



SMRT Link Server Installation

- SMRT Link server software is supported on English-language CentOS 6.x; 7.x and Ubuntu 14.04; 16.04 64-bit Linux® distributions.
- SMRT Link is **not** guaranteed to work on Linux versions that are no longer supported by the Operating Systems' vendors.
- SMRT Link server software **cannot** be installed on macOS® or Windows® systems.
- Several new SMRT Link v5.1.0 features are computationally-intensive and require adherence to the computational and storage requirements listed in the document **SMRT Link Software Installation (v5.1.0)**.
- To install **only** SMRT Tools, use the `--smrttools-only` option with the installation command, whether for a new installation or an upgrade.

Examples:

```
smrtlink-*.run --rootdir smrtlink --smrttools-only  
smrtlink-*.run --rootdir smrtlink --smrttools-only --upgrade
```

Included with the SMRT Link Installation

Barcode Sequences:

- RSII_384_barcodes
- RSII_96_barcodes
- Sequel_RSII_16_barcodes_v1
- Sequel_RSII_96_barcodes_v1
- Sequel_RSII_384_barcodes_v1

Reference Sequences:

- Sequel DNA Internal Control
- HIV_HXB2
- lambdaNEB

New Features

SMRT Link

- A streamlined end-to-end barcoding workflow for automated analysis of multiplexed samples.
 - Enables users to define barcodes and their associated sample names for an experiment at Run Design.
 - Utilizes a new and improved demultiplexing algorithm.
 - Provides extensive QC metrics for evaluation of barcoding performance.
 - The SMRT Link graphical user interface supports up to 384 barcodes per sample. Support for more than 384 samples is available using the command-line.

SMRT Link - SMRT Analysis

- **Structural Variant Calling:** Users can now perform multi-sample structural variant-calling for a group of up to 10 samples using the graphical user interface, and up to 1000 samples using the command-line. This feature improves sensitivity of detection at low coverage.
- **Iso-Seq 2 [Beta]:** Enhanced Iso-Seq[®] analysis application with improved performance and time-to-result for whole transcriptome (high diversity) samples.
- **Assembly (HGAP 4):** Enables users to generate output files that can be run through Falcon Unzip using the command-line.
- **Minor Variants Analysis:** Support for custom target configurations.
- **Resequencing and CCS with Mapping:** Addition of an advanced parameter to generate a consolidated BAM file; the file is then available from the File Downloads page.
- New options for analysis of multiple Data Sets:
 - One analysis on all selected Data Sets merged together.
 - One analysis per Data Set with identical parameters.
 - One analysis per Data Set with custom parameters.
- Enables users to export SMRT Link analysis results and import into a different SMRT Link instance.
- Enables users to copy an analysis and run it again using different parameters.
- Completion stage of in-progress analyses is displayed.
- Enables SMRT Link to send e-mail to users when an analysis finishes.

SMRT Link - Data Management

- Enables users to delete references and barcode sets using the graphical user interface.
- Enables users to edit attributes of existing Data Sets.

SMRT Link - Sample Setup

- Added support for Iso-Seq experiments.
- Added support for the SMRTbell[®] Express Template Prep Kit.
- Added support for the Sequel[®] Binding Kit 2.1.
- Added support for AMPure[®] clean up.
- Added pre-extension guidance.
- Improved support for low-concentration samples.
- Updated protocols supporting all new reagents.
- Numerous usability improvements.
- Removed display of on-plate loading concentration guidance. Loading recommendations are now provided using the Quick Reference Card – Diffusion Loading and Movie Time Recommendations for the Sequel[®] System document.

SMRT Link - Run Design

- Enables users to specify barcoded samples.

SMRT Link - Run QC

- Displays the Chemistry Bundle version.

Fixed Issues

- Improved SMRT Link functionality for small browser windows.
- SMRT Link Chemistry Bundle Update: After applying an update, Run Design part numbers and version information are now visible after a page reload (CTRL-R).
- Sample Setup: When opening multiple saved samples, they are now ordered in the same order as in the summary table.
- Run QC: Productivity metrics from SMRT Link v4.0.0 now display correctly.
- Run QC: The Control Concordance Mode value is no longer missing.
- Run QC: Improved display of empty results for Internal Control when no control reads are found.
- Run QC: The X-axis of the Loading Evaluation plot is now correctly labeled.
- Run QC: In CSV file export, Polymerase Read Length and Longest Subread Length are now correctly displayed and are no longer swapped.
- Data Management: Removing the only project owner is no longer allowed.
- Data Management: Clicking **Export Data Sets**, then selecting a Project in the top right of the page no longer displays a "Bad request" error message.
- SMRT Analysis: In the results page **Data > File Downloads** section, the Analysis Log file size is now correct.
- SMRT Analysis: Import of reference or barcode files now checks for invalid entries - blank lines and carriage returns in FASTA files.
- SMRT Analysis: Importing a genome reference FASTA file with a period in the name does not fail.
- SMRT Analysis: Links included in the `index.xml` files (part of analysis output) are now correct. Additional information is included in the SMRT Tools Reference Guide.
- SMRT Analysis: The "Pipeline Id" field is renamed "Analysis Application" and is searchable on the SMRT Analysis home page.
- SMRT Analysis: Reduced memory consumption for `bam2fastx` and resequencing applications.
- SMRT Analysis: Compressed (GZIP) consensus FASTQ files from LAA output can now be unzipped into separate files.

Known Issues

- In Run Design, clicking **Edit** on a saved barcoded Run Design with a CSV-format Barcoded Sample Name File does not display the previously-saved CSV file, although it is still saved.

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- In the Run Design CSV-format Barcoded Sample Name File, if users omit a comma to separate fields, the Bio Sample Names will be incorrectly assigned.
 - In Run Design, setting “Sample Is Barcoded” to “Yes” **without** specifying a barcode set can cause the run to fail, causing loss of the SMRT Cell. Ensure that you specify a barcode set when setting “Sample Is Barcoded” to “Yes”.
 - Some of the Run Design tooltip help messages are displayed at the bottom of the page instead of next to the fields they describe.
 - In Data Management, copying a demultiplexed child Data Set causes it to lose its association with the parent Data Set.
 - In SMRT Analysis, copying a `Demultiplex Barcodes (Auto)` analysis displays an error message. None of the other analysis applications, including `Demultiplex Barcodes`, have this issue.
 - In SMRT Analysis, copying an analysis launched using multiple Data Sets with the option “One Analysis per Data Set – Identical Parameters” displays an error message.
 - In SMRT Analysis, copying an analysis fails and displays a red error screen if any advanced parameters in the copied analysis were set to non-default values. The workaround is to create a new analysis, then manually reenter the parameter values used in the original analysis. (The original parameter values are located on the **Analysis Results** page, in the **Analysis Overview > Status > Analysis Parameters** list.)
 - In SMRT Analysis, clicking **Export Selected Analysis** after selecting an analysis, then selecting an export location and clicking **Export**, results in the dialog box disappearing and a transparent gray overlay covering the entire page. Refreshing the page removes the overlay.
 - In Sample Setup, when sample concentration is very low and a “low sample concentration” warning displays, the calculation in the Sample Information Table for the final sample concentration may be incorrect. This does not affect the protocol, and if you choose to continue you may follow the protocol.

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