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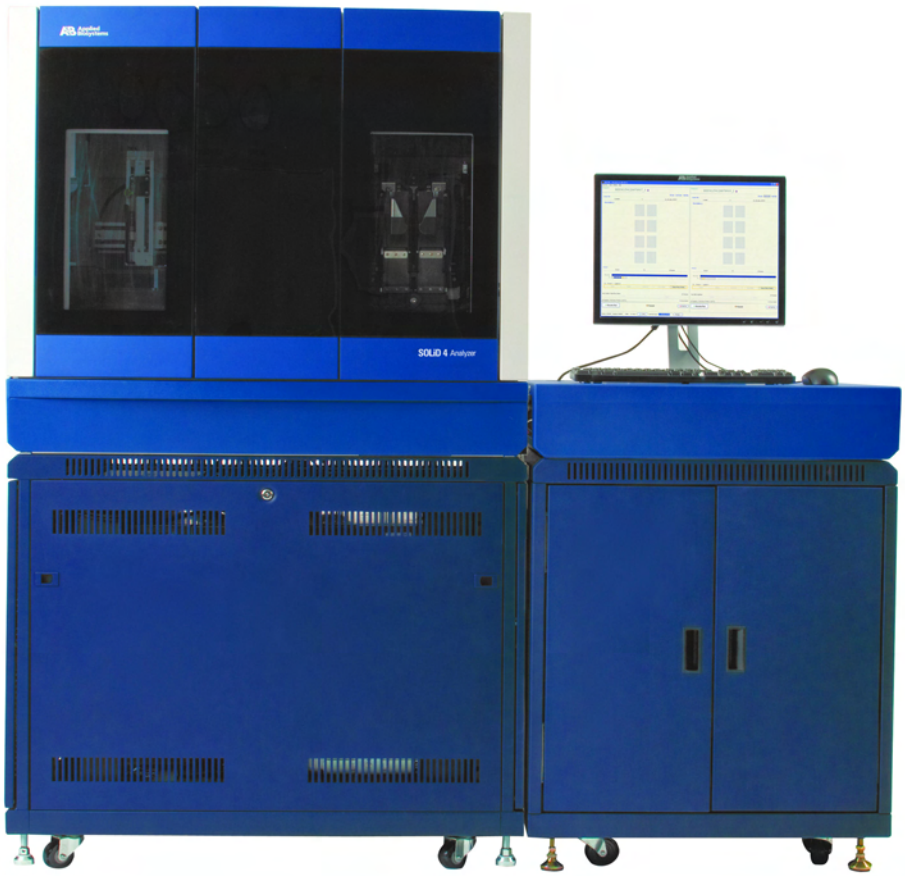
Applied Biosystems SOLiD™ 4 System

SOLiD™ SAGE™ Analysis Software v1.10 Guide

SOLiD™ SAGE™ Tag Preparation | Templated Bead Preparation | Instrument Operation



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Description of the Software

Overview

SOLiD™ SAGE™ Analysis Software v1.10 is a Linux-based program that takes the raw data files from SOLiD™ SAGE™ sequencing reads and matches them to known sequences in your reference database of choice. It is designed for use with the SOLiD™ SAGE™ Kit or the SOLiD™ SAGE™ Kit with Barcoding Adaptor Module, which generates libraries of 27-bp tags for all transcripts in a cell.

Using the simple software interface, you create a project name and location, and then identify the directories containing:

- Sequences from a SOLiD™ System read (csfasta formatted)
- A fasta-formatted database of reference sequences (*e.g.*, from RefSeq).

The software then automatically matches the experimental data with the reference data or compares two experimental data sets. The software identifies tags, tabulates tag abundances, and displays associated gene descriptions. It also sorts multiplexed libraries by their barcodes. It presents the data in a tabular format that is easy to read and export to spreadsheet or other programs for further analysis.

How Tags are Processed

The user selects the folders containing the SOLiD™ SAGE™ reads and reference sequences, and specifies the parameters of tag length and number of mismatches allowed. The software then maps the reads using only a portion of the reference sequences, to greatly reduce processing time. The algorithm extracts tag-length fragments flanking the *Nla* III sites (CATG) in the reference sequences, and concatenates unique fragments together with a spacer of NNN. Only the first tag-length bases of SOLiD™ reads are used in mapping.

Output Files

All output files are in tab-delimited text format. The output files for the individual barcoded libraries in a multiplex SOLiD™ sequencing run are stored in subdirectories of the main project directory. The output files are:

- A mapping output file that lists each tag, its frequency of occurrence, its GenBank Identifier (GI) number, and a description of the identified gene
- A mapping results file that provides more detailed information, including SOLiD™ sequencing read IDs and mismatches
- A file comparing the tags in two different samples
- A file calculating the abundances of repeat reads

To transfer the files to a Windows-based computer for analysis (e.g., by Microsoft Excel), you can use WinSCP or equivalent.

Download and Installation

System Requirements

SOLiD™ SAGE™ Analysis Software v1.10 has the following system requirements:

Linux operating system

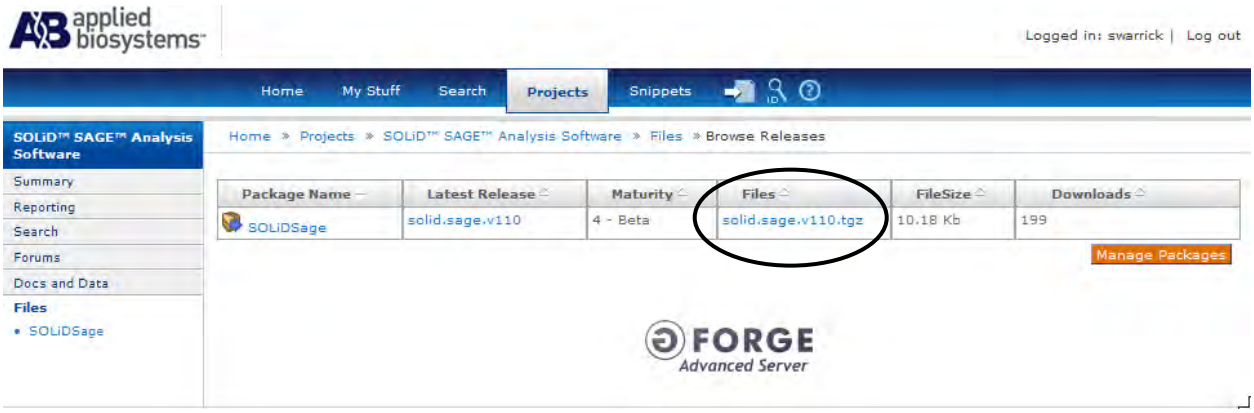
Perl Tk

Reference sequences, in NCBI fasta format (*e.g.*, RefSeq files)

≥8 gigabytes of memory—varies depending on the size of the reference genome, selected tag length, and selected number of allowed mismatches

Downloading and Installing SOLiD™ SAGE™ Software

1. To download the software, go to: http://solidsoftwaretools.com/gf/project/solid-sage
2. Click on the **Files** link in the left navigation bar, and then select the "scripts" tgz file (e.g., **solid.sage.v110.scripts.tgz**) to begin the software download.



Completing the Installation

1. Extract the downloaded **.tgz** file into the directory where you want to install the software.
2. Open **.bash_profile** in your home directory with a text editor program.
3. Add the following line, substituting the actual directory name from Step 1: **PATH=$PATH:/solid_sage_directory**
4. Add the line: **export PATH**
5. Save **.bash_profile** and exit the text editor.
6. Logout and re-login.

Downloading Sample Data

1. To download sample data, go to: http://solidsoftwaretools.com/gf/project/solid-sage
2. Click on the **Docs and Data** link in the left navigation bar, and then select **Sample Data**.
3. Select the "samples" tgz file (e.g., **solid.sage.v110.samples.tgz**) to begin the data file download.
4. The tgz file contains sample data for each mapping and comparison option, and results are provided in a corresponding directory. An example Description File for mapping multiple sequence files with different parameters is provided as well (see page 11).

Getting Started

Launching the Program

To launch the software, type **solid.sage.v110.pl**. (The software should be in your working path, so you can launch it from anywhere.)

If you have a problem launching the software, try entering the full path of the directory where you extracted the package followed by the program name.

Example launch commands:

solid.sage.v110.pl

solid.sage.v110.pl &

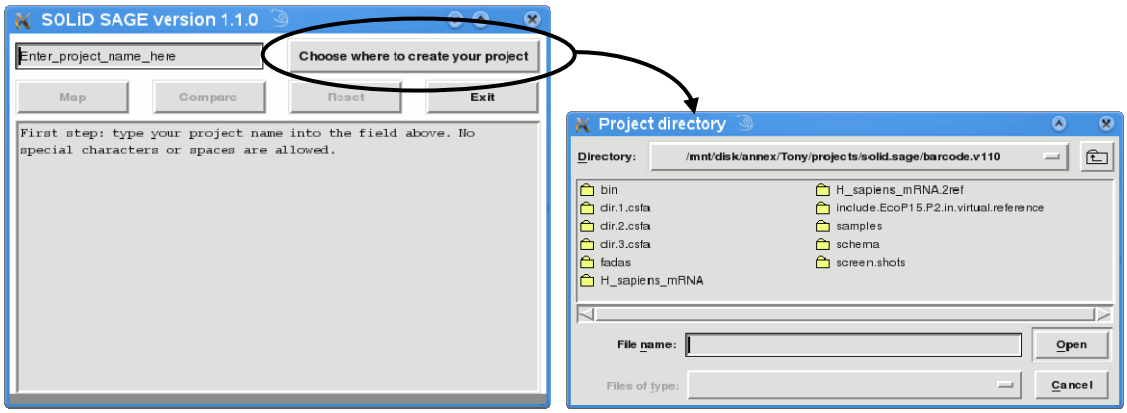
solid_sage_directory/solid.sage.v110.pl &

The program screen will open.

Creating a Project

1. To begin, enter a project name in the top left field, using only alphanumeric characters with no spaces.
2. Next, click on **Choose where to create your project** to specify the file path to the project directory. A directory with the project name you entered will be created containing temporary analysis files and final output files. (Make sure a directory with that project name does not already exist in that location.)

   Note: You must have "write" permission to the location of the project directory.
3. When you have made your selection, the full path will be displayed in the top left box, and the **Map** and **Compare** buttons will become available.



Next Steps

With the project name and location created, you can now proceed to:

Sequence Files and, starting on the next page

Comparing Two Mapped Samples, page 11

Generating Repeat Reads Statistics, page **Error! Bookmark not defined.**

Mapping Sequence Files

Sequence Files from the SOLiD™ System

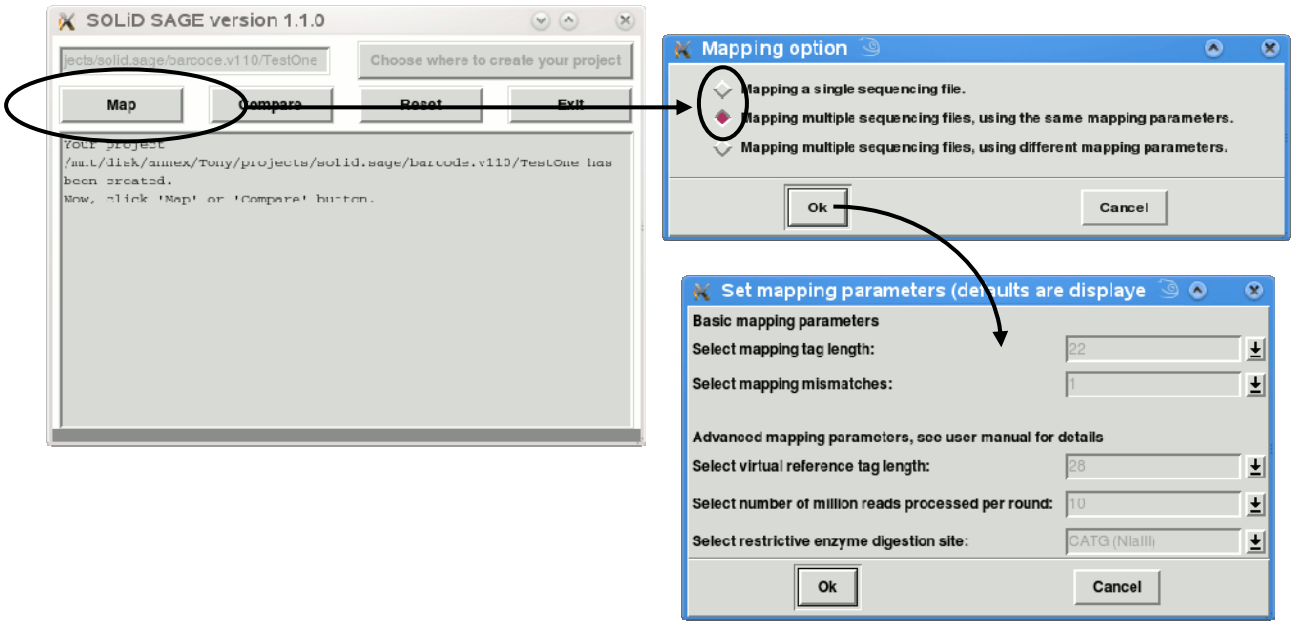
Sequences files generated by the SOLiD™ System are in the color-space fasta format (**.csfasta**). SOLiD™ SAGE™ Analysis Software v1.10 can analyze a single .csfasta results file or multiple files (e.g., from different barcoded libraries in a single run). This section describes mapping sequence files using a single set of mapping parameters.

Note: All csfasta files to be analyzed must be located in the same directory.

Besides read definition lines (beginning with a > sign) and the actual read sequence (beginning with T followed by the digit 0/1/2/3), there may be some commenting lines which begin with a # sign at the beginning of the file. The first read definition line should appear within the first 100 lines of the input reads. If it appears later than 100 lines, you will need to trim the preceding comment lines.

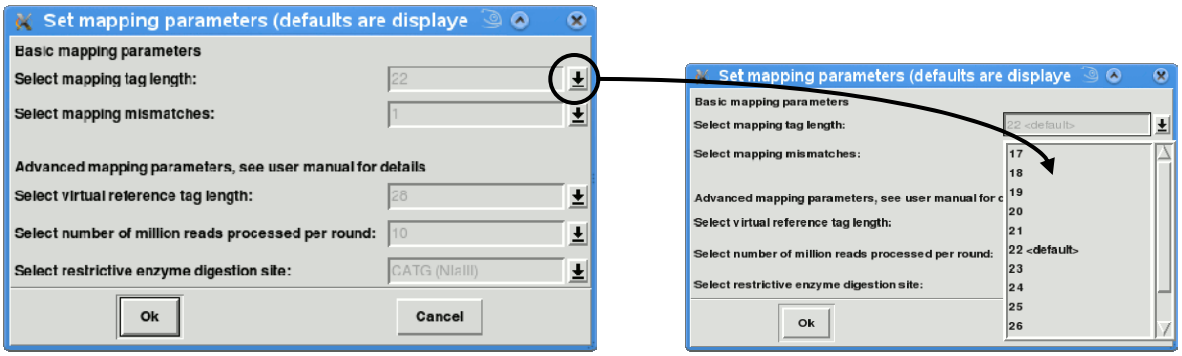
Mapping a Single File or Multiple Files with the Same Parameters

1. Click on the **Map** button.
2. Select either **Mapping a single sequencing file** or **Mapping multiple sequencing files using the same parameters**.
3. Click on **OK**. The **Set mapping parameters** dialog will open.



Selecting Mapping Parameters

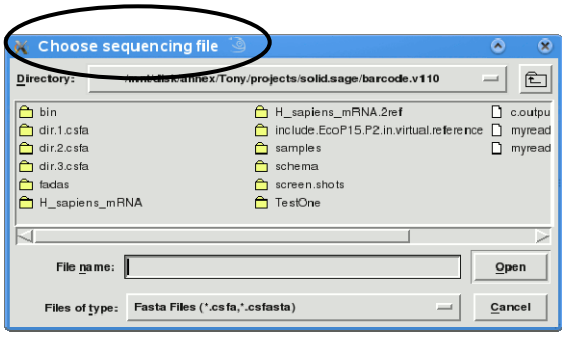
In the **Set mapping parameters** dialog, the default parameters are preselected. Click on the pulldown arrow to change each selection, then click on **OK**.



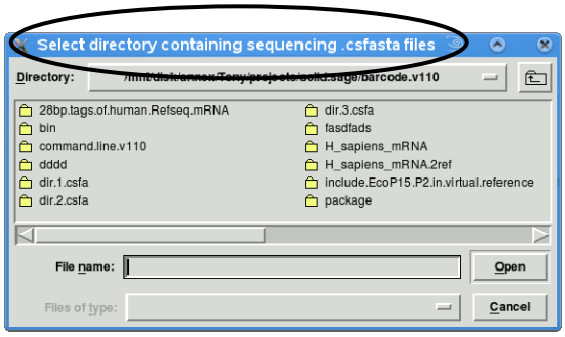
Parameter	Default	Explanation
Mapping tag length	22	The number of bases after the restriction enzyme site (CATG) in each tag used for mapping.
Mapping mismatches	1	The number of base mismatches allowed during mapping. Empirical data have shown that 1 mismatch provides the optimal balance of mapping accuracy and robustness. Increasing the number of mismatches allowed will decrease accuracy and increase processing time.
Virtual reference tag length	28	The length of each reference sequence following a restriction anchor site that is analyzed for mapping purposes.
Number of million reads processed per round	10	The number of tags (in millions) processed at one time by the software. If this number is less than the total number of tags in a run, the software will process the tags in multiple "rounds" of processing and then combine the data into a single result. This enables slower computers to process large numbers of tags without crashing, but also increases processing time.
Restriction enzyme digestion site	CATG (*Nla* III)	The default restriction enzyme used by the SOLiD™ SAGE™ System is *Nla* III. Alternative workflows may use a different restriction enzyme.

Selecting the Sequencing File(s)

After you select the mapping parameters, you will be prompted to select either a single .csfasta file or the directory containing multiple files, depending on your selection in the **Mapping option** dialog box. Multiple files must be located in the same directory, and all the files in that directory will be analyzed.



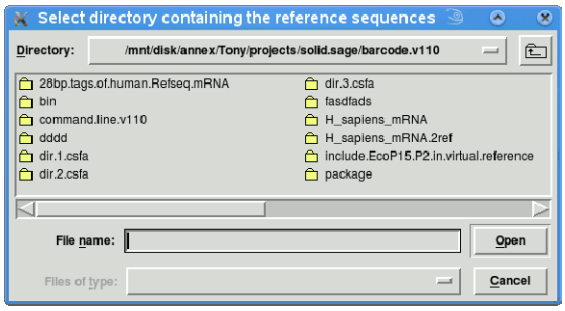
or



Selecting the Reference Directory

After you have selected a sequence file or directory and specified the mapping parameters, you will be prompted to select the directory containing the reference sequences.

Note: You do not select the reference files themselves—only the directory. The software will use every fasta file in that directory for its analysis. See **Reference File Format** below.



Reference File Format

In general, the reference files should have the same format as the fasta files from an NCBI RefSeq database. You can simply download the appropriate gDNA/mRNA database from NCBI RefSeq and use it as your reference.

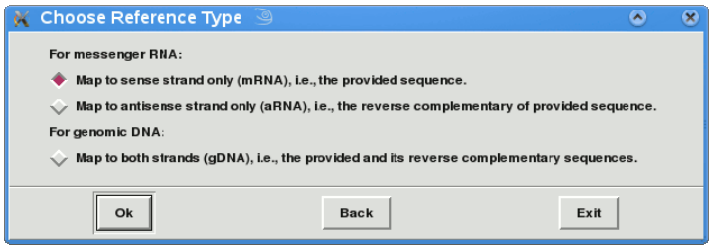
The reference files must be in the fasta format (**.fasta**). The definition lines of the reference sequences should have an NCBI RefSeq sequence format or have the following structure: >gi|xxx|ref|yyy

… where xxx is the GI number and yyy is the gene name. There is no length restriction on the GI number or gene name.

The fasta file can have multiple sequences, but each definition line should follow the structure defined above. No comment lines are allowed.

Selecting the Reference Type

After selecting the reference library, choose whether to map tags to the **sense strand** for mRNA, **antisense strand** for aRNA, or **both strands** for genomic DNA.



Note on Virtual Reference Sequences

The program generates a virtual reference sequence that is much shorter than the actual reference sequence so that the analysis will run faster. The virtual reference sequence consists of fragments of a pre-set length—either the default 28 base pairs that follow the *Nla* III anchor site CATG or a user-selected tag length (as selected under **Select mapping parameters**).

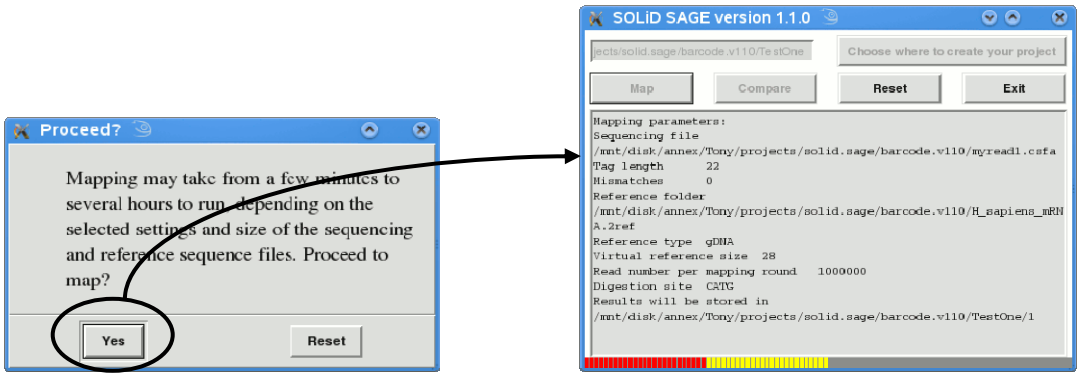
For mRNA or aRNA mapping, only the anchor site CATG and the following bases on the sense or antisense strand will be used as the virtual reference.

If genomic DNA mapping, CATG and the following bases on both strands—the provided sequence and its reverse complementary sequence—will be used as the virtual reference.

Running the Analysis

After you select the reference type and click on **OK**, you will be prompted to proceed to mapping. Click on **Yes**. After a short processing delay, a progress bar will appear at the bottom of the main software window.

The length of time required for mapping will vary depending on the size of the reference database, the selected tag length, and the number of mismatches allowed. Complex analyses may take several hours.



Output and Results Files

For each fasta file in the reference directory, the SOLiD™ SAGE™ Analysis Software generates a corresponding subdirectory in the selected project directory.

In each subdirectory, two files are generated by the mapping program:

A mapping Output file (**output.tab**) that lists each tag, its frequency of occurrence, its associated GenBank Identifier (GI) number, and a brief description of the identified gene

A mapping Results file (**results.tab**) that provides additional information, including SOLiD™ sequencing read IDs and mismatches

Output file example:

	A	B	C	D
1	Tag	Count	GI	Description
2	CATGAAAAAACTCCAAATAAGAGAATC	1	GI33413399	>gi|33413399|ref|NM_001984.1| Homo sapiens esterase D/formylglutathione hydrolase (ESD), mRNA
3	CATGAAAGGGTCACTTCTGTAATAGTG	1	GI81158221::GI81158223::GI81158225	>gi|81158221|ref|NM_001037133.1| Homo sapiens neuronal cell adhesion molecule (NRCAM), transcript variant 3, mRNA::>gi|81158223|ref|NM_005010.3| Homo sapiens neuronal cell adhesion molecule (NRCAM), transcript variant 2, mRNA::>gi|81158225|ref|NM_001037132.1| Homo sapiens neuronal cell adhesion molecule (NRCAM), transcript variant 1, mRNA
4	CATGAACAACCGGCTGGCCGAGACCAG	1	GI11545760	>gi|11545760|ref|NM_022055.1| Homo sapiens potassium channel, subfamily K, member 12 (KCNK12), mRNA
5	CATGAACTTGATACGTCCGTGTGTCCC	1	GI53759150	>gi|53759150|ref|NM_005063.4| Homo sapiens stearoyl-CoA desaturase (delta-9-desaturase) (SCD), mRNA
6	CATGAAGATGATATGAGGCCGGGGCGG	1	GI52145308	>gi|52145308|ref|NM_032808.5| Homo sapiens leucine rich repeat and Ig domain containing 1 (LINGO1), mRNA
7	CATGAAGGAAGATCCCACAGTCTCAGC	1	GI4758483	>gi|4758483|ref|NM_004832.1| Homo sapiens glutathione S-transferase omega 1 (GSTO1), mRNA
8	CATGACAGCCCTCTGCTCTTGAGTACC	1	GI39812204	>gi|39812204|ref|NM_025164.3| Homo sapiens KIAA0999 protein (KIAA0999), mRNA
9	CATGACGGAACAATAGGACTCCCCAGG	2	GI38505192	>gi|38505192|ref|NM_000954.5| Homo sapiens prostaglandin D2 synthase 21kDa (brain) (PTGDS), mRNA
	CATGACGTGTCTATGTCAAAAGTTCTT	1	GI115583669	>gi|115583669|ref|NM_003253.2| Homo sapiens T-cell lymphoma invasion and

Results file example:

	A	B	C	D	E
1	Tag_Seq	GI_num	GI_Pos	Read_ID	Mismatch
2	CATGTGCAAATAAATGTGGCTTAGACT	133908618	3298	>665_1155_42_F3	0
3	CATGGTAATAAAATATGAATGATAAAA	189083841	2125	>665_817_382_F3	0
4	CATGGTAATAAAATATGAATGATAAAA	189083835	2280	>665_817_382_F3	0
5	CATGGTAATAAAATATGAATGATAAAA	189083837	2146	>665_817_382_F3	0
6	CATGGTAATAAAATATGAATGATAAAA	189083839	2128	>665_817_382_F3	0
7	CATGGTAATAAAATATGAATGATAAAA	51593094	2283	>665_817_382_F3	0
8	CATGGTAATAAAATATGAATGATAAAA	132814488	2268	>665_817_382_F3	0
9	CATGTGATGGGCATTGAGCCACACCTC	34147410	1140	>665_1807_1132_F3	0
10	CATGAGGAGCTCGGCTTAAAATGTCTT	40254847	2324	>665_1994_720_F3	0
11	CATGAAGATGATATGAGGCCGGGGCGG	52145308	1899	>668_309_447_F3	0

Transferring Files

To transfer the output files to a Windows-based computer for analysis using a spreadsheet program such as Microsoft Excel, you can use WinSCP or equivalent.

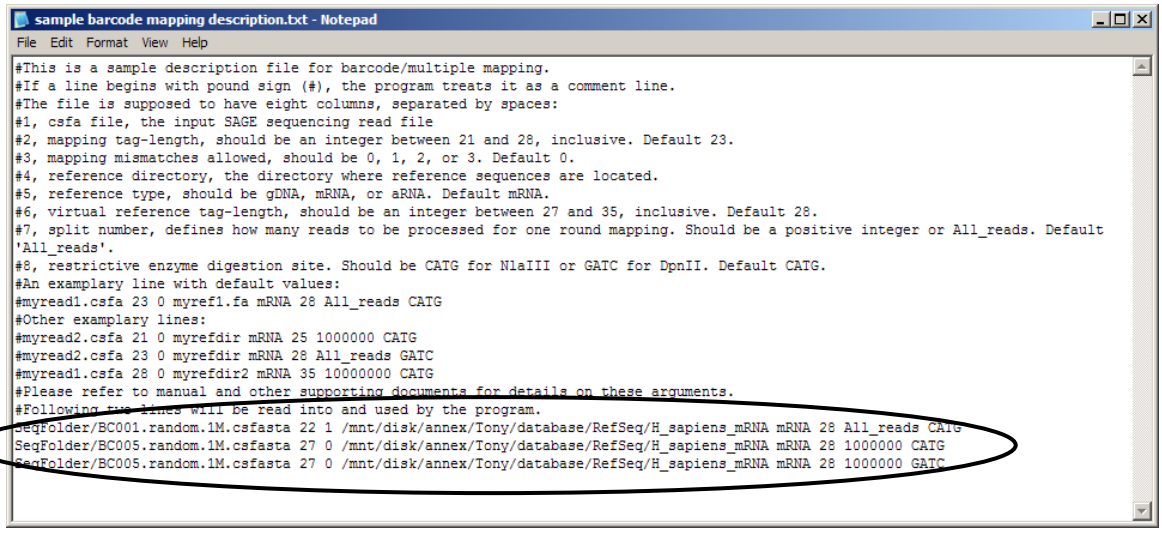
Mapping Multiple Sequence Files with Different Parameters

Description File

You can map multiple sequence files (in .csfasta format) with different mapping parameters using a **Description File** that contains all the details about each sequence file, the parameters to apply, and the reference directories to use.

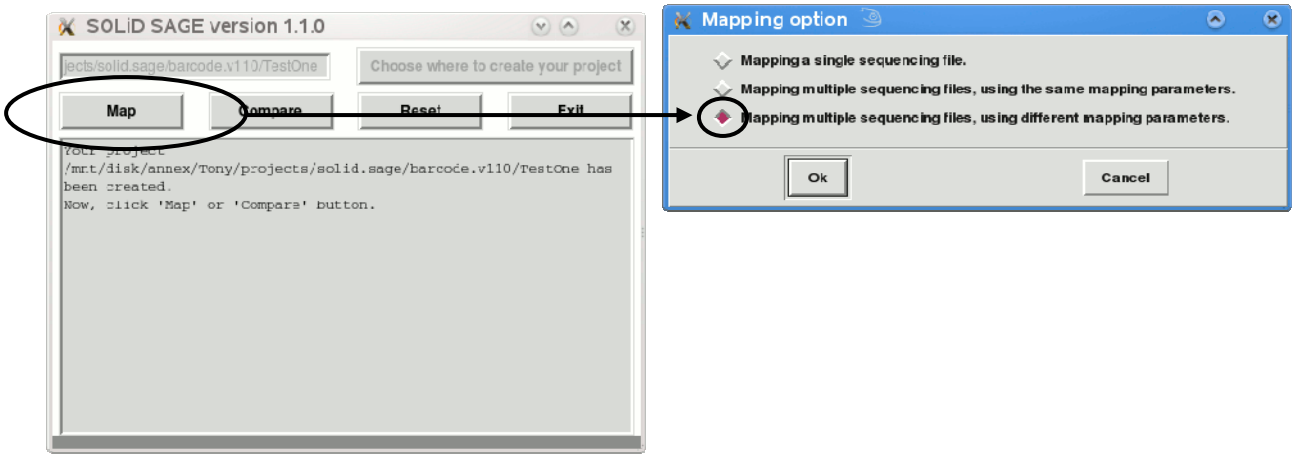
An example Description File is available as part of the sample data download. See **Downloading Sample Data** on page 5.

The example Description File is shown below. The comment lines at the top describe the different elements, and an example format is shown in the circled area, with a single space between each element.



Creating and Selecting the Description File

1. Create the Description File using the example provided with the software and shown above.
2. Launch the program and create a project, then click on the **Map** button.
3. Select **Mapping multiple sequencing files using different mapping parameters**.
4. Click on **OK**. The **Open mapping description file** dialog will open.
5. Select the Description File you created and click on **Open**.
6. The software will proceed to mapping, as described in **Running the Analysis**.



Comparing Two Mapped Samples

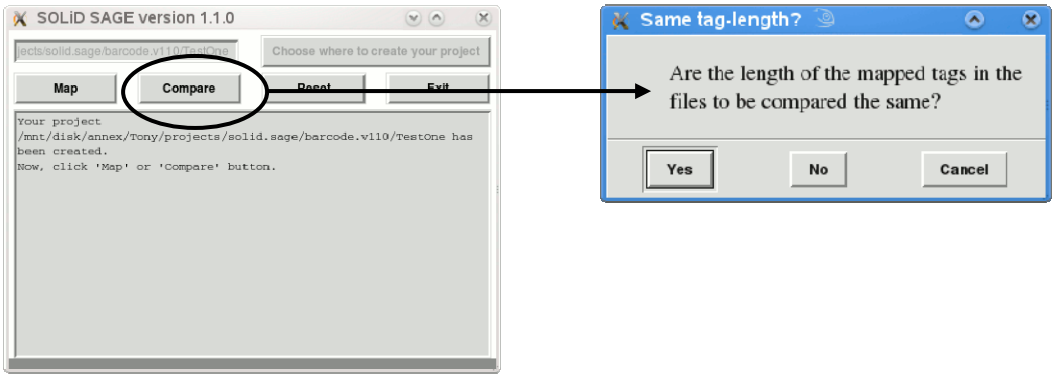
Introduction

You can compare two previously mapped libraries, as described in this section.

Selecting the Sequencing Reads to Compare

1. Launch the program and create a project, as described in **Getting Started**.
2. Click on the **Compare** button in the program screen and follow the on-screen steps to select the files to compare. These will be Output files from previous mapping runs (**output.tab** format; see page 10). Do *not* compare results files.
3. The software will find common tags between the libraries and identify the corresponding tag counts and genes. When the comparison is complete, you will receive an alert.

Note: If the lengths of the tags in the two libraries are different, all tags will be truncated to the shortest length before comparison. Note that the gene description may not match the truncated tags as well as the non-truncated ones. Be careful when comparing libraries with different tag lengths.



Comparison File

The Comparison file generated by the analysis (**comparison.tab**) lists each tag, its frequency of occurrence in each sample, and its GenBank identification.

	A	B	C	D
1	Tag_Sequence	File1	File2	
2	CATGAAGACAGTGGCTGGCGGTGCCTG	4715	5430	(Based on non-truncated tag) >gi|78214519|ref|NM_000998.4| Homo sapiens ribosomal protein L37a (RPL37A), mRNA
3	CATGCACAAACGGTAGTTTTGTGTGTT	2361	2604	(Based on non-truncated tag) >gi|169202035|ref|XM_001726056.1| PREDICTED: Homo sapiens similar to hCG2027326 (LOC100129905), mRNA::>gi|169203086|ref|XM_001725509.1| PREDICTED: Homo sapiens similar to hCG2027326 (LOC100129905), mRNA::>gi|169203606|ref|XM_001726007.1| PREDICTED: Homo sapiens similar to hCG2027326 (LOC100129905), mRNA::>gi|68160923|ref|NM_001030.3| Homo sapiens ribosomal protein S27 (metallopanstimulin 1) (RPS27), mRNA
4	CATGACAACAAAGAAAAAGACCTTGTA	1736	1606	(Based on non-truncated tag) >gi|56550064|ref|NM_001008220.1| Homo sapiens complexin 2 (CPLX2), transcript variant 2, mRNA::>gi|56550103|ref|NM_006650.3| Homo sapiens complexin 2 (CPLX2), transcript variant 1, mRNA
5	CATGAATATGTGGGCTAAGAAATAGTT	1659	1980	(Based on non-truncated tag) >gi|17999531|ref|NM_004374.2| Homo sapiens cytochrome c oxidase subunit VIc (COX6C), mRNA
6	CATGAAGCTGAGGTCTTGAAGCAGCTG	1558	715	(Based on non-truncated tag) >gi|44889961|ref|NM_005563.3| Homo sapiens stathmin 1/oncoprotein 18 (STMN1), transcript variant 3, mRNA::>gi|44890049|ref|NM_203399.1| Homo sapiens stathmin 1/oncoprotein 18 (STMN1), transcript variant 2, mRNA::>gi|44890051|ref|NM_203401.1| Homo sapiens stathmin 1/oncoprotein 18 (STMN1), transcript variant 1, mRNA
7	CATGAACTAATACTACAATAAAGGATG	1533	2163	(Based on non-truncated tag) >gi|142358075|ref|NM_152350.2| Homo sapiens chromosome 17 open reading frame 45 (C17orf45), mRNA
	CATGATCGCTTTCTACACTGTATTACA	1442	1496	(Based on non-truncated tag) >gi|41406053|ref|NM_000484.2| Homo sapiens amyloid beta (A4) precursor protein (APP), transcript variant 1, mRNA::>gi|41406054|ref|NM_201413.1| Homo sapiens amyloid beta

Resetting and Exiting the Program

Reset

Clicking on **Reset** will reset all the options in the program screen, including the project name selection. You will be prompted to complete this action.

Exiting the Program

Click on **Exit** to exit the program. Depending on the state the program is in, there may be a small or lengthy delay before exiting.

You can also end the program by using standard Unix commands (*e.g.*, CTRL-C) from the terminal where you launch the program.

Appendix

Frequently Asked Questions

Is the Program Frozen?

The program will sometimes appear frozen during processing. This often happens when you first load the SOLiD™ reads or the reference sequences, or when you attempt to switch application windows while the SOLiD™ SAGE™ program is actively running. Allow the process to continue for a few minutes before force-quitting.

Why Am I Getting An Error Message?

The program includes various error messages, most of which are self-explanatory. If the error says something about "balloon" or "cancellation", you can ignore it. These errors will not affect your results.

How Long Does Mapping Take?

The running time of the mapping analysis depends on a few factors:

- The number of reads in the input SOLiD™ SAGE™ file
- The size of the reference genome
- Input tag length
- Number of allowed mismatches (more mismatches allowed = longer processing time)
- Number of million reads per round of processing (more rounds required = longer processing time)

A typical read file with octet SOLiD™ data mapped to the human Refseq mRNA dataset with a tag length = 27 and mismatch = 2 takes ~10 minutes on a computer with a 2.33 GHz Duo CPU and 8 GB of memory (note that only one CPU and ~2 GB memory are actually used).

If you are mapping to the human genome with mismatch = 0, it may take 2–3 hours and up to 15 hours with mismatch = 2.

Examples of Files and Formats

Input File Formats

SOLiD™ reads file:

This is a standard .csfasta file, which looks like:

Tue Dec 23 01:04:30 2008 comments follow

Cwd: /home/pipeline

Title: Solid0110_20081218_JMK26_Ribominus_JMK26_C_amp_1_

>443_12_55_F3

T2112001212101120200203200000000

>443_12_118_F3

T0000000000000000000000000000000

>443_12_170_F3

T2130011300121020330330321313 01

>443_12_201_F3

T0030000323000000000000000000000

>443_12_278_F3

T0000000000000000000000000000000

>443_12_294_F3

T0000000000000000000000000000000

>443_12_336_F3

T0000000000000000000000000000000

Reference files:

This is a standard NCBI fasta file, part of a database file set in a single directory.

Files look like:

>gi|155369268|ref|NM_001100917.1| Homo sapiens tetraspanin 19 (TSPAN19), mRNA

AAACAATCTCGATTCTAAATTG… … (bases follow) … … ACTGGTG

>gi|169212695|ref|XM_001716884.1| PREDICTED: Homo sapiens hypothetical protein LOC100132679 (LOC100132679), mRNA

ATGTGTGTATATATATATACACATATATATG… … (bases follow) … … ATGGATGTAT

Comparison input file:

Same as standard output file (see next page), with a file name of the format solidsageread.csfasta.taglength.mismatch.output.tab (sample file: solidsageSampleRead.csfasta.27.0.output.tab)

Output File Names

Output file: File name format: solidsageread.csfasta.taglength.mismatch.output.tab (sample file: solidsageSampleRead.csfasta.27.0.output.tab)

Results file: File name format: solidsageread.csfasta.taglength.mismatch.results.tab (sample file: solidsageSampleRead.csfasta.27.0.results.tab)

Comparison file: File name: comparison.tab

Mapping Output File Format

Tag Count GI Description

CATGAAAAAACTCCAAATAAGAGAATC 1 GI33413399 >gi|33413399|ref|NM_001984.1| Homo sapiens esterase D/formylglutathione hydrolase (ESD), mRNA

CATGAAAGGGTCACTTCTGTAATAGTG 1 GI81158221::GI81158223::GI81158225 >gi|81158221|ref|NM_001037133.1| Homo sapiens neuronal cell adhesion molecule (NRCAM), transcript variant 3, mRNA::>gi|81158223|ref|NM_005010.3| Homo sapiens neuronal cell adhesion molecule (NRCAM), transcript variant 2, mRNA::>gi|81158225|ref|NM_001037132.1| Homo sapiens neuronal cell adhesion molecule (NRCAM), transcript variant 1, mRNA

CATGAACAACCGGCTGGCCGAGACCAG 1 GI11545760 >gi|11545760|ref|NM_022055.1| Homo sapiens potassium channel, subfamily K, member 12 (KCNK12), mRNA

CATGAACTTGATACGTCCGTGTGTCCC 1 GI53759150 >gi|53759150|ref|NM_005063.4| Homo sapiens stearoyl-CoA desaturase (delta-9-desaturase) (SCD), mRNA

CATGAAGATGATATGAGGCCGGGGCGG 1 GI52145308 >gi|52145308|ref|NM_032808.5| Homo sapiens leucine rich repeat and Ig domain containing 1 (LINGO1), mRNA

CATGAAGGAAGATCCCACAGTCTCAGC 1 GI4758483 >gi|4758483|ref|NM_004832.1| Homo sapiens glutathione S-transferase omega 1 (GSTO1), mRNA

CATGACAGCCCTCTGCTCTTGAGTACC 1 GI39812204 >gi|39812204|ref|NM_025164.3| Homo sapiens KIAA0999 protein (KIAA0999), mRNA

CATGACGGAACAATAGGACTCCCCAGG 2 GI38505192 >gi|38505192|ref|NM_000954.5| Homo sapiens prostaglandin D2 synthase 21kDa (brain) (PTGDS), mRNA

CATGACGTGTCTATGTCAAAAGTTCTT 1 GI115583669 >gi|115583669|ref|NM_003253.2| Homo sapiens T-cell lymphoma invasion and metastasis 1 (TIAM1), mRNA

Mapping Results File Format

Tag_Seq GI_num GI_Pos Read_ID Mismatch

CATGTGCAAATAAATGTGGCTTAGACT 133908618 3298 >665_1155_42_F3 0

CATGGTAATAAAATATGAATGATAAAA 189083841 2125 >665_817_382_F3 0

CATGGTAATAAAATATGAATGATAAAA 189083835 2280 >665_817_382_F3 0

CATGGTAATAAAATATGAATGATAAAA 189083837 2146 >665_817_382_F3 0

CATGGTAATAAAATATGAATGATAAAA 189083839 2128 >665_817_382_F3 0

CATGGTAATAAAATATGAATGATAAAA 51593094 2283 >665_817_382_F3 0

CATGGTAATAAAATATGAATGATAAAA 132814488 2268 >665_817_382_F3 0

CATGTGATGGGCATTGAGCCACACCTC 34147410 1140 >665_1807_1132_F3 0

CATGAGGAGCTCGGCTTAAAATGTCTT 40254847 2324 >665_1994_720_F3 0

Comparison File Format

Tag_Sequence File1 File2

CATGAAGACAGTGGCTGGCGGTGCCTG 4715 5430 (Based on non-truncated tag) >gi|78214519|ref|NM_000998.4| Homo sapiens ribosomal protein L37a (RPL37A), mRNA

CATGCACAAACGGTAGTTTTGTGTGTT 2361 2604 (Based on non-truncated tag) >gi|169202035|ref|XM_001726056.1| PREDICTED: Homo sapiens similar to hCG2027326 (LOC100129905), mRNA::>gi|169203086|ref|XM_001725509.1| PREDICTED: Homo sapiens similar to hCG2027326 (LOC100129905), mRNA::>gi|169203606|ref|XM_001726007.1| PREDICTED: Homo sapiens similar to hCG2027326 (LOC100129905), mRNA::>gi|68160923|ref|NM_001030.3| Homo sapiens ribosomal protein S27 (metallopanstimulin 1) (RPS27), mRNA

CATGACAACAAAGAAAAAGACCTTGTA 1736 1606 (Based on non-truncated tag) >gi|56550064|ref|NM_001008220.1| Homo sapiens complexin 2 (CPLX2), transcript variant 2, mRNA::>gi|56550103|ref|NM_006650.3| Homo sapiens complexin 2 (CPLX2), transcript variant 1, mRNA

CATGAATATGTGGGCTAAGAAATAGTT 1659 1980 (Based on non-truncated tag) >gi|17999531|ref|NM_004374.2| Homo sapiens cytochrome c oxidase subunit VIc (COX6C), mRNA

Repeat Reads File Format

Total reads for SOLiD™SAGE™SampleRead.csfasta: 49999
Total unique reads for SOLiD™SAGE™SampleRead.csfasta: 48630
Top 100 abundant reads for SOLiD™SAGE™SampleRead.csfasta:
T1111111111111111111111111111111..1 148
T1111111110111111111111111111111..1 135
T1111011111011111111111111111111..1 127
T3111111111011111111111111111111..1 70
T1111011111111111111111111111111..1 46
T0000000000000000000000000000010..0 39
… …
Total unique reads for SOLiD™SAGE™SampleRead.csfasta at tag 26: 48102
Top 100 abundant reads for SOLiD™SAGE™SampleRead.csfasta at tag 26:
T11110111110111111111111111 172
T11111111110111111111111111 157
T11111111111111111111111111 156
T31111111110111111111111111 90
T00000000000000000000000000 57
T31111110110111111111111111 55
T11110111111111111111111111 55
… …
Total unique reads for SOLiD™SAGE™SampleRead.csfasta at tag 27: 48363
Top 100 abundant reads for SOLiD™SAGE™SampleRead.csfasta at tag 27:
T111111111111111111111111111 150
T111111111101111111111111111 140
T111101111101111111111111111 139
T311111111101111111111111111 73
T000000000000000000000000000 57
T111101111111111111111111111 50
T111101110101111111111111111 37
T311111101101111111111111111 36
… …
Total unique reads for SOLiD™SAGE™SampleRead.csfasta at tag 28: 48412
Top 100 abundant reads for SOLiD™SAGE™SampleRead.csfasta at tag 28:
T111111111111111111111111111 150
T111111111101111111111111111 140
T111101111101111111111111111 138
T311111111101111111111111111 73
T000000000000000000000000000 57
T111101111111111111111111111 49
T311111101101111111111111111 36

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