## Revio™ system Instrument Control Software PacBie release notes (v13.0)

Note: This Instrument Control Software release applies only to Revio systems.

## **New features**

- Add analysis support for the Kinnex<sup>™</sup> library type to enable the new Kinnex kits: Kinnex single-cell RNA kit,
   Kinnex 16S rRNA kit, and Kinnex full-length RNA kit.
- Add scheduler support for 12-hour and 30-hour movies.
- Add support for **run previews** in SMRT<sup>®</sup> Link to provide early information on loading, library fragment size, and representation of barcodes in the pool.
- Add sequencing support for libraries shorter than 3 kb.
- Add support for Revio SMRT® Cell tray 1 SMRT Cell and Revio sequencing plate 1 reaction consumables.
- Enable adaptive loading to prevent overloading SMRT Cells. The Adaptive Loading feature is available at the run level and applies to **all** SMRT Cells from the run.
- Add analysis support for the Adeno-associated virus (AAV) library type to provide adapter calling and consensus read calling appropriate for ssAAV and scAAV libraries.
- Add analysis support for runs that do not perform on-instrument demultiplexing.
- Add analysis support for two consensus modes: Molecule (this was the prior default; it generates one
  consensus read per ZMW) and Strand, which generates separate consensus reads for forward and reverse
  strands.
- Trim the standard adapter stem from all reads prior to BAM file generation. This **deprecates** the concept of the "default" adapter stem and aligns Revio with Sequel<sup>®</sup> II system behavior.
- Update 5mC methylation-calling to include training for Revio system chemistry and use a new model implemented in the jasmine software, which replaces the primrose software in previous releases. Inputs, outputs, and model architecture are unchanged from the previous release. (It is not necessary to reprocess existing datasets already called with primrose.)
- Update basecaller and consensus read calling models to improve accuracy. Inputs, outputs, and model
  architecture are unchanged from the previous release. Note: It is appropriate to combine v13.0 datasets with
  existing datasets.
- Add analysis support for full-resolution (non-binned) base quality values. The full-resolution base quality values are accessible using the Full Resolution Base Qual option when creating new run designs.
- Add analysis support for "rich HiFi" tags that provide a summary of subread-to-consensus read alignment. The rich HiFi tags are accessible using the **Subread to HiFi Pileup** option when creating new run designs.
- Add a feature to the instrument screen to clear disk partitions to restore an instrument if data is left behind
  from a failed run. We recommend that you contact PacBio Technical Support to review data before clearing as
  deleting cannot be undone.
- More efficiently compress output BAM files by sorting records to reduce file sizes approximately 10%.
- Include the run identifier, SMRT Cell part number, SMRT Cell identifier, ICS version, and movie length in the read group header in output BAM files.
- Add a fail flag (ff) bit mask tag to indicate the reasons that reads are included in the fail reads.bam file.
- Include HiFi reads that match the Revio sequencing control in the fail\_reads.bam file tagged with the 0x2 fail flag bit. Previously, sequencing control reads were **not** included in output BAM files.
- Include ZMWs that have at least **one full subread** but do **not** produce a Q10+ consensus read in the output fail\_reads.bam file. A single representative subread for each ZMW is output and tagged with the 0x8 fail flag bit. Kinetics tags are **not** output for these subreads.
- Update the firmware on the Revio system for robustness.
- Enable the Revio workdeck camera for remote troubleshooting with PacBio Technical Support.

- Add **Shut down** and **Reset** options to the Instrument Control Software.
- Collect additional instrument and consumable performance metrics and logs to increase the efficiency of
  instrument support. New metrics cover SMRT Cell loading and compute utilization (GPU, CPU, RAM). New log
  files cover primary and post-primary analysis.
- Add software support for different hardware configurations in analysis servers.

## **Fixed issues**

- Modified the behavior of the instrument's Door button so that the button is not lit during startup diagnostics.
- Fixed an issue that sometimes caused the touchscreen to be resized using multitouch gestures.
- · Fixed an issue that sometimes caused user-initiated self-tests to fail.
- Fixed an issue that sometimes caused warning messages to persist to a subsequent run.
- Fixed an issue that sometimes caused the incorrect SMRT Cell identifier (e.g. EA010390) to be displayed in SMRT Link. This fix applies only to **future** runs.
- Improved the reliability of communications between Revio systems and a connected SMRT Link server over DHCP or static IP network connections. SMRT Link will **automatically** update Revio system IP addresses when they are changed using the instrument network connection settings.
- Added a timeout to the file transfer test performed when loading and launching a run.
- Fixed an issue where run files were sometimes deleted when post-primary analysis failed. Files are now
  retained to allow recovery. Contact PacBio Technical Support when sequencing completes, but post-primary
  analysis fails.
- Individual sequencing stage failures no longer prevent sequencing runs from starting, as long as at least one sequencing stage is available.
- Fixed an issue that sometimes prevented users from pre-loading runs in certain conditions.
- Fixed an issue with compute resource utilization in the primary analysis basecaller that contributed to some collections ending early.

## **Known issues**

• At times the touchscreen may freeze and be unresponsive during initialization. The workaround is to hardreset the instrument to restart initialization by **either** pressing the power button, **or** by unplugging the instrument. Contact PacBio Technical Support if the issue persists.

**Note**: Contact your FSE (Field Service Engineer) or PacBio Technical Support to perform the Instrument Control Software installation.

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