

## Introduction to recent upgrades of the DNA sequencing software at the Plant-Microbe Genomics Facility

### 1) Sequence Scanner ver. 1.0

Applied Biosystems, which makes the DNA sequencing kit and analyzer, has released new software called Sequence Scanner ver. 1.0 which can view, edit and print the electropherogram (.ab1) file as well as generate reports about the sequence data in the selected files. The software is quite useful, and can be obtained for free from the following website: [http://marketing.appliedbiosystems.com/mk/get/sss\\_login](http://marketing.appliedbiosystems.com/mk/get/sss_login). A link to this page can also be found on the DNA sequencing page ([http://www.biosci.ohio-state.edu/~pmgf/services\\_dna\\_sequencing.htm](http://www.biosci.ohio-state.edu/~pmgf/services_dna_sequencing.htm)) of the PMGF website.

### 2) dnaLIMS

a) A new option, "Invoice Summary", is on the menu page that allows you to determine the approximate cost of the DNA sequencing reactions for the selected month.

dnaLIMS User Tools

Sequencing	User Profile	Other Tools
<ul style="list-style-type: none"> <li>■ <a href="#">Enter DNA Sequencing Requests</a></li> <li>■ <a href="#">View Your Requests</a></li> <li>■ <a href="#">Retrieve Your DNA Results</a></li> <li>■ <a href="#">Enter your Sequencing Primers</a></li> <li>■ <a href="#">Enter Fragment Analysis Requests</a></li> <li>■ <a href="#">Retrieve Fragment Analysis Results</a></li> <li>■ <a href="#">96-and-48 well Plate Requests</a></li> <li>■ <a href="#">Invoice Summary</a></li> </ul>	<ul style="list-style-type: none"> <li>■ <a href="#">Change User Password</a></li> <li>■ <a href="#">Change User Profile</a></li> </ul>	<ul style="list-style-type: none"> <li>■ <a href="#">Contact Info.</a></li> <li>■ <a href="#">Sample Preparation Guidelines</a></li> <li>■ <a href="#">Support Tools</a></li> <li>■ <a href="#">Logout</a></li> </ul> <p>Supported Browsers  <a href="#">Learn More</a></p>

b) Three additional files (text format) can be downloaded for each DNA sequencing reaction: FASTA, qual, and Phred. The FASTA file contains the text sequence formatted for uploading to the NCBI data base. The qual file has the quality score for each base. The Phred (phd) file contains the text sequence, the quality score for each base as well as the scan line from the raw data. [Note: The raw data can be viewed with Sequence Scanner ver. 1.0].

Select a Button to download ALL corresponding files listed in the results table.

Sequencer Output		Phred Output			
<a href="#">Text</a>	<a href="#">Trace</a>	<a href="#">Fasta</a>	<a href="#">Qual</a>	<a href="#">Phred</a>	<a href="#">SCF</a>
<a href="#">Text</a>	<a href="#">Chromat</a>	<a href="#">fasta</a>	<a href="#">qual</a>	<a href="#">phred</a>	<a href="#">scf</a>

To download individual files:

Mac: Hold down Control Key, Click Mouse. Select Save Linked File As...

PC: Right Mouse Click, Save Link As...

[Clear](#) [Submit](#)

Invoice 7308

Get: 825

Order#	Req#	Download	View	SeqId	WID	Sample	Primer	Investigator	UID	Date	Phred Q20	Comments
7308	57448	<a href="#">Text</a> <a href="#">Chromat</a>	<a href="#">View</a>	825-8	H1	pPXB	CBDPFD	daniels,chuck	1470	Oct 19 2005	<a href="#">phd</a> <a href="#">qual</a> 1032 <a href="#">fasta</a>	ok
7308	57449	<a href="#">Text</a> <a href="#">Chromat</a>	<a href="#">View</a>	825-9	A2	pPXB	CBDREV	daniels,chuck	1470	Oct 19 2005	<a href="#">phd</a> <a href="#">qual</a> 1081 <a href="#">fasta</a>	ok

There are 2 samples.

c) The number of bases in the reaction that have a quality score of 20 (Q20) or greater is reported on the download page, e.g. 1032 and 1081 in the image below. A score of 20 means the base call has a probability of error of 0.01. For a complete explanation of the quality score see the following link:

[http://www.biosci.ohio-state.edu/~pmgf/services\\_dna\\_sequencing\\_quality\\_value.htm](http://www.biosci.ohio-state.edu/~pmgf/services_dna_sequencing_quality_value.htm).

If no quality score or qual file is present, then the file did not contain any sequence, i.e. no bands.

Date	Phred Q20	Comments
ct 19 2005	<a href="#">phd qual 1032</a> <a href="#">fasta</a>	ok
ct 19 2005	<a href="#">phd qual 1081</a> <a href="#">fasta</a>	ok



d) All of the Q20 scores can be displayed simultaneously, based on their position in a 96-well plate, from the link shown below “[View Phred Data]”:

Clear Submit

Order Number Search

Order Number	Order#	# Samples	Order Date
7308	7308	2	Oct 19 2005
[View Phred Data]	7283	13	Oct 18 2005
	7131	18	Oct 6 2005
	7114	4	Oct 5 2005
	7012	3	Sep 28 2005

Sample Sheet Search

Sample Sheet Number	Sample Sheet	Date
[View Phred Data]	825	Oct 21 2005
	822	Oct 20 2005
	801	Oct 10 2005
	799	Oct 7 2005
	788	Sep 30 2005

If you have any questions or comments, then please contact the Facility: 614/247-6204 or [pmgf@osu.edu](mailto:pmgf@osu.edu).

mrz; 10-05