low Quality) in the Genotype Quality (GQ) column.  

Sample Name	Run Name	Panel	Marker	Dye	Allele 1	Allele 2	AE Com	AE Com	AD	AE	OS	BIN	PHR	LPH	SPU	AN	BD	CC	Ovl	Lo	
1	Sample5	Databasing	Identifiler_v3	CSFIPO	G	24	242														
2	ID_Ladder	Databasing	Identifiler_v3	D21S11	B	24	242														
3	Sample3	Databasing	Identifiler_v3	D18S51	Y	15	18														
4	ID_Ladder	Databasing	Identifiler_v3	D18S51	Y	7	9														
5	ID_Ladder	Databasing	Identifiler_v3	D3S1358	G	12	13														
6	ID_Ladder	Databasing	Identifiler_v3	D16S539	G	5	8														
7	ID_Ladder	Databasing	Identifiler_v3	D2S1338	G	15	16														
8	ID_Ladder	Databasing	Identifiler_v3	TPOX	Y	6	7														
9	ID_Ladder	Databasing	Identifiler_v3	D8S1179	B	8	9														
10	ID_Ladder	Databasing	Identifiler_v3	FGA	R	17	18														
11	ID_Ladder	Databasing	Identifiler_v3	D19S433	Y	9	10														
12	ID_Ladder	Databasing	Identifiler_v3	WVA	Y	11	12														
13	ID_Ladder	Databasing	Identifiler_v3	CSFIPO	B	6	7														
14	ID_Ladder	Databasing	Identifiler_v3	TH01	G	4	5														
15	ID_Ladder	Databasing	Identifiler_v3	D13S317	G	8	9														
16	ID_Ladder	Databasing	Identifiler_v3	AMEL	R	X	Y														
17	ID_Ladder	Databasing	Identifiler_v3	D5S818	R	7	8														
3. Click  (Display Plots) to open the Genotypes Plot window and select the **HID Genotyping** plot from the drop-down list below the menu bar.



To examine data and edit labels: *(continued)*

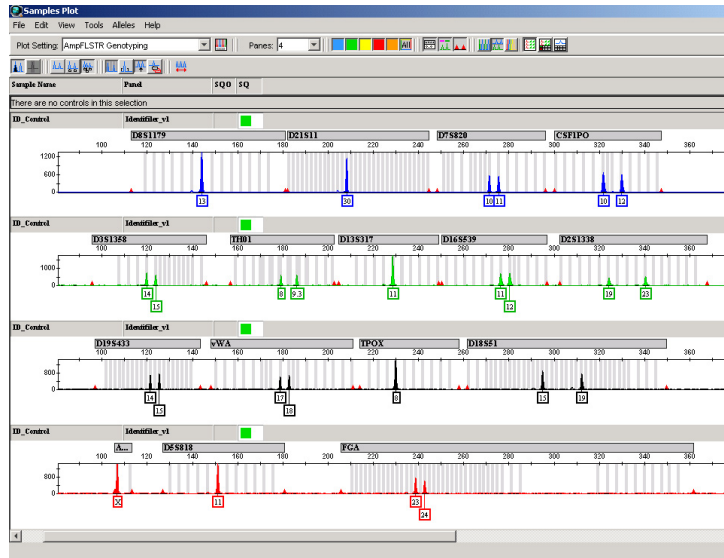
4. Observe that there are three alleles for the marker.



5. Staying within the bounds of the electropherogram, select the three peaks by clicking to the left of the first peak, and then dragging to the right of the last peak before releasing.

## To examine data and edit labels: (continued)
































6. Observe the peaks within the context of a sample:
  - a. With the peaks selected in the Genotypes Plot window, return to the Project window.
  - b. Select the **Samples** tab to open the Samples Plot window displaying the HID Genotyping plot.



**Note:** GeneMapper *ID* software remembers the last plot used in the Samples Plot window and Genotypes Plot window independently.

7. Continue with troubleshooting:
  - a. Observe that there are no other indications of a mixture in the sample.
  - b. Select **View > Raw Data** for further examination.

## To examine data and edit labels: (continued)

8.	<p>Override the genotype for the tri-allelic sample:</p> <ol style="list-style-type: none"> <li>Return to the Samples Plot window and click  (Genotypes Table).</li> <li>Select the row containing the red octagon in the GQ column.</li> <li>Right-click, and then click <b>Yes</b> in the dialog box.</li> <li>Observe that the GQ flag changed from a red octagon (Low Quality) to a green square (Pass) and that the other flags changed to gray triangles to indicate that the sample was edited.</li> </ol> <table border="1" data-bbox="548 618 1157 696"> <thead> <tr> <th>OS</th> <th>BIN</th> <th>PHR</th> <th>LPH</th> <th>SPU</th> <th>AN</th> <th>BD</th> <th>CC</th> <th>OVL</th> <th>GQ</th> </tr> </thead> <tbody> <tr> <td></td> <td></td> <td></td> <td></td> <td></td> <td></td> <td></td> <td></td> <td></td> <td></td> </tr> </tbody> </table>	OS	BIN	PHR	LPH	SPU	AN	BD	CC	OVL	GQ										
OS	BIN	PHR	LPH	SPU	AN	BD	CC	OVL	GQ												
																					
9.	Close the Samples Plot window and save the project.																				

## Exporting the Table (Optional)

You may export the table data as a tab-delimited text file that can be used with spreadsheet software.

**Note:** To export a table for the CODIS database, see Chapter 4.

## To export the table:

1.	In the Genotypes tab view, select <b>File &gt; Export Table</b> .
2.	Select a location for the file.
3.	Enter <b>Database Table</b> for the file name.
4.	Click <b>Export Table</b> .
5.	Using Microsoft® Excel® software or equivalent spreadsheet software, open the exported table file.



This chapter covers:

About CODIS .....	4-2
CODIS Export Manager .....	4-3
CODIS Table Export .....	4-5

## About CODIS

### GeneMapper *ID* Software Features

GeneMapper® *ID* software can export data from the analysis in a format suitable for the FBI Laboratory Combined DNA Index System (CODIS). For more information about CODIS, see:

<http://www.fbi.gov/hq/lab/codis/index1.htm>

### CODIS Requirements

Creation of CODIS CMF files from GeneMapper *ID* software requires that:

- Genotypes and specimen categories for shared markers are identical for each sample tested with:
  - AmpF $\mathcal{L}$ STR® Profiler Plus™ Kit
  - COfiler® PCR Amplification Kit
  - More than one kit with shared markers
- A specimen category is selected for each sample to be exported.
- Samples designated as a positive control, negative control, or allelic ladder cannot be exported.
- A sample must have at least one allele call.
- Allele calls contain acceptable characters: numbers, decimal points, letters X and Y for Amelogenin allele calls, and the symbols less than (<) and greater than (>).

# CODIS Export Manager

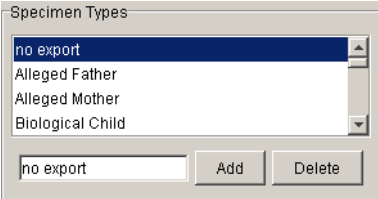
**Overview** In this section, use the CODIS Export Manager to view and set values for three fields required for exporting samples to CODIS:

- Specimen Types
- Source Lab ID
- Destination Lab ID

## Setting CODIS Export Fields

For this tutorial, you create a Source Lab ID and a Destination Lab ID. For future CODIS export procedures, CODIS laboratories should use their assigned Source Lab ID and Destination Lab ID.

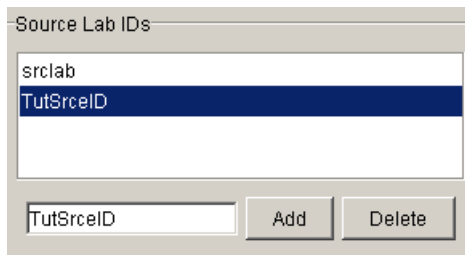
To set CODIS export fields:

1.	Select <b>Tools &gt; CODIS Export Manager</b> .
2.	<p>View the Specimen Types.</p> <p>The specimen types included in GeneMapper <i>ID</i> Software version 3.1 are currently accepted by CODIS.</p> <p>When CODIS accepts a new specimen type, you can add the specimen type to the software by typing it in the text box and then clicking <b>Add</b>.</p> 

To set CODIS export fields: *(continued)*

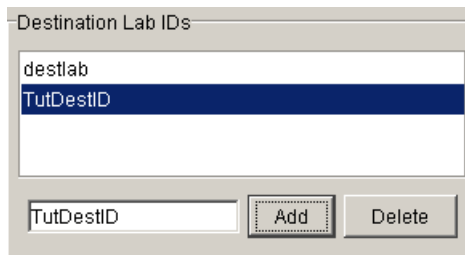
3. Add a Source Lab ID.
  - a. Type **TutSrceID** in the text box.
  - b. Click **Add**.

**Note:** The Source Lab ID cannot exceed nine characters.



4. Add a Destination Lab ID.
  - a. Type **TutDestID** in the text box.
  - b. Click **Add**.

**Note:** The Destination Lab ID cannot exceed nine characters.



5. Click **OK** to save the changes and close the CODIS Export Manager.



# CODIS Table Export

**Overview** In this section:

1. Modify columns for samples that will be exported.
2. Export a table for CODIS.

**CODIS Specimen Number** The CODIS Specimen Number (up to 24 characters) is accessed from:

- The Sample Name field, if the UD1 column is not used
- The UD1 column, if the UD1 column is used

**Note:** For more information about the UD1 column, see page 2-28.

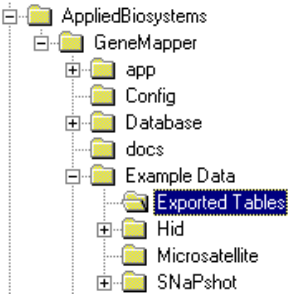
**Modifying Columns** To modify columns:

1.	<p>Open the project:</p> <ol style="list-style-type: none"> <li>a. Select <b>File &gt; Open</b>.</li> <li>b. Select <b>Database Project</b>.</li> </ol>																					
2.	<p>For each row, make the appropriate setting in the Specimen Category column:</p> <ol style="list-style-type: none"> <li>a. Use the default setting of <b>no export</b> for allelic ladders and controls.           <p><b>Note:</b> Exporting sample types designated as allelic ladders or controls generates an error message.</p> </li> <li>b. Select <b>Convicted Offender</b> for Sample3, Sample4, and Sample5.</li> </ol> <table border="1" data-bbox="521 1204 1107 1482"> <thead> <tr> <th>Sample Name</th> <th>Sample Type</th> <th>Specimen Category</th> </tr> </thead> <tbody> <tr> <td>ID_Control</td> <td>Positive Control</td> <td>no export</td> </tr> <tr> <td>ID_Ladder</td> <td>Allelic Ladder</td> <td>no export</td> </tr> <tr> <td>ID_Neg_Cntrl</td> <td>Negative Control</td> <td>no export</td> </tr> <tr> <td>Sample3</td> <td>Sample</td> <td>Convicted Offender</td> </tr> <tr> <td>Sample4</td> <td>Sample</td> <td>Convicted Offender</td> </tr> <tr> <td>Sample5</td> <td>Sample</td> <td>Convicted Offender</td> </tr> </tbody> </table>	Sample Name	Sample Type	Specimen Category	ID_Control	Positive Control	no export	ID_Ladder	Allelic Ladder	no export	ID_Neg_Cntrl	Negative Control	no export	Sample3	Sample	Convicted Offender	Sample4	Sample	Convicted Offender	Sample5	Sample	Convicted Offender
Sample Name	Sample Type	Specimen Category																				
ID_Control	Positive Control	no export																				
ID_Ladder	Allelic Ladder	no export																				
ID_Neg_Cntrl	Negative Control	no export																				
Sample3	Sample	Convicted Offender																				
Sample4	Sample	Convicted Offender																				
Sample5	Sample	Convicted Offender																				

## Exporting the CODIS Table

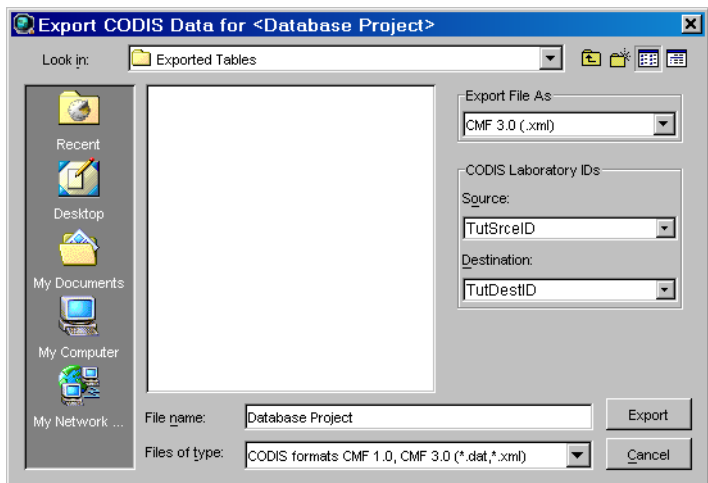
**Note:** GeneMapper ID software exports a composite genotype for samples containing concordant genotypes for shared markers from the same sample. For example, 13 locus STR profile with sample amplified using both AmpF $\mathcal{L}$ STR Profiler Plus and AmpF $\mathcal{L}$ STR COfiler kits. If you attempt to export nonconcordant profiles, the software reports an error message because CODIS will not accept the file. Resolve discrepancies to export a composite profile.

### To export the CODIS table:

1.	<p>Create a new folder called <b>Exported Tables</b>, and place it in the Tutorial Data folder:</p> <p><i>X</i>:\Applied Biosystems\GeneMapper\Example Data\ Exported Tables</p> <p><b>Note:</b> <i>X</i> is the drive where you installed GeneMapper <i>ID</i> software.</p> 
2.	<p>Return to the GeneMapper <i>ID</i> software Project window, and select <b>File &gt; Export Table for CODIS</b>.</p> <p>The Export CODIS Data dialog box opens.</p>

**To export the CODIS table: (continued)**

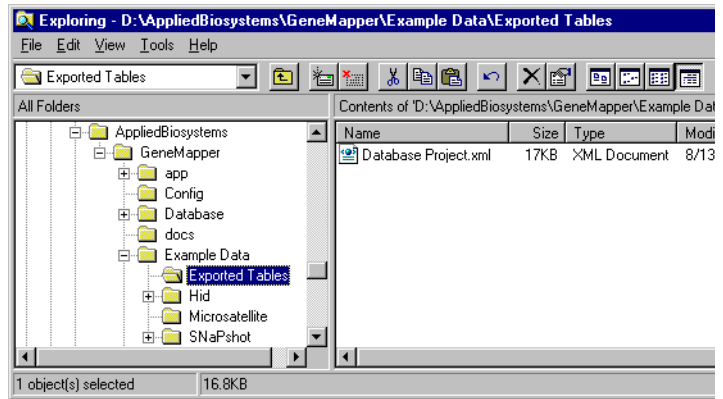
3. Make the following selections:
  - a. Look in: *X*:\GeneMapper\Example Data\Exported Tables  
**Note:** *X* is the drive where you installed GeneMapper *ID* software.
  - b. Export File As: **CMF 3.0 (.xml)**  
**Note:** GeneMapper *ID* software supports CMF 1.0 (.dat) file types for export.
  - c. Source: **TutSrcID**
  - d. Destination: **TutDestID**
  - e. File name (default): **Database Project**  
**Note:** The default file name for CODIS export is the project name.



To export the CODIS table: *(continued)*

4. Click **Export**.

The file is exported to the Exported Tables folder.



# Additional Information

---

# A

This appendix covers:

Size Standard Definitions . . . . .	A-2
Peak Detection Algorithms . . . . .	A-3
Lists of Tables and Procedures in This Tutorial . . . . .	A-4
Genotyping Samples Manually . . . . .	A-6

## Size Standard Definitions

**Size Standard Definitions Provided** The definitions for the following size standards are provided with GeneMapper® ID software version 3.1 for use with the Advanced algorithm:

- 377\_F\_HID\_GS500
- 377\_G5\_HID\_GS500
- CE\_F\_HID\_GS500
- CE\_G5\_HID\_GS500

**Fragment Sizes** The table below lists the fragment sizes for each size standard.

Table A-1 Size standard fragment sizes

Size Standard	Fragment Sizes (bp)
377_F_HID_GS500	75, 100, 139, 150, 160, 200, 250, 300, 340, 350, 400
377_G5_HID_GS500	75, 100, 139, 150, 160, 200, 250, 300, 340, 350, 400, 450
CE_F_HID_GS500	75, 100, 139, 150, 160, 200, 300, 340, 350, 400
CE_G5_HID_GS500	75, 100, 139, 150, 160, 200, 300, 340, 350, 400, 450

---

# Peak Detection Algorithms

- Available Algorithms**
- **Basic** – Contains limited parameters that may not provide enough user control over data analysis for desired results.
  - **Classic** – Includes the same parameters and the same size caller and produces the same results as GeneScan® Software version 3.1.2 designed for use with the Macintosh® operating system.
  - **Advanced** – Provides the most user control over data analysis. Includes the same parameters as, and produces similar results to, GeneScan software designed for use with the Microsoft® Windows NT® operating system, with the exception of the smoothing function. Smoothing in GeneMapper *ID* software applies to both the electropherogram and data table. The Advanced algorithm also includes a new size caller with a quality value based on the fit of the size standard definition to the actual size standard in the sample.

- PQV System Description**
- The Process Component-Based Quality Value (PQV) system monitors the major components of the size-calling and allele-calling process. The quality values:
- Are reported by GeneMapper *ID* software as an aid to flag criteria related to sample preparation, PCR, separation, detection, and analysis for each marker
  - Are weighted by the user and represented as green squares (Pass), yellow triangles (Check), and red octagons (Low Quality)
  - Do not affect the genotypes called by the software and can be manually overridden by the user

The final conclusions made by the examiner of the STR profile override and take precedence over any assignments made by the PQV system.

## Lists of Tables and Procedures in This Tutorial

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## Genotyping Samples Manually

To genotype samples manually:

1.	Select one lane or injection of the allelic ladder to use for genotyping.  <b>Note:</b> Applied Biosystems studies have shown that it does not matter which lane or injection of allelic ladder is selected if the alleles in the allelic ladder samples are within $\pm 0.5$ bp of each other.
2.	Compare the base pair size obtained for each sample allele peak to the sizes obtained for the allelic ladder peaks.
3.	Assign genotypes to those sample allele peaks falling within $\pm 0.5$ bp of the corresponding allelic ladder peak.  <b>Note:</b> For the allele designation for each allelic ladder peak, refer to the appropriate user guide for your AmpF $\Phi$ STR <sup>®</sup> PCR Amplification kit.

# Converting Macintosh Sample Files

# B

## Appendix Overview

This appendix describes how to use sample file conversion programs to prepare Macintosh® computer-generated fragment analysis sample files for transfer to a Microsoft® Windows®-based format and vice versa.

## Converting Macintosh Sample Files

### About Converting Sample Files

Applied Biosystems created two conversion programs that prepare sample files for transfer from a Macintosh computer to computers running Microsoft Windows NT operating systems, and vice versa. These sample file conversion programs run only on a Macintosh computer.

The sample file conversion programs do not perform the file transfer from computer to computer. They set attributes of the files so that they can be used on the destination computer. For example, when transferring a fragment analysis sample file from a Macintosh computer to a computer running the Windows operating system, a file extension is required and the conversion program adds *.fsa* to the sample file name. For more detailed information on how these conversion programs function, refer to the SimpleText file entitled “About Conversion Programs” located in the same folder as the sample file conversion programs.

### Installing Conversion Programs

To install the sample file conversion programs on a Macintosh computer:

- |    |                                                                                                                                                                           |
|----|---------------------------------------------------------------------------------------------------------------------------------------------------------------------------|
| 1. | Insert the GeneMapper <i>ID</i> software CD-ROM into your Macintosh computer's CD-ROM drive.<br><br>An icon displays for the CD-ROM on the right-hand side of the screen. |
|----|---------------------------------------------------------------------------------------------------------------------------------------------------------------------------|

**To install the sample file conversion programs on a Macintosh computer: (continued)**

2.	Double-click the CD-ROM icon. A CD-ROM window displays containing files and folders.
3.	Locate and double-click the <b>CONVFOLD</b> folder. Inside this folder are two files, CONVPROG.HQX and README.TXT, which contain the installation instructions.
4.	Copy the CONVPROG.HQX file to your local hard drive by clicking on the file, dragging the file over to the local hard drive icon, and dropping it in.
5.	Decompress the CONVPROG.HQX file by dragging and dropping it onto a program called “Stuffit Expander.” <b>Note:</b> You can download a free version of Stuffit Expander from <a href="http://www.stuffit.com/expander">http://www.stuffit.com/expander</a> . <b>Note:</b> Decompressing the CONVPROG.HQX file creates a folder on the local hard drive. This folder contains the conversion programs and the SimpleText file “About Conversion Programs.” This file is a seven-page document that describes in detail how to use the conversion programs, why they are necessary, solutions to common problems, and possible alternative programs.

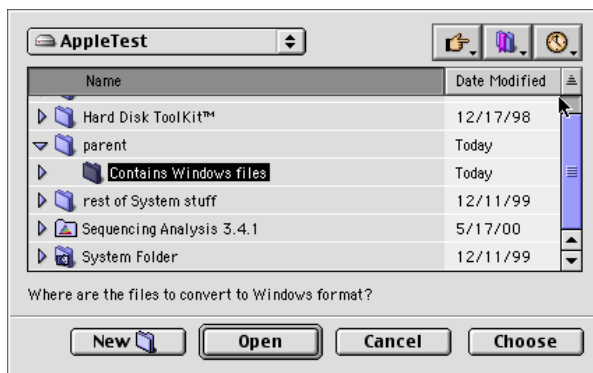
## Converting Macintosh Sample Files to Microsoft Windows Files

To convert Macintosh computer sample files for use on a computer running Microsoft Windows operating system:

1. Double-click the  icon to start the program.

Sample File Win to Mac

The following dialog box opens.



**Note:** On Macintosh computers running operating system 8.0 or less, this dialog box has a different appearance. For more information, refer to the SimpleText file “About Conversion Programs” (see the Note in step 5 above).

2. Using the triangle-shaped icons to the left of the folder names, navigate to the folder that contains the fragment analysis sample files you want to convert.

3. Select the folder by single-clicking its name.

4. Click **Choose** at the bottom of the dialog box.

If there are no problems, the program performs the task and quits automatically. When you open the folder, the sample files have the file extension *.fsa*.

**Note:** To convert sample files created on a computer running the Microsoft Windows operating system for use on a Macintosh computer, follow steps 1-3 above; in Step 1, double-click the **Sample File Win to Mac** icon.



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Printed in USA, 11/2004  
Part Number 4357520 Rev. A

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