

NCSBI Forensic Biology Section	DNA SOP	Effective Date: May 21, 2004
Title: GeneMapper ID Analysis		Revision 00

1. Open File

- 1.1. Open GeneMapper ID v3.1 or current version
- 1.2. Click on File> Add samples to project.
- 1.3. Go to My Network Places
- 1.4. Entire Network> Microsoft Windows Network> NCBCI0094
choose either:
 - 1.4.1. 3100 Workstation: for Abbott
 - 1.4.2. 3100_2: for Costello
- 1.5. Go to "D" on 3100 Workstation (for Abbott).
- 1.6. Go to "Appliedbio">3100>Data Extractor.
- 1.7. Find your run by date and the run number.
- 1.8. Highlight Run Folder or Specific Sample from the run. Add samples to list and click "Add".

2. Check 5 Columns

- 2.1. Labeling samples: Check to make sure:
 - 2.1.1. "Sample Type" and the "Sample Name" match.
 - 2.1.2. Ladder is labeled as "Allelic Ladder".
 - 2.1.3. 9947A is labeled as "Positive control".
 - 2.1.4. Negative Amplification is labeled as "Negative control".
 - 2.1.5. Samples are labeled as "Samples".
- 2.2. Spec. Category: No export
- 2.3. Analysis Method: G5-Advanced
- 2.4. Panel: Identifiler (v1 or CODIS)
- 2.5. Size Standard: G5 500Liz _Adv

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3. To Analyze Samples

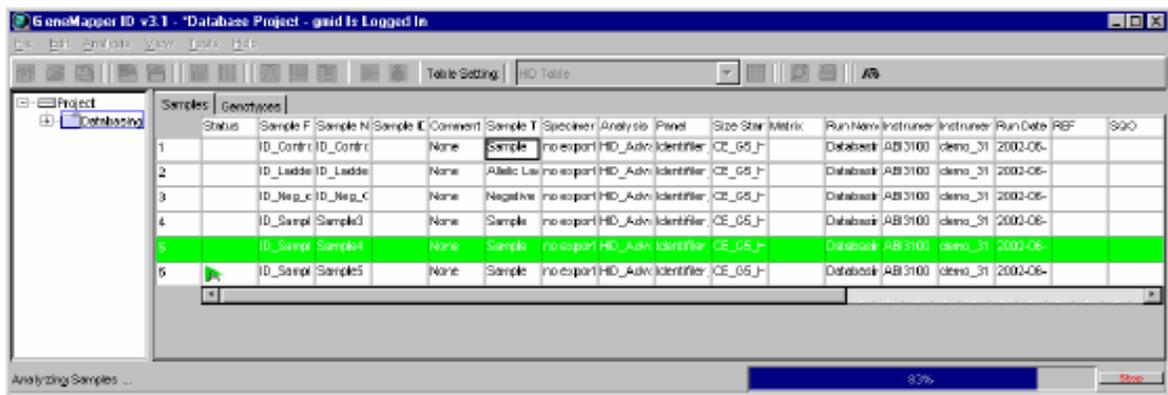
Once all samples are labeled properly,

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 2-1.

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

 - The genotypes tab becomes available after analysis.



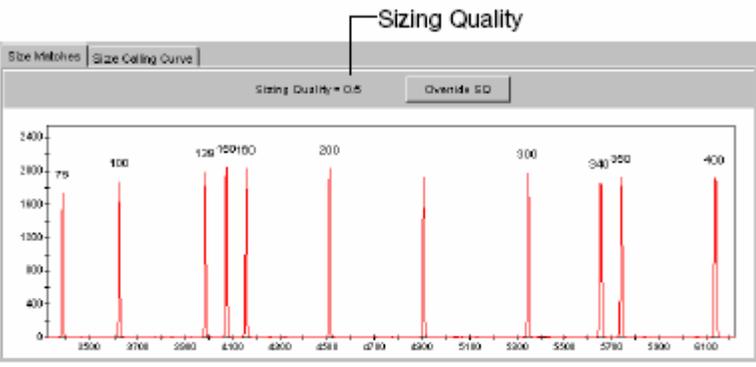
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4. Examination of Samples

- 4.1. Select Header Sample name and go to View>Raw Data.
- 4.2. Review each samples raw data visually.
- 4.3. Duplicated samples (two positive amp. controls or two ladders were run) and samples that are overblown (and are to be diluted re-analyzed later) may be removed from the project. If a sample is deleted from the project the reason for the deletion must be detailed in the analyst's notes. To delete a sample from the project, go to Project Node, select the name of the sample to be deleted, chose Edit>Delete from project .

5. Examine Size Standards

1. Select **Edit > Select All** and click  (Size Match Editor) to view the size standard for all samples.
2. Notice that the Sizing Quality value equals 0.5.


3. 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 LIZ® Size Standard) are correctly assigned for each sample.

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6. Examine the 250-bp peak

- 6.1. Select> All Samples [Ctrl A], click Display Plots icon (Multi-colored icon).
- 6.2. Select the Overlay Liz plot from the plot settings drop-down list. Click the Hide All icon, and then click only the orange square to show only the orange peaks. This is the LIZ size standard.
- 6.3. Drag down the upper box to allow you to view both sets of peaks at the same time.
- 6.4. The upper box contains the size standard peaks for the controls in your project and the lower box contains the size standards for the samples.
- 6.5. Click underneath the 250-bp peak in the upper box. Compare these results then compare the results in the lower box.
- 6.6. The selected peak will be highlighted in the table below the boxes.
- 6.7. You can scroll through the samples with the up and down arrow keys.

NOTE: The 250 bp peak was not defined in the size standard. The purpose of checking the 250-bp peak is to determine if the samples and controls are all within from the ladder. The 250-bp peaks should consistently overlap. In a typical run, the 250-bp peaks all fall within a size window of +/- 0.5 bp. If the samples are too far off (the 250 bp size standard is >+/- 0.5 bp) the sample should be re-injected.

- 6.8. Go back to the project in the Project window.

7. Examining Data

- 7.1. To check the Precision Quality Value of the samples click on the "Genotypes" tab and scroll to the right and check the Precision Quality Value (PQV).
- 7.2. Anything with a **yellow ▲** or a **red stop** sign needs to be reviewed.

8. Examine the Ladder (Note: Ensure the Plot setting is set on "HID Genotype")

- 8.1. Highlight the ladder and click the Display Plots icon  (Multi-colored icon)
- 8.2. Scroll through the ladder double checking it with the key (Separate handout)
- 8.3. If a section of the Ladder comes up with a yellow ▲, make sure the alleles are called correctly and if so you can right click on the yellow ▲ and click "yes" to override the results, and change the yellow ▲ to a green ■.
- 8.4. Continue until all sections of the ladder are green ■s.

9. Examine the controls

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- 9.1. Just as you analyzed the ladder above check your controls next. If your controls did not work properly you must re-inject or re-amp your samples.
- 9.2. There is a key also for the 9947A, K562 and MJB samples on a separate handout.

10. Examination of samples

- 10.1. If the ladder and the controls all worked properly you can continue by analyzing the samples as you did the ladder and the controls.
- 10.2. If there is a problem, look at the data by viewing the Raw Data [Ctrl F2] or view all the samples at once by viewing the samples under **HID Genotyping** (from the pull down menu)
- 10.3. If removing a called allele due to OL (Off Ladder) or pull up, stutter, dye blob, spike, etc. Left click to choose it, then right click to delete/change.
- 10.4. You must note in the comment box that appears the reason you are changing the allele call and type your initials.
- 10.5. The Raw Data showing the primer peaks and Size Standards of samples that are negative (have no observable peaks) should be printed and included in the analysts notes.

11. Save Project

Once all samples have been reviewed save the project. To save click "File">"Save".

12. Export Tables Note: Two tables must be created to export into Excel

- 12.1. Click on the "**Samples Tab**" and from the pull down menu choose "**SBI Casework Table**"
- 12.2. File> Export Table
- 12.3. Place the table in the correct folder
For the second table
- 12.4. Click on the "**Genotypes Tab**" and from the pull down menu choose "**Case Allele Table**".

13. Convert tables to Excel Format

- 13.1. Open MS Excel; then using the open command from the File menu, open the exported table. When you do this, you will be prompted to convert the file to an Excel format using the "Text Import Wizard".
- 13.2. There are 3 steps to this wizard:
 - 13.2.1. Make sure "Delimited" is checked, click "Next".
 - 13.2.2. Make sure "Tab" is checked, click "Next".

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- 13.2.3. Click on “Text” in the Column Data Format section, then click “Finish”.
- 13.3. Use the “Save as” command in the File menu to save your data as an Excel worksheet.

14. To Create the Allele Call Table

- 14.1. Open the GeneMapper Results Worksheet.
- 14.2. Click on the Results Tab at the bottom left corner of the document.
- 14.3. Click on your Exported table document and select all (Ctrl + A).
- 14.4. Copy all the data (Ctrl + C).
- 14.5. Go back to the GeneMapper Results worksheet, click on cell A1 in the “Results” section, and paste your results (Ctrl + V).
- 14.6. Click on the “Allele Call Table” tab at the bottom to see your results displayed in a table format.
- 14.7. Use the “Save as” command in the File menu to save the document, rename it, and put it into your run folder.

15. Make a copy of the folder for Second Reviewer

- 15.1. Right click on the Start Button and open Explore.
- 15.2. Find the original folder
 - 15.2.1. Right click and copy
 - 15.2.2. Right click and paste in the appropriate place for a second review.

Document Revision History		
Revision Number	Date	Reason
00	5/21/04	New Document

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APPROVAL SIGNATURES	Date
Author/Title (Print)	
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